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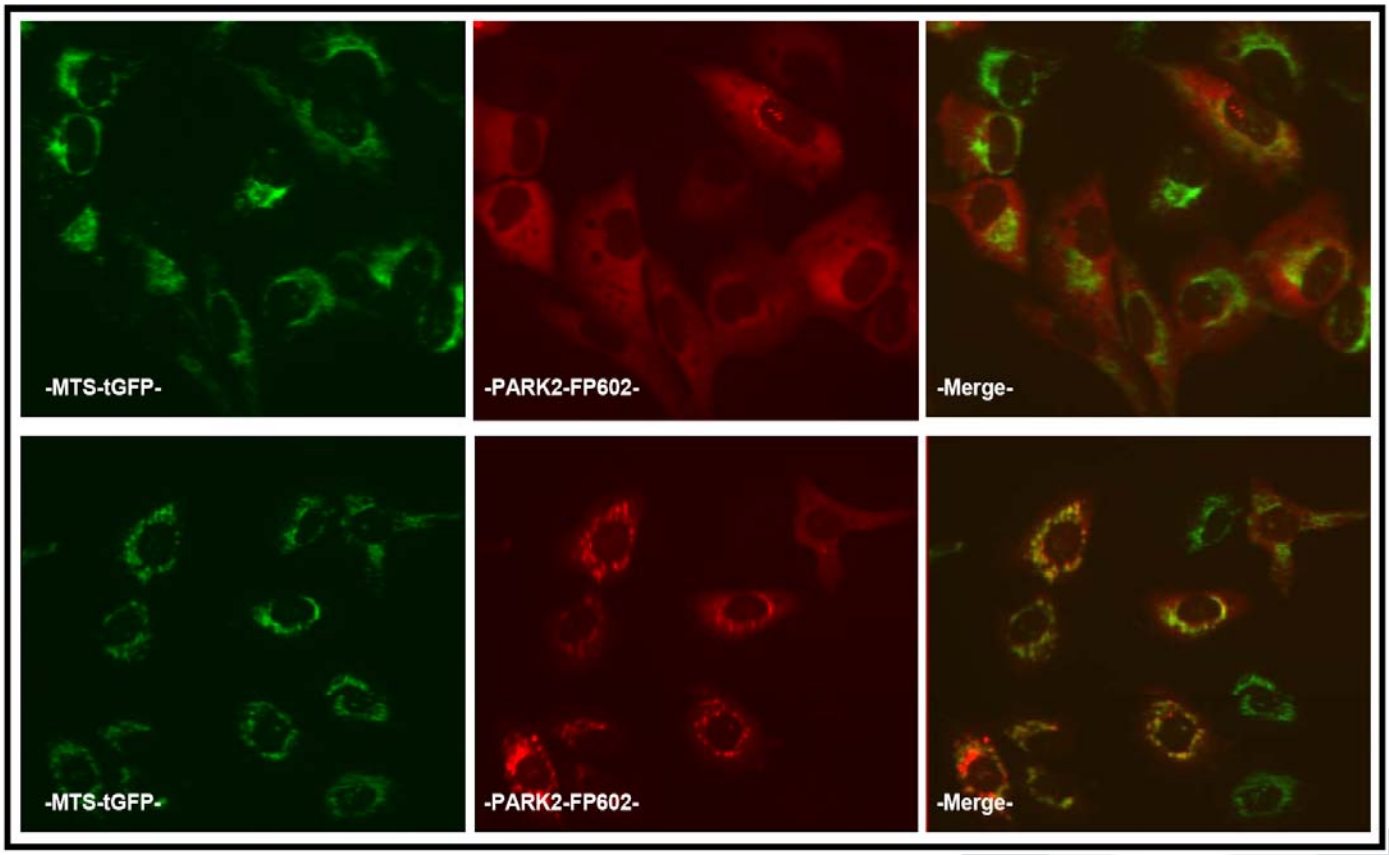
Speeding up drug discovery

REF: P30713

INNOPROT ASSAYS FOR HIGH CONTENT SCREENING

PARKINSON´s DISEASE IN VITRO MODELS

- PARKIN MITOCHONDRIAL RECRUITMENT ASSAY CELL LINE -



Product name: PARK2-FP602+ MTS-TGFP / U2OS cell line

Z´: 0.64

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PARKINSON´s DISEASE *IN VITRO* MODELS

- PARKIN MITOCHONDRIAL RECRUITMENT ASSAY -

Cell Line Name:	PARK2-FP602+MTS-tGFP U2OS Stable Cell line
Pathway:	PARKIN mitochondrial recruitment in Parkison´s disease
HCS Application:	PARK2-FP602 redistribution quantification
Material provided:	P30713: Stable Cell Line (2 vial of cells)
	P30713-DA: Division Arrested cells (2 million cells per vial)

This cell line has been produced with the technology developed within FP7 PASCA EU project, and is 100% certified truly monoclonal.

