“IL LISOSOMA: UN REOSTATO DEL METABOLISMO CELLULARE”

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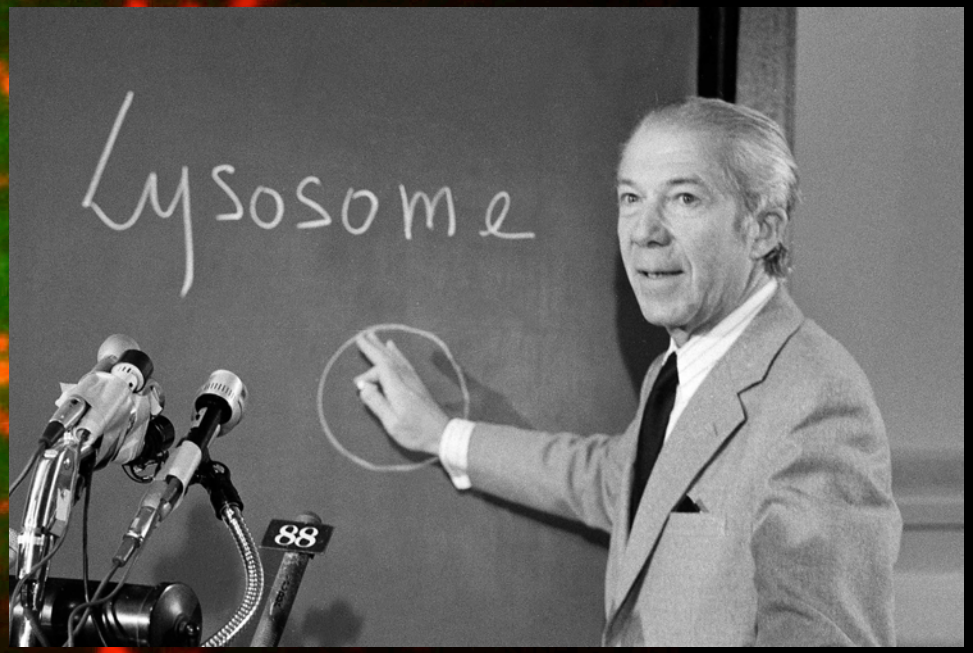
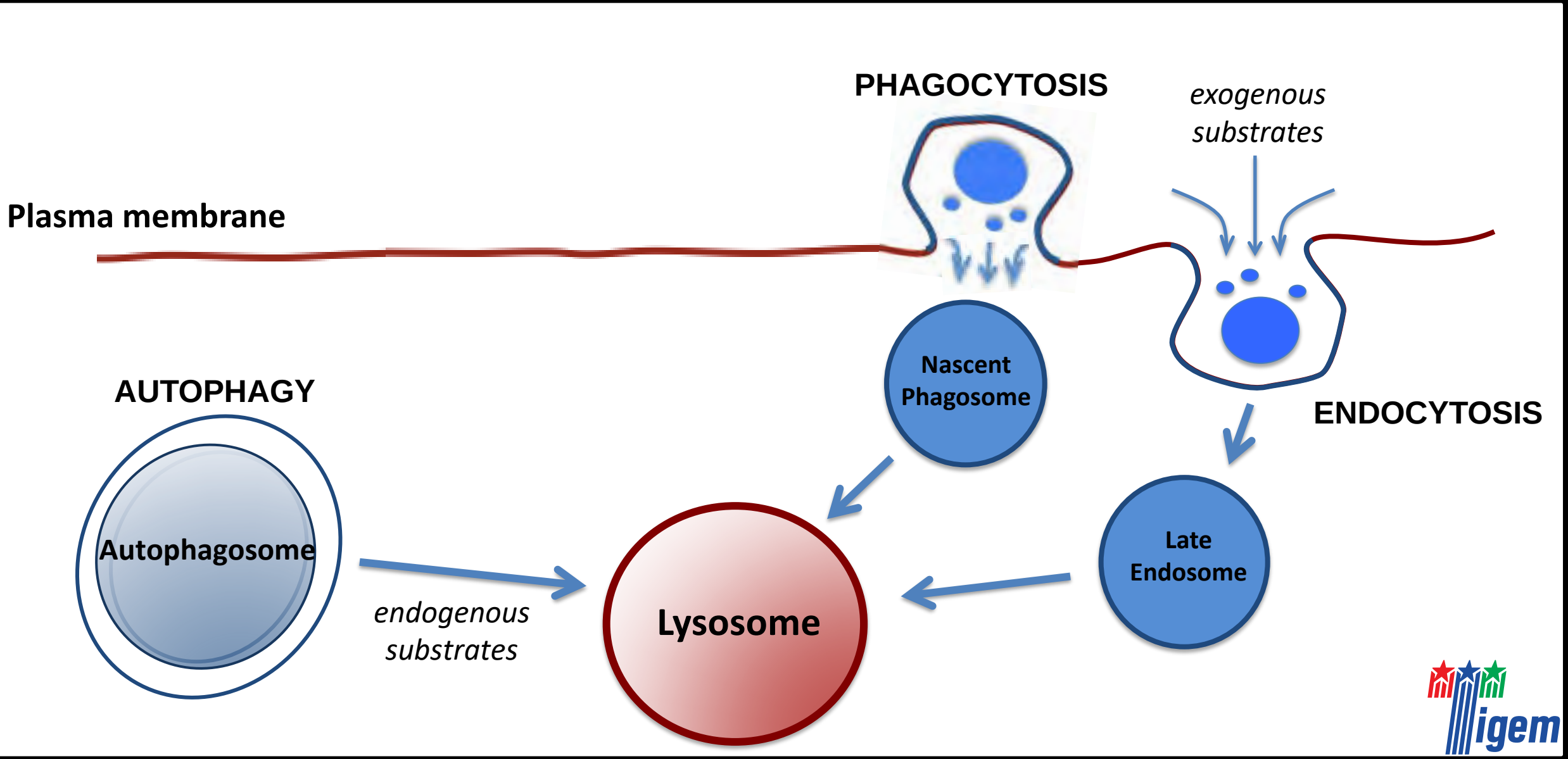
Accademia dei Lincei,
Roma, April 22nd, 2022

LAMP1
TFEB

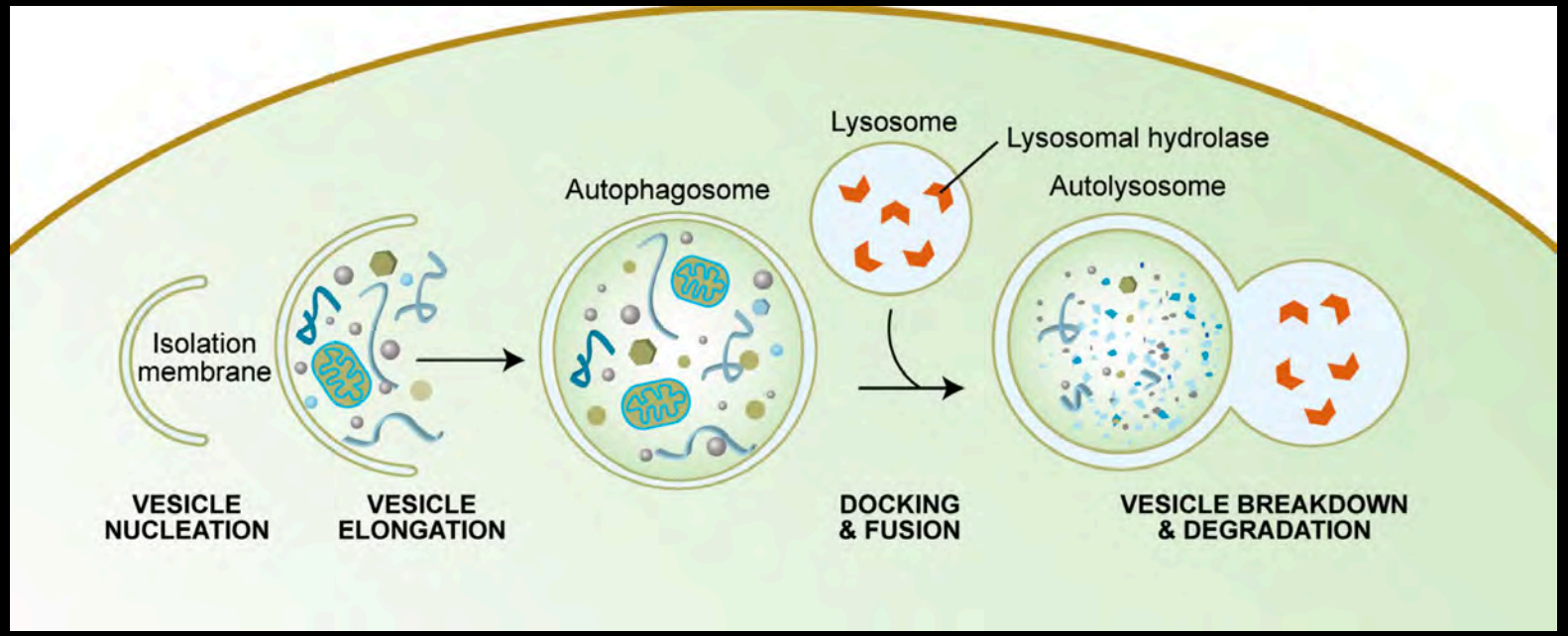


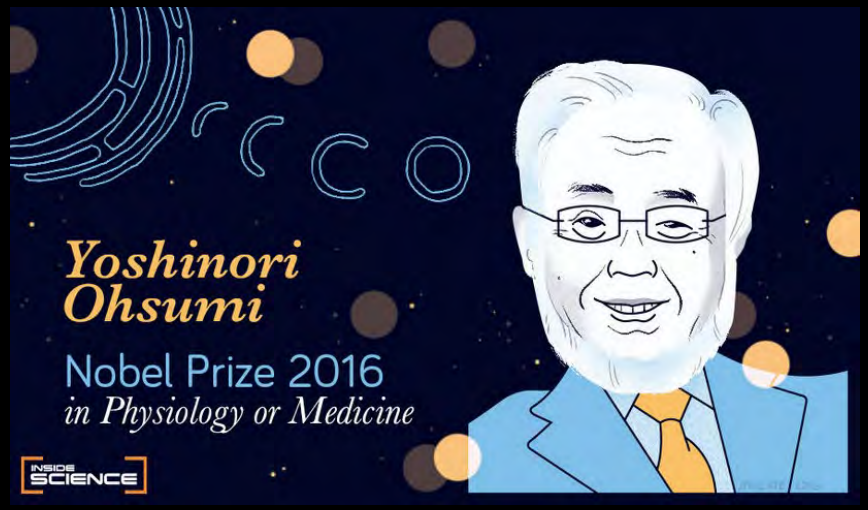
Christian de Duve
1974 Nobel Prize
in Physiology or Medicine

The lysosome in degradation and recycling processes



The autophagy pathway





Traditional view of the lysosome

- the “end” of cellular catabolic pathways (the one who does the dirty job....)
- a static organelle: not subject to regulation, not influenced by environmental cues

