Autophagy

The role in health and disease.

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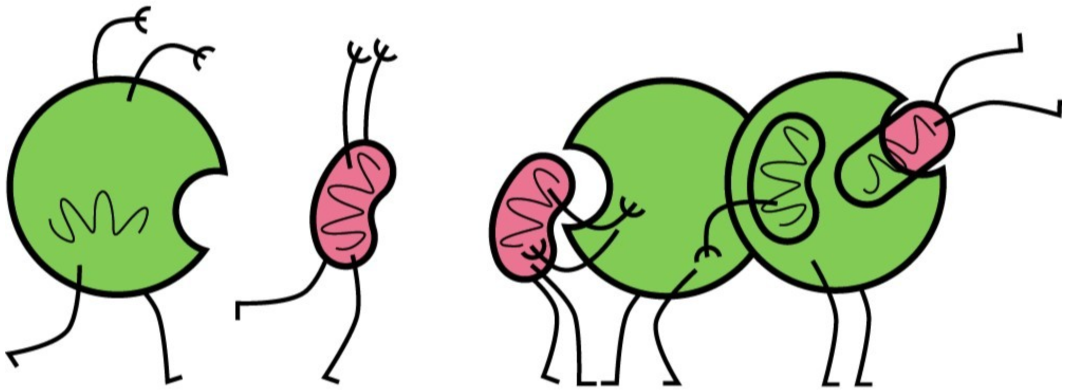
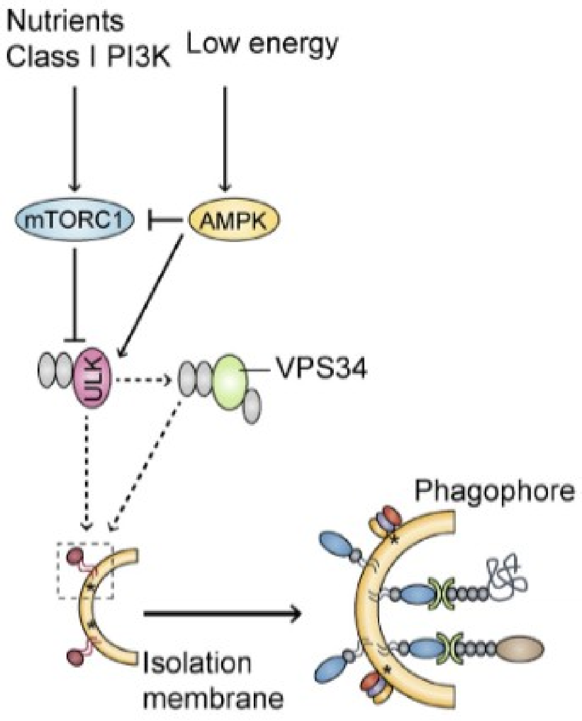


What is Autophagy?

Autophagy is a highly conserved catabolic process induced under various conditions of cellular stress, which prevents cell damage and promotes survival in the event of energy or nutrient shortage and responds to various cytotoxic insults.

Chemical or genetic disturbance of autophagy and the age-dependent decline in autophagic activity have been implicated in the progression of cancer, neurodegeneration and immune diseases, as well as ageing.

The most characterized trigger for induction of autophagy is deprivation of amino acids, which results in inhibition of the master cell growth regulator serine/threonine kinase mTOR.



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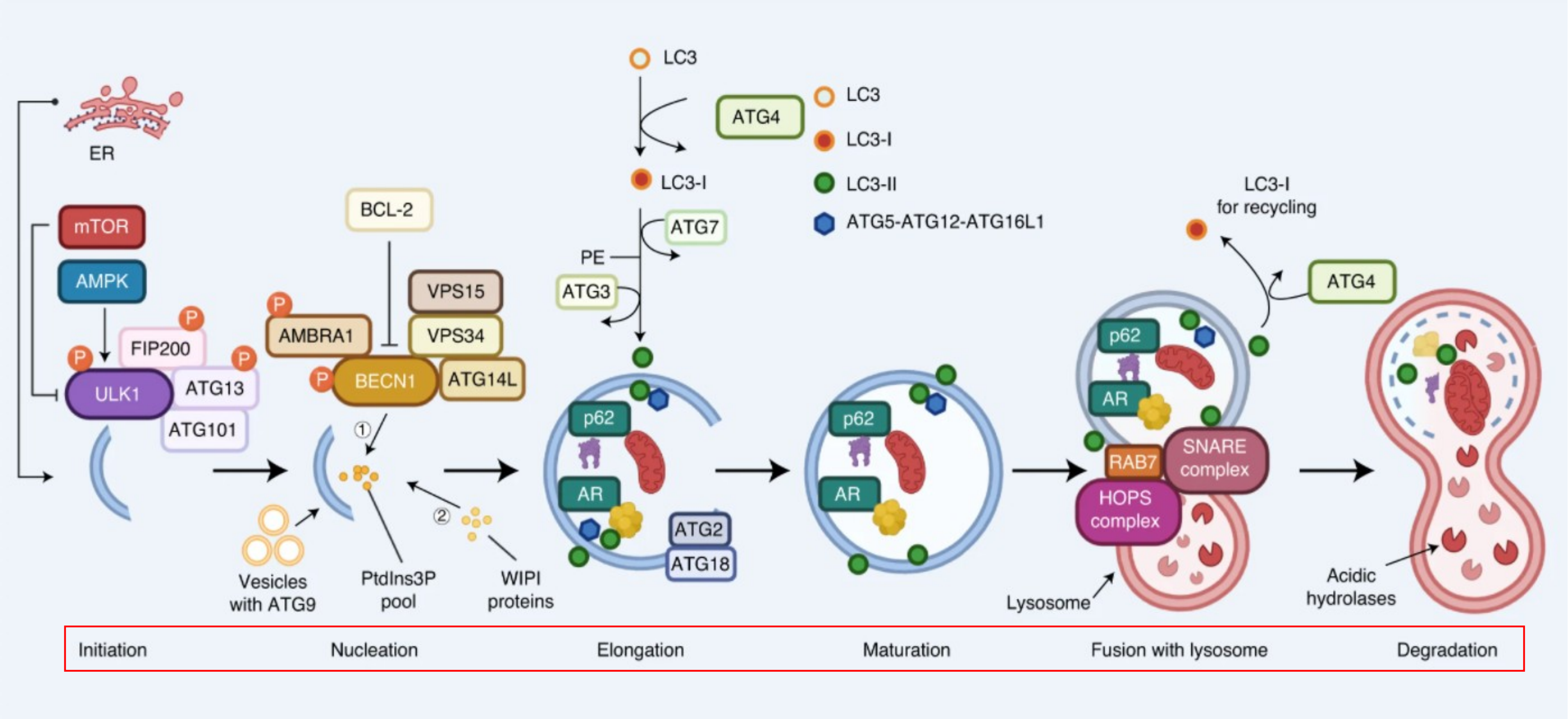
Mechanism of autophagy

Autophagy (from the Greek words *auto*, meaning ‘self’, and *phagein*, meaning ‘to eat’) is a fundamental cellular process that eliminates molecules and subcellular elements, including nucleic acids, proteins, lipids and organelles, via lysosome-mediated degradation to promote homeostasis, differentiation, development and survival.



