Journal of Cell and Molecular Research (2024) 15 (2), 89-99 DOI: 10.22067/jcmr.2024.85650.1082

Review/ Mini-Review

Major Genes Involved in Mitophagy of *Saccharomyces cerevisiae*

Hengameh Hoshyar-Chamanarae, Madjid Momeni Moghaddam *

Department of Biology, Faculty of Sciences, Hakim Sabzevari University, Sabzevar, Iran

Received 1 Dec 2023 Accepted 9 Feb 2024

Abstract

Mitophagy occurs exclusively in the mitochondrial organ, itself considered one of the types of autophagy, and plays a very specific role in cellular functions and controlling tissue expansion. So, knowing this process as much as possible can help us understand many of the cell processes, especially the cell aging process, and the pathways that cause physiological diseases. In the process of mitophagy in the yeast *Saccharomyces cerevisiae*, three genes are directly involved, namely ATG 11, ATG 32, and ATG 8. This process has been researched for many years, but winning the 2016 Nobel Prize in Physiology for his discoveries of mechanisms for Autophagy by Yoshinori Ohsumi caused the world's attention to this cellular mechanism. In recent years, the Saccharomyces cell model has received a lot of attention in understanding the process of cell aging and chronic diseases such as type 2 diabetes, Parkinson's, Alzheimer's, and many types of cancer, and this article reviews the importance of the above genes and specifically examines the pathway in cervical Saccharomyces. The specific Autophagy of each organelle can help cure painful and chronic diseases such as type 2 diabetes, Parkinson's, Alzheimer's, and many types of cancer. They hope that by finding the mechanisms, Autophagy can make it more active or keep it active until the end of life, and in this way, it can cure these diseases or at least help cure a lot. This review article attempts to introduce and overview the role of key genes in the process.

***Keywords*:** mitophagy, *Saccharomyces cerevisiae*, ATG 8, ATG11, ATG 32

Introduction

Autophagy is a Greek word meaning self-devouring. This mechanism is a cellular mechanism that, in 1963, was named by Christian de Duve. It is about recycling damaged or extra protein and organs in cells (Ravikumar et al., 2010). This mechanism exists in all eukaryotes, whether they are unicellular or multicellular (King, 2012).

Based on research, four kinds of autophagy have been found: macroautophagy, microautophagy, chaperone-mediated autophagy, and crinophagy (Csizmadia & Juhász, 2020). usually, when we speak about autophagy, we mean macroautophagy which is the most consummately researched form of autophagy. At the start of the macroautophagy mechanism, a double-layered membrane originates from the endoplasmic reticulum. This membrane wraps around the content to be removed, which, depending on what content or organelle it includes, can be examined more specifically after the vesicle containing Digestible materials is formed. This vesicle, by removing its primary membrane from the lysosome, enters its contents into the lysosome, after which hydrolase enzymes break down the materials and return them to the cell (Eskelinen, 2008). Notably, yeasts and plants have vacuoles instead of lysosomes (Feng et al., 2014)

Mitophagy

Mitochondria-specific macroautophagy is called mitophagy, which was named in 1998. The mechanism of mitophagy occurs for excess or damaged mitochondria in the cell and is essential for cell survival This process is selective (Scott & Klionsky, 1998). The generation of ATP in the cell by the mechanisms of oxidative phosphorylation leads to the production of reactive oxygen species besides the process of aerobic ATP synthesis. This mechanism is being carried out naturally in all cells. Moreover, its accumulation causes damage to the mitochondria, one of the effects of which can be the reduction of ATP production and the release of cytochrome c, which ultimately causes the release of caspases and the initiation of cell apoptosis. The

* Corresponding author's e-mail address:
iranbioman@yahoo.com, m.momeni@hsu.ac.ir

release of cytochrome c in the cytosol can even cause the death of mammals (Scarlett & Murphy, 1997; Wang & Youle, 2009). It should be kept in mind that over time, mitochondrial damage occurs in the cells, and the damage must be removed so as not to cause cell death and disease in multicellular organisms. (Huang & Klionsky, 2021). Since mitophagy is a sub-branch of macroautophagy, its mechanism of action is similar to macroautophagy, which we explain here in more detail. The specialty surrounds the mitochondrion and covers the entire mitochondrion, which can be seen by solid electron microscopes. After the outer membrane of this mitophagosome, it combines with the lysosome and becomes a vesicle with a membrane. This covers the mitochondria and enters the lysosome (vacuole in yeast), where the digestive enzymes clean up the excess or damaged mitochondria, or those that have problems with the mitochondrial membrane, and solve the cell problem at a lower cost than cell death (Figure 1. Mitophagy is a subset of macroautophagy, with the difference that it specifically involves mitochondria.). On the other hand, it causes a certain amount of transformed materials to be available to the cell for reuse. (Delorme-Axford et al., 2015). According to research, damaged mitochondria that are not organized properly can be related to cancer, Parkinson's, diabetes, and Alzheimer's diseases. (Liesa et al., 2009; Vyas et al., 2016; Wang et al., 2009)

What factors cause what changes in the mitochondria that cause it to enter the mitophagy cycle is being investigated and researched.