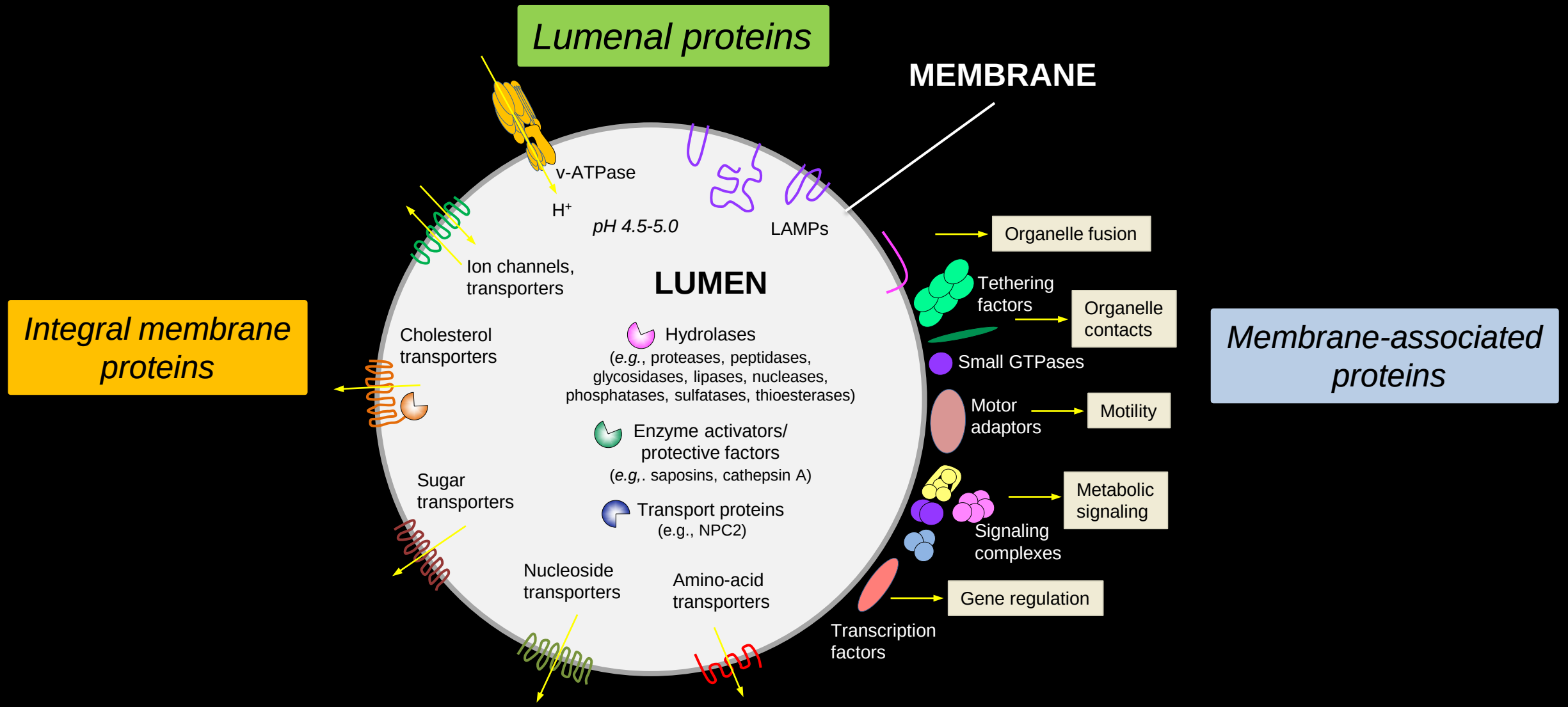
OUR HYPOTHESIS

The lysosome is a dynamic structure, able to adapt to environmental cues.

Lysosomal function is subject to a global control.

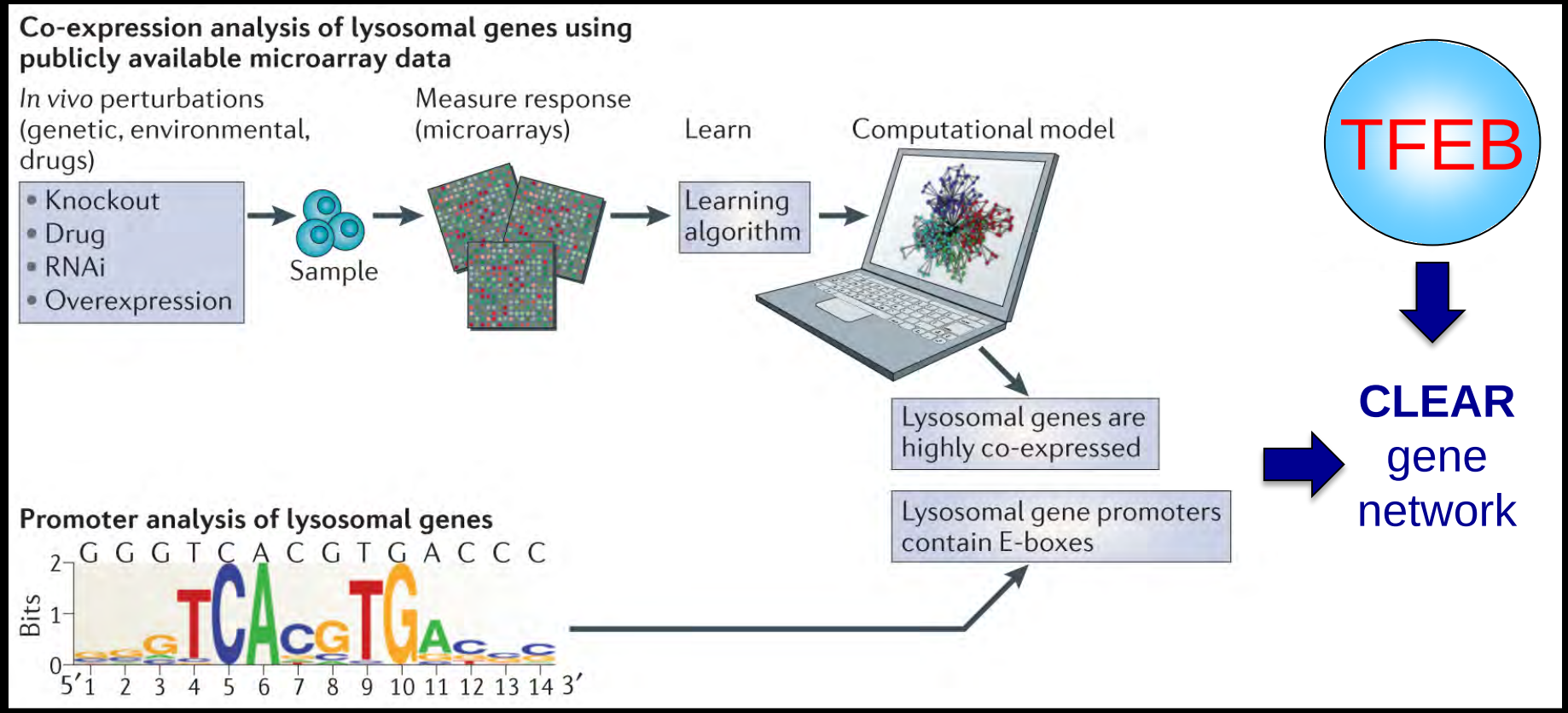
How does the cell control the function of an entire organelle?

LYSOSOMAL PROTEINS



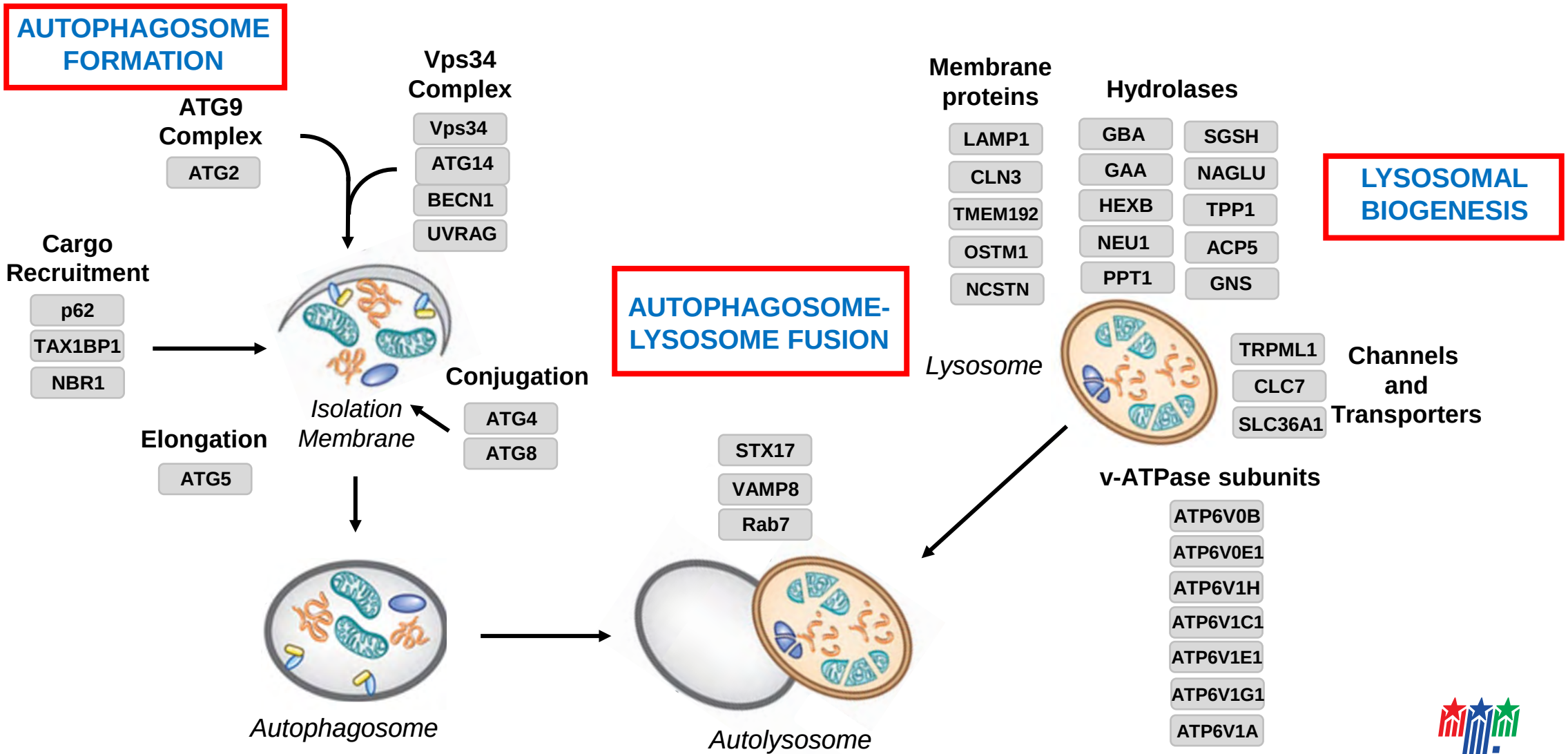
Ballabio and Bonifacino, Nature Reviews Mol Cell Biol 21, 101–118 (2020)

