CREATIVE BIOMART®
Assay Kit

NADPH Fluorimetric Assay Kit (Red)

Product Information

Cat.No. Kit-2050

Product Overview

This Fluorimetric NADPH Assay Kit provides a convenient method for the detection of NADPH. The enzymes in the system specifically recognize NADPH in an enzyme recycling reaction. In addition, this assay has very low background since it is run in the red visible range that significantly reduces the interference from biological samples.

Size

400 assays in 96-well plates

Description

Nicotinamide adenine dinucleotide (NAD+) and nicotinamide adenine dinucleotide phosphate (NADP+) are two important cofactors found in cells. NADH is the reduced form of NAD+, the oxidized form of NADH. NAD forms NADP with the addition of a phosphate group to the 2" position of the adenyl nucleotide through an ester linkage. NADP is used in anabolic biological reactions, such as fatty acid and nucleic acid synthesis, which requires NADPH as a reducing agent. In chloroplasts, NADP is an oxidizing agent important in the preliminary reactions of photosynthesis. The NADPH produced by photosynthesis is used as reducing power for the biosynthetic reactions in the Calvin cycle of photosynthesis. The traditional NAD/NADH and NADP/NADPH assays are done by monitoring the changes in NADH or NADPH absorption at 340 nm. This method suffers low sensitivity and high interference since the assay is done in the UV range that requires expensive quartz microplate.

Applications

The assay can be performed in a convenient 96-well or 384-well microtiter-plate format. Its signal can be easily read by either a fluorescence microplate reader at Ex/Em = 540/590 nm or an absorbance microplate reader at ~576 nm.This kit has been used for screening enzyme activities that use NADP/NADPH as a cofactor. It has also been used for the sensitive detection of NADPH in cell-based assays. Compared to the other commercial kits, this assay has higher signal/background ratio.

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Storage

Keep Component A in freezer (-20°C) and avoid exposure to light; Component C at -20°C; Component B &G at 4°C.

Kit Components

Component A: NADPH Recycling Enzyme Mixture, 2 bottles (lyophilized powder).Component B: NADPH Assay Buffer, 1 bottle (20 ml).Component C: NADPH Standard (FW: 833.36), 1 vial (167 µg).Component G: NADP/NADPH Lysis Buffer, 1 bottle (10 ml).

Detection method Fluorescence microplate reader (Ex/Em = 540/590 nm)

Features & Benefits

Broad application: NADPH detection in solution or cell extracts.Sensitive: Detect as low as 10 nanomoles of NADPH in solution.Continuous: Easily adapted to automation without a separation step.Convenient: Formulated to have minimal hands-on time. No wash required.Non-radioactive: No special requirement for waste disposal.

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