Saccharomyces cerevisiae

Saccharomyces cerevisiae is a single-celled yeast from the eukaryote family, which is in the kingdom of fungi and the division of Ascomycota. The yeast is also known as brewer's yeast or baker's yeast. In nature, this yeast exists naturally in the skin of grapes. This yeast is about 5 to 10 micrometers in diameter (Feldmann, 2010). *Saccharomyces cerevisiae* can exist in both haploid and diploid forms, and in both ways, they can continue the path of vegetative reproduction by budding. This yeast has both sexual and asexual reproduction. In the sexual model, which is rarer, two haploid cells, α and, β enter the path of meiosis together. This reproduction can be vital because it produces genetic diversity (Kruckeber & Dickinson, 2004). Access to the complete genome of *Saccharomyces cerevisiae* was done as the first eukaryote in 1996, and its rapid and low-cost growth and reproduction with minimal problems and ease of genetic manipulation caused a lot of cellular and molecular research on this yeast and made it a eukaryotic model (Engel et al., 2014; King, 2012). Depending on its species, this yeast can have a lifespan of 6 to 15 days, and in this sense, it has become essential for aging studies (Parrella & Longo, 2008).

Budding yeasts in glucose-rich conditions mainly use the glycolysis pathway to provide their required enzymes, even in aerobic conditions (Huang & Klionsky, 2021). One way to induce mitophase in yeast, which has been extensively researched, is to grow the yeast in a medium that has non-fermentable carbon resources and then transfer it to a glucose-rich medium where it grows and in the Finally, in an environment that does not have nitrogen to use, and the yeast is subjected to nitrogen starvation conditions, this leads to the fact that the amino acid sources have been depleted so that the yeast will remove its excess and damaged mitochondria (Kanki, Wang, Cao, et al., 2009).

Autophagy-related genes (Atg)

"ATG" and "Atg" stand for "autophagy-related" genes or proteins, respectively. So," Atg "conveys the concept related to autophagy(Klionsky, 2012). In the 1990s, molecular research on autophagy was carried out, and at that time, 15 autophagy-related genes were discovered in *Saccharomyces cerevisiae*; after that, this research continued until today, when 41(**Error! Reference source not found.**) autophagy-related genes were discovered in the yeast *Saccharomyces cerevisiae*. Fortunately, about half of these genes are homologous in higher eukaryotes (Torggler et al., 2017). The existing genes are directly or indirectly involved in the different stages of autophagy, from the initial formation of the double-layer membrane to the fusion of the autophagosome and lysosome membranes. ATG genes can be divided into core-autophagy genes and non-core autophagy genes. Core autophagy genes are essential and functional for autophagy, while non-core autophagy genes are involved in specific aspects of autophagy. The proteins that play a role in autophagosome formation are called "core ATG proteins" (Faruk et al., 2021).

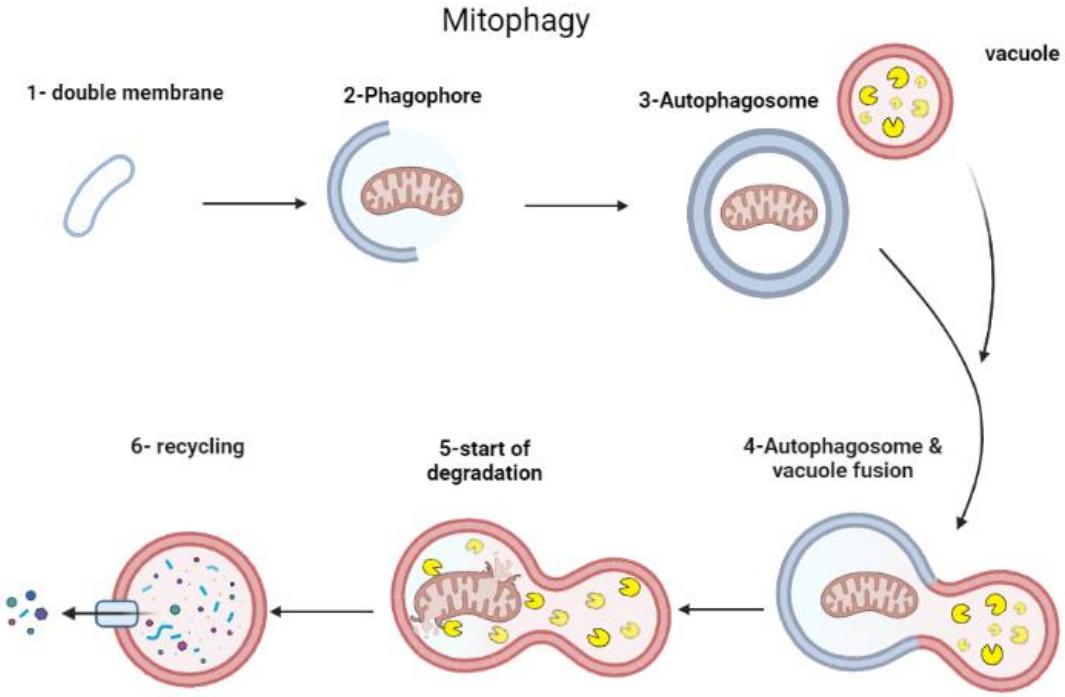

Figure 1. Mitophagy is a subset of macroautophagy, with the difference that it specifically involves mitochondria. This figure was prepared by the authors of this article and using the biorender website.

