

Protocol for Emulate Organ-Chips:

Creatinine Quantification Assay

July 2, 2019

EP170 v1.0



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Goals:	Key Steps:	Other Required Materials:
Quantify creatinine levels from Emulate Organ-Chip effluent samples	 Prepare all reagents, samples, and standards Run the assay Read the plate 	 Invitrogen[™] Creatine Urinary Detection Kit (Thermo Fisher EIACUN) Creatinine (Sigma, C4255) Distilled or deionized water Microtiter plate reader

Introduction

Creatinine, a non-protein nitrogenous (NPN) waste product, is produced from the breakdown of creatine and phosphocreatine. Creatinine levels can be used as an indicator of renal function.

To measure creatinine secretion in the Proximal Tubule Kidney-Chip, the vascular channel (bottom channel) is perfused with medium containing 1mg / dL of human creatinine, while the secretion of creatinine in the epithelial channel (top channel) is measured from chip effluent that collects in outlet reservoirs of the Pod[™] portable module.

This protocol uses the Proximal Tubule Kidney-Chip as a reference point. These methods and assay conditions could change with different Organ-Chips.

Method

Sample type	Organ-Chip effluent	
Sample type	See protocol EP124 Effluent Sampling.	
Recommended assay flow rate (Proximal Tubule Kidney-Chip)	60 μL / h	
	No dilution. Samples are loaded neat.	
Recommended effluent dilution (Proximal Tubule Kidney-Chip)	Note: Creatinine levels will change depending on cell viability and functional status, or based on donor-to-donor variability. Hence, sample dilution may need to be modified to accommodate different experimental conditions or cells from different donors. It is therefore recommended to run a preliminary study in order to define optimal dilution factors and timepoints for media sampling.	
Run assay as described	https://www.thermofisher.com/order/catalog/product/EIACUN	
on supplier site	Note: Store kit at -20°C immediately upon receipt.	
Sample concentration range (Proximal Tubule Kidney-Chip)	TBD	



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