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Product datasheet

Cytochrome P450 2C9 (CYP2C9) Inhibitor Screening Kit (Fluorometric) ab284519

3 Images

Overview

Product name	Cytochrome P450 2C9 (CYP2C9) Inhibitor Screening Kit (Fluorometric)
Detection method	Fluorescent
Sample type	Inhibitor compounds
Assay type	Enzyme activity
Assay duration	Multiple steps standard assay
Product overview	The CYP2C9 Inhibitor Screening Kit (ab284519) (K896) enables rapid screening of drugs and other new chemical entities (NCEs) for compound-CYP2C9 interaction in a reliable, high-throughput fluorescence-based assay. The kit provides a yeast microsomal preparation of human CYP2C9 and human cytochrome P450 reductase (CPR) enzymes. The assay utilizes a non-fluorescent CYP2C9 substrate that is converted into a highly fluorescent metabolite detected in the visible range (Ex/Em = 415/502 nm), ensuring a high signal-to-background ratio with little interference by autofluorescence. The kit contains a complete set of reagents sufficient for performing 100 reactions in a 96-well plate format. General range = 0-20 pmol/well.
Notes	This product is manufactured by BioVision, an Abcam company and was previously called K896 Cytochrome P450 2C9 (CYP2C9) Inhibitor Screening Kit (Fluorometric). K896-100 is the same size as the 100 test size of ab284519.
Platform	Microplate (12 x 8 well strips)

Properties

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

Components	100 tests
7-HFC Standard	1 vial

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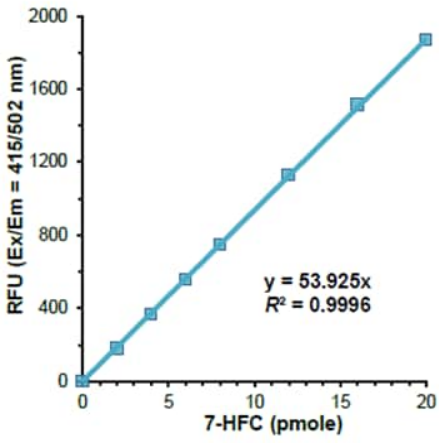
beta-NADP+ Stock (100X)	1 vial
CYP2C9 Assay Buffer	1 x 100ml
CYP2C9 Inhibitor (Sulfaphenazole)	1 vial
CYP2C9 Substrate	1 vial
NADPH Generating System (100X)	1 vial
Recombinant Human CYP2C9	2 vials

Function Cytochromes P450 are a group of heme-thiolate monooxygenases. In liver microsomes, this enzyme is involved in an NADPH-dependent electron transport pathway. It oxidizes a variety of structurally unrelated compounds, including steroids, fatty acids, and xenobiotics. This enzyme contributes to the wide pharmacokinetics variability of the metabolism of drugs such as S-warfarin, diclofenac, phenytoin, tolbutamide and losartan.

Sequence similarities Belongs to the cytochrome P450 family.

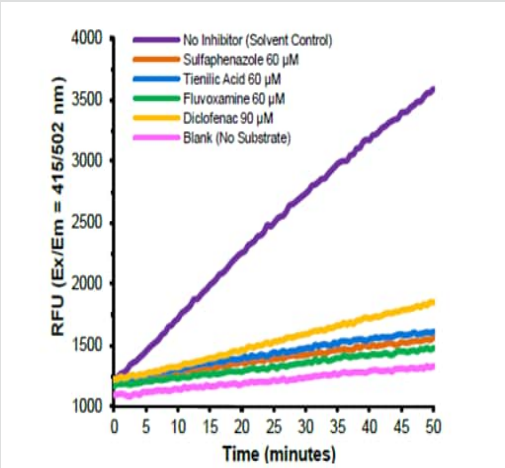
Cellular localization Endoplasmic reticulum membrane. Microsome membrane.

Images



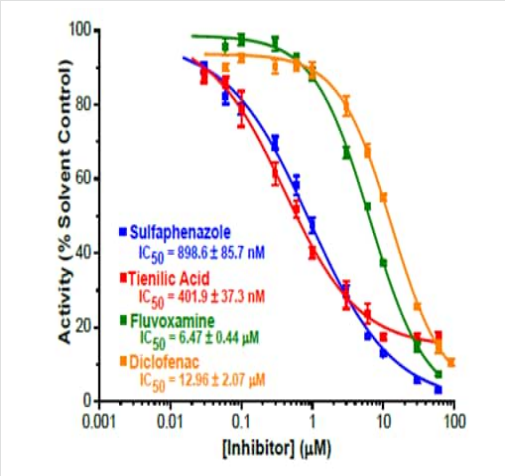
7-HFC standard curve

Relative fluorescence units and 7-HFC (7-hydroxy-4-(trifluoromethyl)coumarin) standard curve. One mole of 7-HFC corresponds to the metabolism of one mole of CYP2C9 substrate.



Reaction kinetics of CYP2C9

Recombinant human CYP2C9 enzyme and its reaction kinetics at 37°C in the presence and absence of the indicated CYP2C9 inhibitors. The solvent control reaction contained assay buffer with 0.3% acetonitrile.



Dose-reponse curve of various CYP2C9 ligands

Dose-response curves for various CYP2C9 ligands of differing structural and mechanistic classes. The results from canonical competitive CYP2C9 inhibitor sulfaphenazole, the mechanism-based irreversible inhibitor tienilic acid, the antidepressant fluvoxamine (a competitive inhibitor of several CYP isoforms) and the CYP2C9-selective substrate diclofenac are presented in the graph. For dose-response curves, percent activity was calculated for each concentration of inhibitor by comparison to activity of reactions containing no inhibitor. For each CYP2C9 inhibitor, IC50 values were derived by 4- parameter logistic curve fitting with each point representing the mean ± SEM of at least three replicates. Assays were performed according to the kit protocol.

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