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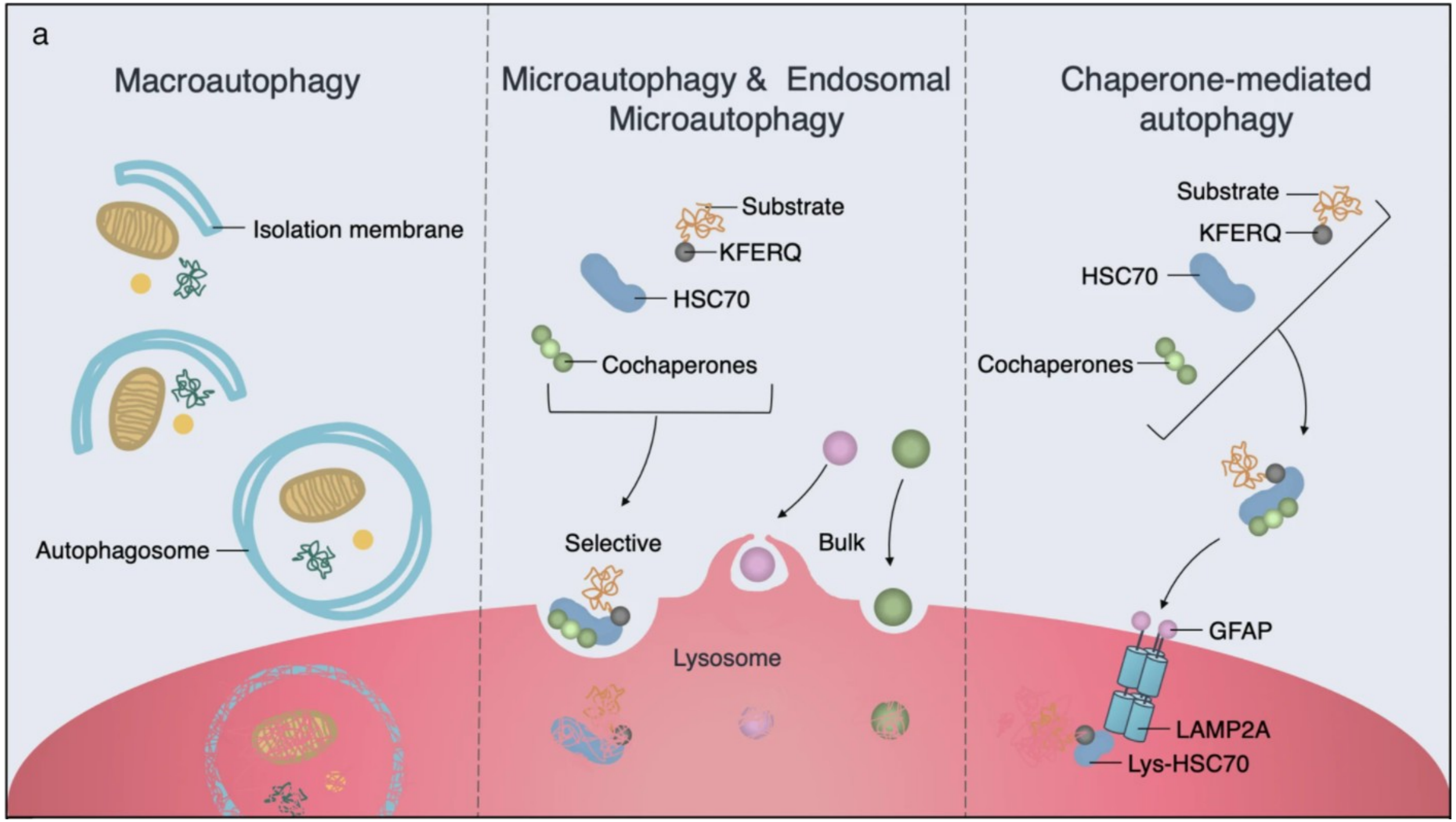
Types of Autophagy

Autophagy process can be distinguished according to how cargo enters the lysosome compartment.

In macroautophagy, a double-membrane isolation membrane elongates, expands, and seals to make an autophagosome around cytoplasmic components before fusing with the lysosome.

Microautophagy and endosomal microautophagy deliver small cargoes directly to the lysosome either without or with chaperones, respectively.



Chaperone-mediated autophagy requires the lysosome-associated membrane protein 2A (LAMP2A), in addition to molecular chaperones.

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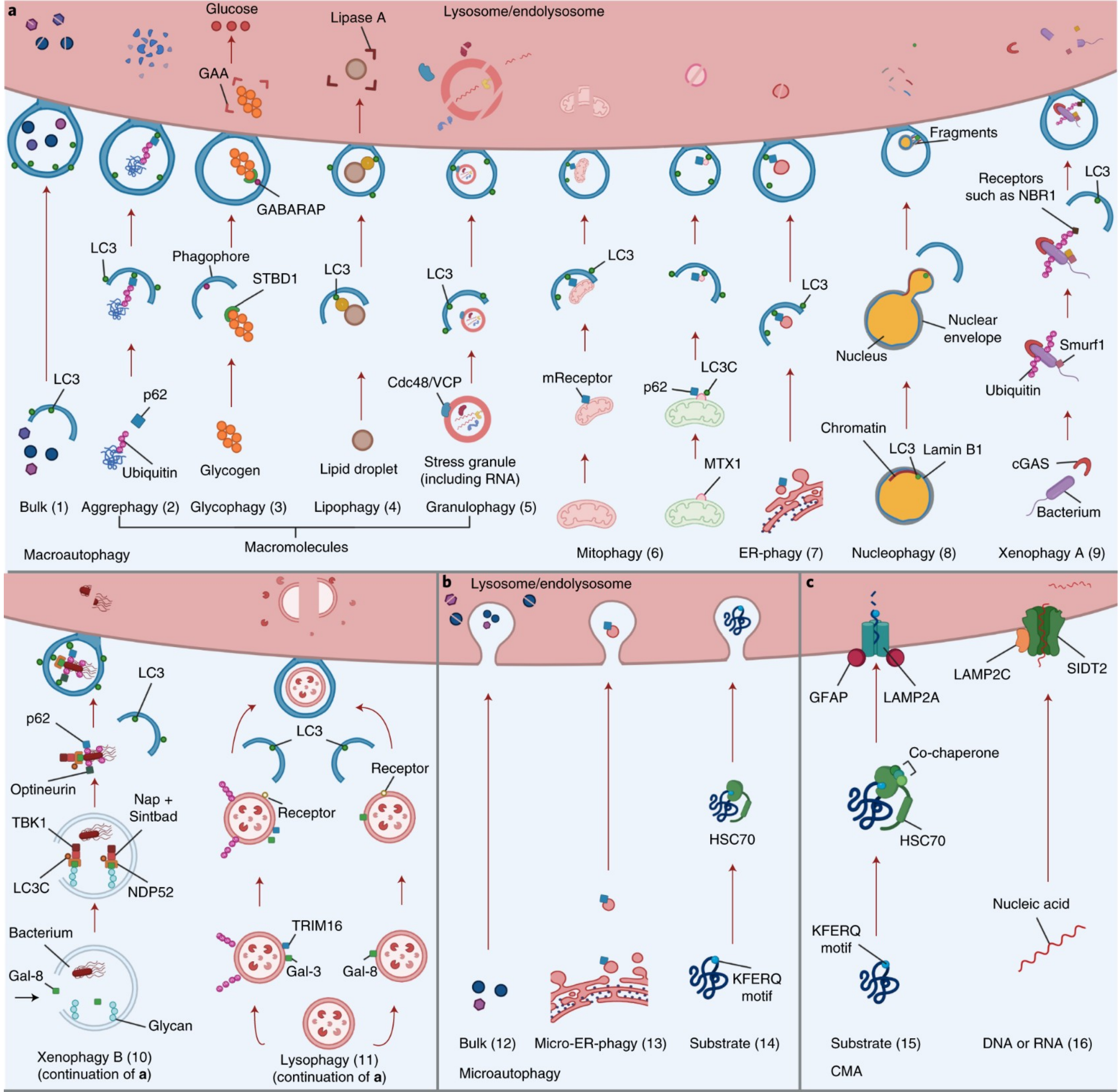
Types of Autophagy

Autophagy is recognized as a highly selective cellular clearance pathway that is associated with the maintenance of cellular and tissue homeostasis.

Selective autophagy can be further divided into many subtypes on the basis of the specific cargos involved.



Aman, Y., Schmauck-Medina, T., Hansen, M. *et al.* Autophagy in healthy aging and disease. *Nat Aging* **1,** 634–650 (2021). https://doi.org/10.1038/s43587-021-00098-4

Physiological Roles of Autophagy

At first autophagy activation was identified as the response to starvation; currently, we know that autophagy is activated in response to different cellular stressors including exercise, endoplasmic reticulum stress, infection, and hypoxia.

Emerging results of the research are highlighting the pivotal role of autophagic response in tissue differentiation, functions, and remodeling after stimuli.

