

Targeting PPA2 dependent mitochondrial fission triggers cisplatin sensitivity in oral squamous cell carcinoma

Sujit Kumar Bhutia

PhD Department of Life Science National Institute of Technology Rourkela

Abstract

The mitochondrial pyrophosphatase PPA2 is a mitochondrial matrix localized protein known for maintaining mitochondrial function. Herein, a critical role of PPA2 in oral squamous cell carcinoma (OSCC) cell survival and therapy resistance is reported. The expression of PPA2 was observed to be significantly increased in human OSCC tissue and cell lines, as well as in a DMBA-induced OSCC hamster model, compared to the respective normal counterparts. Furthermore, cell proliferation, migration, and colony-forming potential were increased in OSCC cells with forced expression of PPA2, whereas in PPA2-depleted conditions, these parameters were reduced. In addition, a significant increase in CDDP-induced apoptotic cell death was observed with PPA2 depletion. The pyrophosphatase activity of PPA2 was required for function, and OSCC cells expressing mutant PPA2-R127A lacking PPA2 pyrophosphatase activity failed to provide a survival advantage during CDDP treatment. Mechanistically, the increased expression of PPA2 countered mitochondrial hyperfusion-induced ROS production and cell death through increased mitochondrial fission. Hence, forced expression of PPA2 failed to protect against CDDP-induced cell death in fission-deficient conditions. Furthermore, PPA2-induced fission was mediated through MTP18; forced expression of PPA2 did not provide cytoprotection without MTP18. A PPA2-mediated increase in mitochondrial DNA content also inhibited apoptosis and enhanced the therapy resistance of OSCC cells. In conclusion, PPA2 expression is increased in OSCC, and PPA2-mediated mitochondrial fission and mitochondrial DNA replication protect OSCC cells against cisplatin-induced apoptosis.

Keywords: Apoptosis, Mitochondrial fission, MTP18, Oral cancer, PPA2

Targeting PPA2-dependent mitochondrial fission triggers cisplatin sensitivity in oral squamous cell carcinoma

Sujit Kumar Bhutia, PhD

Department of Life Science

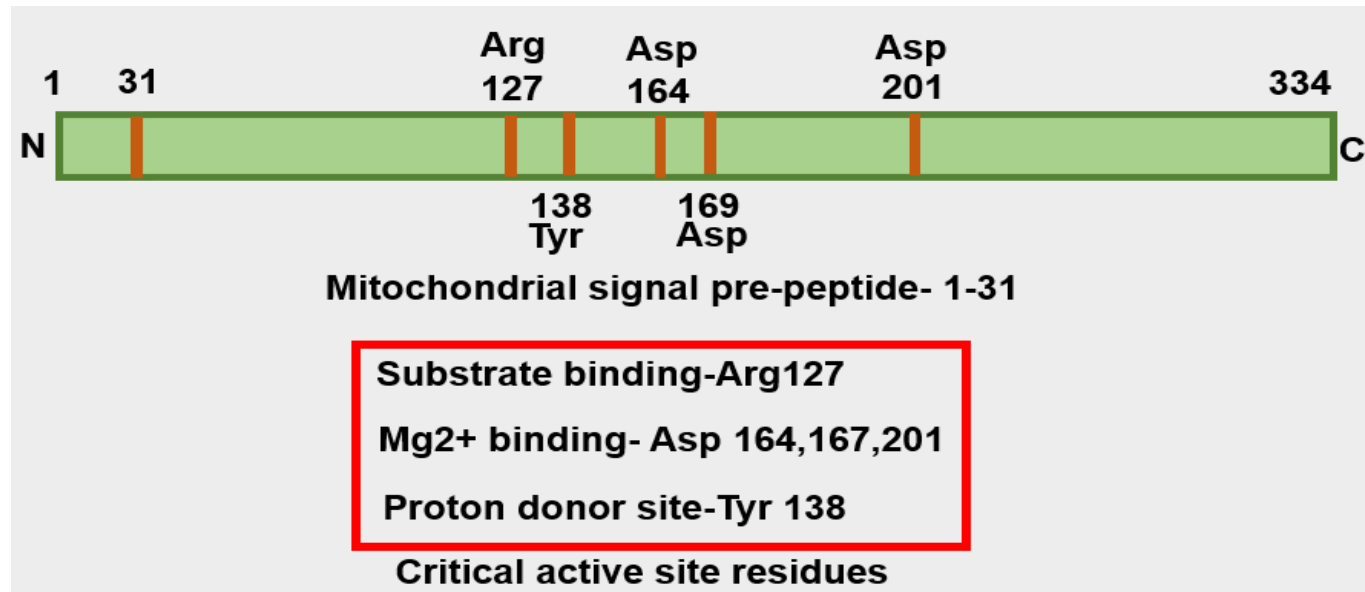
National Institute of Technology Rourkela



**International Symposium on
Mitochondria, Cell death and Human Diseases: Frontiers in Cancer Research**

Inorganic pyrophosphatase 2, PPA2

PPA2 (inorganic pyrophosphatase 2) is a mitochondrial enzyme that helps convert pyrophosphate (PPi) into inorganic phosphate (Pi). It's encoded by nuclear DNA



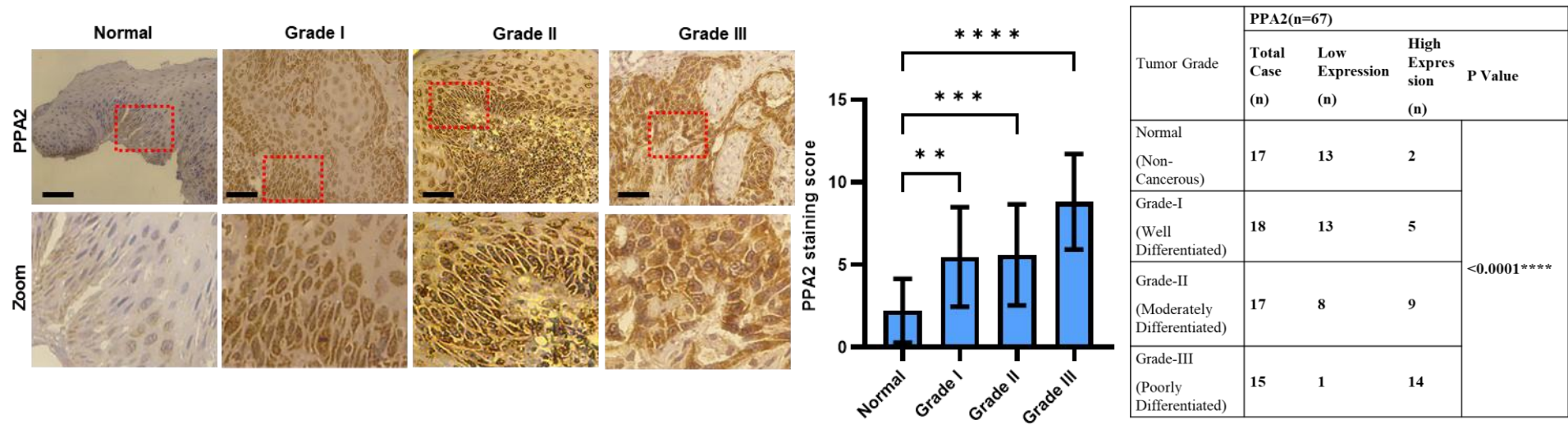
10	20	30	40	50
MSALLRLLRT	GAPAAACRL	GTSAGTGSRR	AMALYHTEER	GQPCSQNYRL
60	70	80	90	100
FFKNVTGHI	SPFHDIPLKV	NSKEENGIPM	KKARNDEYEN	LFNMIVEIPR
110	120	130	140	150
WTNAKMEIAT	KEPMNPIKQY	VKDGLRYVA	NIFPYKGYIW	NYGTLPQTWE
160	170	180	190	200
DPHEKDKSTN	CFGDNDPIDV	CEIGSKILSC	GEVIHVKILG	ILALIDEGET
210	220	230	240	250
DWKLIANAN	DPEASKFHDI	DDVKKFKPGY	LEATLNWFRL	YKVPDGKPEN
260	270	280	290	300
QFAFNGEFKN	KAFALEVIKS	THQCWKALLM	KKCNGGAINC	TNVQISDSPF
310	320	330		
RCTQEEARSL	VESVSSSPNK	ESNEEEQVWH	FLGK	

Essential for Mitochondrial function and protect from oxidative damage
(J Biol Chem 1991;266(19):12168-72; Int J Biol Macromol 2024;290:138908)

