



emulate

Protocol for Emulate Organ-Chips:

Live Staining of CDFDA Uptake into Bile Canaliculi

May 10, 2019

EP195 v1.0

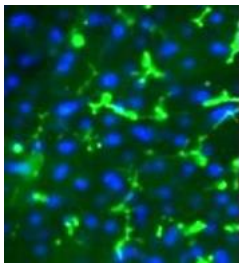
TITLE Live Staining of CDFDA Uptake into Bile Canaliculi	DOCUMENT EP195	VERSION 1.0
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Goals:	Key Steps:	Other Required Materials:
Visualize bile canaliculi structure and MRP2 transporter function in Emulate Organ-Chips	<ul style="list-style-type: none"> Live staining and fluorescent imaging in Liver-Chips 	<ul style="list-style-type: none"> Carboxy-DCFDA (CDFDA) (Invitrogen, Cat # C369) PBSPBS Fluorescence microscope

Introduction

Carboxy-DCFDA (5-(and-6)-Carboxy-2',7'-Dichlorofluorescein Diacetate) is a reagent that passively diffuses into cells. It is colorless / nonfluorescent and is cleaved by intracellular esterases to yield a fluorescent fluorophore, 5-(and-6)-carboxy-2',7'-dichlorofluorescein (CDFDA). This reagent can be used to visualize the formation of bile canaliculi in polarized hepatocytes which is indicative of MRP-2 transporter activity. Carboxy-DCFDA is a substrate of multidrug resistance-associated protein 2 (MRP2), a hepatocyte efflux transporter, thus it can be used to visualize MRP2-mediated canalicular uptake and efflux, as well as to label the bile canaliculi structures.

Method

Sample type	Live Liver-Chip See Protocol EP155 Live Staining of Cells.
Recommended reagent dilution and incubation time	Dilute 5 mM stock CDFDA solution 1:500 in culture medium. Incubate in the dark at 37°C for 30 minutes.
Representative image	 <p>Image of CDFDA (green) and nuclei staining (blue) indicating hepatocyte polarization, establishment of bile canaliculi structures, and active MRP2 transporters in hepatocytes in the human Liver-Chip (top channel).</p>
More information on vendor site	https://www.thermofisher.com/order/catalog/product/C369?SID=srch-hj-c369

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