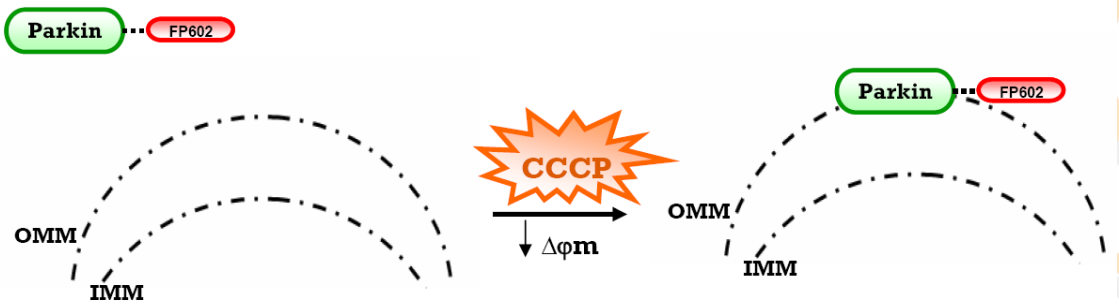
Background

Parkinson´s disease (PD) is a neurodegenerative disorder caused by the death of dopamine producing cells in the substantia nigra in the brain. The main sympton is trembling in the hands, legs or face. Most cases of PD are sporadic, but 10% are inherited either in an autosomal dominant or recessive manner.

Parkin is one of the proteins implicated in one form of familial PD known as autosomal recessive juvenile Parkinson's disease (AR-JP)

Cell Line Characteristics

PARK2-FP062+MTS-tGFP cell line has been obtained by a double transfection of an expression vector that encodes turbo green fluorescent protein (tGFP) fused to mitochondrial targeting sequence (MTS) derived from the subunit VIII of human cytochrome C oxidase and a vector containing PARK2 protein fused to FP602 red fluorescent protein.



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Assay Validation

Parkin is a protein that forms part of E3 ubiquitin ligase complex that mediates the targeting of proteins for degradation.

Parkin is recruited to damaged mitochondria during the early stages of mitophagy induced by CCCP.

This cell line has been designed to ensure the colocalization of PARK2-FP602 and Mitochondria once CCCP has been added.

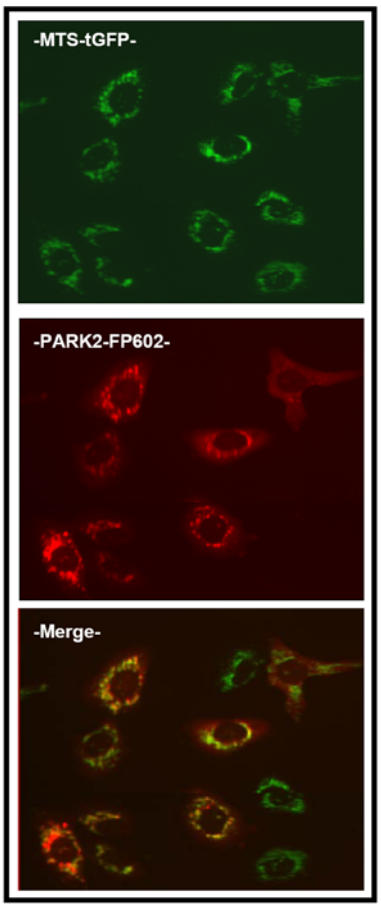

Fig1. Colocalization assay of MTS-tGFP and PARK2-FP602 after a treatment with CCCP 5µM during 2h.

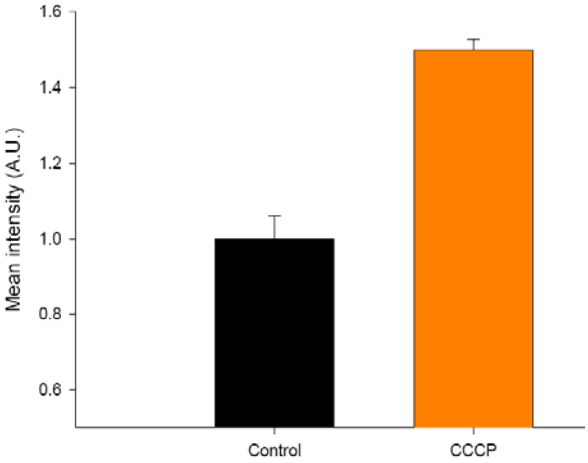

Fig 2. To analyze PARK2 mitochondrial localization, a Region Of Interest (ROI) of PARK2 was delimited and the intensity of mitochondria signal (GFP) was quantified in this ROI. So, when PARK2 was recruited in the mitochondria, the intensity of the GFP was increased.

Applications

The stably transfected PARK2-FP602+MTS-tGFP cell line can be used in drug discovery for compounds that inhibit mitochondrial loss of membrane potential. So it can be a usefull tool to test possible drugs against Parkinson's disease.

This cellular model has been adapted to HCS analyses based on image algorithms to test PARK2 protein redistribution process.

Use Restriction

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