

Nota applicativa

ACQUITY™ UPLC™ I- Class/Xevo™ TQ-S micro IVD System: Analytical Performance for an Organic Acid

Waters Corporation

For *in vitro* diagnostic use. Not available in all countries.

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

Abstract

This document describes a test of the analytical performance of the ACQUITY UPLC I-Class/Xevo TQ-S micro IVD System for the analysis of methylmalonic acid (MMA) in serum.

Introduction

The Waters™ ACQUITY UPLC I-Class/Xevo TQ-S micro

IVD System enables the quantification of organic compounds in human biological liquid matrices.



Figure 1. The Waters ACQUITY UPLC I-Class System and Xevo TQ-S micro Mass Spectrometer.

Experimental

The ACQUITY UPLC I-Class/Xevo TQ-S micro IVD System was controlled by MassLynx™ IVD (v4.2) and the data processed using the TargetLynx™ Application Manager. Calibrators were prepared by spiking commercially available material in 1% (w/v) bovine serum albumin (BSA) in phosphate buffered saline (PBS), and quality controls (QCs) in both plasma and serum. Samples were processed using the following conditions:

Sample Preparation Conditions

A 100- μ L sample and internal standard were added to an Ostro™ protein precipitation and phospholipid removal plate. Samples were precipitated with 1% formic acid in

acetonitrile, mixed in-well and then eluted into a collection plate, evaporated, and reconstituted prior to analysis.

LC Conditions

Column:	ACQUITY UPLC CSH™ C ₁₈ , 1.7 μm, 2.1 mm × 100 mm, with in-line filter
Mobile phase A:	Water with 0.2% formic acid
Mobile phase B:	Acetonitrile with 0.2% formic acid
Flow rate:	0.45 mL/min
Gradient:	1% B over 0.3 minutes, 1–20% B over one minute, 20% over 0.2 minutes, 95% B over 0.5 minutes

MS Conditions

Resolution:	MS1 (0.75 FWHM), MS2 (0.75FWHM)
Acquisition mode:	MRM
Polarity:	ESI-

Results and Discussion

Performance characteristics of MMA using the Waters ACQUITY UPLC I-Class/Xevo TQ-S micro IVD system is shown in Table 1.

Analytical sensitivity of the system for analyzing extracted MMA serum samples is illustrated in Figure 1.

Compound	Range (nmol/L)	LLOQ (nmol/L)	%RSD at LLOQ	Total precision	Repeatability	Mean bias
MMA	21-1270	21	11.1%	≤6.3%	≤5.5%	-0.8%

Table 1. Performance characteristics of MMA. Range defined by linear fit where $r^2 > 0.99$. LLOQ defined by $S/N (PtP) > 10$ and $\%RSD \leq 20\%$. % RSD at LLOQ determined through analytical sensitivity experiments performed over three occasions ($n=30$). Total Precision and repeatability of plasma and serum QCs. performed over five occasions ($n=25$). Mean bias determined through Bland-Altman comparison of calculated concentrations to an independent LC-MS/MS method for MMA. (Note: To convert SI units to conventional mass units, divide by 8.47 for MMA (nmol/L to ng/mL).)

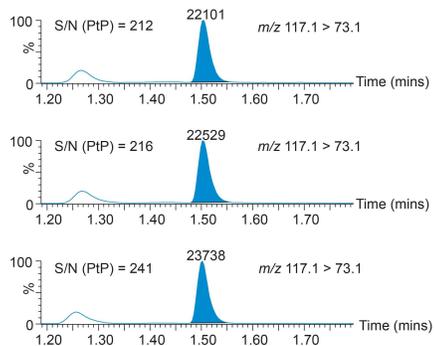


Figure 2. Chromatogram to show S/N (PtP) of extracted MMA control samples at 264 nmol/L using the ACQUITY UPLC I-Class/Xevo TQ-S micro IVD System.

Conclusion

The Waters ACQUITY UPLC I-Class/Xevo TQ-S micro IVD System has demonstrated the capability to deliver analytically sensitive and selective performance with excellent precision and accuracy for the analysis of methylmalonic acid.

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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