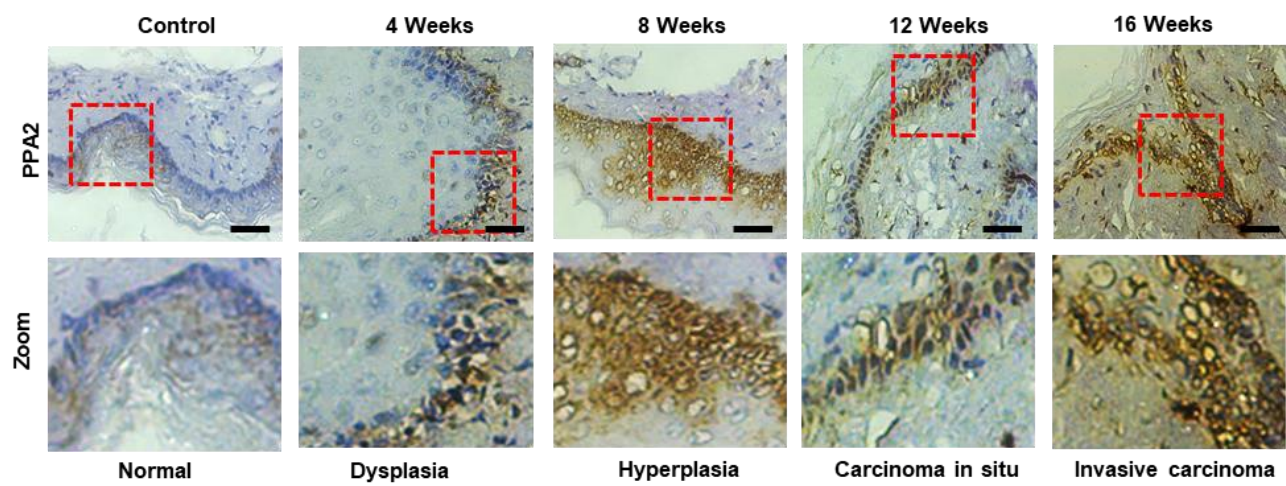
Case study on
PPA2-associated sudden cardiac death
(Am J Hum Genet. 2016;99(3):674-682) and many other

Multi-omics pan-cancer analysis
(Front Immunol 2024;15:1435502)

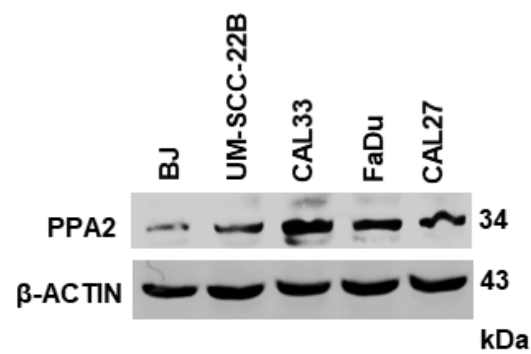
PPA2 expression is increased in OSCC and associated with histological grade



DMBA-Induced Oral Carcinoma in Syrian Hamster

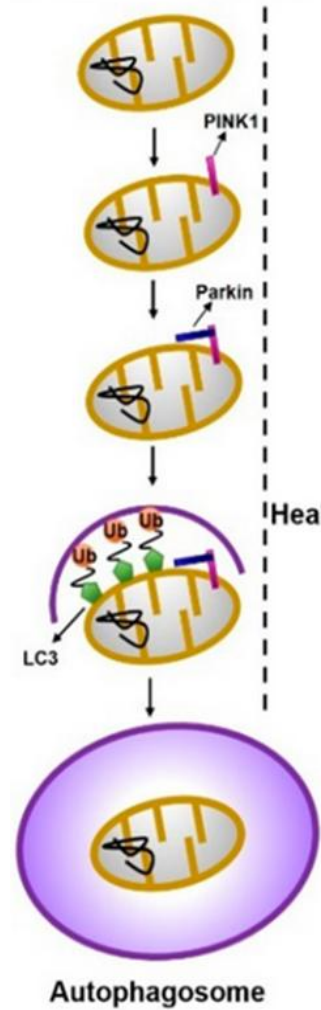
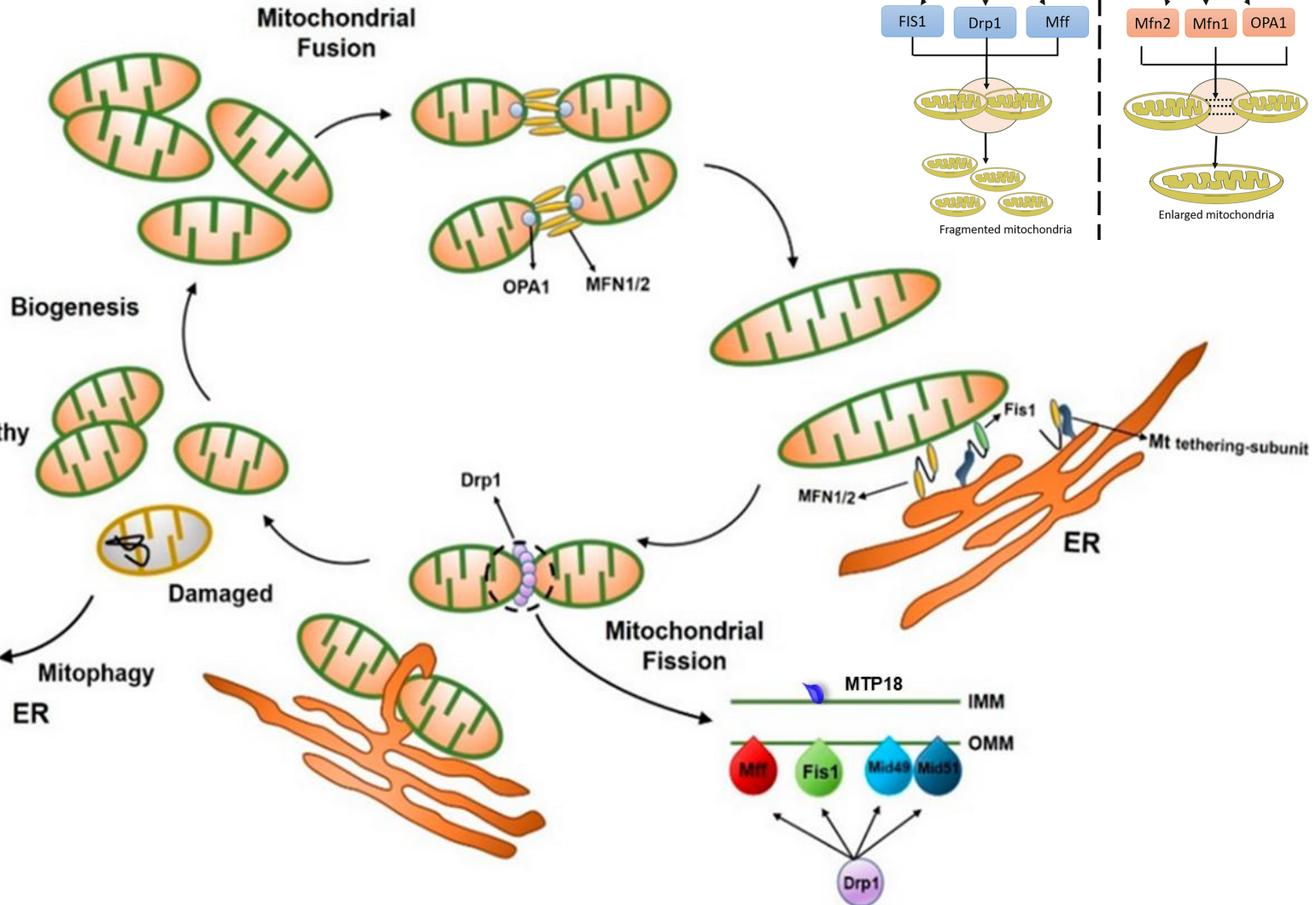


