abcam

Product datasheet

LDL Uptake Assay Kit (Cell-Based) ab133127

13 References 2 Images

Overview

Product name LDL Uptake Assay Kit (Cell-Based)

Detection method Fluorescent
Sample type Adherent cells
Assay type Cell-based
Assay time 6h 30m

Species reactivity Reacts with: Mammals, Other species

Product overview LDL Uptake Assay Kit (Cell-Based) (ab133127) provides a convenient tool for studying LDL

uptake and regulation at the cellular level. This kit employs Human LDL conjugated to DyLight™ 550 as a fluorescent probe for detection of LDL uptake into cultured cells. An LDL receptor-specific polyclonal antibody and a DyLight™ 488-conjugated secondary antibody are included for

identifying the distribution of LDL receptors.

LDL uptake assay protocol summary:

- remove culture medium from experimentally treated cells
- add LDL-Dylight 550 solution and incubate for 3-4 hrs
- replace solution with culture medium
- analyze LDL uptake with fluorescent microscope
- wash cells and fix for 10 min
- wash 3 times for 5 min
- add assay blocking solution and incubate for 30 min
- add anti-LDL receptor antibody and incubate for 1 hr
- wash 3 times for 5 min
- add Dylight 488 secondary antibody and incubate for 1 hr
- wash 3 times for 5 min
- analyze staining with fluorescence microscope

Notes LDL is the major carrier of cholesterol in the blood, accounting for more than 60% of total plasma

cholesterol. LDL is taken up by hepatic and extra-hepatic tissues through receptor mediated endocytosis triggered by apoB-100-LDL receptor interaction. The internalized LDL particle is

transported to lysosomes where it is degraded to free cholesterol and amino acids.

Platform Fluorescence microscope

Properties

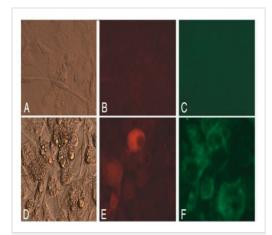
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Storage instructions

Please refer to protocols.

Components	1 kit
Cell-Based Assay Blocking Solution	1 vial
Cell-Based Assay Fixative	1 vial
DyLight 488-Conjugated Goat Anti-Rabbit Secondary Antibody	1 unit
LDL-DyLight 550	1 vial
Rabbit Anti-LDL Receptor Primary Antibody	1 vial

Images



Functional Studies - LDL Uptake Assay Kit (Cell-Based) (ab133127)

LDL uptake in pre-adipocytes and adipocytes.

Panel A: Light image of undifferentiated pre-adipocytes.

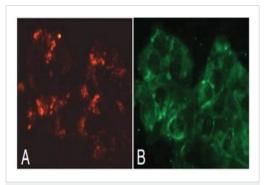
Panel B: Undifferentiated pre-adipocytes do not show any LDL uptake.

Panel C: Undifferentiated pre-adipocytes show little LDL receptor expression.

Panel D: Light image of differentiated adipocytes.

Panel E: Corresponding cells to those in Panel D showing uptake of LDL (red).

Panel F: Corresponding cells to those in Panel D showing expression of LDL receptor (green).



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LDL uptake in HepG2 cells.

HepG2 cells were cultured at a density of 3 x 10⁴ cells/well in a 96 well plate for two days then treated with 32.5µM EGCG overnight. LDL-Dylight™ 549 (10µg/ml) was added and the cells were incubated for an additional four hours. Cells were fixed and stained for LDL receptor using a Rabbit anti-LDL receptor primary antibody and Dylight™ 488-conjugated secondary antibody.

Panel A: LDL-Dylight™ 549 taken into cells appears in red.

Panel B: LDL receptors (in green) show a distribution pattern that matches the cells in the Panel A containing LDL-Dylight™ 549.

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