A Gene Network Regulating Lysosomal Biogenesis and Function

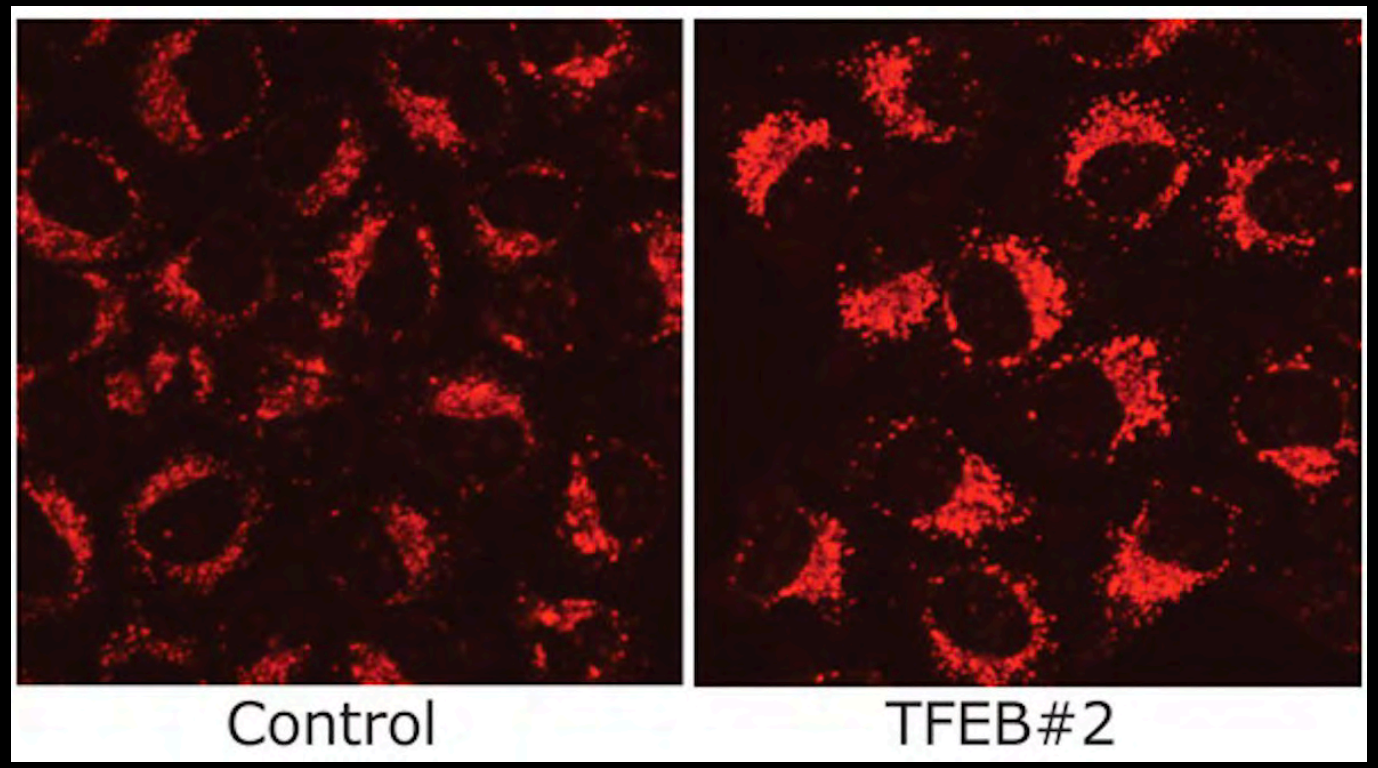


Sardiello et al. Science 325: 473-476, 2009

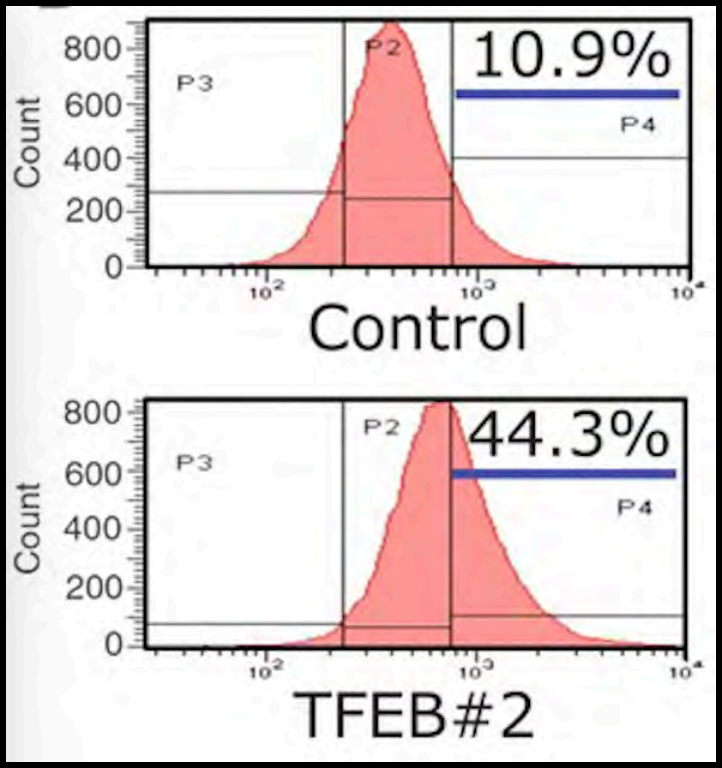
TFEB controls the lysosomal-autophagic pathway



TFEB OVEREXPRESSION INDUCES LYSOSOMAL BIOGENESIS



LAMP1 immunofluorescence



Lysotracker staining (FACS)

Science 325: 473-476, 2009

Transcription Factor EB
A Master Regulator of Lysosomal Biogenesis and Autophagy

Lysosomal biogenesis

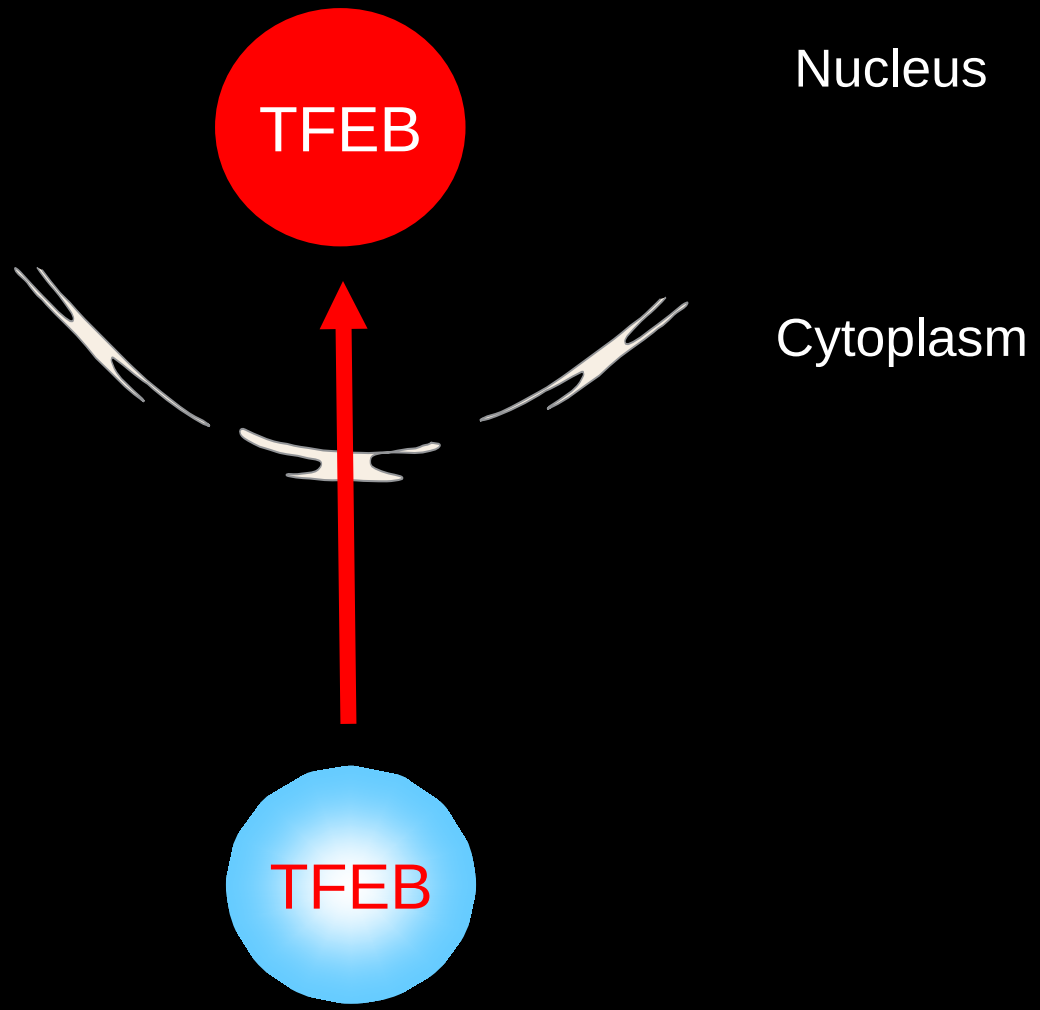
Autophagy

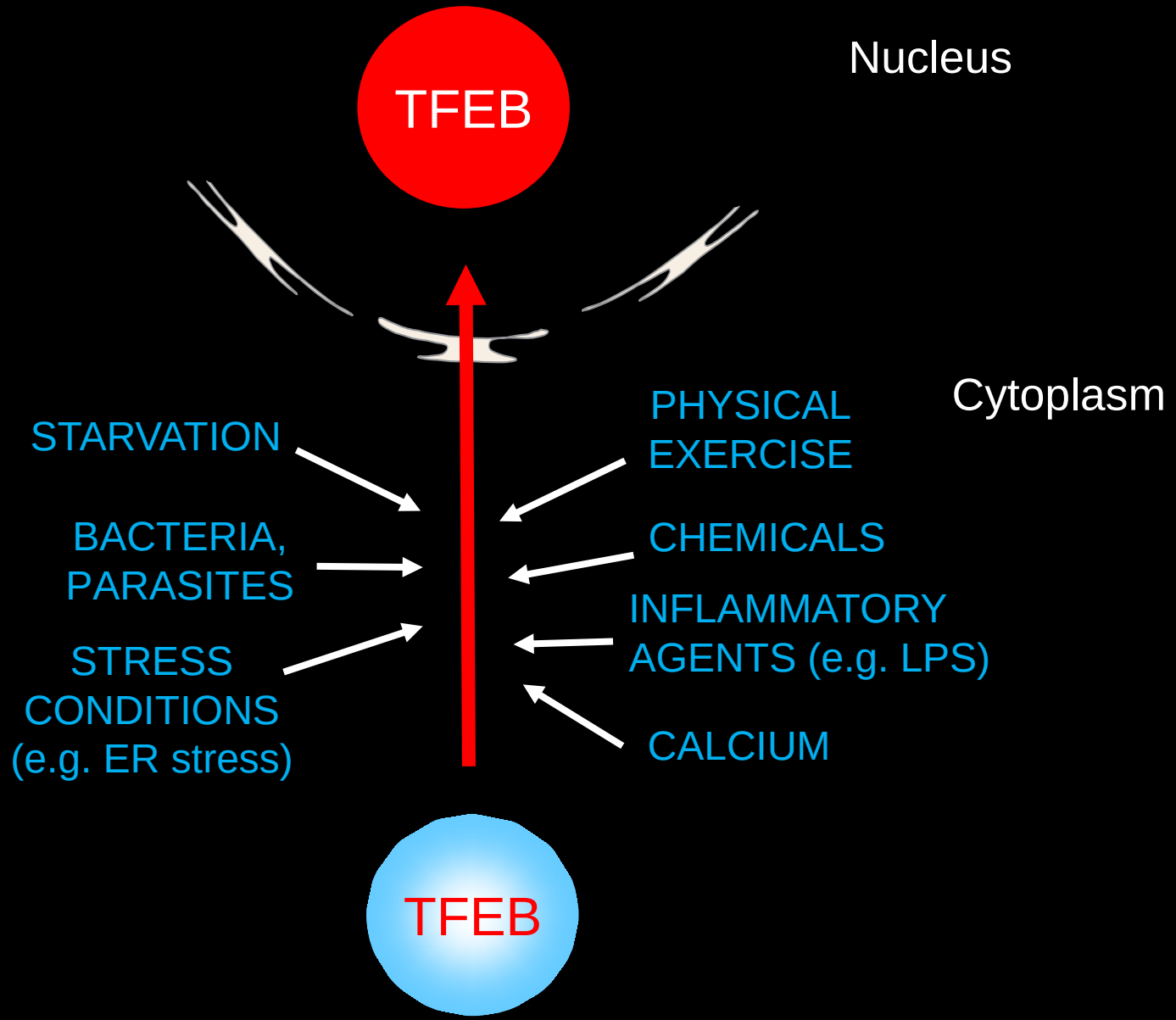
Sardiello et al. ***Science*** 325: 473-476, 2009

Settembre et al., ***Science*** *332: 1429-1433, 2011*

Who regulates TFEB?