PPA2 expression in OSCC cell lines

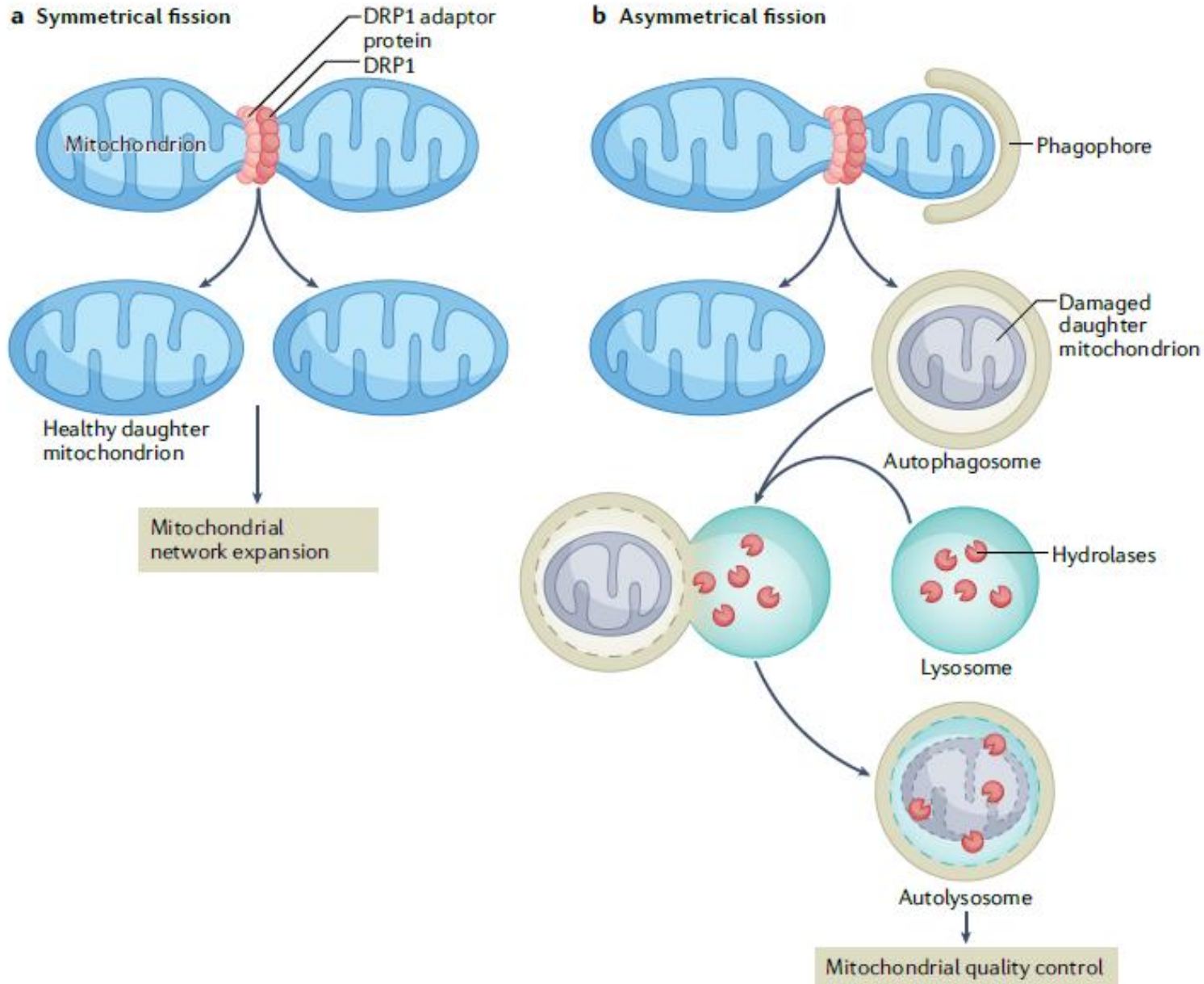


Mitochondrial dynamics and mitophagy

Sensing of damaged mitochondria by PINK1

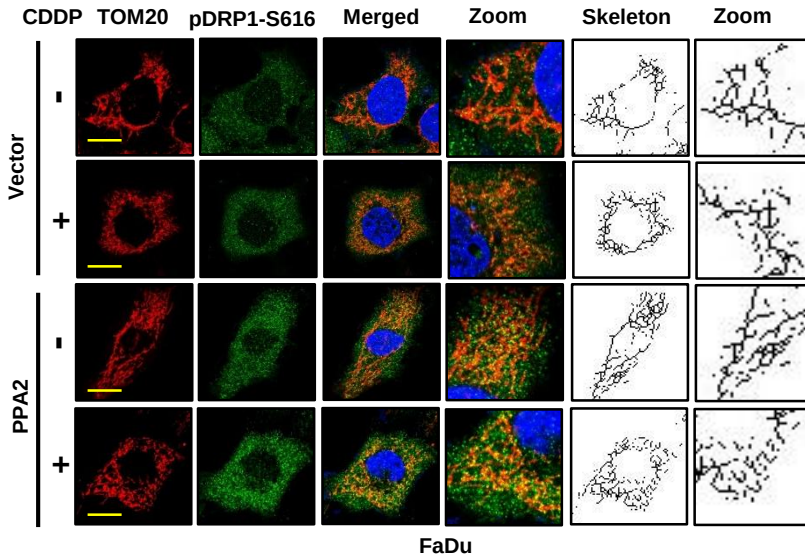
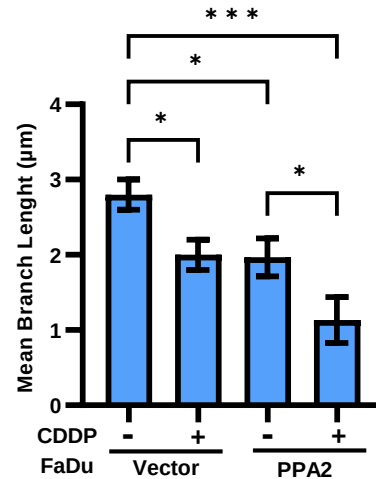
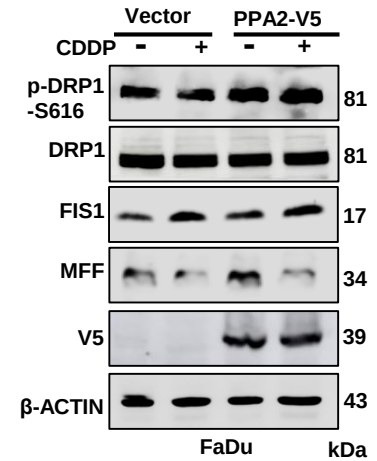


Mitochondrial fission

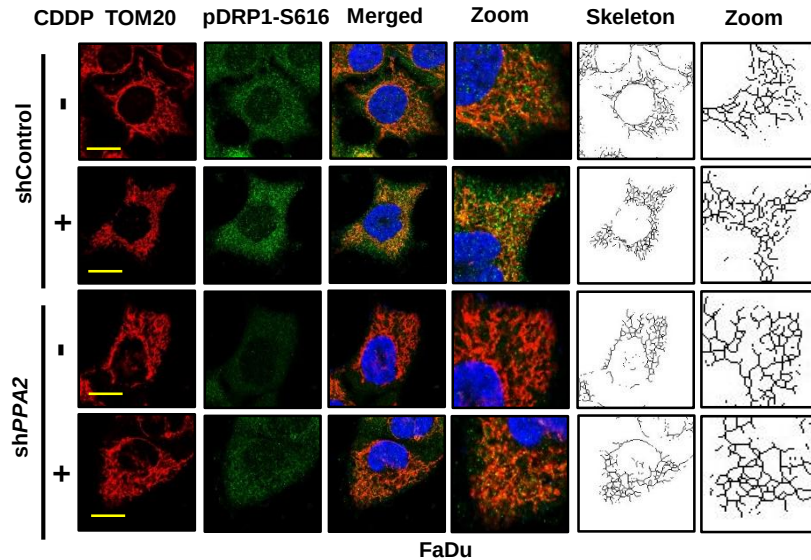
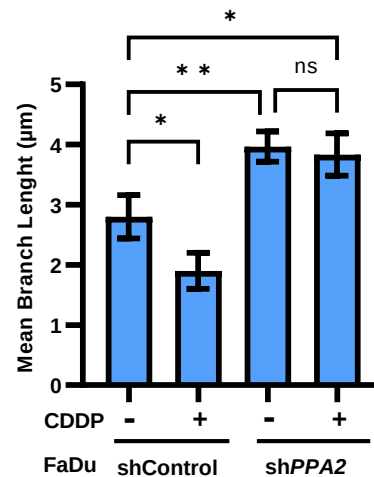
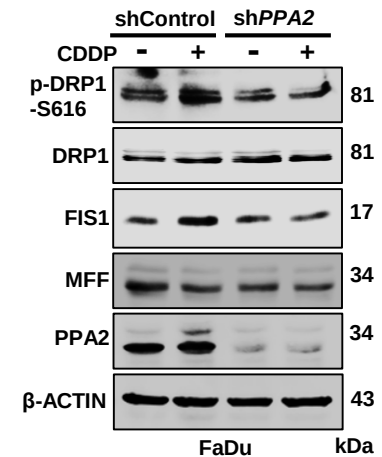