Table 1. 41 genes related to autophagy

Name of ATG	FUNCTION	RESOURCE
ATG 1	**serine/threonine kinase -It is vital for the initial building of the autophagosome and mitophagy**	**(Straub et al., 1997)**
ATG 2	**autophagosome formation**	**(Osawa et al., 2019)**
ATG 3	**Atg8 conjugation system, contributing to phagophore elongation**	**(Fang et al., 2021)**
ATG 4	**Atg8 conjugation system**	**(Maruyama & Noda, 2018)**
ATG 5	**Forms a complex with ATG12 and ATG16, facilitating ATG8 conjugation.**	**(Noda et al., 2013)**
ATG 6	**Necessary for autophagy, endocytosis, and protein secretion**	**(Shravage et al., 2013)**
ATG 7	**like Atg 4 has a connection with Atg12 and Atg8 for conjugation**	**(Martens & Fracchiolla, 2020)**
ATG 8	**intercede membrane fusion events leading to autophagosome biogenesis**	**(Shpilka et al., 2011b)**

ATG 9	**participate in the formation of the phagophore, the precursor cistern of autophagosomes**	**(Ungermann & Reggiori, 2018)**
ATG 10	**mediates the formation of the Atg12p-Atg5p conjugate**	**(Meijer et al., 2007)**
ATG 11	**protein scaffolds**	**(Zientara-Rytter & Subramani, 2020)**
ATG 12	**autophagosome formation**	**(Popelka et al., 2021)**
ATG 13	**regulation of Atg1 catalytic activity and consequently in autophagy activation**	**(Miller-Fleming et al., 2014)**
ATG 14	**Autophagy-specific subunit of complex I/required for localizing additional ATG proteins to the PAS**	**(Sun et al., 2008)**
ATG 15	**degrade autophagic bodies by its lipase activity**	**(Hirata et al., 2021)**
ATG 16	**ATG16 is needed for the correct locating of the ATG12~5 conjugate with the pre-autophagosomal structure**	**(Hirata et al., 2021)**
ATG 17	**protein scaffolds**	**(Zientara-Rytter & Subramani, 2020)**
ATG 18	**elongation of phagophores / the recycling of Atg9**	**(Lei et al., 2021)**
ATG 19	**Atg19 mediates the transport of Ape1 to the vacuole/cvt pathway**	**(Pfaffenwimmer et al., 2014)**
ATG 20	**Sorting nexin family member/cvt pathway/Phox homology**	**(Popelka et al., 2017)**
ATG 21	**cvt pathway/micronucleophagy /mitophagy**	**(Yamamoto et al., 2023)**
ATG 22	**A vacuolar amino acid transporter involved in autophagic degradation**	**(Yang et al., 2006)**
ATG 23	**Peripheral membrane protein needed for autophagy and CVT pathway**	**(Leary et al., 2022)**
ATG 24	**pre-autophagosomal structure/ phagophore assembly site (PAS)**	**(Wang et al., 2009)**
ATG 25	**Macropexophagy/ (Cvt) pathway, mitophagy, and pexophagy.**	**(Monastryska et al., 2005)**

ATG 26	**(Cvt) pathway, mitophagy, and pexophagy**	**(Lynch-Day & Klionsky, 2010)**
ATG 27	**regulate cvt pathway /needed for autophagy-dependent cycling of ATG9**	**(Yen et al., 2007)**
ATG 28	**autophagic degradation/pexophagy**	**(Stasyk et al., 2006)**
ATG 29	**preautophagosomal structure (PAS)**	**(Barve & Manjithaya, 2021)**
ATG 30	**pexophagy/ micropexophagy/ macropexophagy**	**(Farre et al., 2008)**
ATG 31	**Hase a part in starvation-induced autophagy. / mitophagy/ the pre autophagosomal structure (PAS)**	**(Kanki, Wang, Baba, et al., 2009)**
ATG 32	**special receptor for mitophagy**	**(Ko & Tan, 2023)**
ATG 33	**mitophagy via autophagy during starvation and at the post-log phase**	**(Kanki, Wang, Baba, et al., 2009)**
ATG 34	**Cargo-receptor protein involved (Cvt pathway) and in autophagy. Recognizes cargo proteins, and delivers them to the pre-autophagosomal structure**	**(Watanabe et al., 2010)**
ATG 35	**micropexophagy-specific protein**	**(Nazarko et al., 2011)**
ATG 36	**pexophagy**	**(Motley et al., 2012)**
ATG 37	**Cvt pathway**	**(Motley et al., 2012)**
ATG 38	**is required for autophagy-specific phosphatidylinositol 3-kinase complex integrity**	**(Araki et al., 2013)**
ATG 39	**nucleophagy receptor**	**(Mochida et al., 2022)**
ATG 40	**reticulophagy**	**(Liu et al., 2022)**
ATG 41	**regulates the rate of autophagosome formation/**	**(Yao et al., 2015)**

ATG 8

ATG 8 gene is a nuclear gene in *Saccharomyces cerevisiae* that makes the Atg 8 protein, a ubiquitin-like protein that acts as an autophagosome marker in yeasts (Galluzzi et al., 2017). Atg8 is known as a monomer which has 117 amino acids and a molecular weight of 13,6kDa. It also consists of a 5-stranded β-sheet that is surrounded by two α-helices at one side and one α-helix at the other side (Geng & Klionsky, 2008). Atg8 proteins are one of the 62 super-protected eukaryote-specific protein families

(Wood et al., 2002). Interestingly, yeast and other fungal species have a single Atg8 gene, while multicellular animals, green plants, and even some protists have several. According to these, genes were made many years ago, so Atg8 genes passed a long way until now; they have been duplicated and even lost through evolution, leading to the extinction and enlargement of some subfamilies in the special phylum. There are three subfamilies of ATG 8 genes. There are three subfamilies of ATG 8 genes, and in the human genome, there are samples without introns from all three subfamilies, which have been deactivated due to repeated mutations over time. (Shpilka et al., 2011a).

