HPLC Application ID No.: 21874

Human Plasma TMP & Thiamine by Gemini-NX 3u C18 100x3mm with Impact Plate_200nmol

Column:	Gemini [®] 3 µm NX-C18 110 Å, LC Column 100 x 3 mm, Ea							
Dimensions:	100 x 3 mm ID							
Order No:	00D-4453-Y0							
Elution Type:	Gradient							
Eluent A:	25mM Na2HPO4, 10% methanol							
Eluent B:	25mM Na2HPO4, 70% methanol							
Gradient	Step No.	Time (min)	Pct A	Pct B				
Profile:	1	0	3	97				
	2	0.25	25	75				
	3	0.75	25	75				
	4	3	35	65				
	5	4	100	0				
	6	5	100	0				
	7	5.1	3	97				
	8	8	3	97				
Flow Rate:	0.75 mL/mi	n						
Col. Temp.:	ambient							
Detection:	Fluorescence (FLUOR) @ Ex 375 nm (ambient)							
Detector Info:								
21874	Shimadza i Eb							
	_							
mAU _	мт	P						
68 -								
-								
66 -		Thiamine						
64 -								
-								
62 -								
60—								
-								
58 -								
-								
56 _								
54 -								
	<u> </u>		m.	_				
52 _		1 1 1						
0	2	4	6	8 min				



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Products used in this application:



ANALYTES:

1 TMP

Retention Time: 2.517 min

2 Thiamine

Retention Time: 4.128 min

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PRODUCT DESCRIPTION:

Impact[™] Protein Precipitation, 2mL Square Well Filter Plate, 2/Pk

Order No.: CE0-7565

SOLID PHASE EXTRACTION (SPE) PRODCEDURE:

Note: The solvent volumes shown below are for a Proprietary bed mass.

The solvent volumes will need to be adjusted for a smaller or larger bed mass.

Condition:

Load: Wash: Dry: Elute:

Final Prep and Analysis:

Inject: 20 µL on HPLC Fluorescence (FLUOR) @ Ex 375 nm (ambient)

ANALYTES:	Spiked Conc. (ng/mL)	Log P	рКа	% Rec	%RSC (n=0)
1 TMP	200				
2 Thiamine	200				

Note: This method is designed as a convenient starting point for further investigation and can be tailored to meet your extraction goals. Call your local Phenomenex Representative for assistance in method development and optimization techniques.

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