Structure of PPA2 (AF-Q9H2U2-F1)

PPA2 induces mitochondrial fission during cisplatin treatment

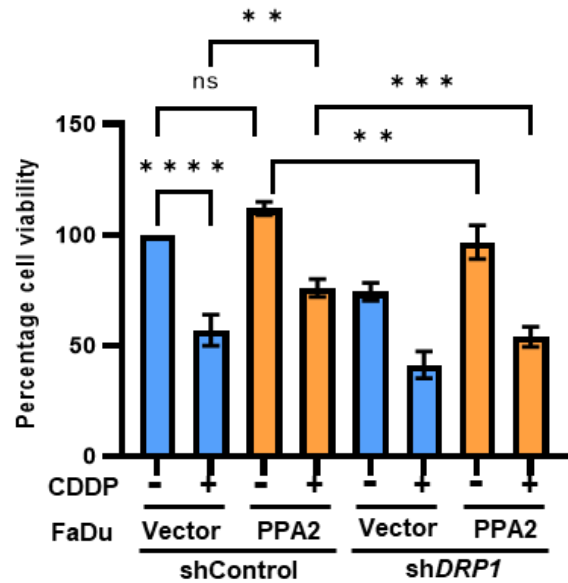
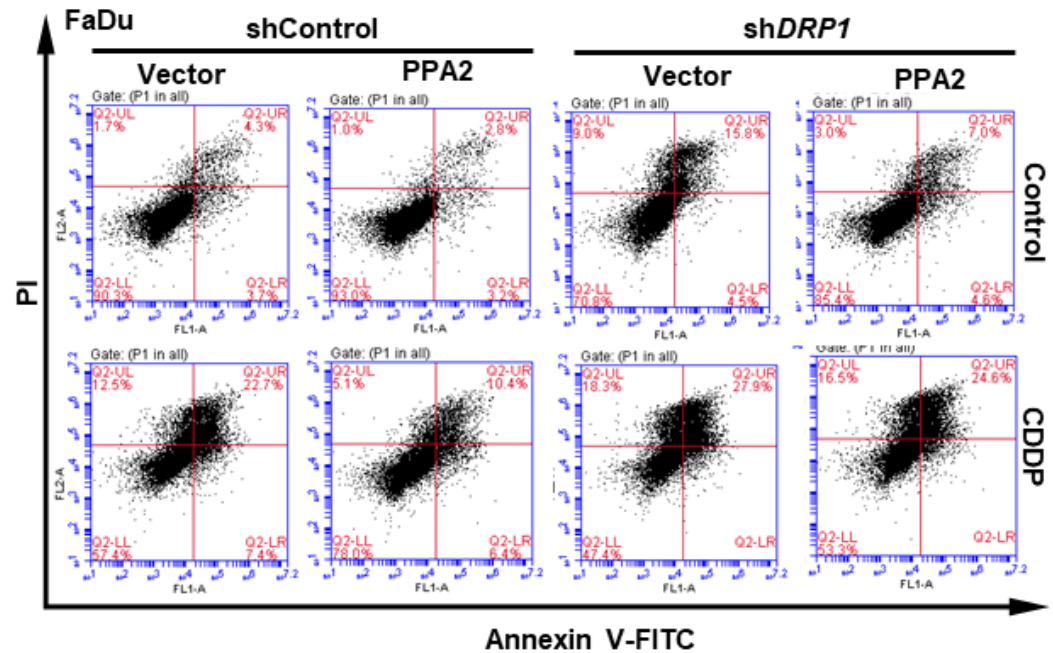
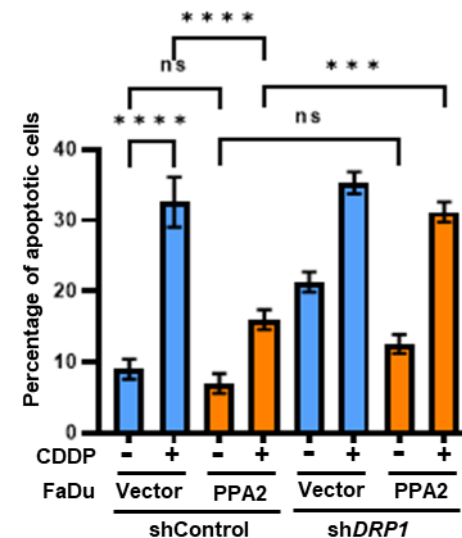


PPA2 overexpression-Mitochondrial fragmentation and increase in the mitochondrial translocation of p-DRP1-S616

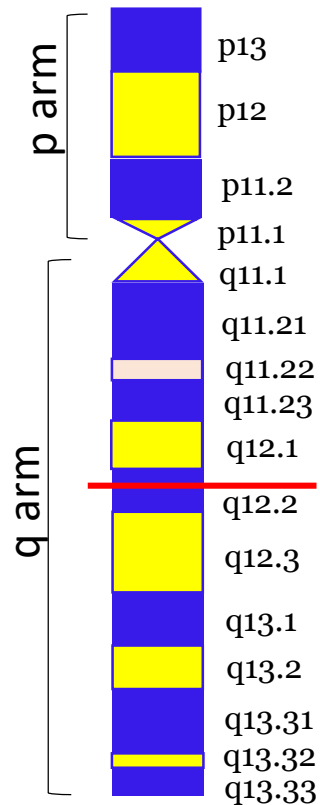


A hyperfused mitochondrial network with decrease in p-DRP1-S616-upon PPA2 depletion

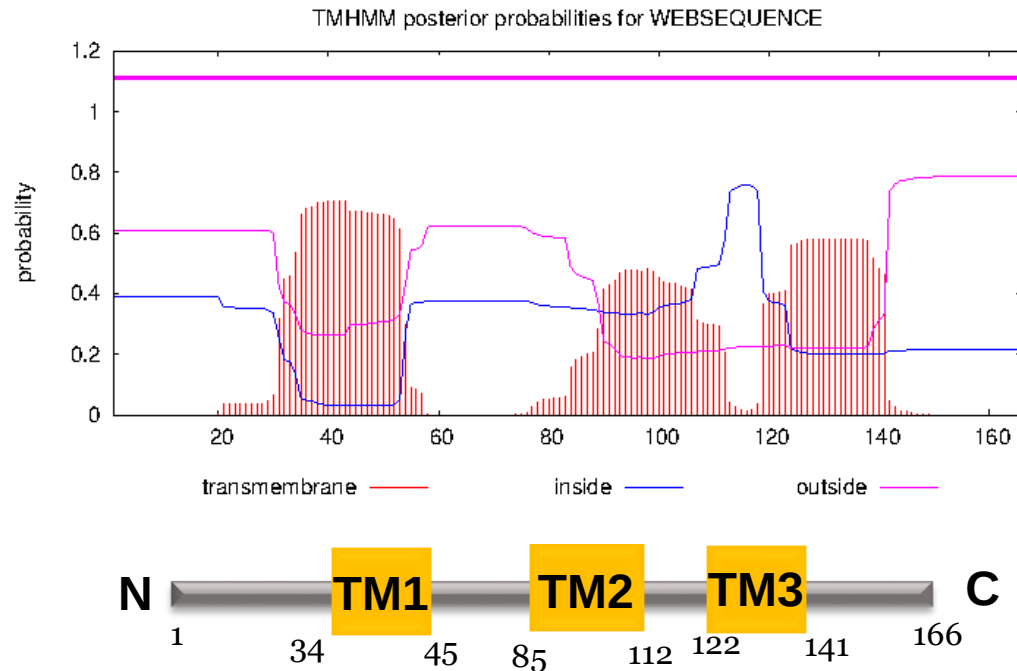
PPA2-induced mitochondrial fission promotes cell viability through DRP1-dependent pathway in OSCC cells



Mitochondrial fission process protein 1 (MTP18)