During this process of autophagy, Atg8 plays an important role, especially for autophagosome maturation (lipidation) (Eskelinen, 2008). Atg8 is localized in the cytoplasm and at the PAS under nutrient-rich situations; when autophagy is induced, this protein will be associated with the membrane of autophagosome. It is then located at the site of autophagosome nucleation, the phagophore-assembly site (PAS) (Geng & Klionsky, 2008). Therefore, the Atg8 proteins can be seen both in the cytoplasm and attached to the membrane. Being attached to the membrane is obtained as a result of the Atg8 protein binding with phosphatidylethanolamine, which is one of the lipid components of the membrane (Kirisako et al., 2000). ATG 8 functions with the help of ATG 4. This gene is particularly essential in nucleophagy, mitophagy, and autophagy related to the endoplasmic reticulum. This gene is important in terms of involvement in conjugation and deconjugation in the autophagy process. (Kirisako et al., 1999)

ATG 11

ATG 11 is responsible for producing Atg 11, which is a sizeable cytosolic protein. This protein has important roles in the cell, such as involvement in the Cvt pathway and plays a role in various types of autophagy, such as mitophagy, pexophagy, and nucleophagy, and is like a protein scaffold that causes the absorption of ATG proteins to the PAS (He et al., 2006; Kanki, Wang, Cao, et al., 2009).

ATG 32

ATG 32 gene is a specialized gene for mitophagy in yeast by producing Atg32(Autophagy-related protein 32), and it has been shown that this mechanism is simpler in yeast than in mammals. Atg32 protein is an external protein of the mitochondrial membrane. (Camougrand et al., 2020). In *Saccharomyces cerevisiae*, Atg32 has 529 amino acids single-transmembrane protein. when the mitophagy-inducing situation starts, Atg32 creates complexes with Atg11, and these parts are named mitophagy initiation sites (Margolis et al., 2020). Atg32 can Atg11 and Atg8 through its N terminus; however, it is necessary to know that only the associated with Atg11 is needed for autophagy to be inducted (Huang & Klionsky, 2021). Nevertheless, how is this happening? Absorbing, Atg32 is phosphorylated at serines 114 and 119 when the condition for inducing mitophagy is available, and serine 114 phosphorylation causes the striking growth of the binding affinity between Atg32 and Atg 11 (Aoki et al., 2011). Casein kinase 2 (CK2) is responsible for serine 114 phosphorylation in other ways; the cleavage of the Atg32 C terminus ends with rising binding affinity in the middle of Atg32 and Atg11; in this situation, promoting mitophagy induction will happen (Wang et al., 2013).

There are other methods besides phosphorylation to activate this mechanism, such as post-translational modifications (Camougrand et al., 2020; Wang et al., 2013). Atg 32 protein exists on the outer membrane of mitochondria as a single-pass membrane protein and the vacuole membrane as a single-pass membrane protein, as well as the pre-autophagosomal structure membrane as a single-pass membrane protein. It is also necessary to know that this protein enters the Pre-autophagosomal structure membrane during mitophagy and enters the vacuole along with mitochondria.(Kanki, Wang, Cao, et al., 2009; Okamoto et al., 2009). As we said earlier, nitrogen starvation is effective in increasing mitophagy; research has shown that under nitrogen starvation conditions, the level of ATG32 mRNA increases up to 10 times.(Aihara et al., 2014). According to our knowledge, we have Atg 32 is the only specific receptor for mitophagy in *Saccharomyces cerevisiae*. (Huang & Klionsky, 2021).

How do these proteins relate to mitophagy?

When mitophagy is induced into the cell based on the received mechanisms, a two-layered membrane of the endoplasmic reticulum or Golgi begins to form. Mitochondria is called a vesicle forming a phagophore, and a completed vesicle is called an autophagosome in general. In the outer membrane,

the outer layer and the inner part of the inner membrane are covered with eight proteins. On the other hand, we know that the 32 proteins are generally located on the outer membrane of the mitochondria. It is necessary to remember that the mitochondrion is a double-membrane organelle. Research has shown that the higher the ROS content, the more intense the mitophagy mechanism. Phosphorylation of serine 114 in Atg 32 causes protein complexes of 11, and the possibility of starting the mitophagy mechanism is provided. Then, the Atg11 homodimer attracts other autophagy proteins to the stage, which stimulates the expansion of the phagophore membrane. The same thing that is needed to cover the mitochondria at that moment and the connection of the proteins of ATG 8 and 32 and complex 11 eventually leads to the closing of the mitochondria and the completion of the phagophore, which is now called the autophagosome, and in the next step, the outer membrane of the autophagosome with the vacuole membrane. For this, other proteins are involved, which are not discussed in our discussion, and cause the autophagosome, which has a membrane, to enter the vacuole, and the mitochondria in the lumen of the vacuole are converted, and its contents are returned. (Figure 2) To save energy and to save consumption, damaged mitochondria must be removed from the cell so as not to cause problems for the cell.(Delorme-Axford et al., 2015; Innokentev & Kanki, 2021; Nair & Klionsky, 2005).

What mechanisms are involved in the binding of these proteins, what factors increase the amount of mitophagy, and what will be the consequences are some of the things that continue to challenge researchers.