Table 1. Physiological Functions of Autophagy in Mammals.*

Function	Mechanism
Adaptive metabolic response to starvation and exercise	Enhanced degradation to maintain protein synthesis and energy production
Development	
Embryonic development	Degradation of maternal proteins to produce zygotic proteins, degradation of paternal mitochondria
Differentiation and tissue development	Adipose tissues, lymphocytes, erythrocytes, heart, intestine, and other organs (e.g., testis and ovary)
Homeostasis†	
Basal turnover	Continuous bulk degradation of cytoplasmic contents (e.g., proteins, nucleic acids, and glycogen)
Protein quality control	Active degradation of misfolded proteins or condensates and aggregates
Organellar homeostasis	Elimination of excess, damaged, harmful, or ruptured organelles
Lipid homeostasis	Degradation of membrane lipids and lipid droplets (lipophagy) and regulation of PPARα
Redox homeostasis	Degradation of damaged mitochondria (mitophagy)
Nrf2 regulation	Degradation of the KEAP1-binding protein SQSTM1/p62
Iron homeostasis	Degradation of ferritin
Immunity or inflammation	
Control of pathogen replication	Selective elimination of pathogens (xenophagy)
Regulation of innate immunity	Regulation of inflammasome activation, innate immune signaling, and cytokine secretion
Regulation of B- and T-cell responses	Lymphocyte differentiation and antigen presentation
Other functions	
Antiaging	Homeostatic roles of autophagy
Stem-cell maintenance	Homeostatic roles of autophagy
Genomic integrity	Homeostatic roles of autophagy
Conventional secretion	Enhancement of regulated or constitutive secretion
Unconventional secretion	Fusion of autophagosomes (or related structures) with the plasma membrane
Cell death	Various mechanisms, including autosis

* A reference list for the information in this table and in Table S1 is provided in the Supplementary Appendix, available with the full text of this article at NEJM.org. Nrf2 denotes nuclear factor erythroid 2–related factor 2, and PPARα peroxisome proliferator–activated receptor α.

† The following types of autophagic degradation are named according to their specific substrate: aggrephagy (protein aggregates), ER-phagy or reticulophagy (endoplasmic reticulum), ferritinophagy (ferritin), glycophagy (glycogen), lipophagy (lipid droplets), lysophagy (lysosomes), mitophagy (mitochondria), nucleophagy (nucleus), pexophagy (peroxisomes), ribophagy (ribosomes), and xenophagy (microbes).

Mizushima, N. and B. Levine, *Autophagy in Human Diseases.* New England Journal of Medicine, 2020. **383**(16): p. 1564-1576.

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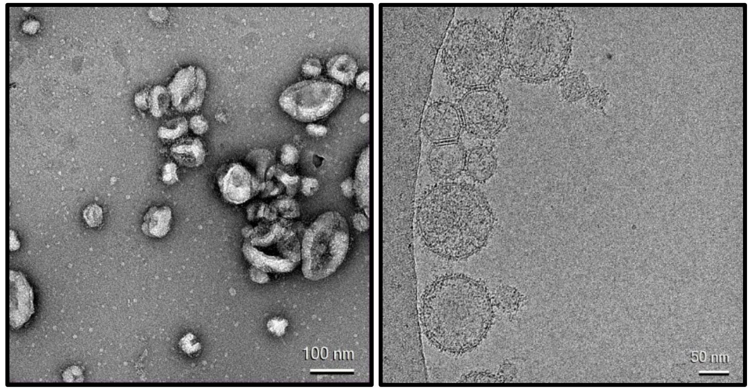
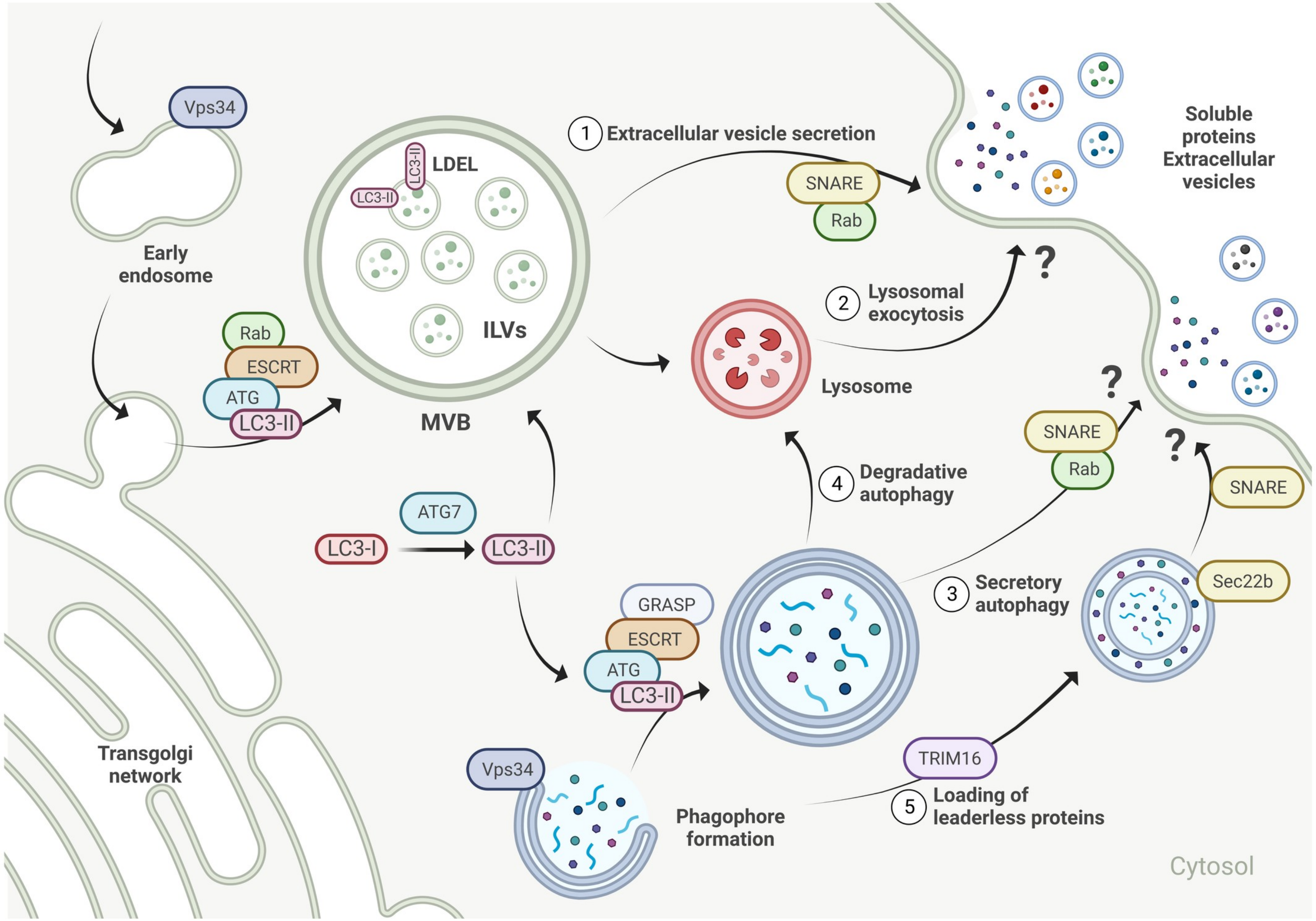
Secretory or non-cell autonomous autophagy

Secretory or non-cell autonomous autophagy is an umbrella term for autophagy-mediated extracellular secretion through autophagosomes/endosomes, including conventional and unconventional secretion of proteins, extracellular vesicle (EV) production, and egress of secretory lysosomes



Micrographs of **extracellular vesicles**. Negative stain EM (left) and Cryo EM (right).



