

Steroids in Serum

Waters Corporation



This is an Application Brief and does not contain a detailed Experimental section.

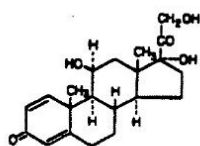
Abstract

This application brief demonstrates analysis of steroids in serum.

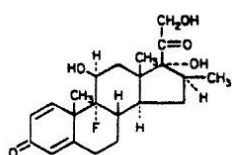
Introduction

The compounds used in this study are –

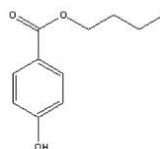
1. Prednisolone
2. Betamethasone
3. Butyl Paraben (I.S.)
4. Betamethasone Valerate



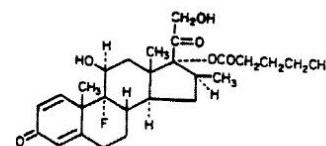
1. Prednisolone



2. Betamethasone



3. Butyl Paraben (I.S.)



4. Betamethasonevalerate

Experimental

HPLC Method

Column:	Symmetry Shield RP ₈ , 3.9 x 150 mm, 5 μm
Guard column:	Sentry Guard Column 3.9 x 20 mm, 5μm
Part numbers:	Column - WAT200655, Guard - WAT200675
Mobile phase:	20 mM potassium phosphate, pH 7/methanol 41:59
Flow rate:	1.0 mL/min

Injection volume:

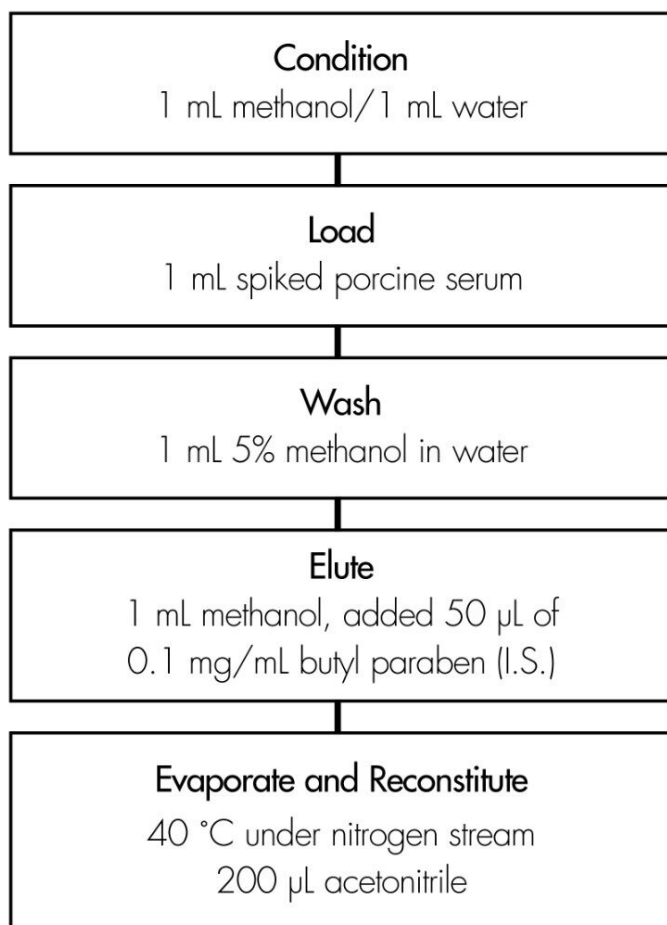
5 μ L of reconstituted porcine serum extract

Detection:

