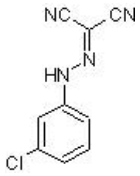
abcam

Product datasheet

CCCP, Mitochondrial oxidative phosphorylation uncoupler ab141229

14 References 5 Images

Overview

Product name	CCCP, Mitochondrial oxidative phosphorylation uncoupler
Description	Potent mitochondrial oxidative phosphorylation uncoupler
Biological description	Potent mitochondrial oxidative phosphorylation uncoupler. Renders mitochondrial inner membrane permeable to protons. Induces apoptosis *in vitro*.
Purity	> 99%
CAS Number	555-60-2
Chemical structure	

Properties

Chemical name	2-[2-(3-Chlorophenyl)hydrazinylyidene]propanedinitrile
Molecular weight	204.62
Molecular formula	$C_9H_5ClN_4$
PubChem identifier	2603
Storage instructions	Store at -20°C. Store under desiccating conditions. The product can be stored for up to 12 months.
Solubility overview	Soluble in DMSO to 100 mM and in ethanol to 100 mM
Handling	Wherever possible, you should prepare and use solutions on the same day. However, if you need to make up stock solutions in advance, we recommend that you store the solution as aliquots in tightly sealed vials at -20°C. Generally, these will be useable for up to one month. Before use, and prior to opening the vial we recommend that you allow your product to equilibrate to room temperature for at least 1 hour. Toxic, refer to SDS for further information. Need more advice on solubility, usage and handling? Please visit our **frequently asked questions (FAQ) page** for more details.

SMILES	Clc1cc(N\N=C(/C#N)C#N)ccc1
Source	Synthetic

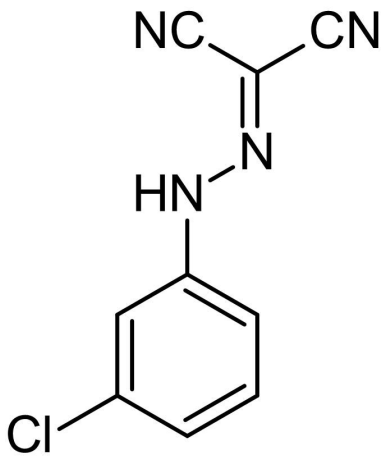
Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab141229 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

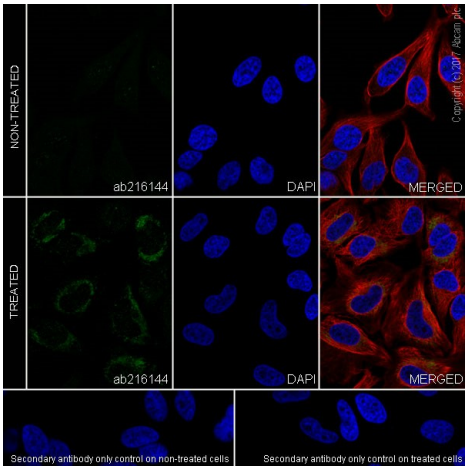
Application	Abreviews	Notes
Functional Studies		Use at an assay dependent concentration.

Images



Chemical Structure - CCCP, Mitochondrial oxidative phosphorylation uncoupler (ab141229)

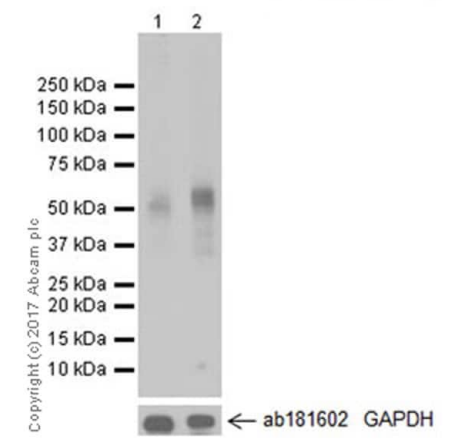
2D chemical structure image of ab141229, CCCP, Mitochondrial oxidative phosphorylation uncoupler



Immunocytochemistry/ Immunofluorescence - CCCP, Mitochondrial oxidative phosphorylation uncoupler (ab141229)

Immunofluorescent analysis of 4 % paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (human epithelial cell line from cervix adenocarcinoma)(+/- treatment with 10µM carbonyl cyanide 3-chlorophenylhydrazone (CCCP, ab141229) for 24 hours) cells labeling PINK1 with **ab216144** at 1/500 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/1000 dilution (green). Confocal image showing cytoplasmic staining on HeLa cells treated with 10µM carbonyl cyanide 3-chlorophenylhydrazone (CCCP, ab141229) for 24 hours. The nuclear counter stain is DAPI (blue). Tubulin is detected with Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) (**ab195889**) at 1/200 dilution (red).