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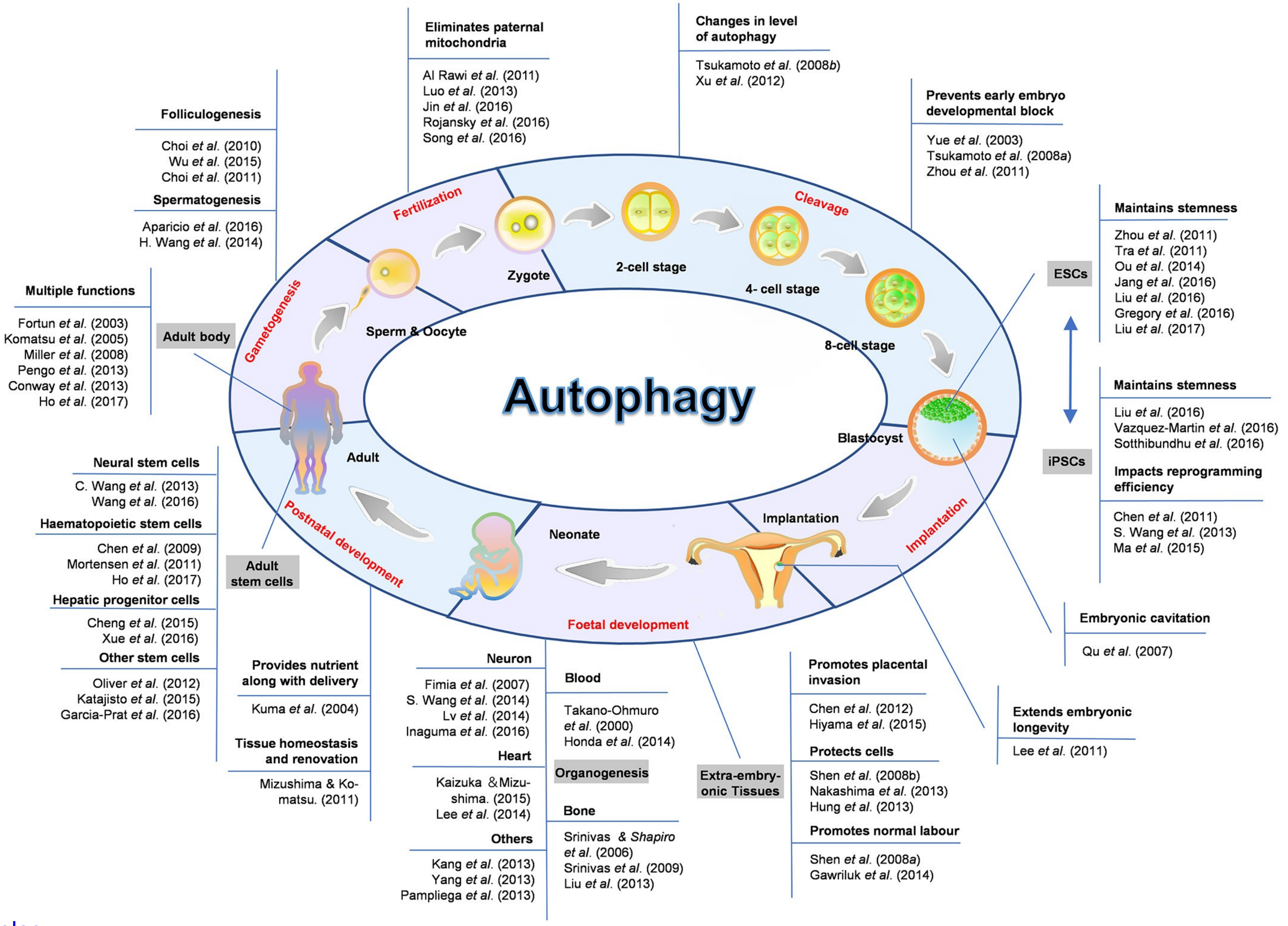
Function of Autopahgy

Autophagy plays an essential role during the mammalian life cycle. These functions span from fertilization, cleavage, implantation, fetal development, and postnatal development, to the next generation of gametogenesis, indicating that autophagy has important functions during the whole mammalian life cycle.

At birth the trans-placental nutrient supply is suddenly interrupted, and neonates face severe starvation until supply can be restored through milk nutrients. Neonates adapt to this adverse circumstance by inducing autophagy.

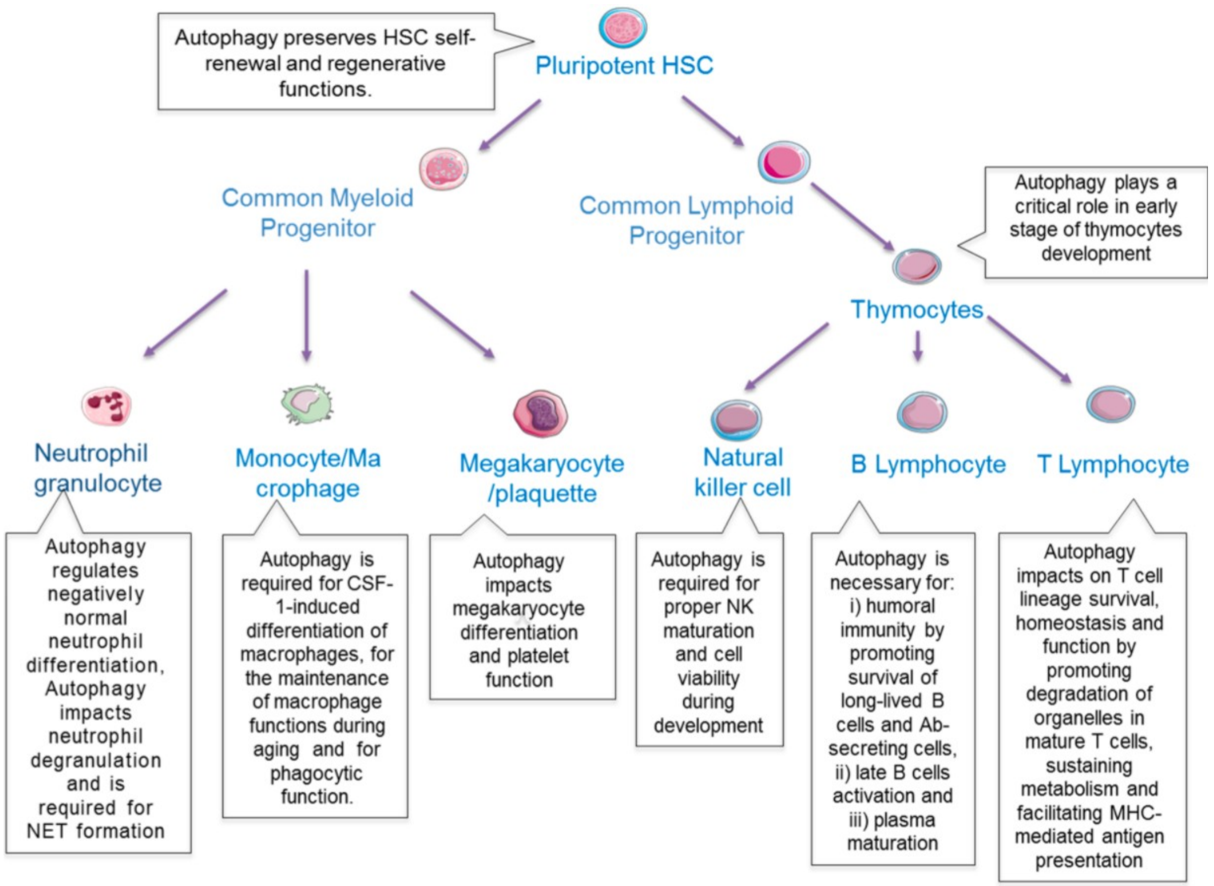


Autophagy and immunity

Autophagy plays a crucial role in initiating and supporting several processes in both innate and adaptive immunity.

ATG proteins directly contribute to pathogen clearance through selective autophagy of microorganisms, coordinated response with pattern recognition receptors, inflammasome formation, antigen presentation, and LC3-associated phagocytosis.

In adaptive immunity, autophagy modulates antigen processing and presentation and regulates the development of lymphocytes. Regulation of the organelle content of lymphocytes, especially mitochondria, by autophagy is crucial during differentiation.



The importance of autophagy for these functions is highlighted by the susceptibility of autophagy-deficient animals to infection and the implication of autophagy defects in autoimmune diseases, such as systemic lupus erythematosus, rheumatoid arthritis, psoriasis, diabetes and multiple sclerosis

Metur, S.P., Klionsky, D.J. Adaptive immunity at the crossroads of autophagy and metabolism. *Cell Mol Immunol* **18,** 1096–1105 (2021). https://doi.org/10.1038/s41423-021-00662-3

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Autophagy and Aging

Aging is a biological process that is characterized by time-dependent cellular and functional decline, resulting in reduced quality of life for the organism.