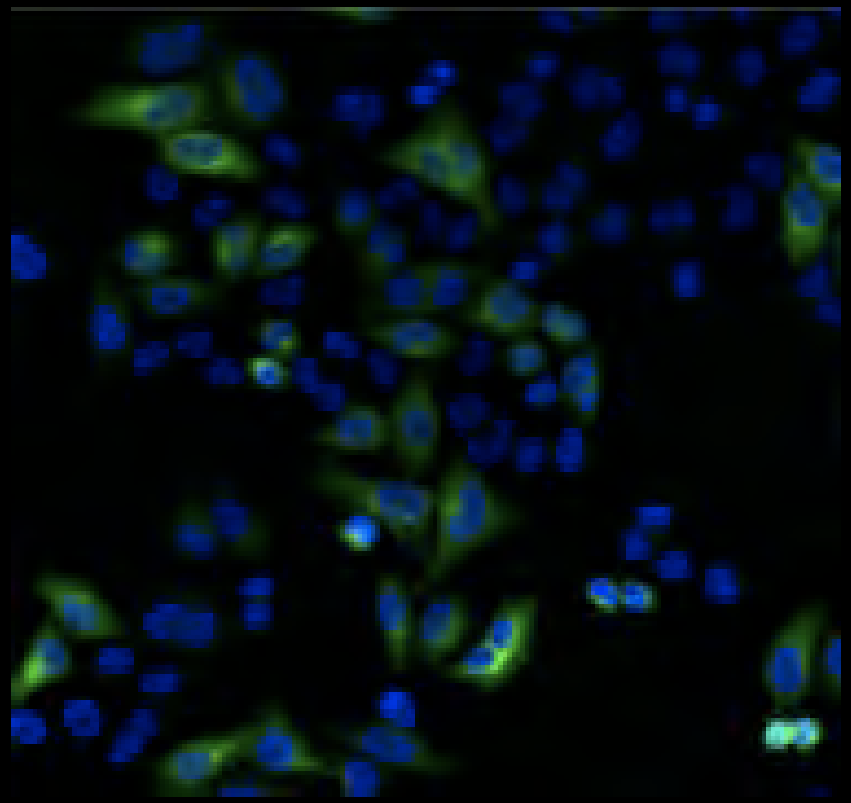
TFEB ACTIVITY IS REGULATED BY ITS NUCLEO-CYTOPLASMIC SHUTTLING



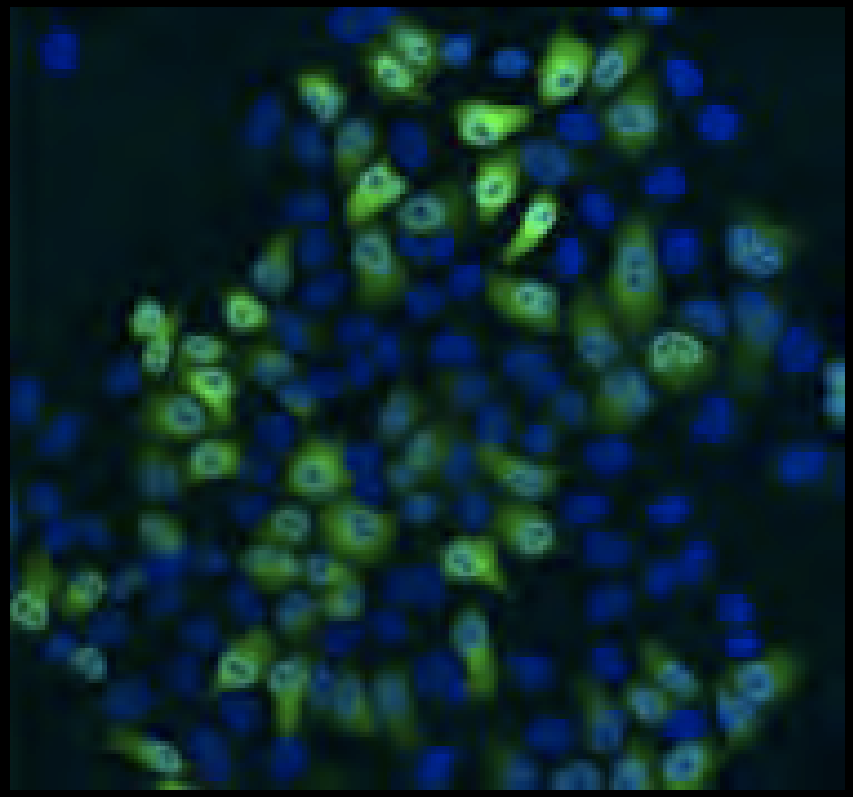

TFEB
Nucleus
Cytoplasm
STARVATION
BACTERIA,
PARASITES
STRESS
CONDITIONS
(e.g. ER stress)
PHYSICAL
EXERCISE
CHEMICALS
INFLAMMATORY
AGENTS (e.g. LPS)
CALCIUM
TFEB

STARVATION INDUCES TFEB NUCLEAR TRANSLOCATION

HeLa TFEB-3XFLAG transfectants



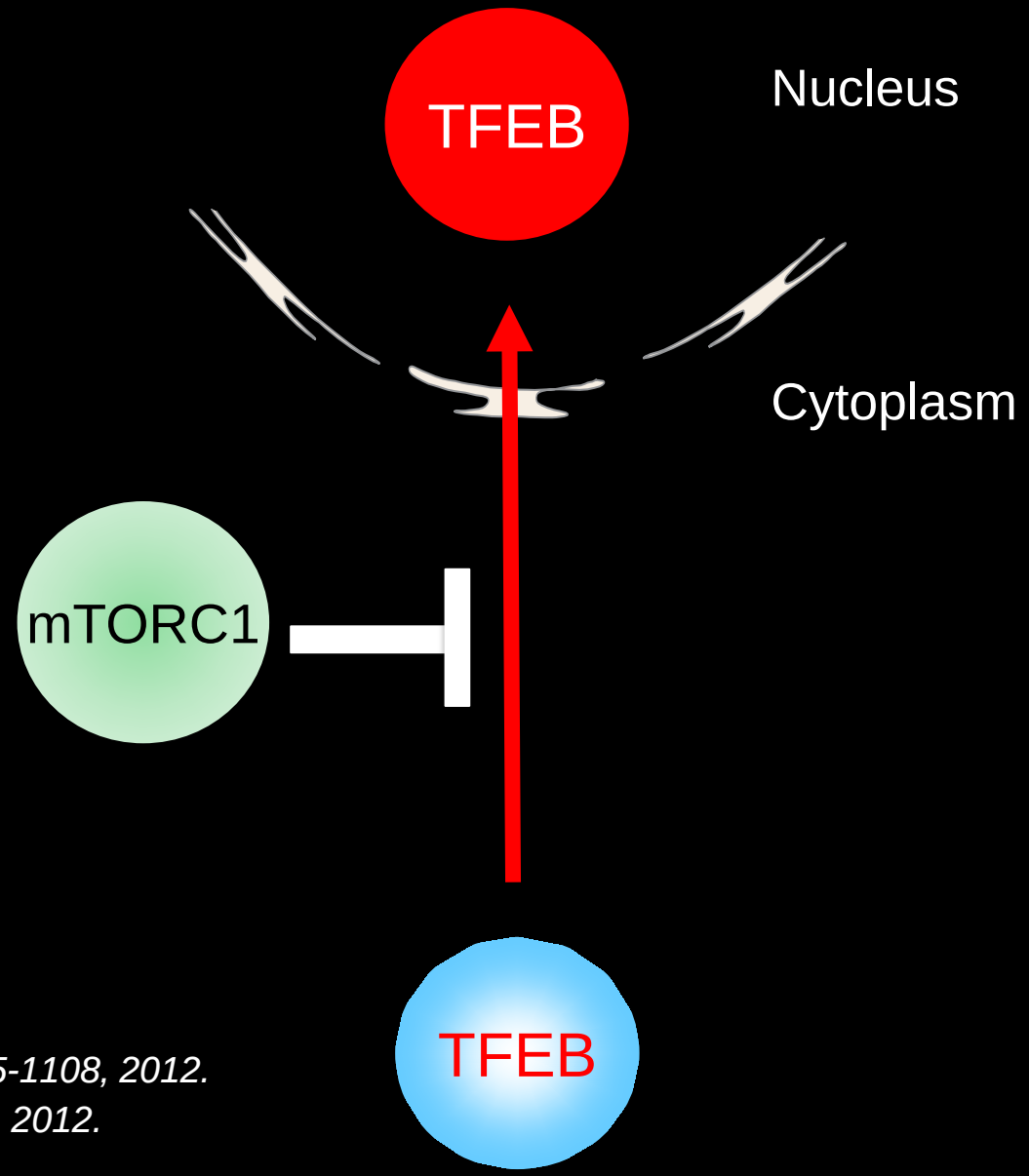
Standard medium



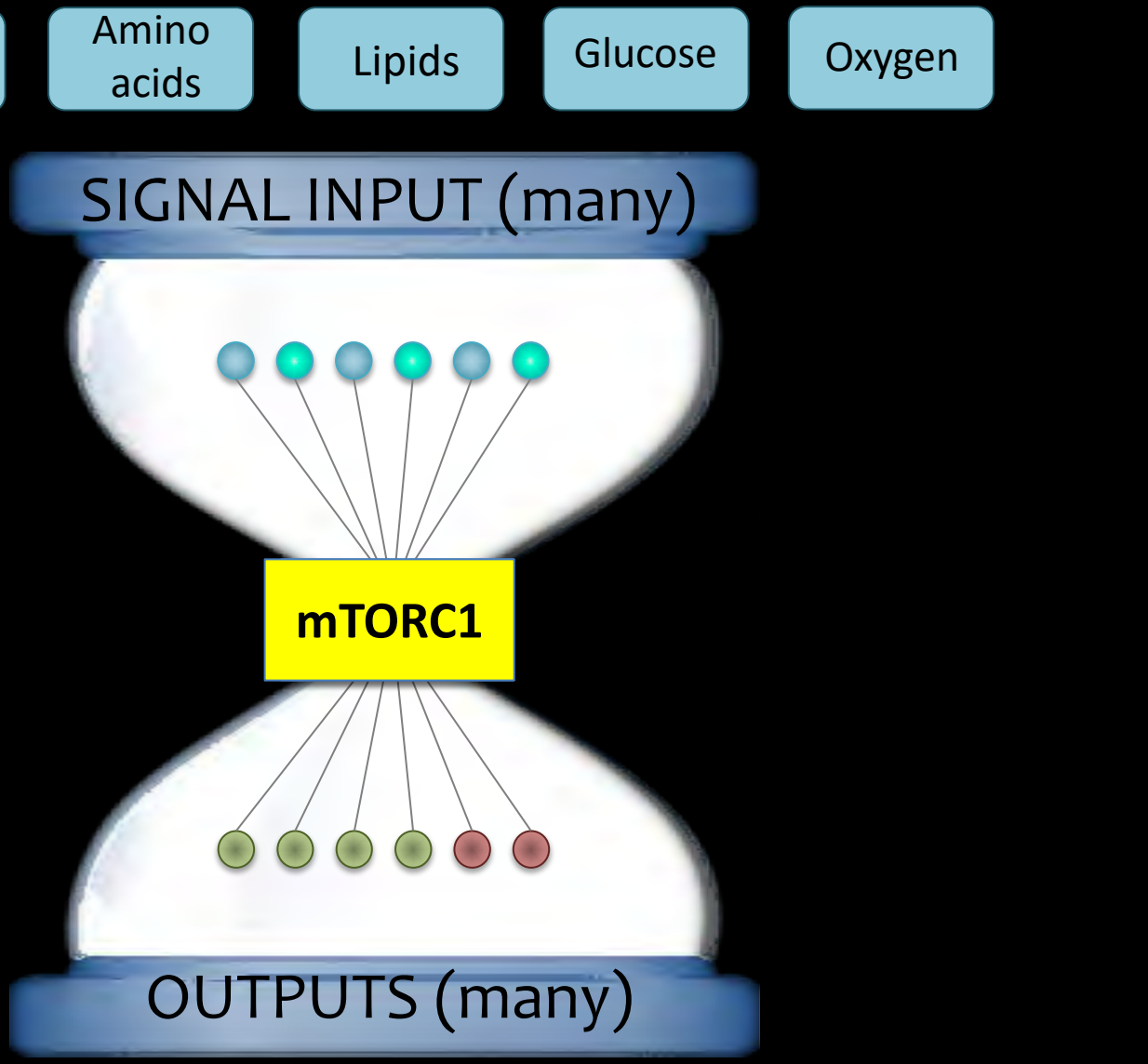
HBSS

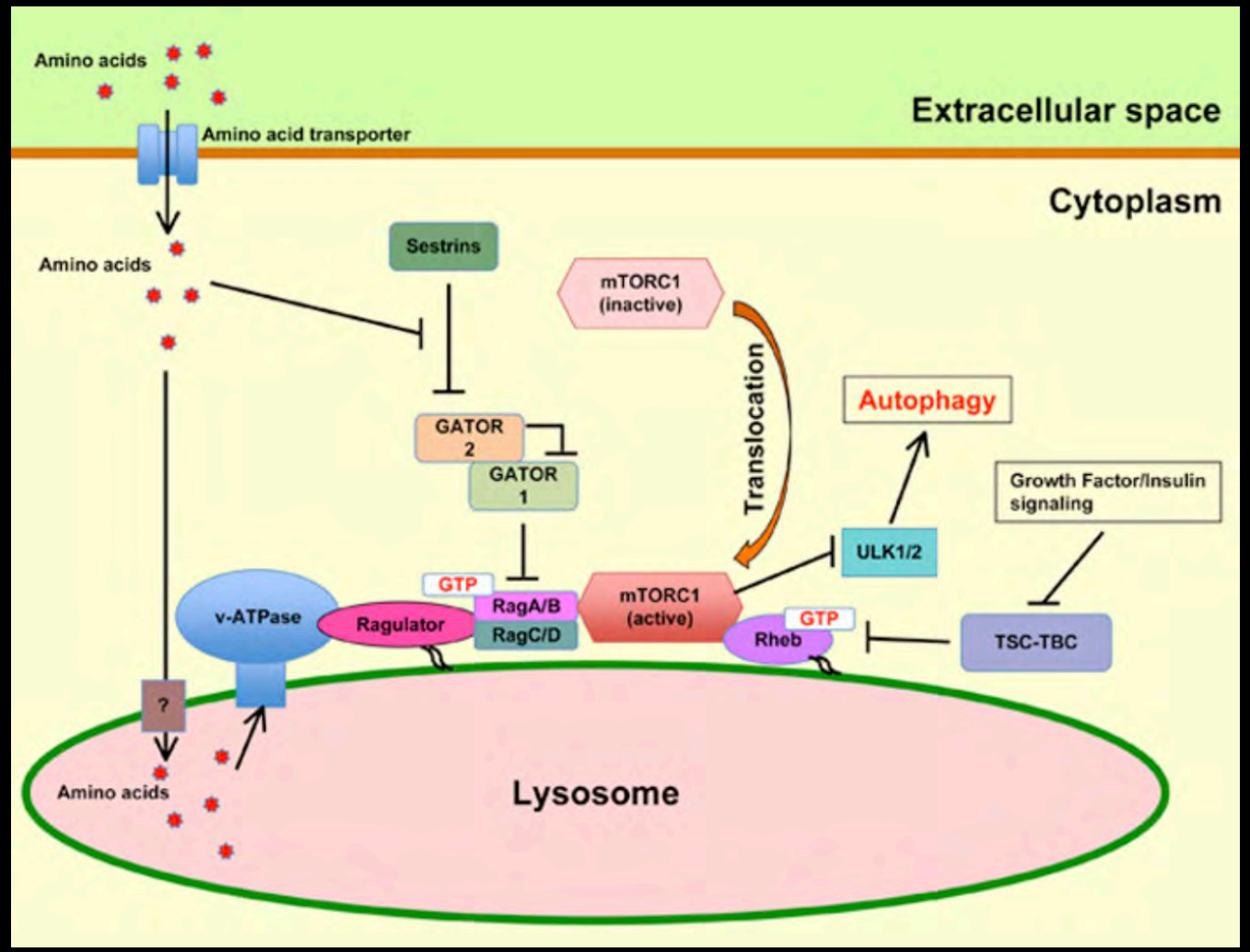
Settembre et al., Science 332: 1429-1433, 2011

mTORC1 phosphorylates TFEB and inhibits its nuclear translocation



Settembre, C., Zoncu, R., et al. EMBO J. 31, 1095-1108, 2012.
Roczniak-Ferguson, A., et al. Sci. Signal. 5, ra42, 2012.
Martina, J. A., et al. Autophagy 8, 903-914, 2012.