The negative controls are as follows:
-ve control: PBS, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/1000 dilution.



Western blot - CCCP, Mitochondrial oxidative phosphorylation uncoupler (ab141229)

All lanes : Anti-PINK1 antibody [EPR20730] (**ab216144**) at 1/1000 dilution

Lane 1 : HeLa (human epithelial cell line from cervix adenocarcinoma) whole cell lysate
Lane 2 : HeLa cells (treated with 10uM carbonyl cyanide 3-chlorophenylhydrazone (CCCP, ab141229) for 24 hours) whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution

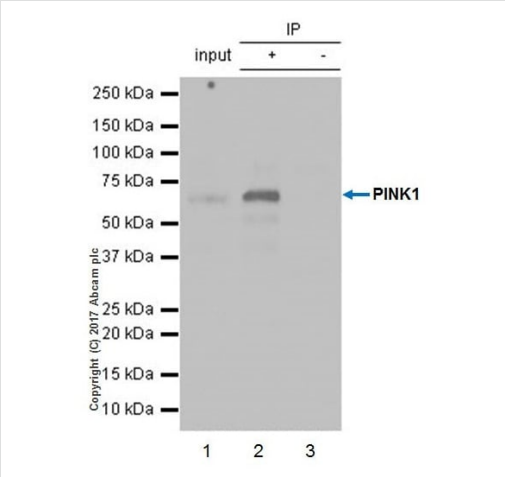
Developed using the ECL technique.

Observed band size: 62 kDa

Exposure time: 5 seconds

Blocking and dilution buffer: 5% NFDM/TBST

PINK1 can be induced by CCCP treatment (PMID: 24184327).

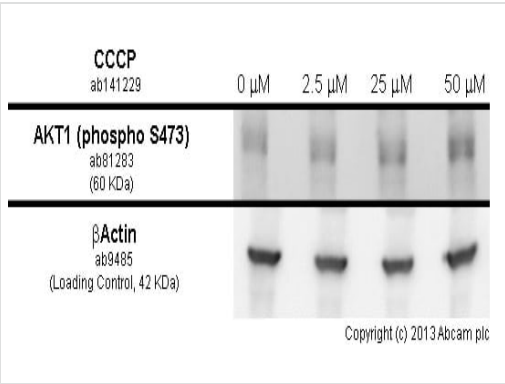


Immunoprecipitation - CCCP, Mitochondrial oxidative phosphorylation uncoupler (ab141229)

PINK1 was immunoprecipitated from 0.35 mg of HeLa (human epithelial cell line from cervix adenocarcinoma) (treated with 10uM carbonyl cyanide 3-chlorophenylhydrazone (CCCP. ab141229) for 24 hours) whole cell lysate with **ab216144** at 1/30 dilution. Western blot was performed from the immunoprecipitate using **ab216144** at 1/500 dilution. VeriBlot for IP Detection Reagent (HRP) (**ab131366**), was used for detection at 1/1000 dilution.

Lane 1: HeLa (CCCP-treated, ab141229) lysate 10 µg (Input).
Lane 2: **ab216144** IP in HeLa (CCCP-treated, ab141229) lysate.
Lane 3: Rabbit monoclonal IgG (**ab172730**) instead of **ab216144** in HeLa (CCCP-treated, ab141229) whole cell lysate.

Blocking and dilution buffer: 5% NFDM/TBST.



Functional Studies - CCCP, Mitochondrial oxidative phosphorylation uncoupler (ab141229)

MCF7 cells were incubated at 37°C for 2 hours with vehicle control (0 µM) and different concentrations of CCCP (ab 141229). Increased expression of AKT1 (phospho S473) (**ab81283**) in MCF7 cells correlates with an increase in CCCP concentration, as described in literature.

Whole cell lysates were prepared with RIPA buffer (containing protease inhibitors and sodium orthovanadate), 10 µg of each were loaded on the gel and the WB was run under reducing conditions. After transfer the membrane was blocked for an hour using 5% BSA before being incubated with **ab81283** at 2 µg/ml and **ab8227** at 1 µg/ml overnight at 4°C. Antibody binding was detected using an anti-rabbit antibody conjugated to HRP (**ab97051**) at 1/10000 and visualised using ECL development solution.

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