Aging is the primary risk factor for the development of many disorders, including cardiovascular disease (for example, stroke), cancer and neurodegenerative disease (for example, Alzheimer’s disease (AD)).

Among the many molecular changes associated with old age, altered autophagy has emerged as a feature of aging across diverse species.

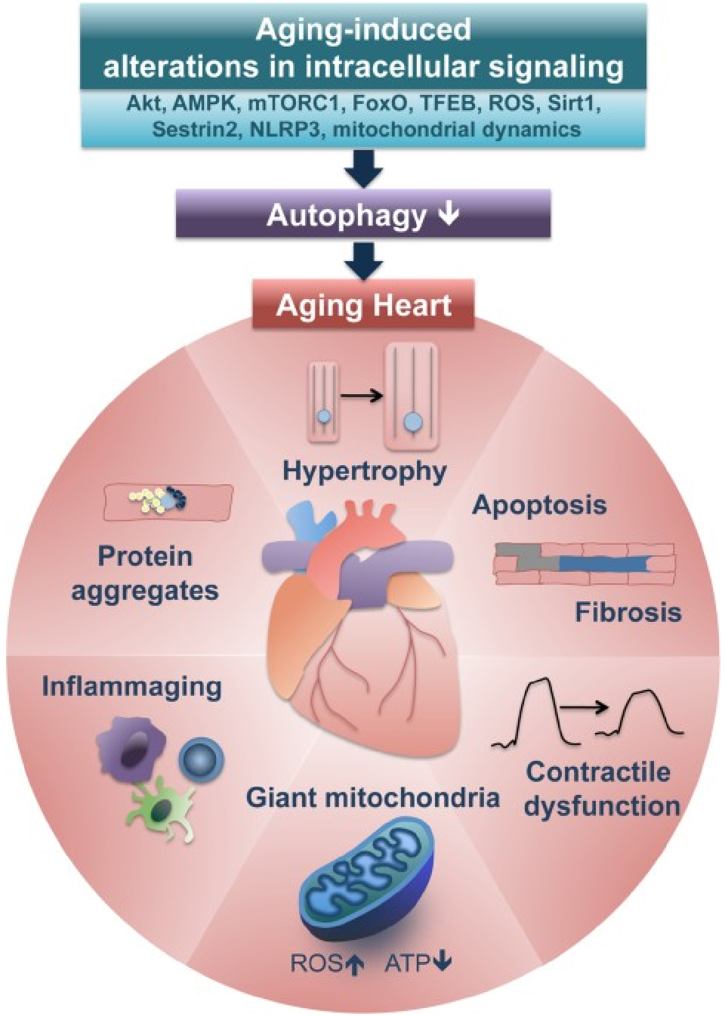
Compromised autophagy is a hallmark of aging

Autophagy and Aging

Aging is associated with an accumulation of damage to subcellular organelles. Selective autophagy is the common mechanism underlying the clearance of damaged and/or superfluous subcellular organelles such as mitochondria (mitophagy), the ER (reticulophagy or ER-phagy), the nucleus (nucleophagy) and lysosomes (lysophagy).

Gradual decline in the abundance of autophagy-related proteins and reduced delivery of cargo to lysosomes occur with age, implicating compromised autophagy as a cardinal feature of organismal aging.

The mounting evidence that an imbalance of autophagy is an important age-associated characteristic has driven extensive research into the development of compounds that can stimulate autophagy to promote longevity.



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Miyamoto, S. Autophagy and cardiac aging. *Cell Death Differ* **26**, 653–664 (2019). https://doi.org/10.1038/s41418-019-0286-9

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Autophagy and Aging

Summary of autophagy inducers that extend healthspan and increase lifespan in laboratory animals.

Pharmacological agent	Health benefit	Mode of action
Metformin	W, M: increase in lifespan and healthspan	Activates AMPK and other mechanisms[241] (also reviewed in ref. [242])
Rapamycin	W, F, M: increase in lifespan and different healthspan parameters	Direct autophagy induction via mTOR inhibition[243] (reviewed in ref. [242])
Resveratrol	Y, W, F, M: increase in lifespan and different healthspan parameters[a]	SIRT1-dependent induction of autophagy and non-autophagy pathways[112] (reviewed in ref. [68])
Spermidine	W, F, M, R: increase in median lifespan and different healthspan parameters	Autophagy, anti-inflammation, and arginine and nitric oxide metabolism[196,199]
NR/NMN	W, F, M: increase in lifespan; W, F, M: increase in healthspan; M: increased memory	Pathways dependent and independent of autophagy/mitophagy (reviewed in ref. [185,244])
Urolithin A	W: increase in lifespan and healthspan; W, M: increased memory	Autophagy/mitophagy induction[138,207,208]
Actinonin	W, M: increased memory	Autophagy/mitophagy-dependent pathway[138]
Tomatidine	W: increase in lifespan and healthspan	Mitophagy induction via the SKN-1–Nrf2 pathway[142]
Trehalose	W: increase in lifespan and healthspan[245]	?
MI	W: increase in lifespan and healthspan	PINK1-dependent mitophagy induction[246]
XPO1 inhibitors	W, F: increase in lifespan and improved conditions in neurodegenerative models	Induction of nuclear localization of HLH-30/TFEB[47]

Y, yeast; W, worms; F, flies; M, mice; R, rats; MI, myoinositol; NR, nicotinamide riboside; NMN, nicotinamide mononucleotide.

[a]No extension was found in wild-type mice with normal diet, but extended lifespan was observed in mice fed a high-fat diet[112].

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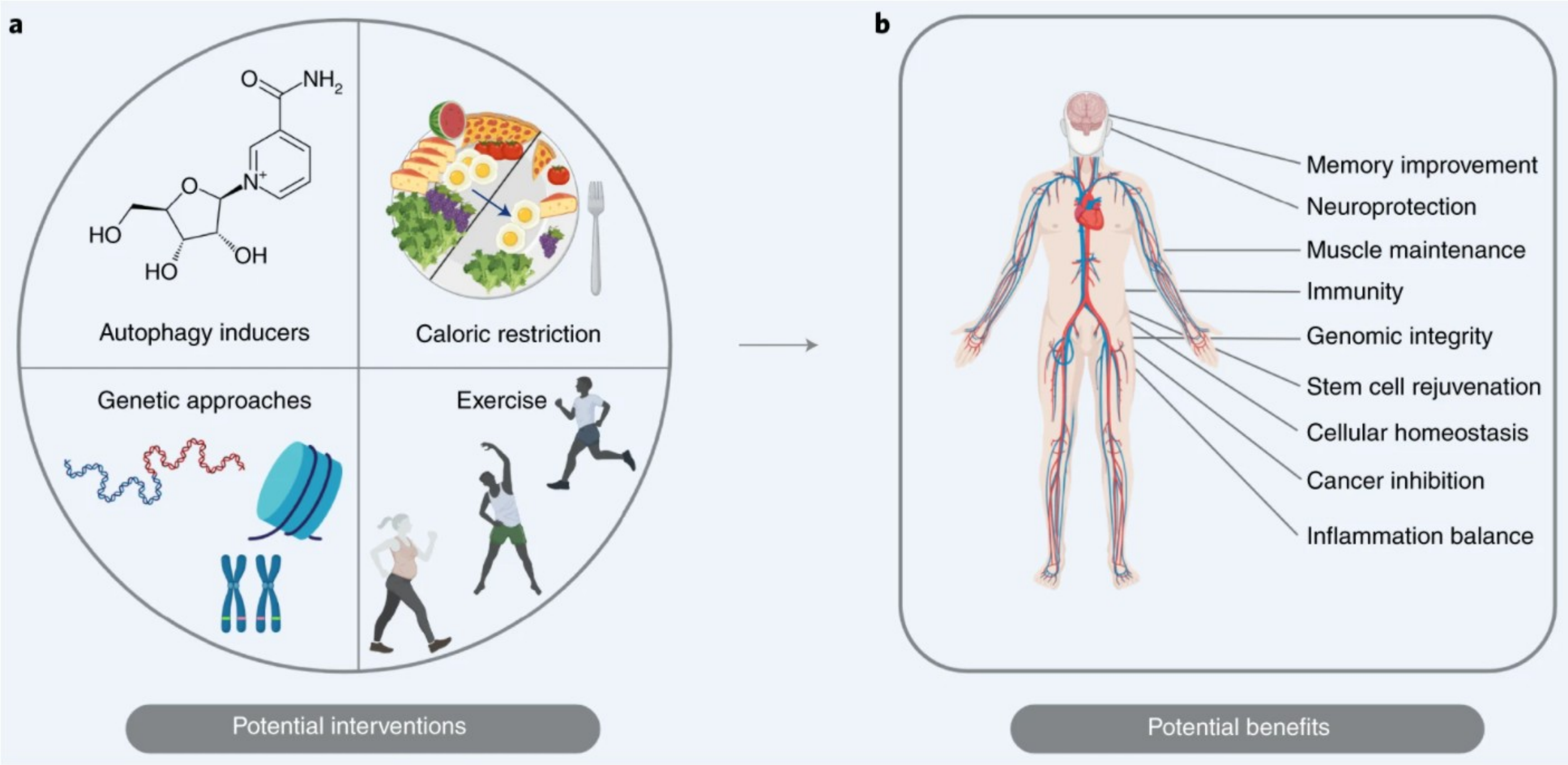
Autophagy and Aging