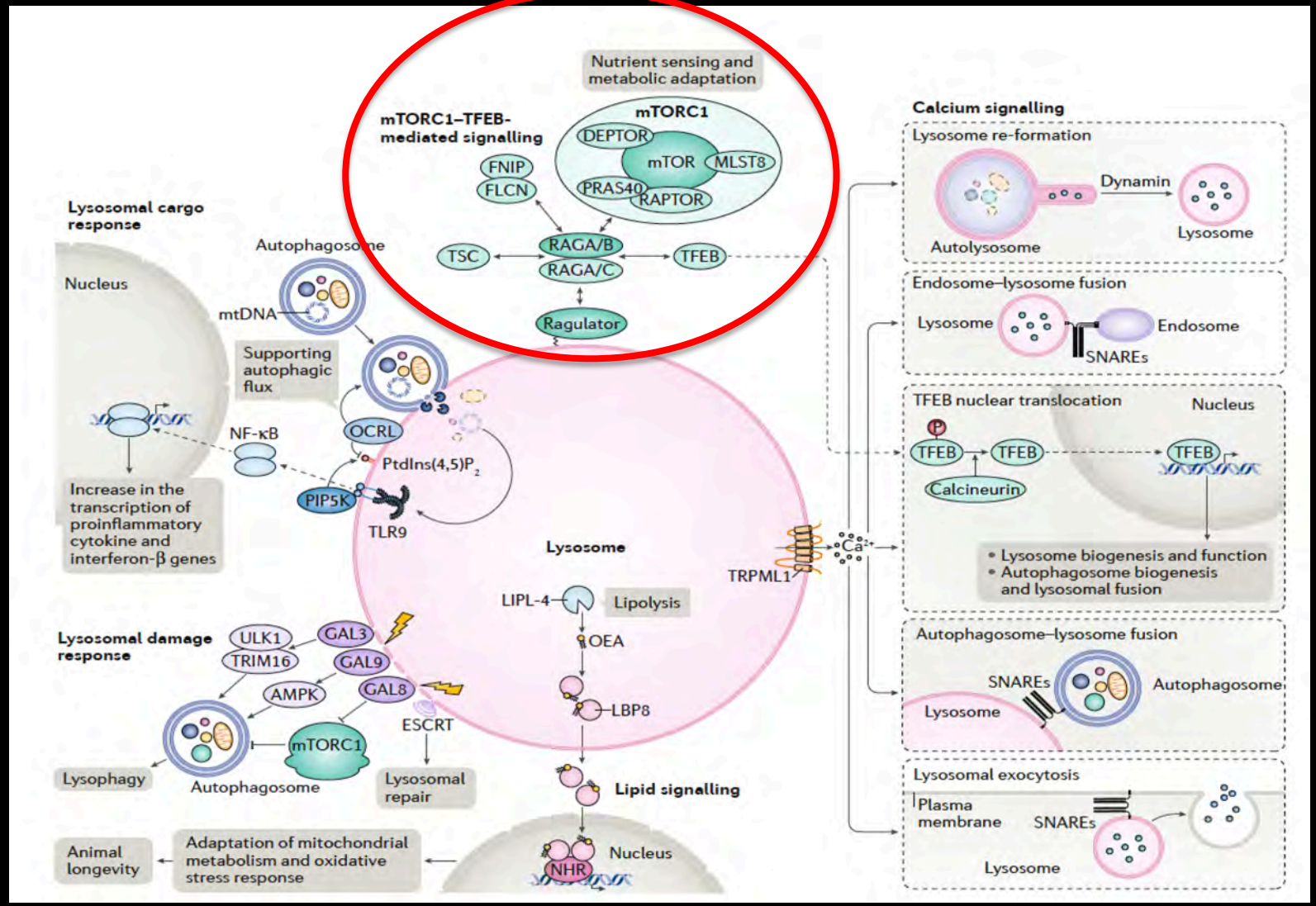

Growth factors
Amino acids
Lipids
Glucose
Oxygen
SIGNAL INPUT (many)
mTORC1
OUTPUTS (many)
Protein synthesis
S6K - 4E-BP1
Nucleotide synthesis
S6K – ATF4
Lipid synthesis
Lipin1 – SREBPs
Aerobic glycolysis
HIF1α
Autophagy
TFEB and ULK1
Lysosome biogenesis
TFEB



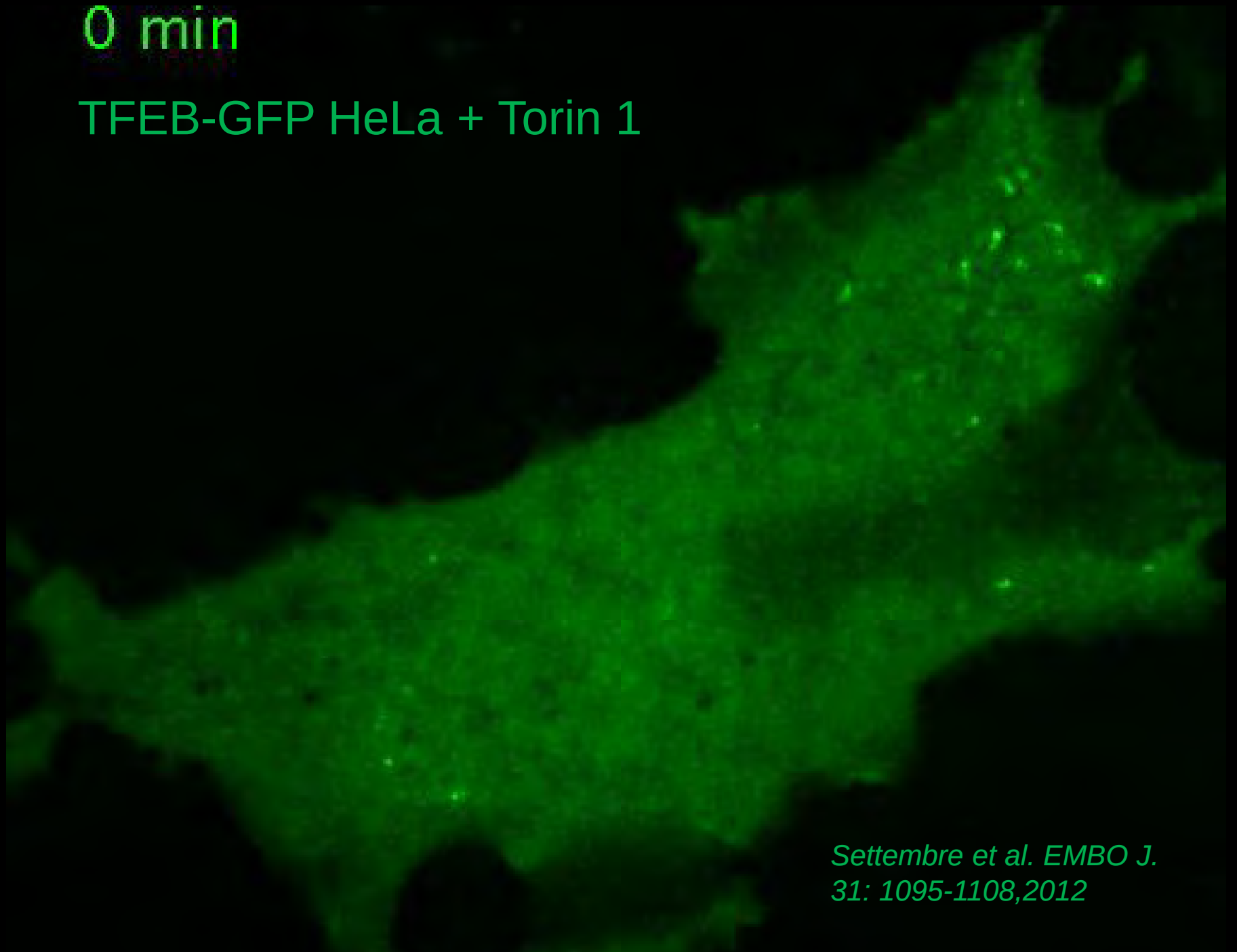
mTORC1 is activated on the lysosome and is part of the lysosomal nutrient sensing machinery

D. Sabatini's group
Cell 141, 290–303, 2010

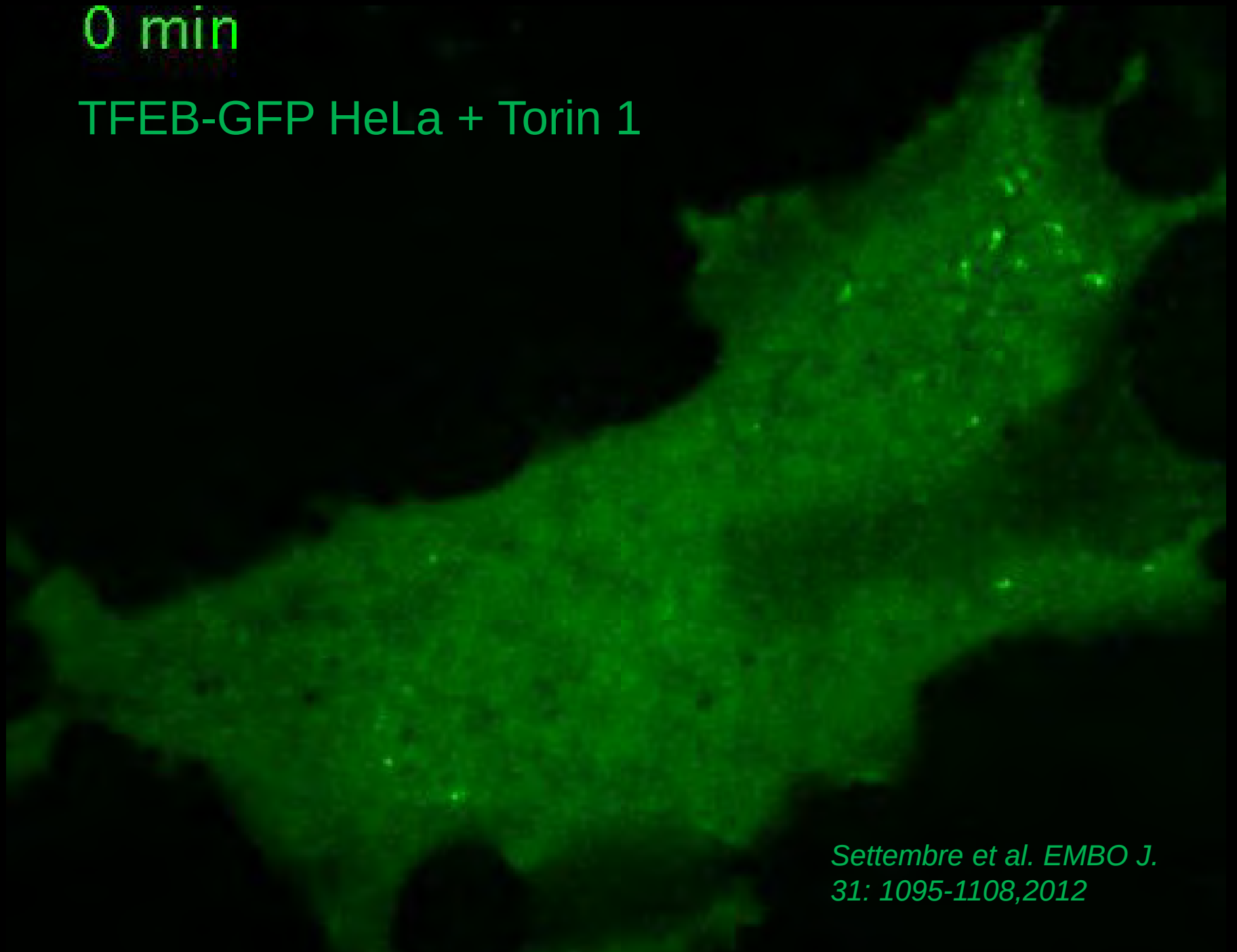
Lysosome signaling



Ballabio and Bonifacino, Nat Rev Mol Cell Biol 21, 101–118 (2020)



