

COX Fluorescent Activity Assay Kit

Product Information

Cat.No.

Kit-0253

Product Overview

COX Fluorescent Activity Assay provides a convenient fluorescence-based method for detecting COX-1 or COX-2 activity in both crude (cell lysates/tissue homogenates) and purified enzyme preparations. The assay utilizes the peroxidase component of COXs. In this assay, the reaction between PGG₂ and ADHP (10-acetyl-3,7-dihydroxyphenoxazine) produces the highly fluorescent compound resorufin that can be analyzed using an excitation wavelength of 530-540 nm and an emission wavelength of 585-595 nm. The kit includes isozyme-specific inhibitors for distinguishing COX-2 activity from COX-1 activity.

Description

Cyclooxygenase (COX, also called Prostaglandin H Synthase or PGHS) is a bifunctional enzyme exhibiting both COX and peroxidase activities. The COX component converts arachidonic acid to a hydroperoxy endoperoxide (PGG₂), and the peroxidase component reduces the endoperoxide to the corresponding alcohol (PGH₂), the precursor of PGs, thromboxanes, and prostacyclins. It is now well established that there are two distinct isoforms of COX. COX-1 is constitutively expressed in a variety of cell types and is involved in normal cellular homeostasis. A variety of mitogenic stimuli such as phorbol esters, lipopolysaccharides, and cytokines lead to the induced expression of a second isoform of COX, COX-2. COX-2 is responsible for the biosynthesis of PGs under acute inflammatory conditions. This inducible COX-2 is believed to be the target enzyme for the anti-inflammatory activity of nonsteroidal antiinflammatory drugs.

Usage

Please read these instructions carefully before beginning this assay. For research use only. Not for human or diagnostic use.

Storage

Stability: 1 year; Storage: -80°C; This kit will perform as specified if stored as directed in the Materials

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Supplied section, and used before the expiration date indicated on the outside of the box.

Kit Components

Kit components may be stored at -80°C prior to use. After opening kit, we recommend each kit component be stored according to the temperature listed below. Assay Buffer (10X): 2 vials, -20°C; Heme: 1 vial, -80°C; COX Positive Control: 1 vial, -80°C; Arachidonic Acid (substrate): 1 vial, -20°C; Potassium Hydroxide: 1 vial, -20°C; ADHP Assay Reagent: 4 vials, -20°C; DuP-697: 1 vial, -20°C; SC-560: 1 vial, -20°C; DMSO Assay Reagent: 1 vial, Room temperature; Resorufin Standard: 100 µl, -20°C; 96-Well Solid Plate (black): 2 plates, Room temperature; Plate Cover: 2 covers, Room temperature