# abcam

## Product datasheet

## ROS/Superoxide Detection Assay Kit (Cell-based) ab139476

69 References 画像数 3

#### 製品の概要

製品名 ROS/Superoxide Detection Assay Kit (Cell-based)

サンプルの種類 Adherent cells, Suspension cells

アッセイタイプ Cell-based

全工程の試験時間 Oh 90m

製品の概要 ROS/Superoxide Detection Assay Kit ab139476 is designed to directly monitor real time reactive oxygen species (ROS) production in live cells using fluorescence microscopy, flow cytometry, or

microplate reader.

The ROS/superoxide assay protocol is based on two fluorescent dyes: Oxidative Stress Detection Reagent (Green, Ex/Em 490/525 nm) for the detection of total ROS, and Superoxide Detection Reagent (Orange, Ex/Em 550/620 nm).

Through the combination of these two specific fluorescent probes, the kit provides a simple and specific assay for the real-time measurement of global levels of total reactive oxygen species (ROS), and of superoxides, in living cells. The kit is compatible with the major components of tissue culture media (phenol red, FBS and BSA).

ROS/superoxide assay protocol summary:

- add ROS inhibitor to negative control cells and incubate for 30 min
- add ROS/superoxide detection mix (with the addition of ROS inducer for positive control cells) and incubate for 30-60 min
- remove and discard the detection mix liquid
- wash samples and analyze with fluorescence microscope, or analyze by flow cytometry or microplate reader without washing

Reagents provided in the kit are sufficient for at least 200 tests using Fluorescent Microscopy or 50 tests using Flow Cytometry.

#### Related products

Review the <u>oxidative stress marker and assay guide</u>, or the full <u>metabolism assay guide</u> to learn about more assays for metabolites, metabolic enzymes, mitochondrial function, and oxidative stress, and also how to assay metabolic function in live cells using your plate reader.

To measure reactive oxygen species within cells, we recommend <u>DCFDA / H2DCFDA - Cellular ROS Assay Kit ab113851</u>. Alternative ROS assays are available in orange (<u>ab186028</u>), red (<u>ab186027</u>), and deep red (<u>ab186029</u>). <u>ab238535</u> is used to measure ROS in biofluids, culture

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supernatants and cell lysates.

For assays designed to differentiate ROS, superoxides, and reactive nitrogen species: to assay ROS and superoxides use ab139476; to assay ROS, superoxides, and reactive nitrogen species use **ab139473**; to assay superoxides use **ab219943**.

#### 試験プラットフォーム

Flow cytometer, Fluorescence microscope

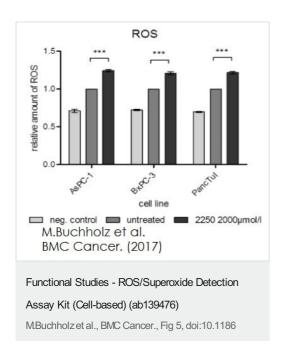
#### 製品の特性

#### 保存方法

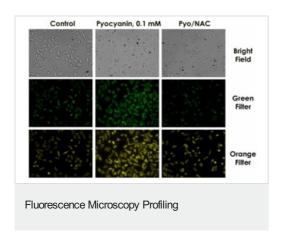
Please refer to protocols.

内容	1 kit
Oxidative Stress Detection Reagent (Green)	1 x 300nmole
ROS Inducer (Pyocyanin 1 μmole)	1 vial
ROS Inhibitor (N-acetyl-L-cysteine)	2 x 10mg
Superoxide Detection Reagent (Orange)	1 x 300nmole
Wash Buffer Salts	1 pack

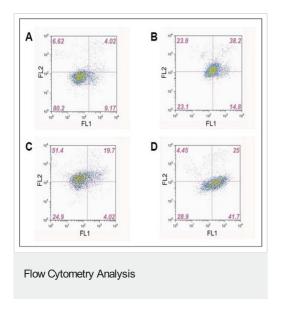
#### 画像



The level of ROS was analyzed in untreated compared to 2250 treated cells, additional NAC treatment served as a neg. Control



Profiling of reactive oxygen species formation by Fluorescence Microscopy was achieved in HeLa cells loaded with ROS/Superoxide detection reagents (ab139476) and treated with pyocyanin. General oxidative stress levels were monitored in the green channel, while superoxide production was detected in the orange channel. Pretreatment with NAC, a general ROS inhibitor, prevents formation of ROS.



Jurkat cells were induced with 100  $\mu$ M pyocyanin (general ROS inducer, panel B), 200  $\mu$ M antimycin A (superoxide inducer, panel C) or 1  $\mu$ M of t-butylhydroperoxide (peroxide inducer, panel D), stained with two color ROS Detection Kit and analyzed using flow cytometry. Untreated cells (panel A) were used as a control. Cell debris were ungated and compensation was performed using single stained pyocyanin-treated samples. Red numbers reflect the percentage of the cells in each quadrant.

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