a, Potential interventions to stimulate autophagy: autophagy inducers, dietary restriction, exercise and genetic approaches. **b**, Autophagy induction could positively impact human health.

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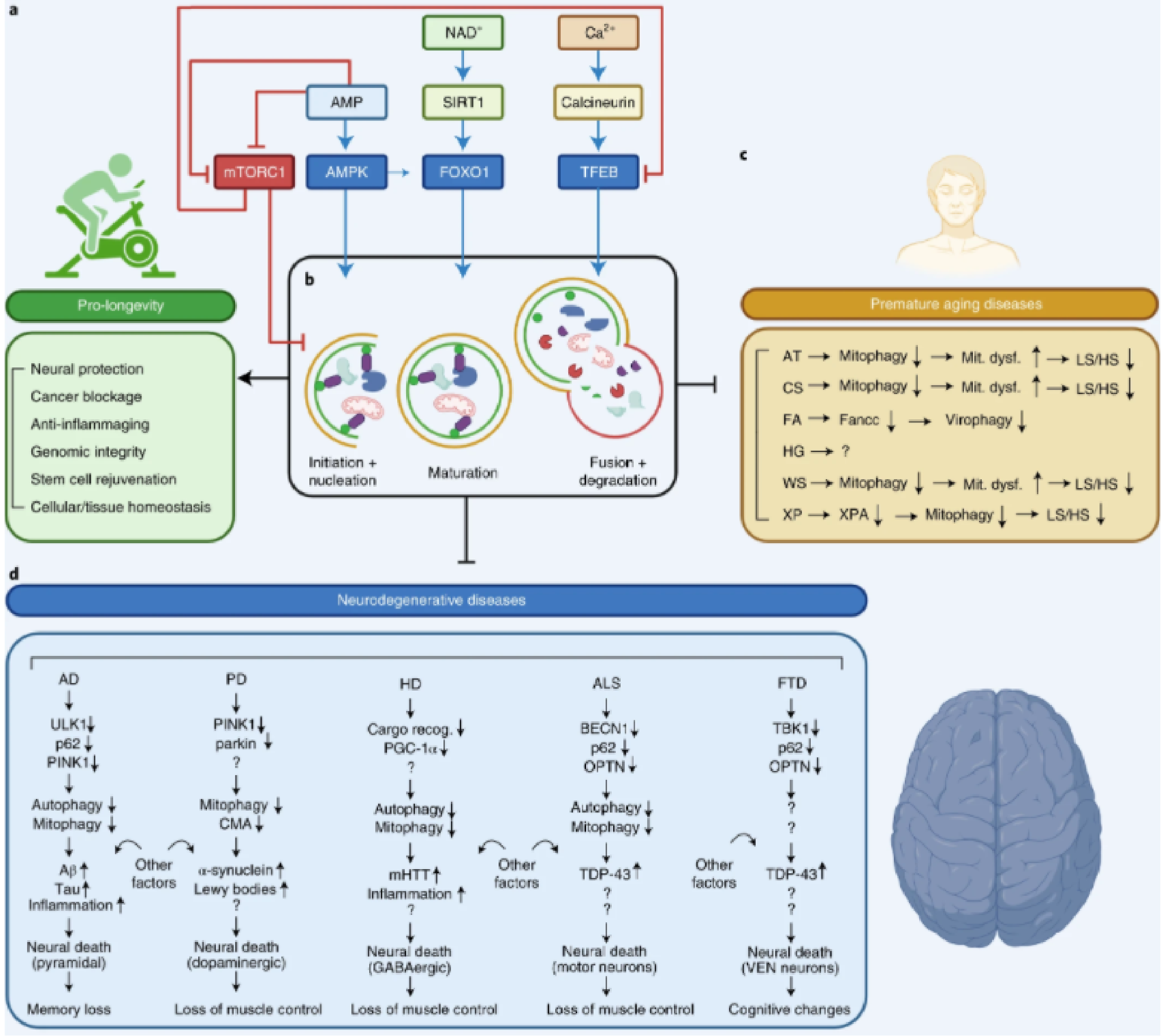
Autophagy in Health and Disease



Premature aging diseases with impaired mitophagy as a cause of mitochondrial dysfunction, which contributes to short lifespan (LS) and healthspan (HS).

ataxia telangiectasia (AT),
Cockayne syndrome (CS),
Fanconi anemia (FA),
Hutchinson–Gilford syndrome (HG),
Werner syndrome (WS) and xeroderma pigmentosum (XP; especially group A).

Autophagy (including subtypes of selective autophagy, such as mitophagy) is impaired in broad neurodegenerative diseases, where impairment may drive or exacerbate disease progression.

Alzheimer's disease (A)D,
Parkinson’s disease (PD),
Huntington’s disease (HD),
amyotrophic lateral sclerosis (ALS),
frontotemporal dementia (FTD).

We emphasize that these are not the only drivers of the diseases and other processes may have roles leading to pathology and symptomatology.

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Autophagy in Health and Disease

Mutations in autophagy-related genes are now known to cause mendelian disorders, and autophagy gene polymorphisms have been found to be associated with susceptibility to some diseases.

In the future, because autophagy has a waste disposal function, its activation and inhibition could be a novel therapeutic strategy for neurodegenerative diseases and cancers.

