

HPLC-UV Method for the Determination of Procainamide from Human Plasma Using SOLA CX

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Abstract

A HPLC-UV method has been developed for the determination of procainamide and sulfanilamide in human plasma. This application note demonstrates the use of the Thermo Scientific SOLA CX cartridges which allow for faster methods and more reproducible results compared with conventional loose-packed cartridges.

Introduction

SOLA™ products are a revolutionary new Solid Phase Extraction (SPE) product range. This first in class SPE product range introduces next-generation, innovative technological advancements, giving unparalleled performance characteristics compared to conventional SPE, phospholipid and protein precipitation products.

This includes:

- Higher levels of reproducibility
- Higher levels of extract cleanliness
- Reduced solvent requirements
- Increased sensitivity

SOLA products have significant advantages for the analyst when processing compounds in complex matrices particularly in high throughput bioanalytical and clinical laboratories where reduced failure rate, higher analysis speed and lower sample/solvent requirements are critical.

The increased performance from SOLA products provides higher confidence in analytical results and lowers cost without compromising ease of use or requiring complex method development.

One of the key goals for the chromatographer is to achieve a consistent, reproducible separation. The selection of a highly reproducible HPLC column is essential if this goal is to be attained. The Thermo Scientific Synchronis column range has been engineered to provide exceptional reproducibility due to its highly pure, high surface area silica, dense bonding and double endcapping, all controlled and characterized through the use of rigorous testing.

Procainamide is an antiarrhythmic agent and is prescribed for the treatment of severe irregular heartbeats. It helps the heart to maintain a regular rhythm by slowing down the rate nerve impulses are conducted through the heart.

This application note demonstrates the successful extraction of procainamide and sulfanilamide from human plasma using SOLA CX cartridges and subsequent separation using a Synchronis C18 column.



Experimental Details

Chemicals and Reagents	Part Number
Fisher Scientific HPLC grade ammonium acetate	A/3446/50
Fisher Scientific HPLC grade water	W/0106/17
Fisher Scientific HPLC grade methanol	M/4056/17
Procainamide and sulfanilamide	

Sample Handling Equipment	Part Number
Thermo Scientific HyperSep glass block manifold	60104-232
Thermo Fisher Scientific Ultra Vap	CLS-229070
NSC Mass Spec Certified 2 mL clear vial with blue bonded PTFE silicone cap	MSCERT4000-34W

Sample Preparation - SOLA CX	Part Number
Compound(s):	procainamide and sulfanilamide (IS)
Matrix:	Human plasma
Cartridge type:	Thermo Scientific SOLA CX 10 mg / 1 mL
Conditioning stage:	1 mL methanol, 1 mL water
Application stage:	250 µL spiked human plasma
Washing stage 1:	250 µL water + 2 % formic acid
Washing stage 2:	250 µL methanol + 2 % formic acid
Elution stage:	250 µL methanol + 5 % ammonia
Additional stage:	Dry down under a gentle stream of nitrogen and reconstitute in 250 µL water

Sample Preparation – Loose-Packed	Part Number
Compound(s):	Procainamide and sulfanilamide (IS)
Matrix:	Human plasma
Cartridge type:	Loose-packed SPE products
Conditioning stage:	1 mL methanol, 1 mL water
Application stage:	500 µL spiked human plasma
Washing stage 1:	500 µL water + 2 % formic acid
Washing stage 2:	500 µL methanol + 2 % formic acid

Key Words

- Procainamide
- Sulfanilamide
- SOLA CX Cartridges and Plates
- Synchronis C18

Elution stage:	500 µL methanol + 5 % ammonia
Additional stage:	Dry down under a gentle stream of nitrogen and reconstitute in 500 µL water

Separation Conditions	Part Number
Instrumentation: Thermo Scientific HPLC system	
Column: Synchronis C18 5 µm, 100 x 4.6 mm	97105-104630

Mobile Phase

A:	20 mM ammonium acetate	
B:	methanol	
Gradient:	Time (minutes)	% B
	0.0	5
	1.0	5
	5.0	95
	5.5	95
	8.0	5

Flow rate:	1 mL/min
Column temperature:	30 °C
Injection details:	10 µL partial loop
Injection wash solvent:	20 mM ammonium acetate
UV detector wavelength:	277 nm

Solutions

Stock standard contained 1 mg/mL of procainamide and sulfanilamide in acetonitrile.
Working standard contained 10 µg/mL of procainamide and sulfanilamide in human plasma.

Results

The analysis was performed on a Synchronis C18 5 µm, 100 x 4.6 mm column. As shown in Figure 1 sulfanilamide (IS) and procainamide were separated in under 5 minutes. SOLA CX 10 mg / 1 mL cartridges were used for the extraction of sulfanilamide (IS) and procainamide from human plasma. Table 1 shows the results from six replicate extractions using the SOLA CX cartridges compared with conventional loose-packed SPE products.

	Sulfanilamide (IS)	Procainamide
% Recovery SOLA CX	77.8	119
% Recovery Loose Packed SPE	97.3	106
% RSD SOLA CX	11.0	4.03
% RSD Loose Packed SPE	21.0	10.7

Table 1: Recovery and precision (% RSD) results for sulfanilamide and procainamide extracted from human plasma using SOLA CX 10 mg / 1 mL cartridges and loose packed SPE products (data calculated from six replicate injections)

Conclusion

This application note has shown that SOLA CX cartridges can be successfully used to extract procainamide and sulfanilamide from human plasma. The SOLA CX cartridges produce reproducible results with greater precision obtained compared to the loose-packed products. SOLA CX cartridges require lower elution volumes in comparison to the traditional loose-packed products demonstrating that SOLA CX cartridges uses less solvent, reduces analysis time and increases sample throughput.

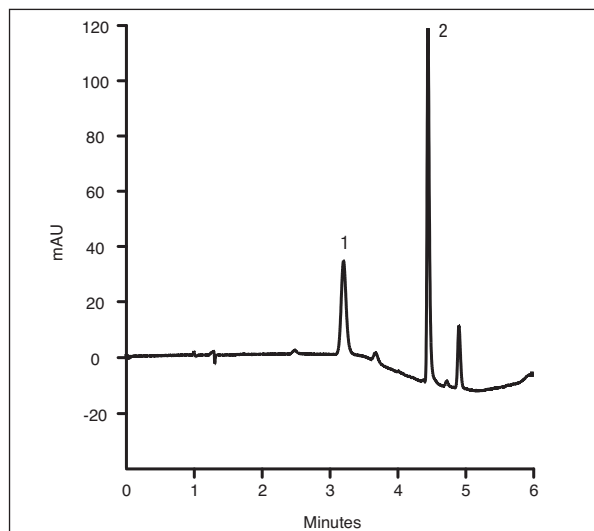


Figure 1: Chromatogram of sulfanilamide (1) and procainamide (2) extracted from human plasma using SOLA CX 10 mg / 1 mL cartridges and separated using a Synchronis C18 5 µm, 100 x 4.6 mm column

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