Chromosome 22



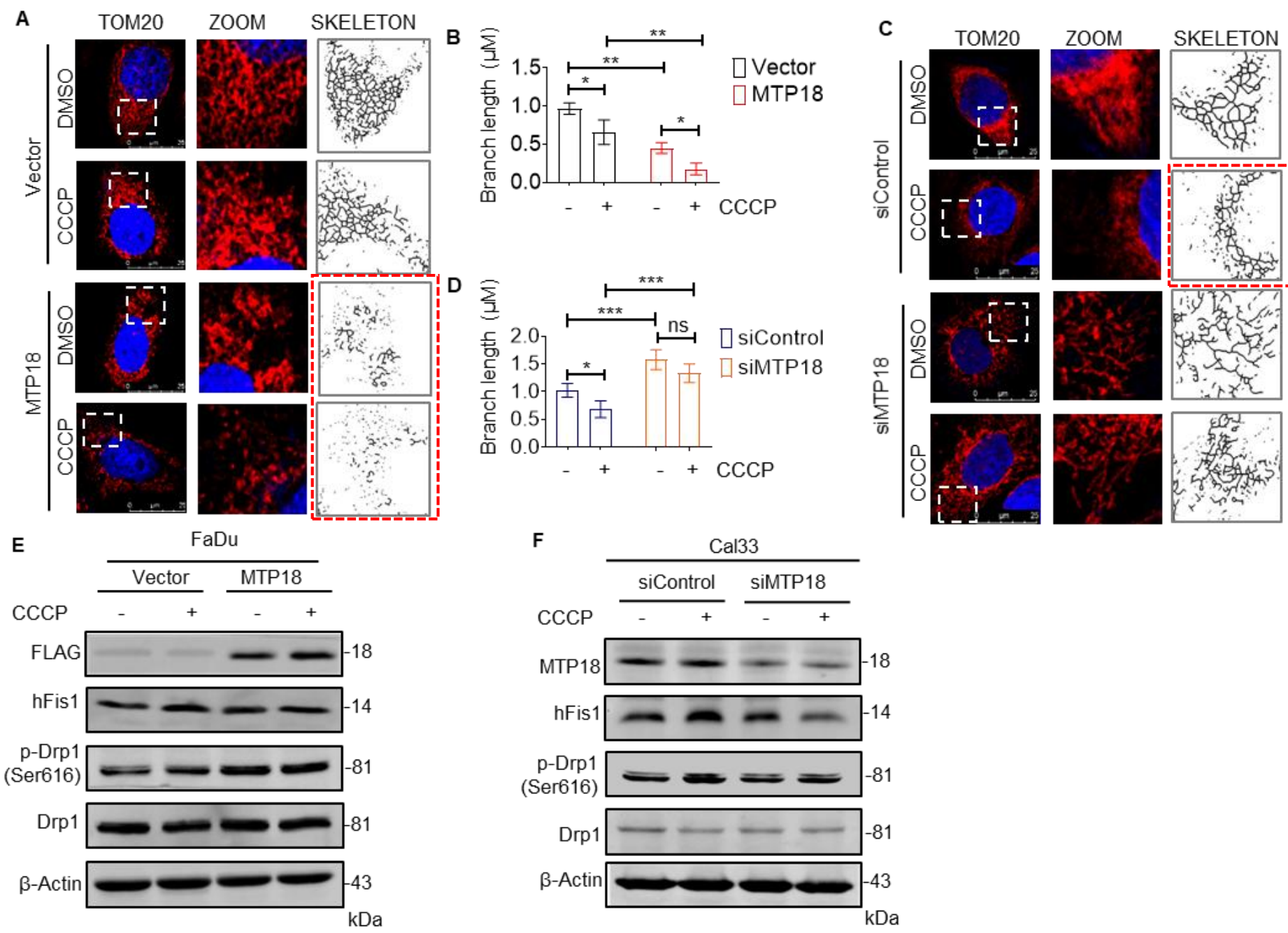
General Features

Localization	Mitochondrial Inner Membrane
Molecular weight	18 kDa
Amino Acid	166

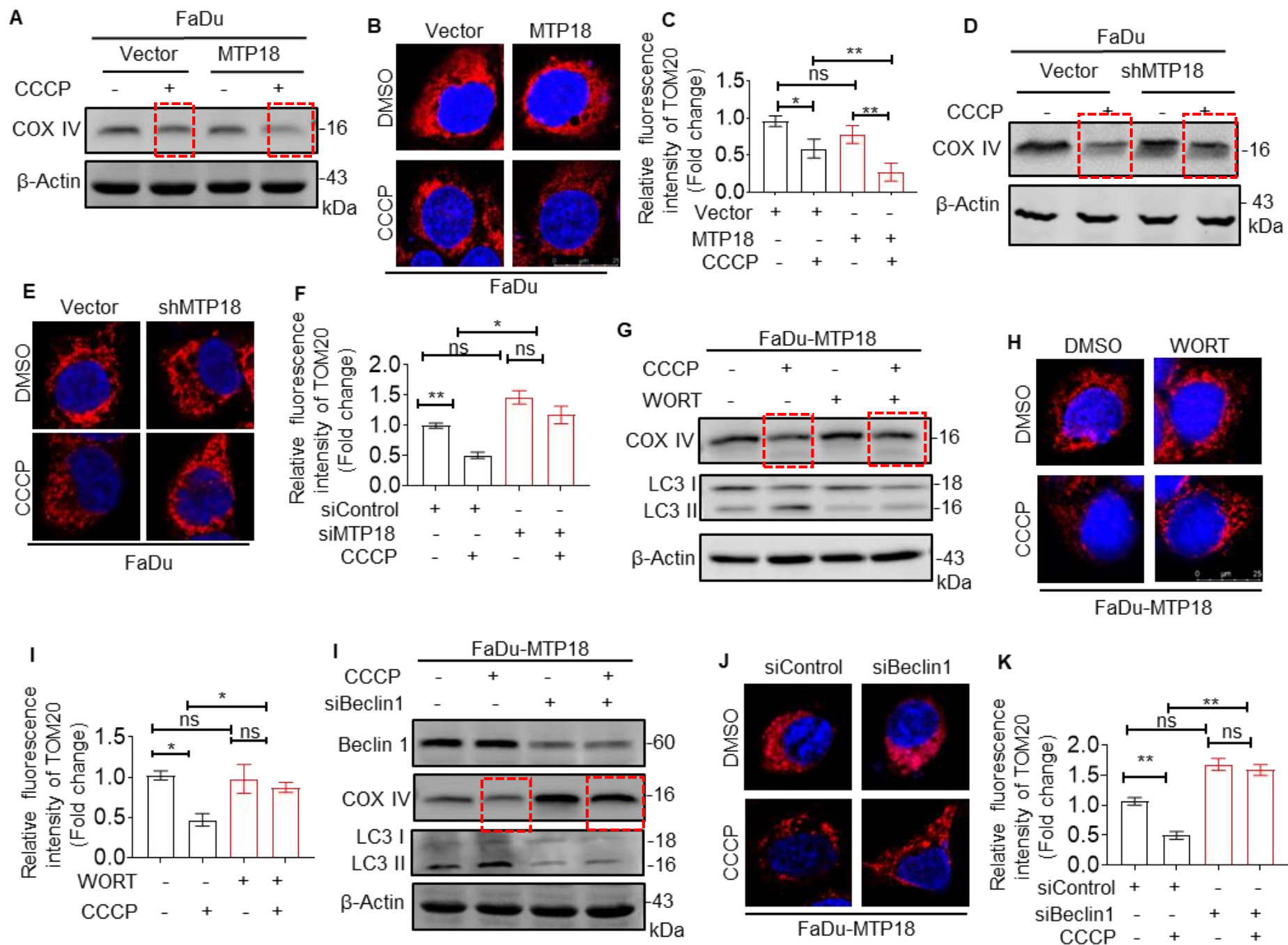
Other Features

Cell Proliferation	High
Apoptosis	Antiapoptotic
Metastasis	High
Mitochondrial dynamics	Fission

