

HDAC fluorometric cellular activity assay Kit

Cat. No. Kit-0415 **Lot. No.** (See product label)

SPECIFICATION

Product Overview

Size 96 wells

Description

The HDAC fluorometric cellular activity assay allows the determination of deacetylase activity within an undisturbed cellular environment and provides accurate activity information that is reflective of endogenous regulation. It also allows the study of the effects of upstream regulators on deacetylase activity and the detection of inhibitors or activators that act indirectly to affect deacetylase activity. The FLUOR DE LYS HDAC substrate is cell permeable. It is deacetylated by cellular HDACs and thus suitable for high-throughput cell-based deacetylase assays. This kit provides the reagents and protocols needed for determining rates of intracellular deacetylase activity with cultured cells. Deacetylation can be quantitated by addition of developer to the media and lysed cells.

Applications

Fluorescence microscopy, HTS

Storage

-80°C

Kit Components

Nuclear Extracts from Hela Cells (human cervical cancer cell line) (50 µl 2 mg protein/ml in 0.1M potassium chloride, 20mM HEPES/sodium hydroxide, pH 7.9, 20% (v/v) glycerol, 0.2mM ethylenediaminetetraacetic acid, 0.5M dithiothreitol, 0.1mM PMSF; Prepared according to a modification of J.D. Dignam et al (1983) and S.M. Abmayr et al. (1988))Storage: -70°C, avoid freeze/thaw cycles!FLUOR DE LYS Substrate (50 µl 50mM in DMSO)Storage: -70°CFLUOR DE LYS Developer

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Concentrate (20x) (300 μ l 20x stock solution, dilute in assay buffer before use)Storage: -70°C
Trichostatin A (HDAC Inhibitor) (100 μ l 0.2mM in DMSO) Storage: -70°C
Nicotinamide (Sirtuin Inhibitor) (500 μ l 50mM nicotinamide in 50mM TRIS/Cl, pH 8.0, 137mM sodium chloride, 2.7mM potassium chloride, 1mM magnesium chloride)Storage: -70°C
FLUOR DE LYS Deacetylated Standard (30 μ l 10mM in DMSO)Storage: -70°C
HDAC Assay Buffer (50mM TRIS/Cl, pH 8.0, 137mM sodium chloride, 2.7 mM potassium chloride, 1mM magnesium chloride) (20 ml)Storage: -70°C
Cell Lysis Buffer (20 ml; 1.0% NP-40 in HDAC Assay Buffer)Storage: -70°C
(2) 1/2 volume microplatesStorage: Room temperature (One sterile, One non-sterile)

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