UV @ 254 nm

Oasis[®] HLB Extraction Method

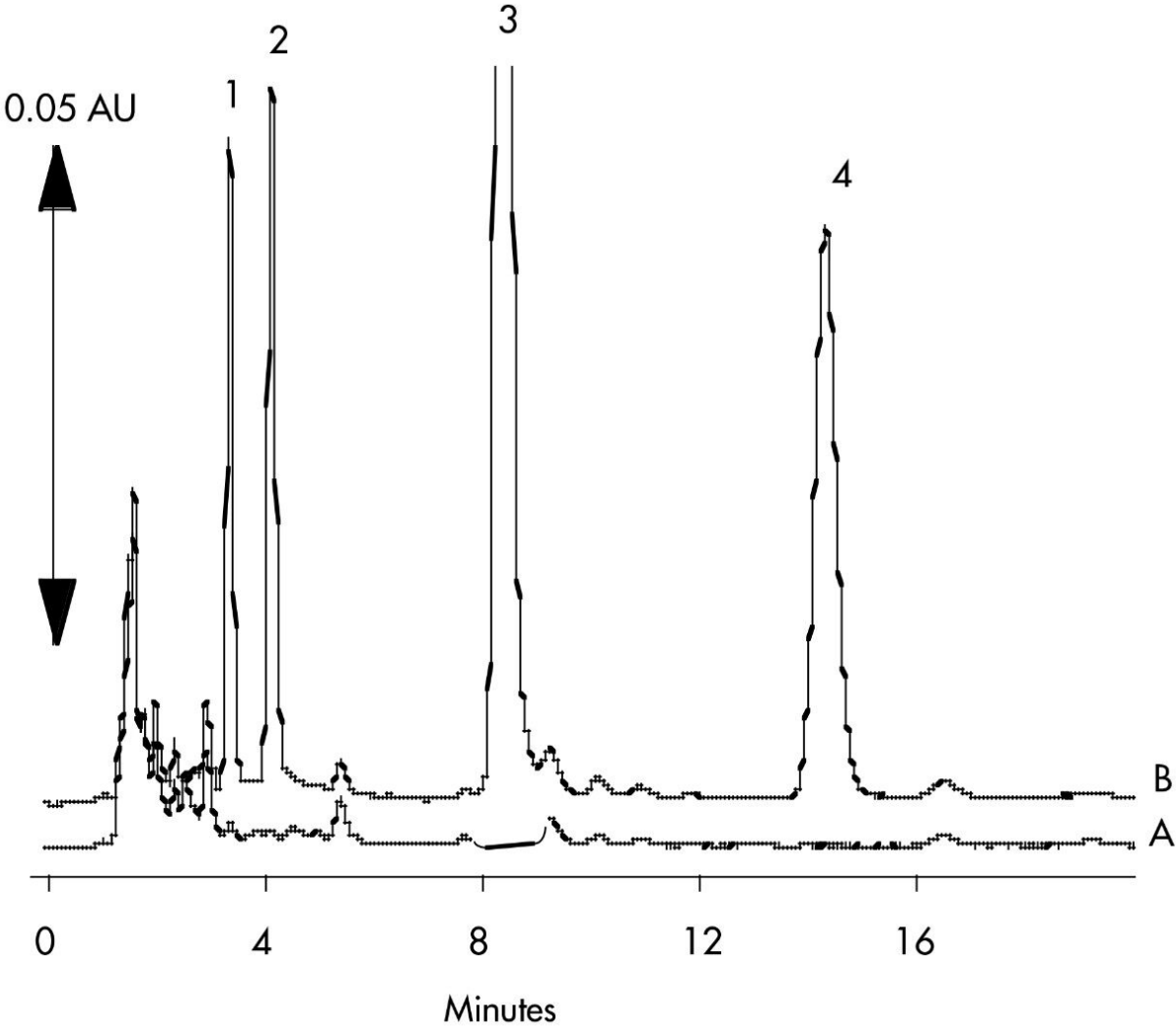
Oasis[®] HLB 1 cc/30 mg Extraction Cartridge
Part Number WAT094225



Results and Discussion

Compound	Concentration $\mu\text{g/mL}$	% Recovery	%RSD (n=6)
Prednisolone	1.00	93.8%	4.7%
	5.00	92.0%	1.4%
Betamethasone	1.00	85.7%	2.3%
	5.00	91.3%	1.5%
Betamethasone	4.00	95.4	2.2%
Valerate	20.00	9.06	1.3%

Chromatogram of Serum Extracts: A) Blank B) Spiked Sample



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