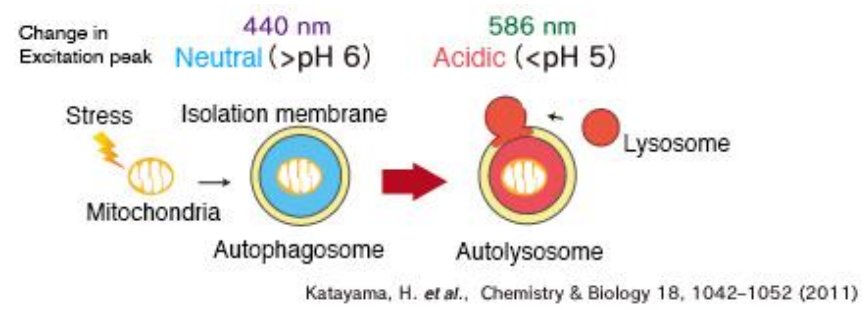
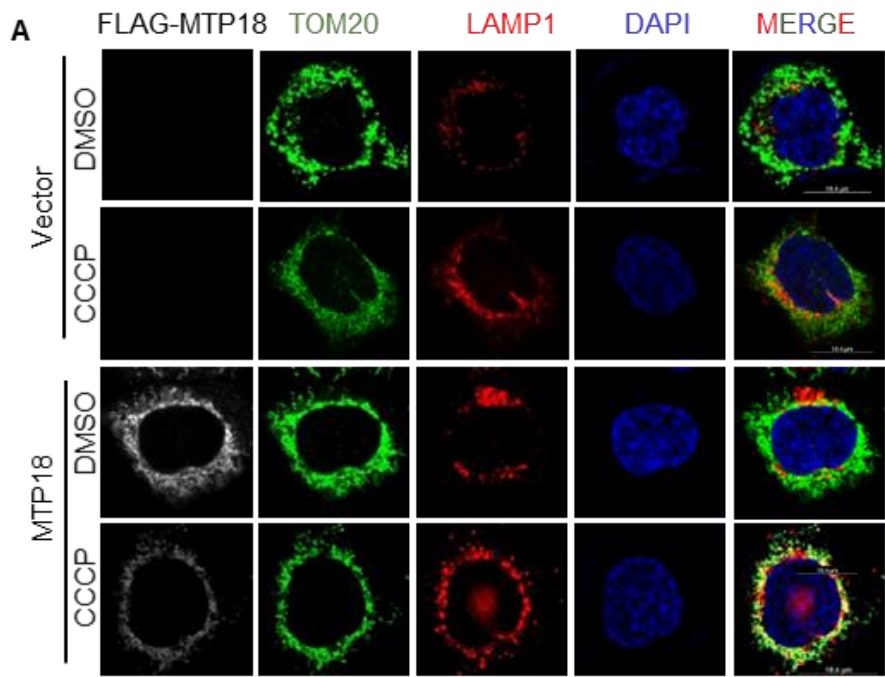
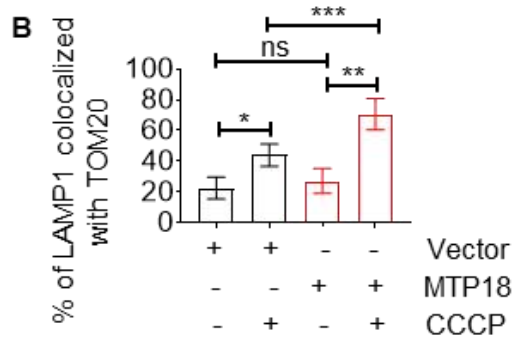
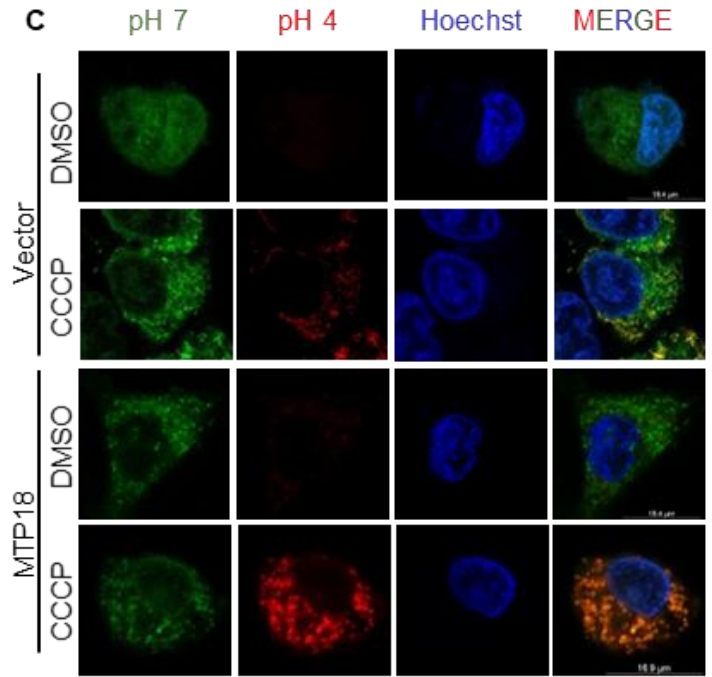
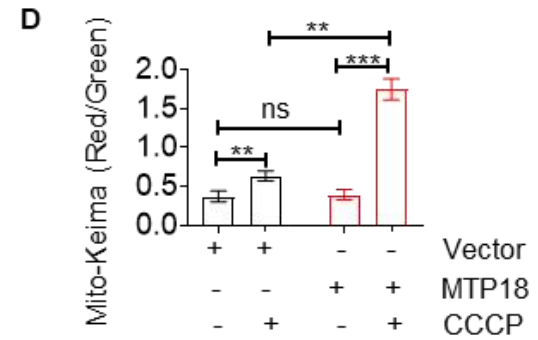
MTP18 in mitochondrial fragmentation and fission



MTP18 induces mitophagy



MTP18-induced mitophagosomes fuse with lysosomes for mitochondrial degradation: mitophagy flux



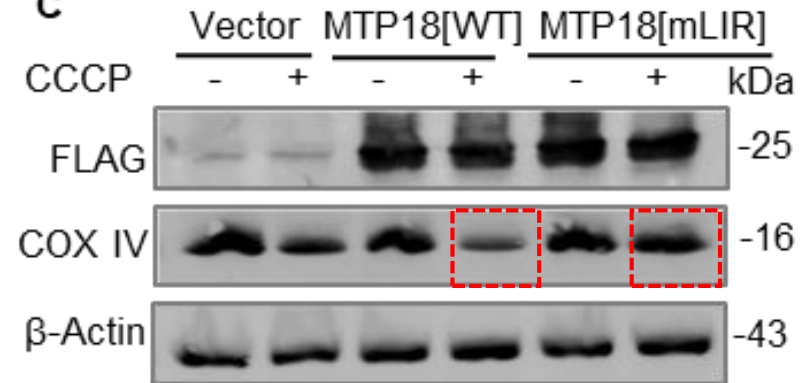
The LC3-interacting region of MTP18 is necessary for it to interact with LC3 for mitophagy induction

A

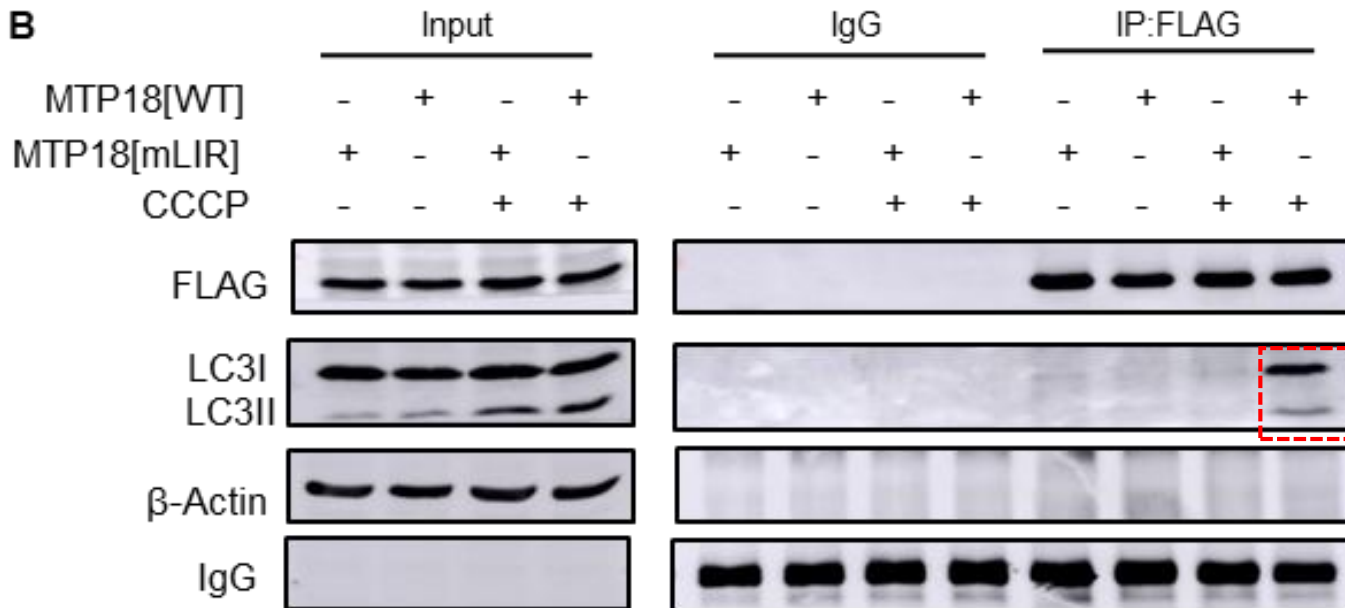
MSEPQPRGAERDLYRDTWVRYLGYANEVGE 30
 AFRSLVPAAVVWLSYGVASSYVLADAIDKG 60
 KKAGEVPSPEAGRSARVTVAVVDTFVWQAL 90
 ASVAIPGFTINRVCAASLYVLGTATRWPLA 120
 VRKWTTTALGLLTIPIIHPIDRSVDFLLD 150
 SSLRKL^{YPTV}GKPSSS 166
 LIR