Conclusion

Mitophagy is the specific autophagy of mitochondria, which is a crucial organelle with a double membrane that produces ATP for the cell. Furthermore, during the life of the cell and depending on the conditions of the cell, there can be more than one in the cell. In *Saccharomyces cerevisiae*, it is controlled by three genes that produce proteins.

Atg 32 protein is a specific protein that is located on the outer membrane of mitochondria. In case of accumulation of reactive oxygen species, the amount of these proteins increases. Finally, due to the accumulation of reactive oxygen species, the function of mitochondria becomes problematic; the cell must remove mitochondria. Slow, and the phagophore is formed around the mitochondria (this is in a situation where the mitochondria have accumulated many active species due to the passage of time; in another situation where the cell has produced many mitochondria and does not need them at that moment, the same procedure continues). Atg 8 proteins are located on the inner and outer surface of the phagophore, and in the presence of an intermediary protein that is a protein scaffold and is concerned with other types of autophagy, called Atg 11, the mitochondria and phagophore membrane are attached, and the autophagosome is completed. In yeast, the vacuole comes into action and eventually causes the digestion of excess mitochondria or damage.

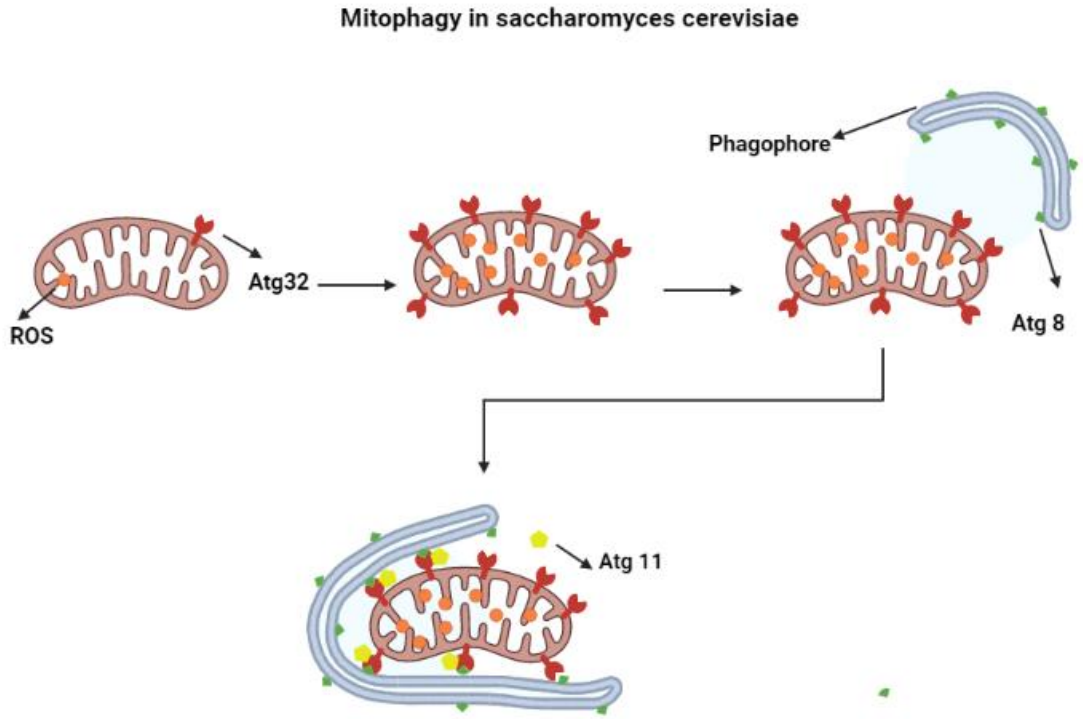

Figure 2. Reactions occurring in yeast mitophagy. This figure was prepared by the authors of this article, using the biorender website.

References

Aihara, M., Jin, X., Kurihara, Y., Yoshida, Y., Matsushima, Y., Oku, M., Hirota, Y., Saigusa, T., Aoki, Y., & Uchiumi, T. (2014). Tor and the Sin3–Rpd3 complex regulate expression of the mitophagy receptor protein Atg32 in yeast. *Journal of cell science*, *127*(14), 3184-3196.

Aoki, Y., Kanki, T., Hirota, Y., Kurihara, Y., Saigusa, T., Uchiumi, T., & Kang, D. (2011). Phosphorylation of Serine 114 on Atg32 mediates mitophagy. *Molecular biology of the cell*, *22*(17), 3206-3217.

Araki, Y., Ku, W.-C., Akioka, M., May, A. I., Hayashi, Y., Arisaka, F., Ishihama, Y., & Ohsumi, Y. (2013). Atg38 is required for autophagy-specific phosphatidylinositol 3-kinase complex integrity. *Journal of Cell Biology*, *203*(2), 299-313.

Barve, G., & Manjithaya, R. (2021). Cross-talk between autophagy and sporulation in *Saccharomyces cerevisiae*. *Yeast*, *38*(7), 401-413.

Biorender.com, https://www.biorender.com

Camougrand, N., Vigié, P., Gonzalez, C., Manon, S., & Bhatia-Kiššová, I. (2020). The yeast mitophagy receptor Atg32 is ubiquitinated and degraded by the proteasome. *PLoS One*, *15*(12), e0241576.

Csizmadia, T., & Juhász, G. (2020). Crinophagy mechanisms and its potential role in human health and disease. *Progress in Molecular Biology and Translational Science*, *172*, 239-255.

