



emulate

Protocol for Emulate Organ-Chips

Immunofluorescence Staining of Vimentin

April 23, 2019

EP150 v1.0

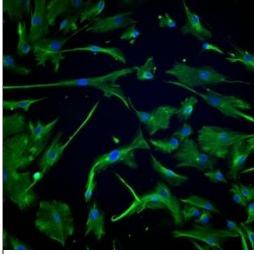
TITLE Immunofluorescence Staining of Vimentin	DOCUMENT EP150	VERSION 1.0
	DATE 23-APR-2019	PAGE 2 OF 3

Goals:	Key Steps:	Other Required Materials:
Visualize vimentin via fluorescent imaging in fixed Emulate Organ-Chips	<ul style="list-style-type: none"> Immunofluorescence staining in Organ-Chips 	<ul style="list-style-type: none"> Anti-vimentin antibody [SP20] (Abcam ab16700) 4% paraformaldehyde (PFA) 10% Saponin PBS BSA Normal goat serum (or other serum from the species the secondary antibody was raised in) Alexa Fluor® 488-conjugated goat anti-rabbit IgG secondary antibody (or other anti-rabbit secondary) Fluorescence microscope

Method

Sample type	Fixed Organ-Chip See Emulate Protocol EP137 Fixation and Immunofluorescence (IF) Staining.
Recommended fixative and incubation time	4% PFA, 15 mins at room temperature
Recommended permeabilization and incubation time	1% Saponin in PBS, 30 minutes at room temperature
Recommended blocking buffer and incubation time	1% BSA, 10% goat serum in PBS, overnight at 4°C
Recommended primary antibody dilution and incubation time	Anti-Vimentin [SP20] (Abcam ab16700) 1:1000 dilution in blocking buffer, overnight at 4°C
Recommended antibody host	Rabbit
Recommended secondary antibody dilution and incubation time	Alexa Fluor® 488-conjugated goat anti-rabbit IgG secondary antibody 1:500 dilution in blocking buffer, 2 hours at room temperature in the dark.

TITLE Immunofluorescence Staining of Vimentin	DOCUMENT EP150	VERSION 1.0
	DATE 23-APR-2019	PAGE 3 OF 3

Representative image		Image of human stellate cells stained by vimentin (green) in the human Liver-Chip (bottom channel)
More information on vendor site	https://www.abcam.com/vimentin-antibody-sp20-ab16700.html	

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