abcam

Product datasheet

Fluo-8 Calcium Flux Assay Kit - No Wash ab112129

42 References 画像数 1

製品の概要

製品名 Fluo-8 Calcium Flux Assay Kit - No Wash

検出方法 Fluorescent

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 1h 00m

製品の概要 Fluo-8 Calcium Flux Assay Kit (ab112129) is a no-wash, fluorescence-based assay for detecting intracellular calcium mobilization in cells from mammals and other species.

In the calcium flux assay protocol, cells are pre-loaded with Fluo-8 which can cross the cell membrane. Once inside the cell, the lipophilic blocking groups of Fluo-8 are cleaved by an esterase, resulting in a negatively charged fluorescent dye that stays inside the cell. Its fluorescence is greatly enhanced upon binding to calcium.

If intracellular calcium levels increase, the fluorescence of Fluo-8 is significantly increased. The characteristics of its long wavelength, high sensitivity, and >100 times fluorescence enhancement make Fluo-8 the brightest green calcium indicator available, and it is an ideal tool for the measurement of cellular calcium through HTS screening.

This calcium flux assay provides an optimized assay method for monitoring G-protein-coupled receptors and calcium channels using HTS instrumentation. The assay can be performed in a convenient 96-well or 384-well microtiter-plate format.

Calcium flux assay protocol summary:

- add Fluo-8 dye solution to cells
- incubate for 30 min at 37°C, and then for 30 min at room temp
- analyze fluoresence at Ex/Em 490/525 nm

Fluo-8 products available

Fluo-8 Calcium Flux Assay Kits are available in No Wash and Medium Removal variants:

- No wash kit Fluo-8 Calcium Flux Assay Kit ab112129 is generally preferred as the workflow is simpler. It is also recommended for cell lines that are only weakly adherent and risk losing adherence when washing.

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- Medium removal kit Fluo-8 Calcium Flux Assay Kit <u>ab112128</u> is recommended for the testing of compounds, receptors and calcium channels that are sensitive to the presence of cell culture media or serum. Alternatively, for cells that can tolerate it, consider using a low serum % in culture media and use No Wash Fluo-8 kit ab112129.

Fluo-8 is also available as a standalone biochemical ab142773.

特記事項

This product is intended to be used for monitoring calcium fluctuations *in vivo* in live cells using the following HTS imaging plate readers: FLIPR™, FDSS, BMG NOVOstar™, FLexStation, ViewLux, IN Cell Analyzer or Arrayscan.

Readers with an in built pipetter are essential for measuring calcium response accurately due to transient property of the calcium signal. Depending on dose of the agonist, the peak response is few seconds to 20 seconds, so one will need to add the compound by instrument and read the fluorescent signal simultaneously every second for ~1-2 min.

If you would like to quantify calcium concentration *in vitro* using cell extracts, we recommend using <u>Calcium Detection Kit (Colorimetric) (ab102505)</u> or <u>Calcium Quantification Assay Kit</u> (ab112115).

試験プラットフォーム

Microplate reader

製品の特性

保存方法

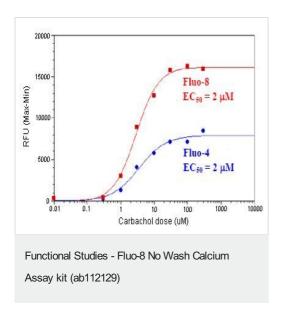
Store at -20°C. Please refer to protocols.

内容	96 tests
10X Pluronic® F127 Plus	1 x 1ml
Fluo-8	1 vial
HHBS	1 x 9ml

関連性

Calcium is essential for all living organisms, where Ca²⁺ sequestration and release into and out of the cytoplasm functions as a signal for many cellular processes. 99% of calcium is found in bones and teeth with the remaining 1% found in the blood and soft tissue. Serum calcium levels are tightly controlled (8.4-11.4 mg/dL) and any variation outside this range can have serious effects. Calcium plays a role in mediating the constriction and relaxation of blood vessels, nerve impulse transmission, muscle contraction, and hormone secretion. Calcium ion channels control the migration of calcium ions across cell membranes, permitting the activation and inhibition of a wide variety of enzymes. Causes of low calcium levels include chronic kidney failure, vitamin D deficiency, and low blood magnesium levels that can occur in severe alcoholism.

画像



Carbachol Dose Response was measured in HEK293 cells with ab112129 and a Fluo-4 calcium assay kit. HEK293 cells were seeded overnight at 40,000 cells/100 μ L/well in a black wall/clear bottom 96-well plate. The cells were incubated with 100 μ L of dyeloading solution using the ab112129 or the Fluo-4 kit for 1 hour at room temperature. Carbachol (50 μ L/well) was added to achieve the final concentrations indicated.

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