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アプリケーションノート

ACQUITY™ UPLC™ I-Class SM-FL/Xevo™ TQ Absolute System: Analytical Performance for Free Testosterone

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本書はアプリケーションブリーフであり、詳細な実験方法のセクションは含まれていません。

Abstract

The Waters™ ACQUITY UPLC I-Class SM-FL/Xevo TQ Absolute IVD System enables the quantification of organic compounds in human biological liquid matrices.

This document describes a test of the analytical performance of the ACQUITY UPLC I-Class SM-FL/Xevo TQ Absolute IVD System for the analysis of free testosterone in serum.

Introduction

The Waters™ ACQUITY UPLC I-Class SM-FL/Xevo TQ Absolute IVD System enables the quantification of organic compounds in human biological liquid matrices.

This document describes a test of the analytical performance of the ACQUITY UPLC I-Class SM-FL/Xevo TQ Absolute IVD System for the analysis of free testosterone in serum.

Experimental

The ACQUITY UPLC I-Class SM-FL/Xevo TQ Absolute IVD System was controlled by MassLynx™ Software (v4.2) and the data processed using the TargetLynx™ XS Application Manager. Calibrators were prepared in a 52.75 mM HEPES buffer adjusted to pH 7.4, to match the constitution of the diasylate obtained during sample preparation. Samples were processed using the following conditions:

Sample Preparation Conditions

200 µL sample was processed using an equilibrium dialysis incubation, then treated with methyl tert-butyl ether, and a liquid-liquid extraction procedure was performed to obtain the final extract for analysis.

LC Conditions

Sample manager:	SM-FL (PL, Load Ahead disabled)
Solvent manager:	BSM
Column:	ACQUITY UPLC BEH™ C_{18} 1.7 µm, 2.1 mm x 100 mm
Mobile phase A:	0.2 mM Ammonium fluoride in water
Mobile phase B:	0.2 mM Ammonium fluoride in methanol
Flow rate:	0.30 mL/min
Gradient:	50% B for 0.5 minutes, 55% B at 0.5 minutes, 55–75% B over 2.7 minutes, 75–98% B over 0.05 minutes, hold 98% B for 0.45 minutes, equilibrate with 50% B for 0.7 minutes

MS Conditions

Resolution:	MS1 (0.75 FWHM), MS2 (0.75FWHM)

Acquisition mode:	MRM
Polarity:	ESI (+)

Results and Discussion

Performance characteristics of free testosterone on the ACQUITY UPLC I-Class SM-FL/Xevo TQ Absolute IVD System are shown in Table 1. Analytical sensitivity of the chromatographic separation is illustrated in Figure 1.

Compound	Range (pg/mL)	LLOQ (pg/mL)	%RSD at LLOQ	Total precision	Repeatability	EQA mean bias
Free testosterone	1–500	0.5	17.6	≤8.4%	≤7.9%	-7.6%

Table 1. Performance characteristics of free testosterone. Range defined by linear fit where r^2 >0.995. LLOQ defined by S/N (PtP) >10 and %RSD ≤20%. %RSD at LLOQ determined through analytical sensitivity experiments performed over five occasions (n=50). Total precision and repeatability of QCs performed over five occasions in serum (n=25). EQA mean bias determined by comparison of obtained values to the all laboratory trimmed mean (ALTM) value (n=45).

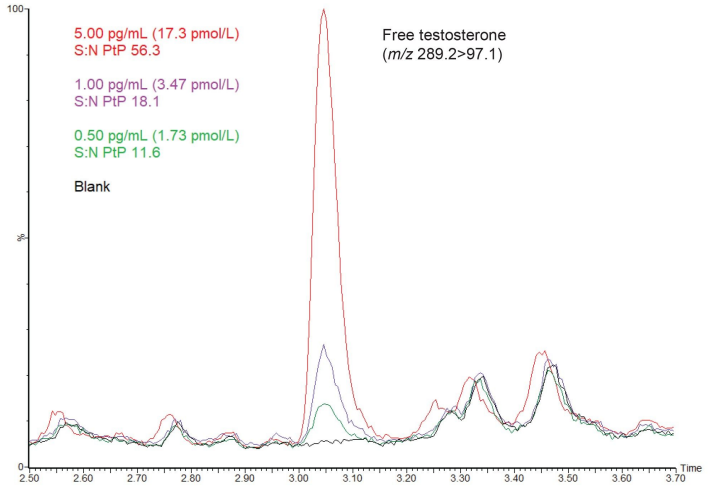

Figure 1. Chromatogram showing the analysis of free testosterone using the ACQUITY UPLC I-Class SM-FL/ Xevo TQ Absolute IVD System.

Conclusion

The Waters ACQUITY UPLC I-Class SM-FL/Xevo TQ Absolute IVD System has demonstrated the capability to analyse free testosterone in serum samples whilst delivering analytical sensitivity and precision.

Disclaimer

The analytical performance data presented here is for illustrative purposes only. Waters does not recommend or suggest analysis of the analytes described herein. These data are intended solely to demonstrate the performance capabilities of the system for analytes representative of those commonly analyzed using liquid chromatography and tandem mass spectrometry. Performance in an individual laboratory may differ due to a number of factors, including laboratory methods, materials used, intra-operator technique, and system conditions. This document does not constitute a warranty of merchantability or fitness for any particular purpose, express or implied, including for the testing of the analytes in this analysis.

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