Table 2. Diseases Associated with Autophagy-Related Gene Mutations.*

Category	Examples of Diseases (Related Genes)
Adult neurodegenerative disorders	Parkinson's disease (**PRKN/PARK2** [AR], **PINK1/PARK6** [AR], **LRRK2/PARK8** [AD], **ATP13A2/PARK9** [AR], *GBA*, *TMEM175*), amyotrophic lateral sclerosis (**OPTN** [AD?], **VCP** [AD], **SQSTM1/p62** [AD], **TBK1** [AD], **UBQLN2** [XLD], **CHMP2B** [AD], **SPG11** [AR], **VAPB** [AD], *C9orf72*), frontotemporal dementia (**OPTN** [AD?], **VCP** [AD], **SQSTM1/p62** [AD], **TBK1** [AD], **UBQLN2** [XLD], **CHMP2B** [AD], **GRN** [AD], *C9orf72*), neuronal ceroid lipofuscinosis (**GRN** [AD]), fulminant neurodegeneration (**ATP6AP2** [XLR]), dementia with Lewy bodies (*C9orf72*)
Pediatric neurodevelopmental disorders	Spinocerebellar ataxia (**ATG5** [AR], **RUBCN** [AR]), cortical atrophy and epilepsy (**PIK3R4/VPS15** [AR]), childhood-onset neurodegeneration (**SQSTM1/p62** [AR]), BPAN (**WDR45/WIPI4** [XLD]), spastic quadriplegia and brain abnormalities (**WDR45B/WIPI3** [AR]), primary microcephaly (**WDFY3/ALFY** [AD]), hereditary spastic paraplegia (**SPG49/TECPR2** [AR], **SPG11** [AR], **SPG15/ZFYVE26** [AR], **ATP13A2** [AR]), ataxia with spasticity (**VPS13D** [AR]), Rett's syndrome (**WDR45/WIPI4** [XLD], **MECP2** [XLD]), Joubert's syndrome (**INPP5E** [AR]), leukoencephalopathy (**VPS11** [AR]), adolescence-onset dystonia (**VPS16** [AR]), CEDNIK syndrome (**SNX14** [AR]), Pelizaeus–Merzbacher–like disorder (**SNAP29** [AR]), West's syndrome (**WDR45/WIPI4** [XLD], **SNAP29** [AR])
Hereditary neuropathies	Sensory and autonomic neuropathy type II (**FAM134B** [AR]), Charcot–Marie–Tooth disease (**RAB7A** [AD], **LRSAM1** [AD,AR], **VCP** [AD], **SPG11** [AR], **HSPB8** [AD]), sensory and autonomic neuropathy type IF (**ATL3** [AD]), distal hereditary motor neuronopathy (**HSPB8** [AD])
Ophthalmologic diseases	Primary open-angle glaucoma (**OPTN1** [AD]), cataracts (**CHMP4B** [AD])
Cardiac and skeletal myopathies	Danon's cardiomyopathy (**LAMP2** [XLD]), distal myopathy with rimmed vacuole (**SQSTM1/p62** [AD]), dilated cardiomyopathy (**PLEKHM2** [AR]), sporadic inclusion-body myositis (**VCP** [AD]), X-linked myopathy with excessive autophagy (**VMA21** [XLR])
Inflammatory disorders	Crohn's disease (*ATG16L1*, *ULK1*, *CALCOCO/NDP52*, *IRGM*, *LRRK2*, *ATG9A*), ulcerative colitis (*LRRK2*, *ATG9A*, *MTMR3*, *SMURF1*, *GPR65*), childhood asthma (*ATG5*)
Autoimmune diseases	Systemic lupus erythematosus (*ATG16L2*, *ATG5*, *DRAM1*, *CLEC16A*), diabetes (*CLEC16A*), other autoimmune diseases (*CLEC16A*)
Infectious diseases	*Mycobacterium tuberculosis* (*IRGM*, *LRRK2*), *M. leprae* (*PRKN/PARK2*, *LRRK2*)
Skeletal disorders	Osteopetrosis (**TCIRG1/ATP6V0A3** [AR], **PLEKHM1** [AD,AR]), Paget's disease of the bone (**SQSTM1/p62** [AD], **VCP** [AD], *OPTN*), Kashin–Beck disease (*ATG4C*)
Congenital multisystem disorders	Global developmental abnormalities (**WIPI2** [AR]), Vici's syndrome (**EPG5** [AR]), Zellweger syndrome (**PEX13** [AR]), glycosylation disorder with autophagy defects (**ATP6AP2** [XLR]), Zimmerman–Laband syndrome (**ATP6V1B2** [AD]), Hermansky–Pudlak syndrome (**VPS33A** [AR]), multisystem proteinopathy (**VCP** [AD], **SQSTM1/p62** [AD])
Cancer (frequently mutated genes)	Breast and ovarian cancer (somatic: *BECN1*, *RB1CC1*, *PRKN/PARK2*, Fanconi anemia pathway genes, *FAM134B*, *EI24*), colorectal cancer (somatic: *ULK1*, *ULK2*, *UVRAG*, *PRKN/PARK2*, *FAM134B EI24*), HBV-related hepatocellular carcinoma (germline: *ATG5*), other solid tumors (somatic: *PRKN/PARK2*, Fanconi anemia pathway genes, *FAM134B*, *EI24*), hematopoietic cancers (germline: *ATG2B*; somatic: Fanconi anemia pathway genes)

* Boldface type indicates causative mutations in mendelian diseases; regular type indicates risk variants or predisposing mutations (identified by genomewide association studies or large-scale analyses). AD denotes autosomal dominant, AR autosomal recessive, BPAN beta-propeller protein–associated neurodegeneration, CEDNIK cerebral dysgenesis, neuropathy, ichthyosis, and palmoplantar keratoma, HBV hepatitis B virus, XLD X-linked dominant, and XLR X-linked recessive. A reference list for the information in this table and in Table S2 is provided in the Supplementary Appendix.

Mizushima, N. and B. Levine, *Autophagy in Human Diseases.* New England Journal of Medicine, 2020. **383**(16): p. 1564-1576.

Complex roles of autophagy in cancer development and progression

Autophagy is an important process during cancer progression, but the exact roles of autophagy in cancer cells are strongly context-dependent.

Its cytoprotective function is believed to have tumour-suppressive potential before the onset of tumorigenesis, and loss of autophagy has been associated with increased risk of cancer.

autophagy provides cancer cells with metabolic plasticity, allowing them to thrive in suboptimal environments and to exploit the pro-survival activity of autophagy to cope with therapy-induced stresses

autophagy induction is a side effect of many cancer therapies, and thus, pharmacological inhibition of autophagy has been proposed as a valid strategy to enhance the efficacy of therapies and to avoid resistance to treatment in certain cancers

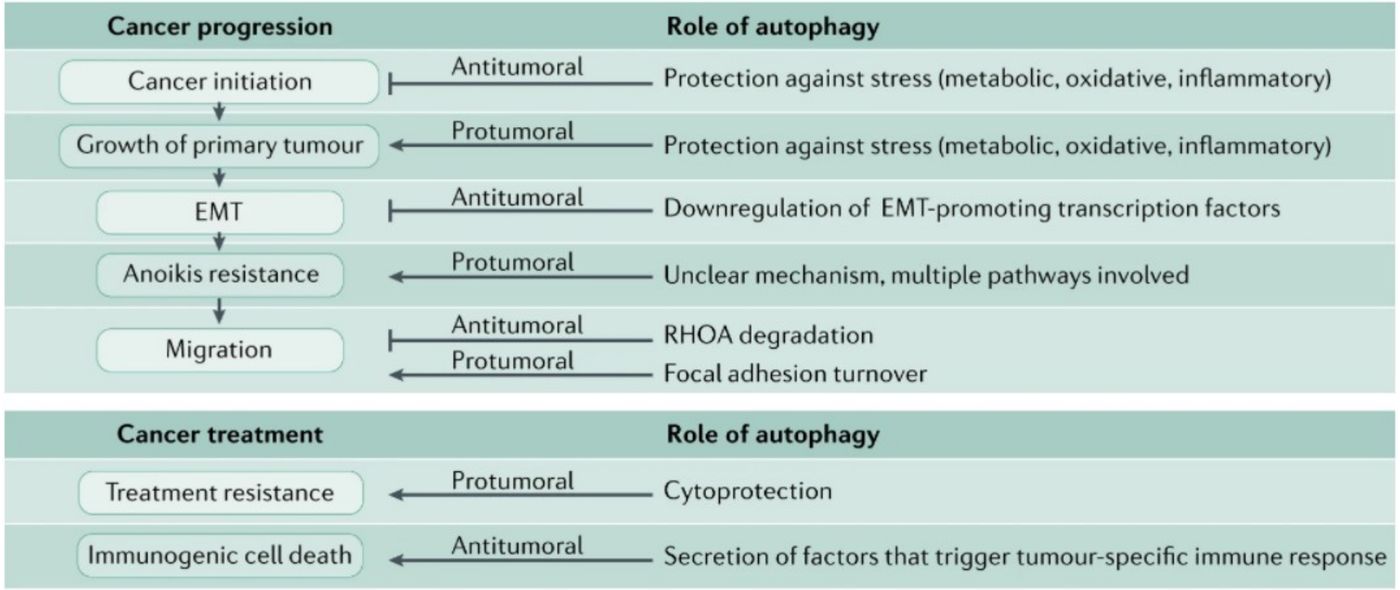

Table 3. Roles of Autophagy in Tumors.*

Role	Cell-Autonomous Effects	Cell-Nonautonomous Effects
Antitumorigenic	Increased chromosome or genome stability Decreased metabolic stress Decreased oxidative stress (e.g., through mitophagy) Decreased NRF2 activity (through p62 degradation) Increased cellular senescence Increased anticancer immunogenicity Decreased metastasis	Decreased cell death–induced inflammation Increased anticancer immunity
Protumorigenic	Increased metabolic, energy, and redox homeostasis Decreased p53 Decreased surface MHCI Granzyme degradation Decreased recruitment of NK cells Decreased endoplasmic reticulum stress Increased metastatic dormancy	Increased nutrient supply from nontumor cells in the microenvironment Increased systemic arginine levels (decreased degradation by arginase) Decreased anticancer T-cell immunity

* A reference list for the information in this table and in Table S3 is provided in the Supplementary Appendix. MHCI denotes major histocompatibility complex class I, and NK natural killer.