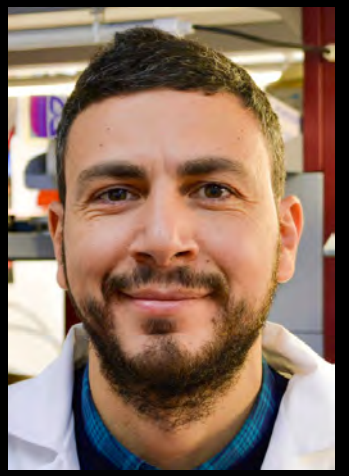
Settembre et al. EMBO J. 31: 1095-1108,2012



Settembre et al. EMBO J. 31: 1095-1108,2012

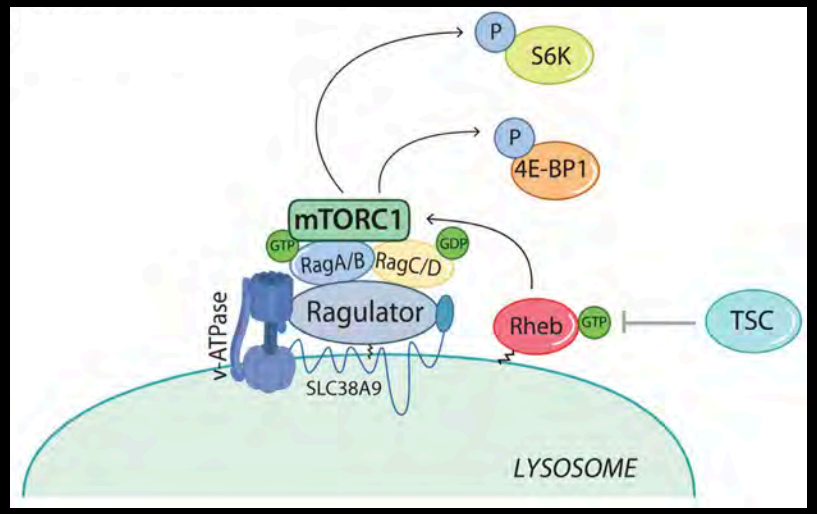
A Substrate-Specific mTORC1 Pathway

Nature, 585: 597-602, 2020

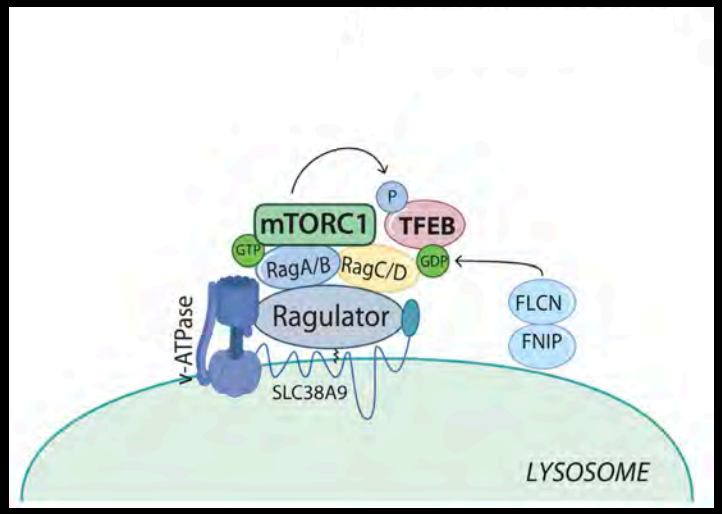


Gennaro Napolitano

canonical mTORC1



non-canonical mTORC1



TFEB IN HUMAN DISEASES

Intracellular Clearance

Accellerated metabolism

Therapeutic effect: on mouse models of diseases associated with accumulation of undegraded substrates (e. g Lysosomal Storage Diseases, common neurodegenerative diseases etc...).

Cancer: Pancreatic cancer, breast cancer, renal cell carcinoma, inherited cancer syndromes (e.g. Birth-Hogg-Dube' syndrome and Tuberous Sclerosis.

OLD

Lysosome

NEW

Luisa Lanfrancone
Giovanni Bertalot
IEO, Milan, Italy

Lisa Henske
Nicola Alesi
Harvard Medical School, Boston MA

Lukas Huber
Mariana De Araujo
Taras Stasyk
Medical Univ. of Innsbruck, Austria

Jim Hurley
Zhicheng Cui
Berkeley Univ., CA

Marston Linehan
Laura Schmidt
Urologic Oncology branch
National Cancer Institute
NIH, Bethesda, USA

Leon Murphy
Jon Goodwin
CASMA Therapeutics, Boston MA

Gennaro Napolitano
Chiara Di Malta
Alessandra Esposito
Jlenia Monfregola
Diletta Siciliano
Maria Matarese
Angela Zampelli
Claudia Vilardo
Alessandro Venuta
Valerio Benedetti
Alessia Calcagni'
Marcella Cesana
Edoardo Nusco