Delorme-Axford, E., Guimaraes, R. S., Reggiori, F., & Klionsky, D. J. (2015). The yeast *Saccharomyces cerevisiae*: an overview of methods to study autophagy progression. *Methods*, *75*, 3-12.

Engel, S. R., Dietrich, F. S., Fisk, D. G., Binkley, G., Balakrishnan, R., Costanzo, M. C., Dwight, S. S., Hitz, B. C., Karra, K., & Nash, R. S. (2014). The reference genome sequence of *Saccharomyces cerevisiae*: then and now. *G3: Genes, Genomes, Genetics*, *4*(3), 389-398.

Eskelinen, E.-L. (2008). New insights into the mechanisms of macroautophagy in mammalian cells. *International review of cell and molecular biology*, *266*, 207-247.

Fang, D., Xie, H., Hu, T., Shan, H., & Li, M. (2021). Binding features and functions of ATG3. *Frontiers in Cell and Developmental Biology*, *9*, 685625.

Farre, J.-C., Manjithaya, R., Mathewson, R. D., & Subramani, S. (2008). PpAtg30 tags peroxisomes for turnover by selective autophagy. *Developmental cell*, *14*(3), 365-376.

Faruk, M. O., Ichimura, Y., & Komatsu, M. (2021). Selective autophagy. *Cancer Sci*, *112*(10), 3972-3978. https://doi.org/10.1111/cas.15112

Feldmann, H. (2010). Horst Yeast. *Molecular and Cell Bio, Wiley-Blackwell, Germany*.

Feng, Y., He, D., Yao, Z., & Klionsky, D. J. (2014). The machinery of macroautophagy. *Cell Research*, *24*(1), 24-41.

Galluzzi, L., Kroemer, G., & Bravo-San Pedro, J. M. (2017). *Molecular Characterization of Autophagic Responses Part B*. Academic Press.

Geng, J., & Klionsky, D. J. (2008). The Atg8 and Atg12 ubiquitin-like conjugation systems in macroautophagy. *EMBO reports*, *9*(9), 859-864.

He, C., Song, H., Yorimitsu, T., Monastyrska, I., Yen, W.-L., Legakis, J. E., & Klionsky, D. J. (2006). Recruitment of Atg9 to the preautophagosomal structure by Atg11 is essential for selective autophagy in budding yeast. *The Journal of cell biology*, *175*(6), 925-935.

Hirata, E., Shirai, K., Kawaoka, T., Sato, K., Kodama, F., & Suzuki, K. (2021). Atg15 in *Saccharomyces cerevisiae* consists of two functionally distinct domains. *Mol Biol Cell*, *32*(8), 645-663. https://doi.org/10.1091/mbc.E20-07-0500

Huang, Y. J., & Klionsky, D. J. (2021). Yeast mitophagy: Unanswered questions. *Biochimica et Biophysica Acta (BBA)-General Subjects*, *1865*(8), 129932.

Innokentev, A., & Kanki, T. (2021). Mitophagy in yeast: Molecular mechanism and regulation. *Cells*, *10*(12), 3569.

Kanki, T., Wang, K., Baba, M., Bartholomew, C. R., Lynch-Day, M. A., Du, Z., Geng, J., Mao, K., Yang, Z., & Yen, W.-L. (2009). A genomic screen for yeast mutants defective in selective mitochondria autophagy. *Molecular biology of the cell*, *20*(22), 4730-4738.

Kanki, T., Wang, K., Cao, Y., Baba, M., & Klionsky, D. J. (2009). Atg32 is a mitochondrial protein that confers selectivity during mitophagy. *Developmental cell*, *17*(1), 98-109.

King, J. S. (2012). Autophagy across the eukaryotes: is S. cerevisiae the odd one out? *Autophagy*, *8*(7), 1159-1162.

Kirisako, T., Baba, M., Ishihara, N., Miyazawa, K., Ohsumi, M., Yoshimori, T., Noda, T., & Ohsumi, Y. (1999). Formation process of autophagosome is traced with Apg8/Aut7p in yeast. *The Journal of cell biology*, *147*(2), 435-446.

Kirisako, T., Ichimura, Y., Okada, H., Kabeya, Y., Mizushima, N., Yoshimori, T., Ohsumi, M., Takao, T., Noda, T., & Ohsumi, Y. (2000). The reversible modification regulates the membrane-binding state of Apg8/Aut7 essential for autophagy and the cytoplasm to vacuole targeting pathway. *The Journal of cell biology*, *151*(2), 263-276.

Klionsky, D. J. (2012). Look people,"Atg" is an abbreviation for "autophagy-related." That's it. In (Vol. 8, pp. 1281-1282): Taylor & Francis.

Ko, T. K., & Tan, D. J. Y. (2023). Is Disrupted Mitophagy a Central Player to Parkinson's Disease Pathology? *Cureus*, *15*(2).

Kruckeber, A., & Dickinson, J. (2004). The metabolism and molecular physiology of *Saccharomyces cerevisiae*. *Carbon Metabolism, 1st ed.; Dickinson, JR, Schweizer, M., Eds*.

Leary, K. A., Hawkins, W. D., Andhare, D., Popelka, H., Klionsky, D. J., & Ragusa, M. J. (2022). Atg23 is a vesicle-tethering protein. *Autophagy*, *18*(10), 2510-2511.

Lei, Y., Tang, D., Liao, G., Xu, L., Liu, S., Chen, Q., Li, C., Duan, J., Wang, K., & Wang, J. (2021). The crystal structure of Atg18 reveals a new binding site for Atg2 in *Saccharomyces cerevisiae*. *Cellular and Molecular Life Sciences*, *78*, 2131-2143.