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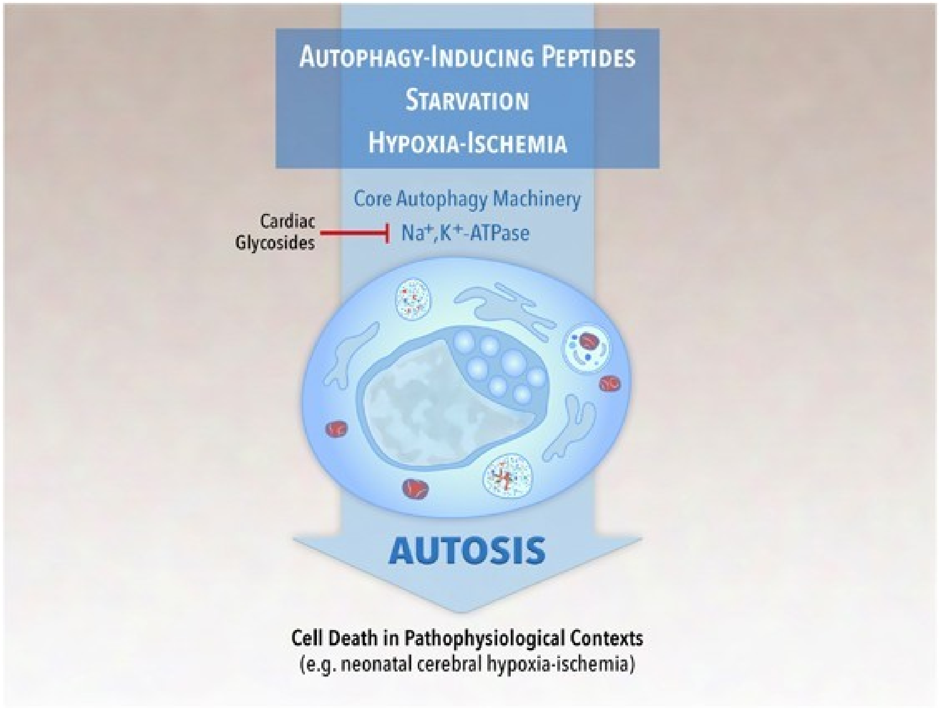
Autophagy as a cell death modality?

It is controversial whether cells truly die via autophagy or whether — in dying cells — autophagy is merely an innocent bystander or a well-intentioned 'Good Samaritan' trying to prevent inevitable cellular demise.

An aggressive form of autophagy termed autosis has been described in cells following either ischemia/reperfusion injury or in response to autophagy-inducing proteins like Tat-Beclin 1.

While it has not been demonstrated unambiguously that autophagy drives cell death during development (Kroemer and Levine, 2008), there is increasing evidence that ADCD is a bona fide cell death program. Nevertheless, there are still major gaps in our understanding of this mechanism in development and pathogenesis.

Genetic models where an absolute requirement for the autophagic machinery has been shown to be essential for cell death: *Dictyostelium discoideum, Drosofila melanogaster*



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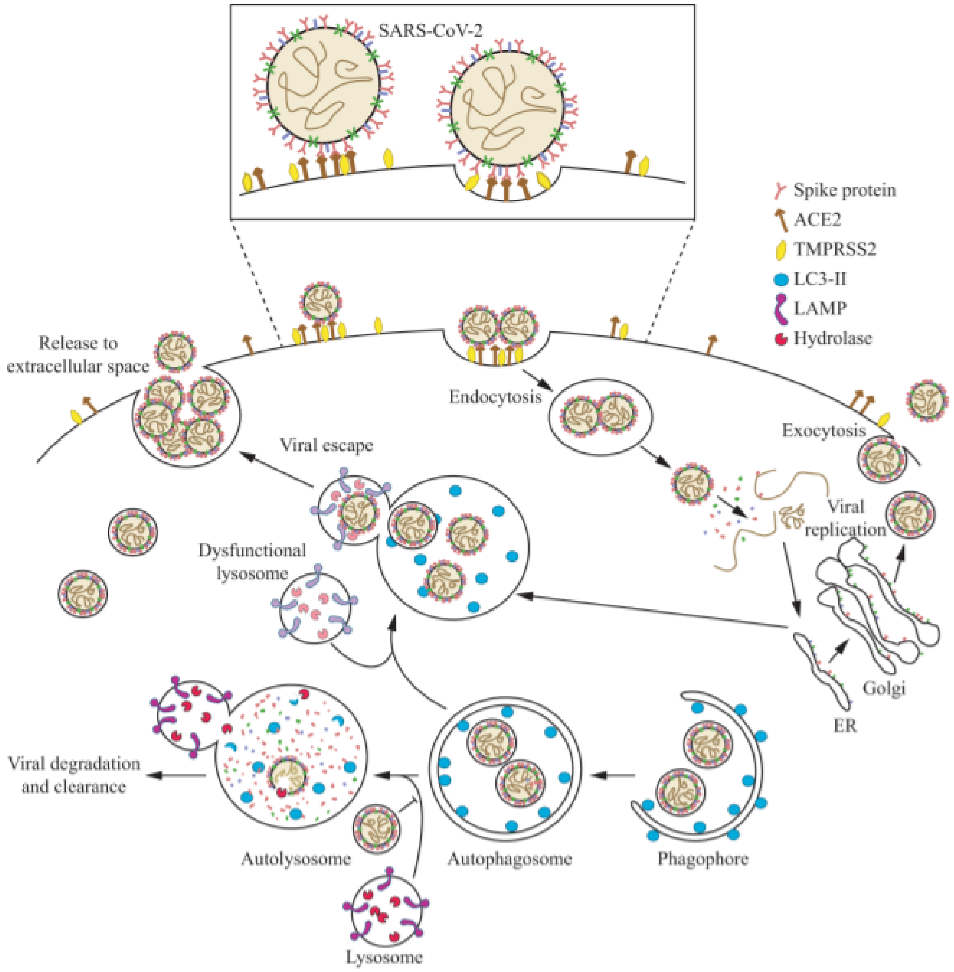
Coronavirus interactions with the cellular autophagy machinery

The viruses enter the host cell via endocytosis (they can do that also by different mechanisms) and release their RNA into the cytosol;
this is followed by replicative translation with the membrane proteins being made in the endoplasmic reticulum.

The newly formed viral particles may be released from the cell via exocytosis, or they may then intersect with autophagy.
In general, autophagy plays an “antiviral” role, sequestering viral structural proteins or even completely assembled viral particles within autophagosomes; these will bind with lysosomes leading to degradation of the cargo by lysosomal hydrolytic enzymes.

However, recent studies suggest that SARS-COV-2 disrupts and hijacks the autophagy-lysosomal pathway and subverts it.
For example, the viral ORF3a protein may block autophagosome-lysosome fusion.
In addition, viral proteins may be delivered to a de-acidified lysosome from which they can be released from the cell. This “proviral” role of the disrupted autophagy process leads to extensive production and release of the virus to the extracellular space causing the infection to spread to non-infected cells.



Nastaran Samimi, Mojtaba Farjam, Daniel J. Klionsky & Nima Rezaei (2021) The role of autophagy in the pathogenesis of SARS-CoV-2 infection in different cell types, Autophagy, DOI: [10.1080/15548627.2021.1989150](https://doi.org/10.1080/15548627.2021.1989150)

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Thank you for your attention!