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THE TELETHON INSTITUTE OF GENETICS AND MEDICINE

Quality research to understand the mechanisms behind rare genetic diseases

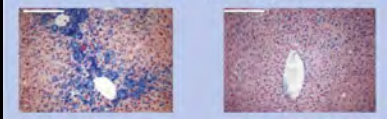
Screenshot

TFEB PROMOTES INTRACELLULAR CLEARANCE

Lysosomal storage diseases

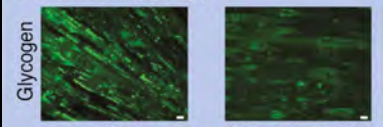
Multiple sulfatase deficiency

Clearance of glycosaminoglycans



Pompe disease

Clearance of glycogen



Batten disease

Clearance of lipofuscin

Cystinosis

Clearance of cystine

Mucopolysaccharidosis Type 3A

Clearance of glycosaminoglycans

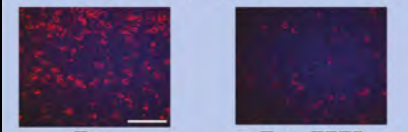
Neurodegenerative diseases

Parkinson's disease

Clearance of alpha-synuclein

Alzheimer's disease

Clearance of Tau proteins



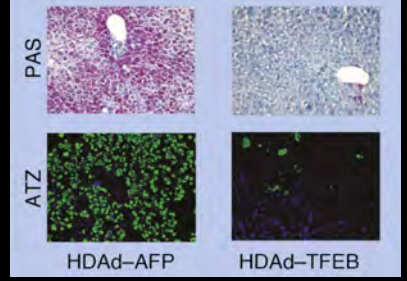
Huntington's disease

Clearance of mutant huntingtin

Other diseases

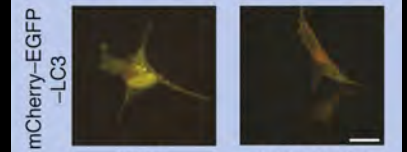
α-1-antitrypsin deficiency

Clearance of α-1-antitrypsin



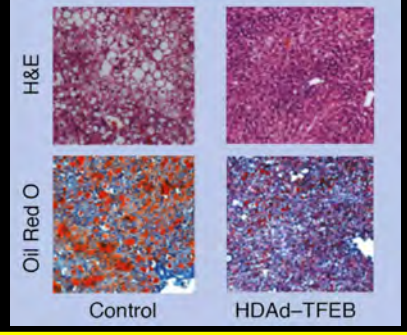
Spinal-bulbar muscular atrophy

Autophagy rescue

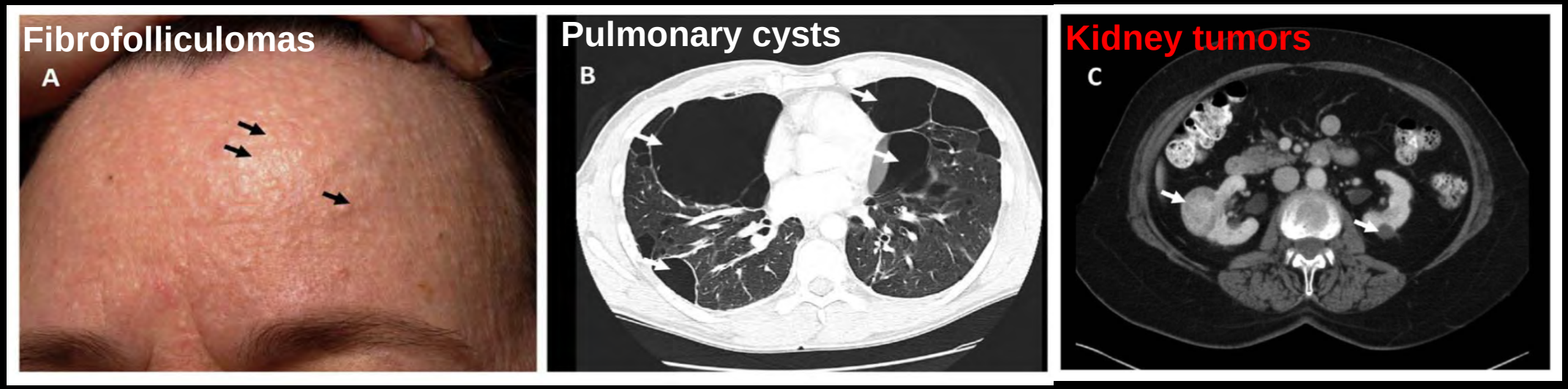


Diet-induced obesity

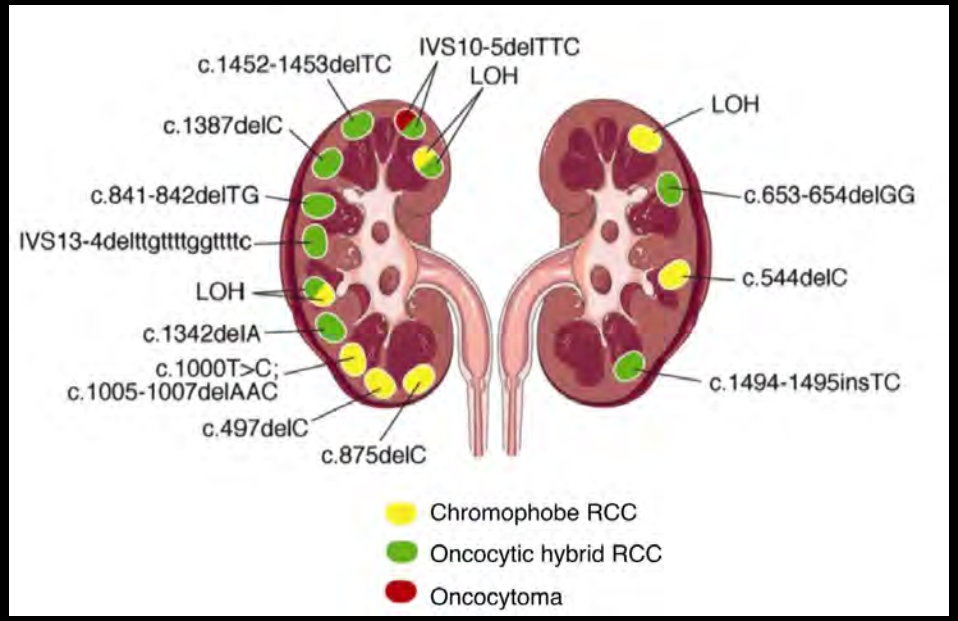
Clearance of lipid droplets

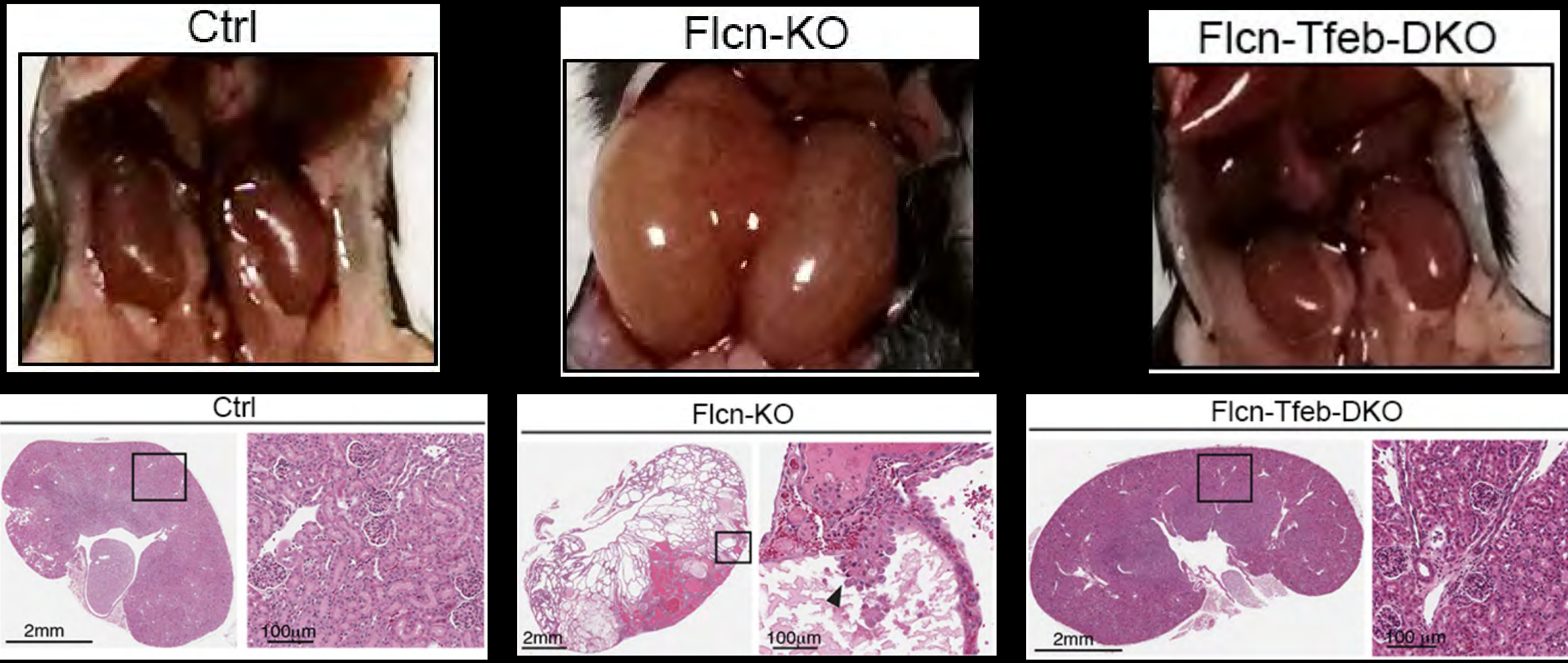


Birt-Hogg-Dubé (BHD) syndrome is due to germline mutations in *FLCN* gene



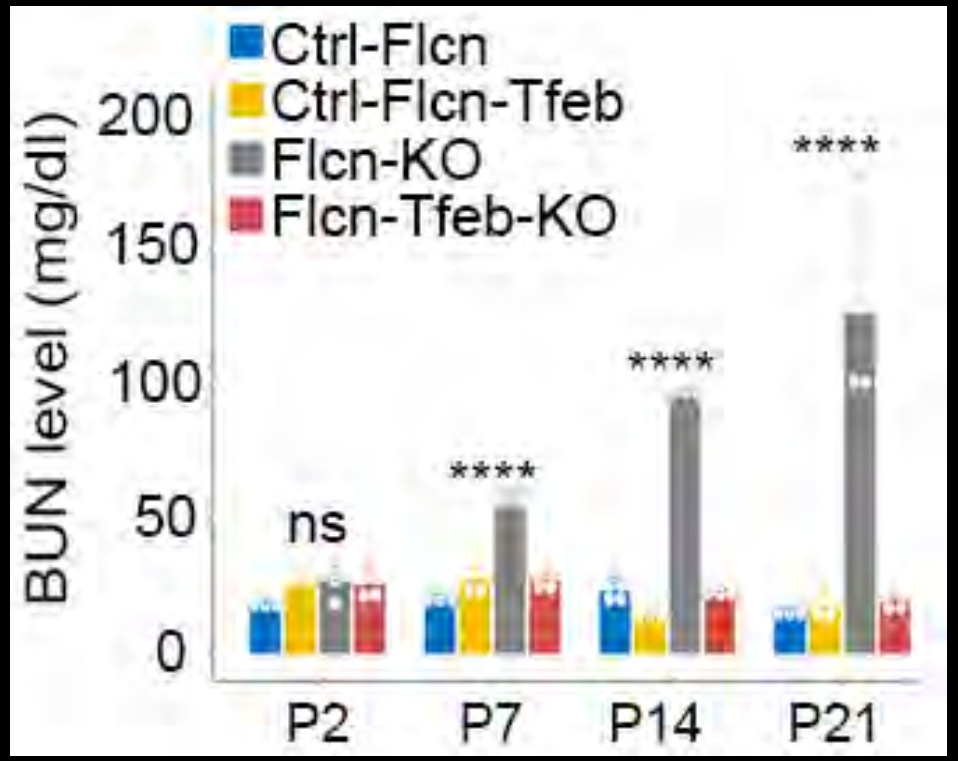
Schimdt et al.,
Gene 2018

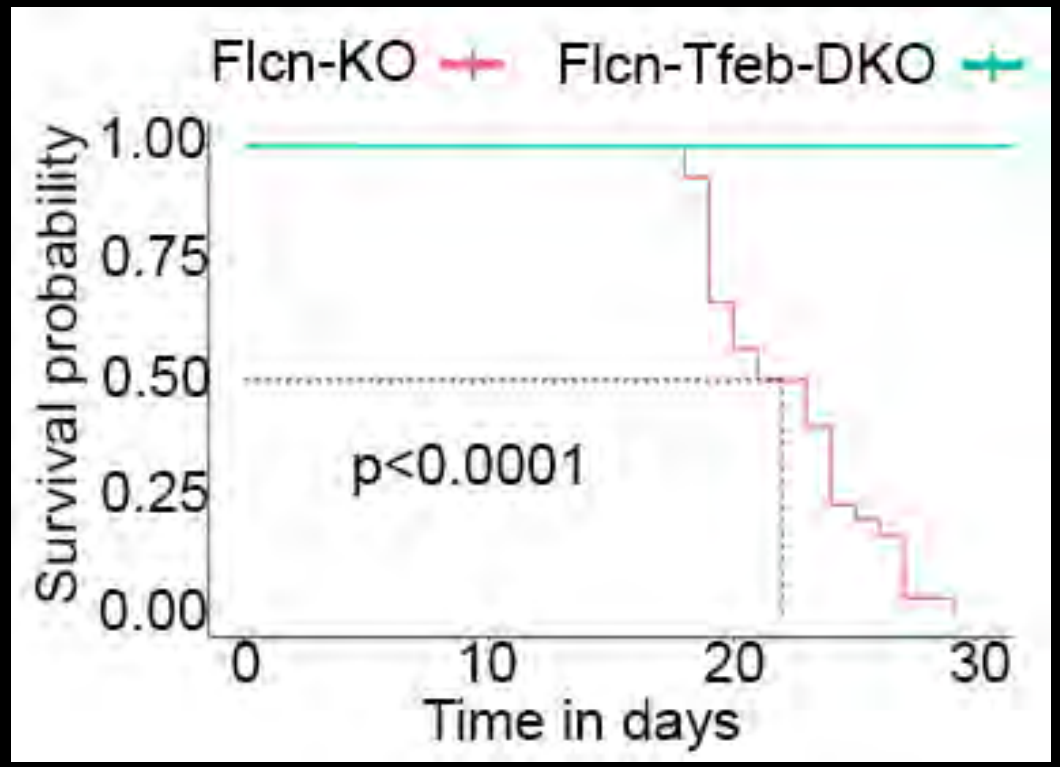




Napolitano, Di Malta* et al., Nature, 585: 597-602, 2020*

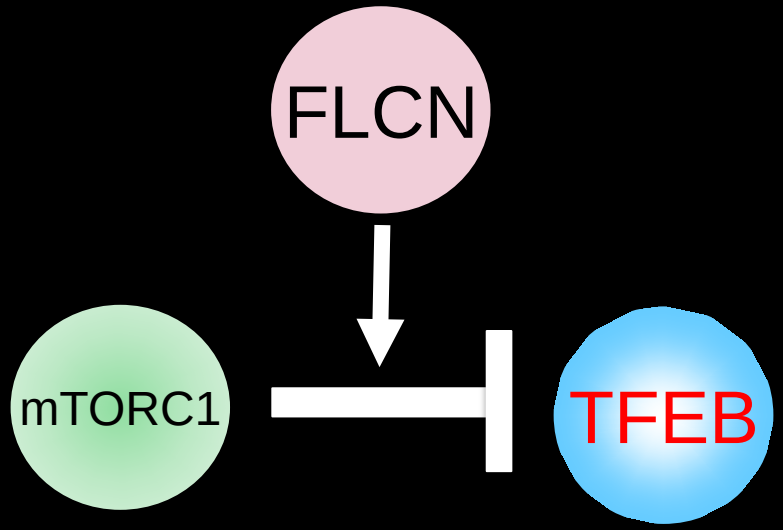
TFEB DEPLETION RESCUES KIDNEY DYSFUNCTION AND LETHALITY IN FLCN KO MICE





Napolitano, Di Malta* et al., Nature, 585: 597-602, 2020*

FLCN is a negative regulator of TFEB

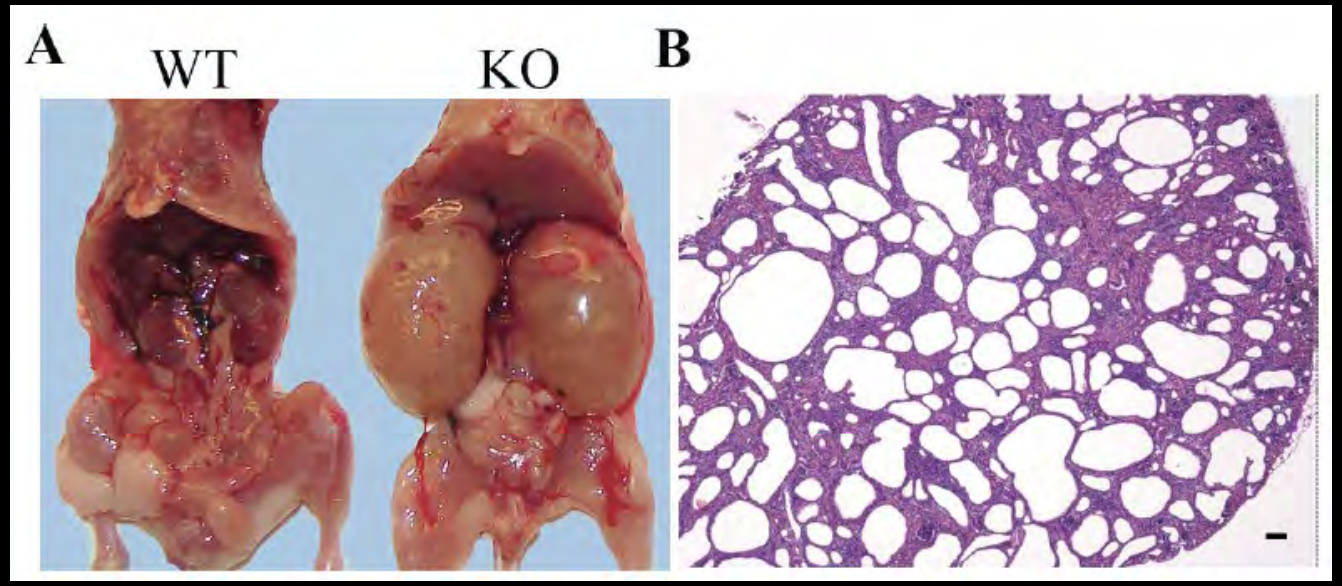


Is TFEB activation involved in BHD syndrome?