Liesa, M., Palacín, M., & Zorzano, A. (2009). Mitochondrial dynamics in mammalian health and disease. *Physiological reviews*, *89*(3), 799-845.

Liu, D., Mari, M., Li, X., Reggiori, F., Ferro-Novick, S., & Novick, P. (2022). ER-phagy requires the assembly of actin at sites of contact between the cortical ER and endocytic pits. *Proceedings of the National Academy of Sciences*, *119*(6), e2117554119.

Lynch-Day, M. A., & Klionsky, D. J. (2010). The Cvt pathway as a model for selective autophagy. *FEBS letters*, *584*(7), 1359-1366.

Margolis, H. K., Katzenell, S., Leary, K. A., & Ragusa, M. J. (2020). The third coiled coil domain

of Atg11 is required for shaping mitophagy initiation sites. *Journal of molecular biology*, *432*(21), 5752-5764.

Martens, S., & Fracchiolla, D. (2020). Activation and targeting of ATG8 protein lipidation. *Cell discovery*, *6*(1), 23.

Maruyama, T., & Noda, N. N. (2018). Autophagy-regulating protease Atg4: structure, function, regulation and inhibition. *The Journal of antibiotics*, *71*(1), 72-78.

Meijer, W. H., van der Klei, I. J., Veenhuis, M., & Kiel, J. A. (2007). ATG genes involved in non-selective autophagy are conserved from yeast to man, but the selective Cvt and pexophagy pathways also require organism-specific genes. *Autophagy*, *3*(2), 106-116.

Miller-Fleming, L., Cheong, H., Antas, P., & Klionsky, D. J. (2014). Detection of *Saccharomyces cerevisiae* Atg13 by western blot. *Autophagy*, *10*(3), 514-517. https://doi.org/10.4161/auto.27707

Mochida, K., Otani, T., Katsumata, Y., Kirisako, H., Kakuta, C., Kotani, T., & Nakatogawa, H. (2022). Atg39 links and deforms the outer and inner nuclear membranes in selective autophagy of the nucleus. *Journal of Cell Biology*, *221*(2), e202103178.

Monastryska, I., Kiel, J. A., Krikken, A. M., Komduur, J. A., Veenhuis, M., & Klei, I. J. v. d. (2005). The Hansenula polymorpha ATG25 gene encodes a novel coiled-coil protein that is required for macropexophagy. *Autophagy*, *1*(2), 92-100.

Motley, A. M., Nuttall, J. M., & Hettema, E. H. (2012). Pex3-anchored Atg36 tags peroxisomes for degradation in *Saccharomyces cerevisiae*. *The EMBO journal*, *31*(13), 2852-2868.

Nair, U., & Klionsky, D. J. (2005). Molecular mechanisms and regulation of specific and nonspecific autophagy pathways in yeast. *Journal of Biological Chemistry*, *280*(51), 41785-41788.

Nazarko, V. Y., Nazarko, T. Y., Farré, J. C., Stasyk, O. V., Warnecke, D., Ulaszewski, S., Cregg, J. M., Sibirny, A. A., & Subramani, S. (2011). Atg35, a micropexophagy-specific protein that regulates micropexophagic apparatus formation in Pichia pastoris. *Autophagy*, *7*(4), 375-385. https://doi.org/10.4161/auto.7.4.14369

Noda, N. N., Fujioka, Y., Hanada, T., Ohsumi, Y., & Inagaki, F. (2013). Structure of the Atg12–Atg5 conjugate reveals a platform for stimulating Atg8–PE conjugation. *EMBO reports*, *14*(2), 206-211.

Okamoto, K., Kondo-Okamoto, N., & Ohsumi, Y. (2009). Mitochondria-anchored receptor Atg32 mediates degradation of mitochondria via selective autophagy. *Developmental cell*, *17*(1), 87-97.

Osawa, T., Kotani, T., Kawaoka, T., Hirata, E., Suzuki, K., Nakatogawa, H., Ohsumi, Y., & Noda, N. N. (2019). Atg2 mediates direct lipid transfer between membranes for autophagosome formation. *Nature Structural & Molecular Biology*, *26*(4), 281-288.

Parrella, E., & Longo, V. D. (2008). The chronological life span of *Saccharomyces cerevisiae* to study mitochondrial dysfunction and disease. *Methods*, *46*(4), 256-262.

Pfaffenwimmer, T., Reiter, W., Brach, T., Nogellova, V., Papinski, D., Schuschnig, M., Abert, C., Ammerer, G., Martens, S., & Kraft, C. (2014). Hrr25 kinase promotes selective autophagy by phosphorylating the cargo receptor Atg19. *EMBO Rep*, *15*(8), 862-870. https://doi.org/10.15252/embr.201438932

Popelka, H., Damasio, A., Hinshaw, J. E., Klionsky, D. J., & Ragusa, M. J. (2017). Structure and function of yeast Atg20, a sorting nexin that facilitates autophagy induction. *Proceedings of the National Academy of Sciences*, *114*(47), E10112-E10121.

Popelka, H., Reinhart, E. F., Metur, S. P., Leary, K. A., Ragusa, M. J., & Klionsky, D. J. (2021). Membrane binding and homodimerization of Atg16 via two distinct protein regions is essential for autophagy in yeast. *Journal of molecular biology*, *433*(5), 166809.

