# abcam

## Product datasheet

## Cytochrome c Release Assay Kit ab65311

★★★★★ 3 Abreviews 48 References 画像数 2

#### 製品の概要

製品名 Cytochrome c Release Assay Kit

サンプルの種類 Tissue, Adherent cells, Suspension cells

 アッセイタイプ
 Direct

 全工程の試験時間
 3h 00m

**種交差性 交差種:** Mouse, Rat, Human

製品の概要 Cytochrome c Release Assay Kit ab65311 provides an effective means for detecting

Cytochrome c Release Assay Kit ab65311 provides an effective means for detecting cytochrome c translocation from mitochondria into cytosol during apoptosis.

The kit provides reagents to isolate a highly enriched mitochondria fraction from cytosol. The procedure is simple and easy to perform; no ultracentrifugation is required and no toxic chemicals are involved.

Cytochrome c release from mitochondria into the cytosol is determined by Western blotting using the cytochrome c antibody provided in the kit.

The anti-Cytochrome c antibody is a mouse monoclonal antibody that reacts with denatured human, mouse, and rat cytochrome c.

Cytochrome c release assay protocol summary:

- collect cells, centrifuge and wash with PBS
- resuspend in cytosol extraction buffer mix
- homogenize cells with a dounce tissue grinder
- centrifuge homogenate at 700 x g for 10 min
- collect supernatant and centrifuge at 10,000 g for 30 min, collect supernatant as cytosolic fraction
- resuspend pellet in mitochondrial extraction buffer mix and save as mitochondrial fraction
- analyze cytosolic and mitochondrial fractions in western blotting with cytochrome c antibody

This product is manufactured by BioVision, an Abcam company and was previously called K257 Cytochrome c Releasing Apoptosis Assay Kit. K257-100 is the same size as the 100 test size of ab65311.

Cytochrome c plays an important role in apoptosis. The protein is located in the space between the inner and outer mitochondrial membranes. An apoptotic stimulus triggers the release of cytochrome c from the mitochondria into cytosol where it binds to Apaf-1. The cytochrome c/Apaf-

特記事項

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1 complex activates caspase-9, which then activates caspase-3 and other downstream caspases

#### Other apoptosis assays

For more apoptosis assays, review the apoptosis assay and apoptosis marker guide.

#### 製品の特性

#### 保存方法

Store at -20°C. Please refer to protocols.

内容	100 tests	100 tests
5X Cytosol Extraction Buffer I	1 x 20ml	1 x 20ml
Anti-Mouse Cyt C Antibody	1 x 100µl	1 x 100µl
DTT II	1 x 110µl	1 x 110µl
Mitochondria Extraction Buffer I	1 x 10ml	1 x 10ml
Protease Inhibitor Cocktail I	1 vial	1 vial

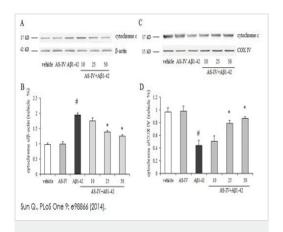
#### 関連性

Cytochrome c plays an important role in apoptosis. The protein is located in the space between the inner and outer mitochondrial membranes. An apoptotic stimulus triggers the release of cytochrome c from the mitochondria into cytosol where it binds to Apaf-1. The cytochrome c/Apaf-1 complex activates caspase-9, which then activates caspase-3 and other downstream caspases.

#### 細胞内局在

Mitochondrial

## 画像

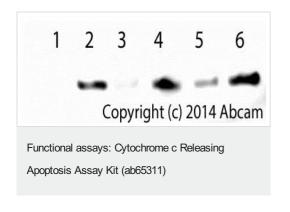


Inhibition of cytochrome c release from mitochondria

in SK-N-SH cells

Sun Q., PLoS One 9(6), Fig 4. doi: 10.1371/journal.pone.0098866. Reproduced under the Creative Commons license http://creativecommons.org/licenses/by/4.0/

Cytochrome C release was measured using Cytochrome C releasing apoptosis assay kit (ab65311). Blots showing immunoreactive bands for cytochrome c in cytosol (Image A). Data was expressed in fold-increase of cytochrome c compared to vehicle. Protein expression levels were normalized to  $\beta$ -actin (Figure B). Blots (Image C) of immunoreactive bands for cytochrome C in mitochondria. Figure D shows a fold-increase of cytochrome C compared to vehicle. Protein expression levels were normalized to COX IV.



5x10e7 Jurkat cells were cultured in the absence (1-2) or presence of 2 uM Camptothecin (CPT) (ab120115) for 24 hours (3-4) or with 10 uM CPT for 4 hours (5-6). 30 uL cytosolic (1, 3, 5) and mitochondrial (2, 4, 6) extracts were loaded onto the gel. Membranes were probed with anti-Cytochrome C Mouse MAb (ab65311) (dilution 1:200) followed by Goat Anti-Mouse IgG (HRP) (ab97040) (dilution 1:2000).

Bands were detected at the prediced size of 12 kDa.

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