Chiara Di Malta

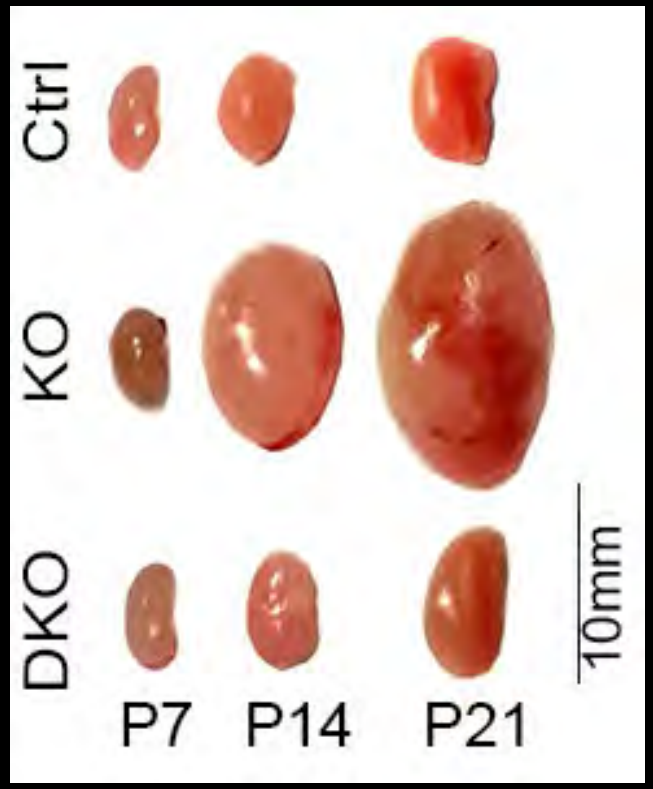
A mouse model of BHD syndrome

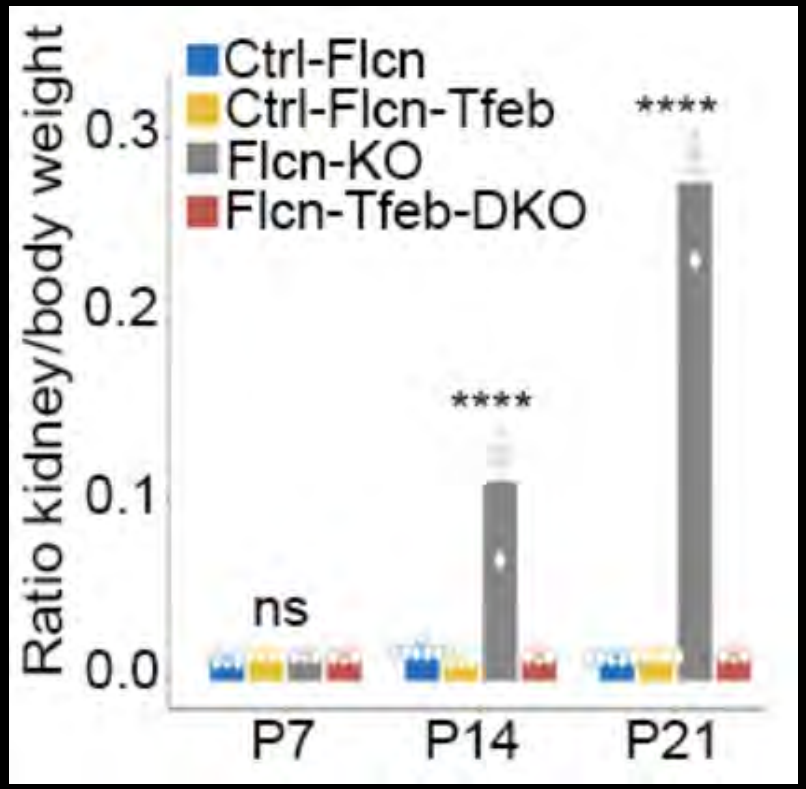
Kidney-specific
(Cdh16 CRE)
FLCN KO mice



Wu M. et al. Oncotarget. 2015; 6: 32761-32773.

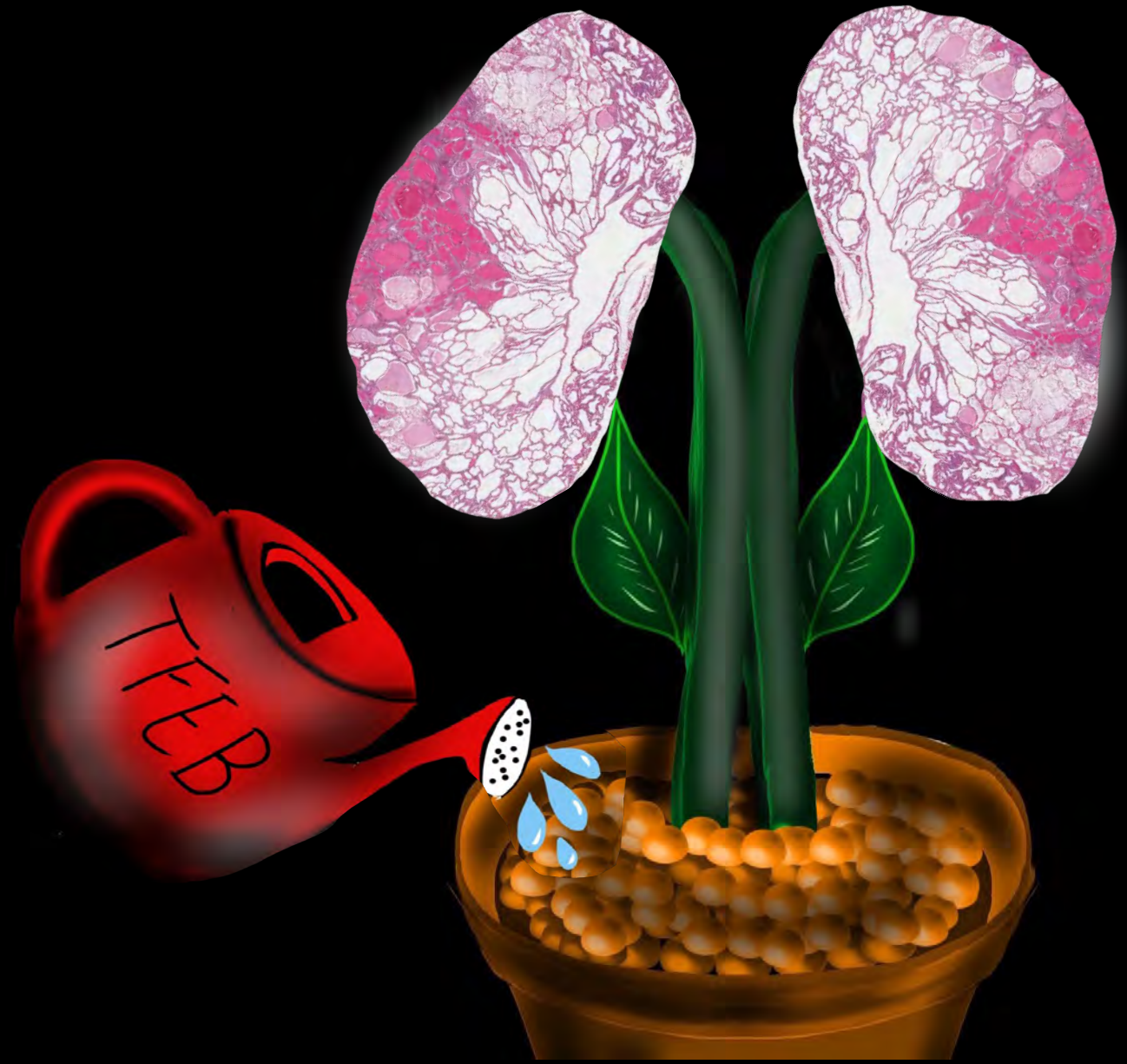
TFEB DEPLETION RESCUES THE KIDNEY PHENOTYPE OF FLCN KO MICE



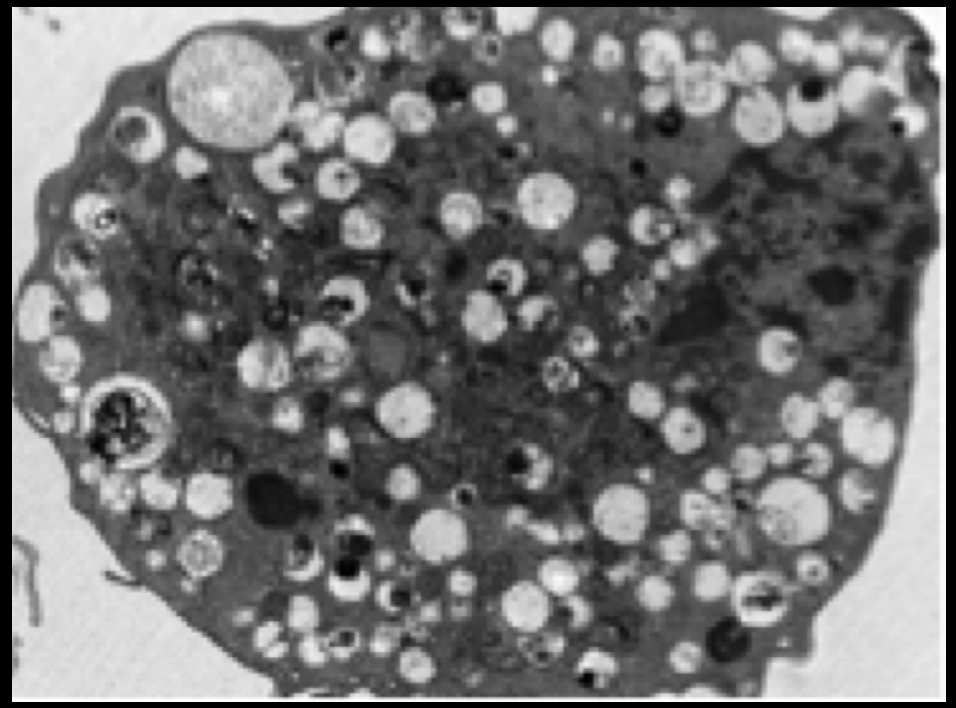


Napolitano, Di Malta* et al., Nature, 585: 597-602, 2020*

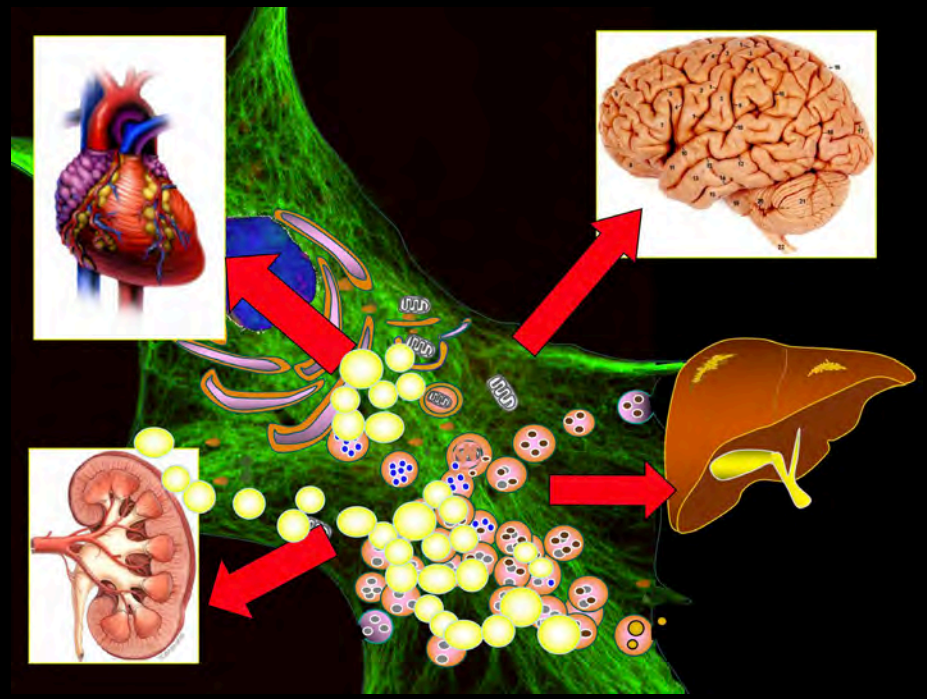
TFEB drives renal cystogenesis and tumorigenesis



anche le cellule devono smaltire i rifiuti…



Malattie associate a difetti del processo di smaltimento dei rifiuti cellulari



DIVERSI TIPI DI MALATTIE GENETICHE RARE

MALATTIE NEURODEGENERATIVE
(ALZHEIMER, PARKINSON ETC…)

MALATTIE METABOLICHE
(OBESITA’, DIABETE ETC…)

DIVERSI TIPI DI CANCRO

LYSOSOMES AND DISEASE

IMPAIRED LYSOSOMAL FUNCTION

Lysosomal storage diseases
(e.g. Mucopolysaccharidoses, Sphingolipidoses, Ceroid lipofuscinoses etc..)

Common neurodegenerative diseases
(e.g. Alzheimer's, Parkinson's, Huntington's)

Others
(e.g. metabolic diseases, infections,….aging)

INCREASED LYSOSOMAL FUNCTION

CANCER

TFEB subcellular localization is regulated by phosphorylation of specific serine residues