Ravikumar, B., Sarkar, S., Davies, J. E., Futter, M., Garcia-Arencibia, M., Green-Thompson, Z. W., Jimenez-Sanchez, M., Korolchuk, V. I., Lichtenberg, M., & Luo, S. (2010). Regulation of mammalian autophagy in physiology and pathophysiology. *Physiological reviews*, *90*(4), 1383-1435.

Scarlett, J. L., & Murphy, M. P. (1997). Release of apoptogenic proteins from the mitochondrial intermembrane space during the mitochondrial permeability transition. *FEBS letters*, *418*(3), 282-286.

Scott, S. V., & Klionsky, D. J. (1998). Delivery of proteins and organelles to the vacuole from the

cytoplasm. *Current opinion in cell biology*, *10*(4), 523-529.

Shpilka, T., Weidberg, H., Pietrokovski, S., & Elazar, Z. (2011a). Atg8: an autophagy-related ubiquitin-like protein family. *Genome Biology*, *12*(7), 226. https://doi.org/10.1186/gb-2011-12-7-226

Shpilka, T., Weidberg, H., Pietrokovski, S., & Elazar, Z. (2011b). Atg8: an autophagy-related ubiquitin-like protein family. *Genome Biology*, *12*(7), 1-11.

Shravage, B. V., Hill, J. H., Powers, C. M., Wu, L., & Baehrecke, E. H. (2013). Atg6 is required for multiple vesicle trafficking pathways and hematopoiesis in Drosophila. *Development*, *140*(6), 1321-1329.

Stasyk, O. V., Stasyk, O. G., Mathewson, R. D., Farré, J.-C., Nazarko, V. Y., Krasovska, O. S., Subramani, S., Cregg, J. M., & Sibirny, A. A. (2006). Atg28, a novel coiled-coil protein involved in autophagic degradation of peroxisomes in the methylotrophic yeast Pichia pastoris. *Autophagy*, *2*(1), 30-38.

Straub, M., Bredschneider, M., & Thumm, M. (1997). AUT3, a serine/threonine kinase gene, is essential for autophagocytosis in *Saccharomyces cerevisiae*. *Journal of bacteriology*, *179*(12), 3875-3883.

Sun, Q., Fan, W., Chen, K., Ding, X., Chen, S., & Zhong, Q. (2008). Identification of Barkor as a mammalian autophagy-specific factor for Beclin 1 and class III phosphatidylinositol 3-kinase. *Proceedings of the National Academy of Sciences*, *105*(49), 19211-19216.

Torggler, R., Papinski, D., & Kraft, C. (2017). Assays to monitor autophagy in *Saccharomyces cerevisiae*. *Cells*, *6*(3), 23.

Ungermann, C., & Reggiori, F. (2018). Atg9 proteins, not so different after all. *Autophagy*, *14*(8), 1456-1459. https://doi.org/10.1080/15548627.2018.1477382

Vyas, S., Zaganjor, E., & Haigis, M. C. (2016). Mitochondria and cancer. *Cell*, *166*(3), 555-566.

Wang, C., & Youle, R. J. (2009). The role of mitochondria in apoptosis. *Annual review of genetics*, *43*, 95-118.

Wang, K., Jin, M., Liu, X., & Klionsky, D. J. (2013). Proteolytic processing of Atg32 by the mitochondrial i-AAA protease Yme1 regulates mitophagy. *Autophagy*, *9*(11), 1828-1836.

Wang, X., Su, B., Lee, H.-g., Li, X., Perry, G., Smith, M. A., & Zhu, X. (2009). Impaired balance of mitochondrial fission and fusion in Alzheimer's disease. *Journal of neuroscience*, *29*(28), 9090-9103.

Watanabe, Y., Noda, N. N., Kumeta, H., Suzuki, K., Ohsumi, Y., & Inagaki, F. (2010). Selective transport of α-mannosidase by autophagic pathways: structural basis for cargo recognition by Atg19 and Atg34. *Journal of Biological Chemistry*, *285*(39), 30026-30033.

Wood, V., Gwilliam, R., Rajandream, M.-A., Lyne, M., Lyne, R., Stewart, A., Sgouros, J., Peat, N., Hayles, J., & Baker, S. (2002). The genome sequence of Schizosaccharomyces pombe. *Nature*, *415*(6874), 871-880.

Yamamoto, H., Zhang, S., & Mizushima, N. (2023). Autophagy genes in biology and disease. *Nature Reviews Genetics*, 1-19.

Yang, Z., Huang, J., Geng, J., Nair, U., & Klionsky, D. J. (2006). Atg22 recycles amino acids to link the degradative and recycling functions of autophagy. *Mol Biol Cell*, *17*(12), 5094-5104. https://doi.org/10.1091/mbc.e06-06-0479

Yao, Z., Delorme-Axford, E., Backues, S. K., & Klionsky, D. J. (2015). Atg41/Icy2 regulates autophagosome formation. *Autophagy*, *11*(12), 2288-2299.

Yen, W.-L., Legakis, J. E., Nair, U., & Klionsky, D. J. (2007). Atg27 is required for autophagy-dependent cycling of Atg9. *Molecular biology of the cell*, *18*(2), 581-593.

Zientara-Rytter, K., & Subramani, S. (2020). Mechanistic Insights into the Role of Atg11 in Selective Autophagy. *J Mol Biol*, *432*(1), 104-122. https://doi.org/10.1016/j.jmb.2019.06.017