(W/F/Y)xx(L/I/V)-x can be amino acid

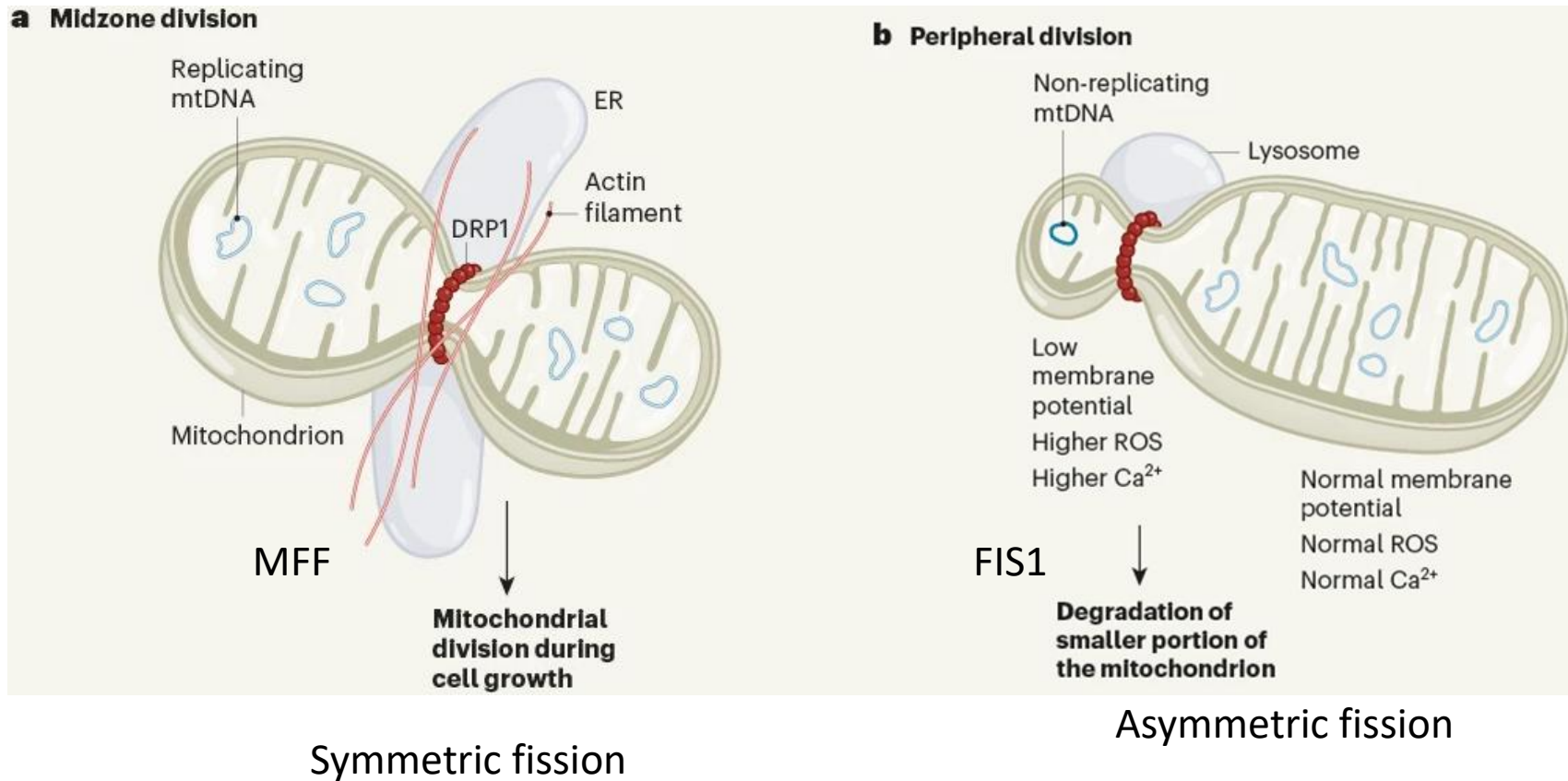
C



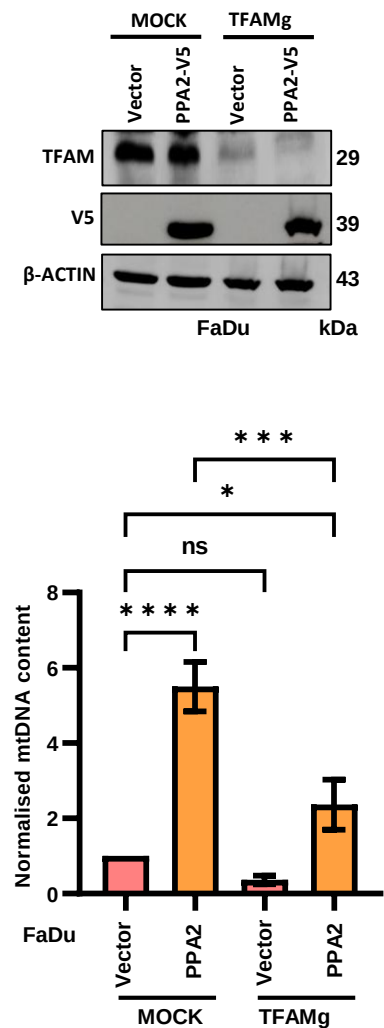
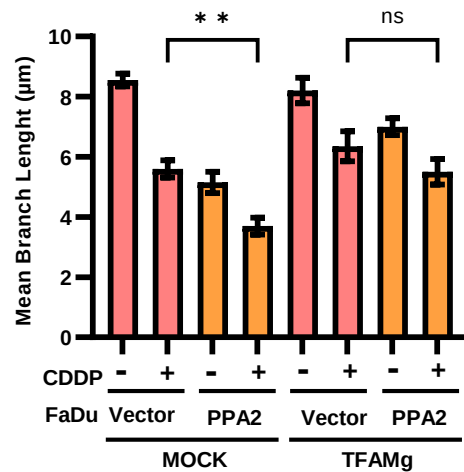
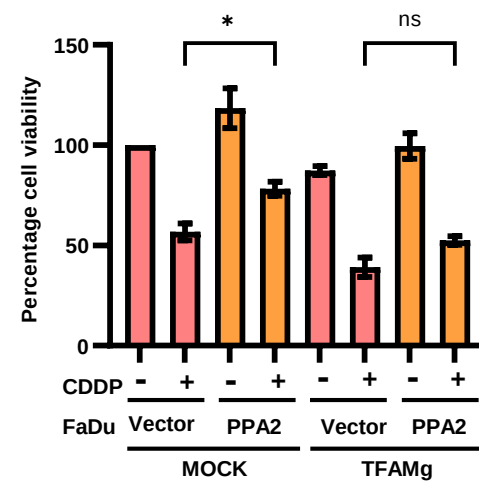
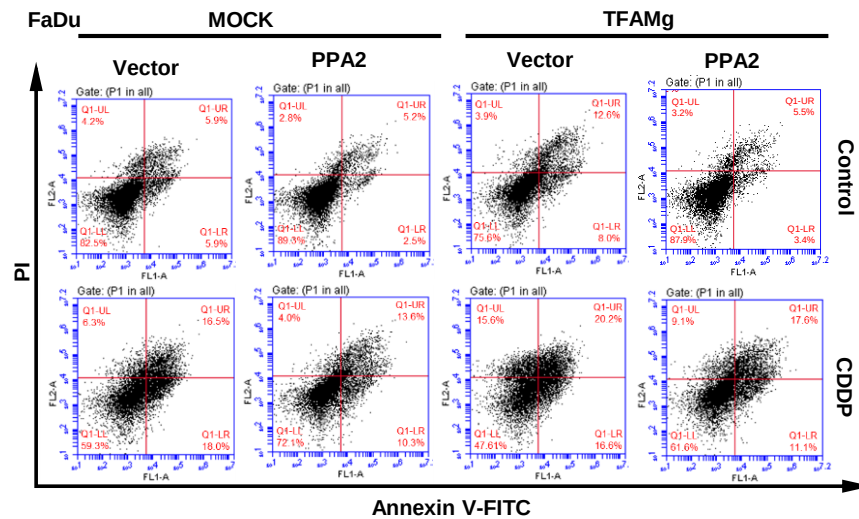
B



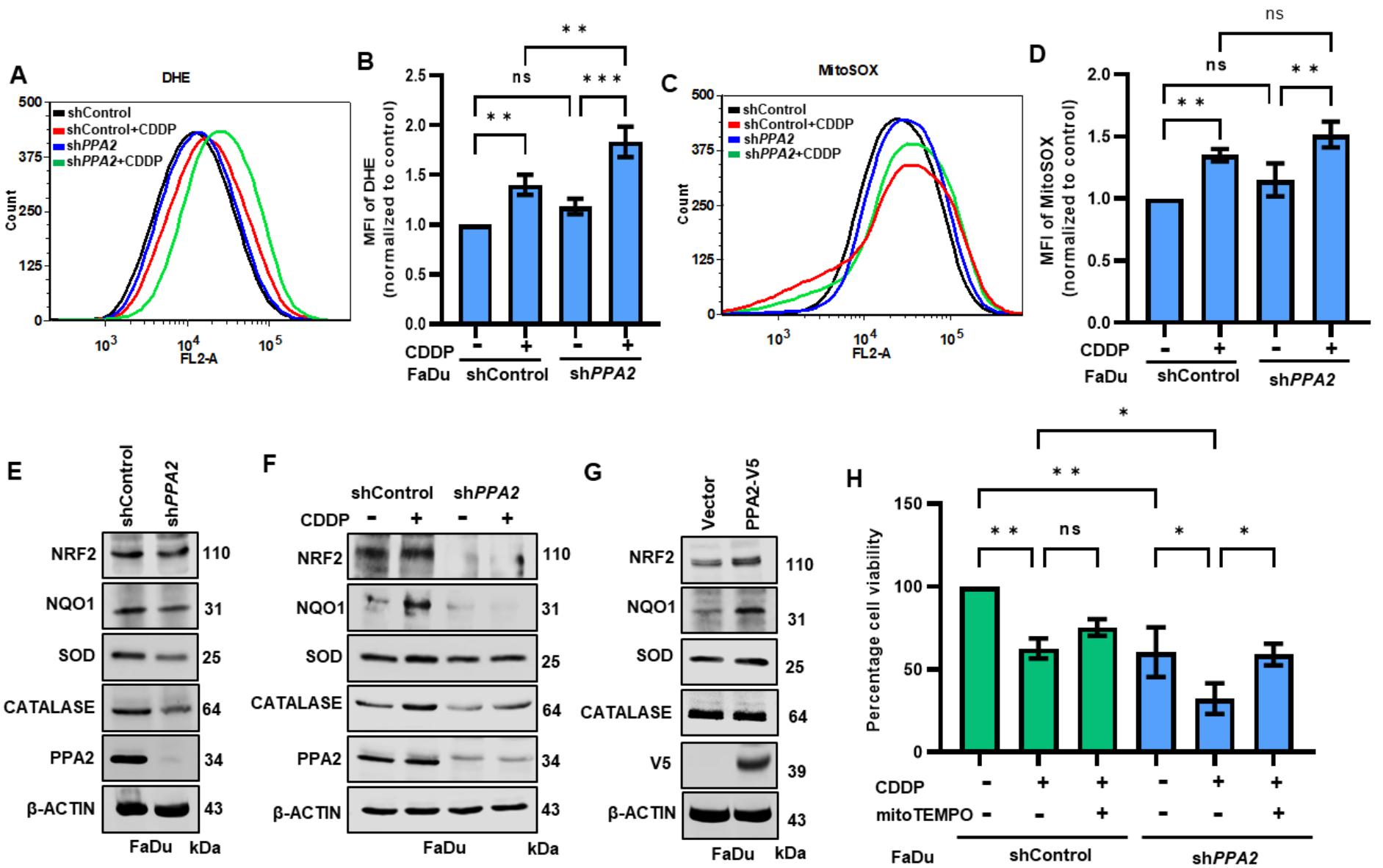
Distinct fission signatures predict mitochondrial degradation or biogenesis



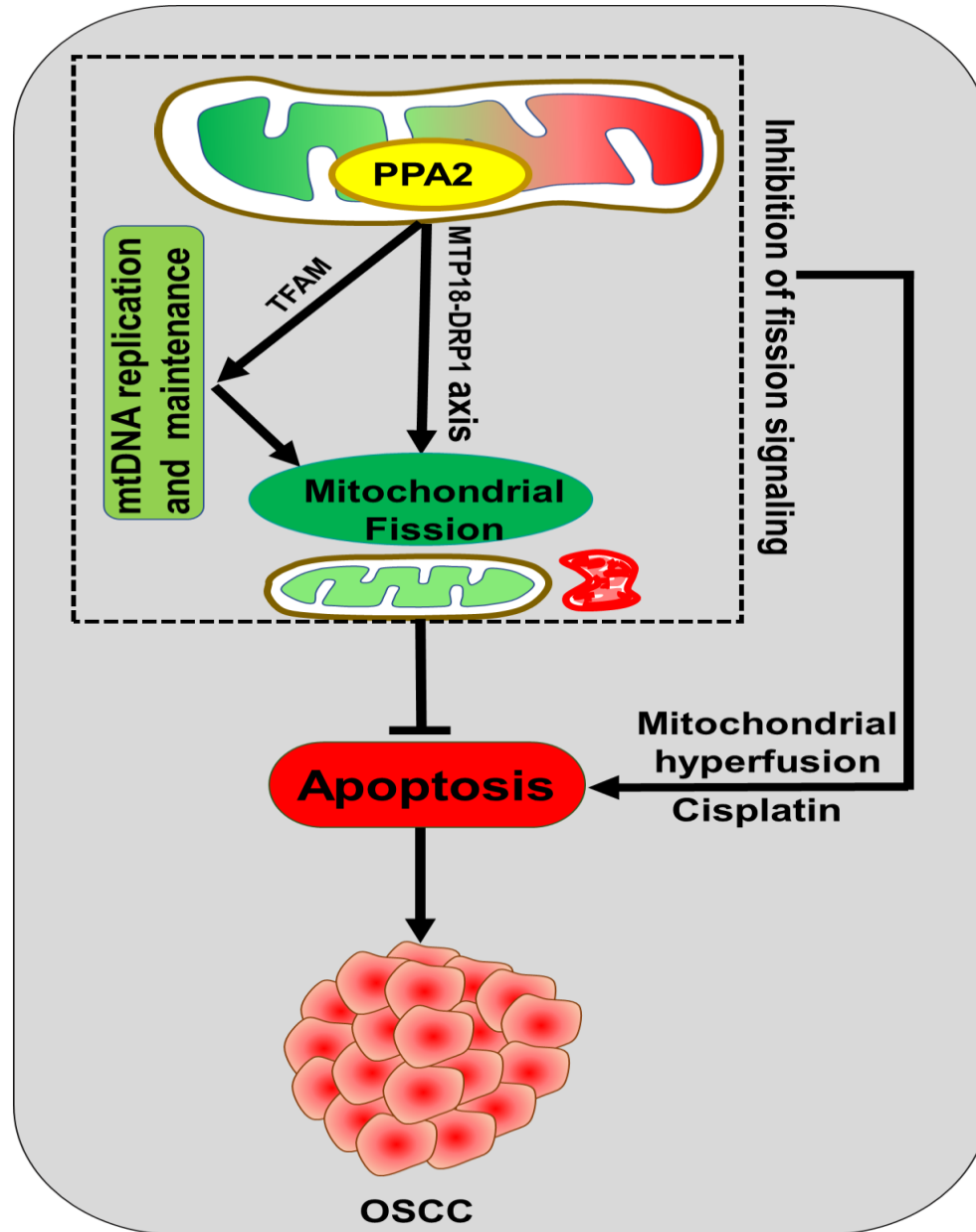
PPA2-mediated mtDNA replication provides a survival advantage to OSCC cells



PPA2 deficiency causes ROS generation to trigger cell death in OSCC cells



Summary



Acknowledgement

Current PhD students

Bishnu P Behera

Amruta Singh

Rakesh Kumar Kar

Priyadarshini Mishra

Sushmita Patra

Prakash Kumar Senapati

Pratyasha Mahadani

Previous PhD students

Soumya R Mishra

Srimanta Patra

Kewal K Mahapatra

Prakash P Praharaj

Chandra S Bhol

Debasna P Panigrahi

Prajna P Naik

Prashanta K Panda

Niharika Sinha

Subhadip Mukhopadhyay

Durgesh Nandini Das

Collaborators

Prof. B S Patro, BARC

Prof. Thomas Efferth, Germany

Prof. H J Chae, South Korea

Dr Rajakishore Mishra, CUJ

Dr. Gautam Sethi, Singapore

Dr. Vijay Pandey, China

Dr Alexander Birbrair, USA

Sponsor





Thank You