Waters™

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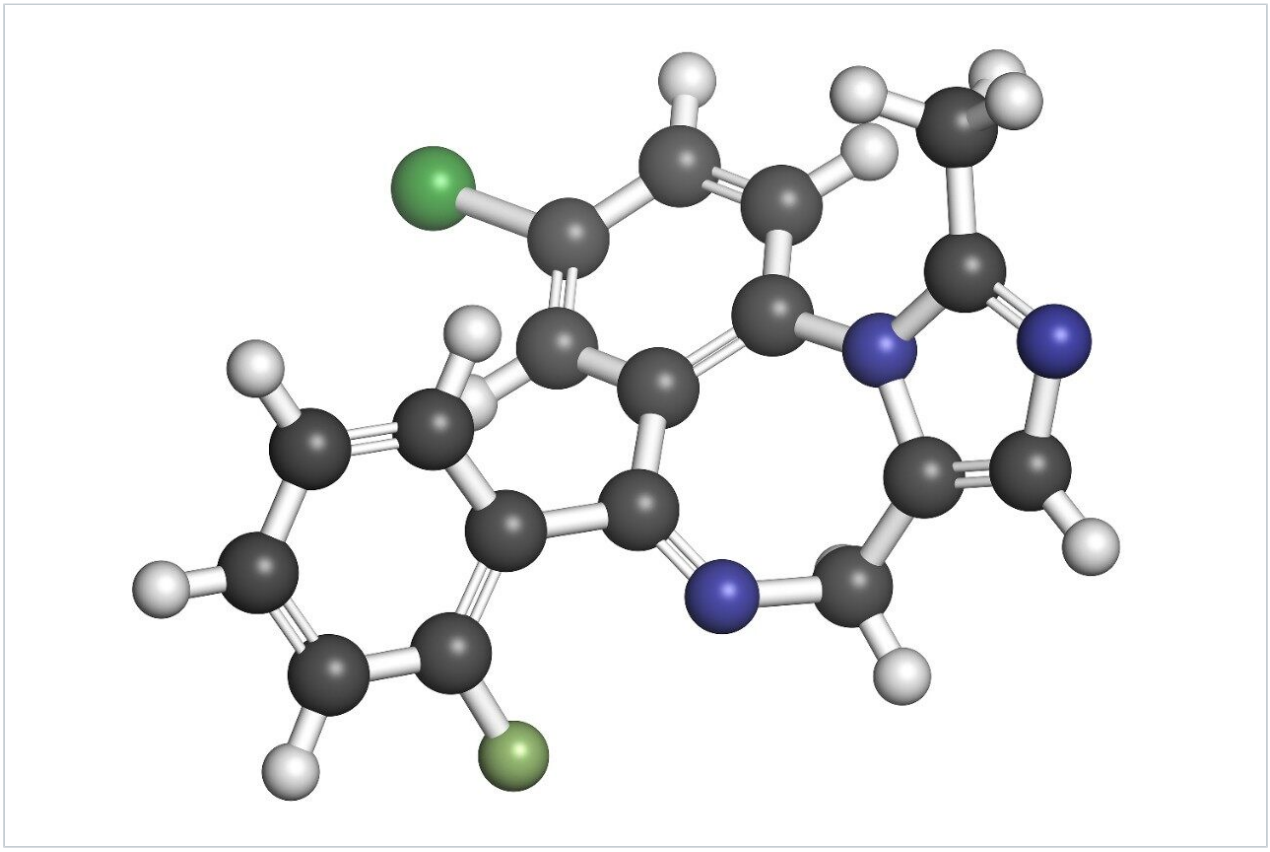
Midazolam on Oasis MCX

Waters Corporation



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

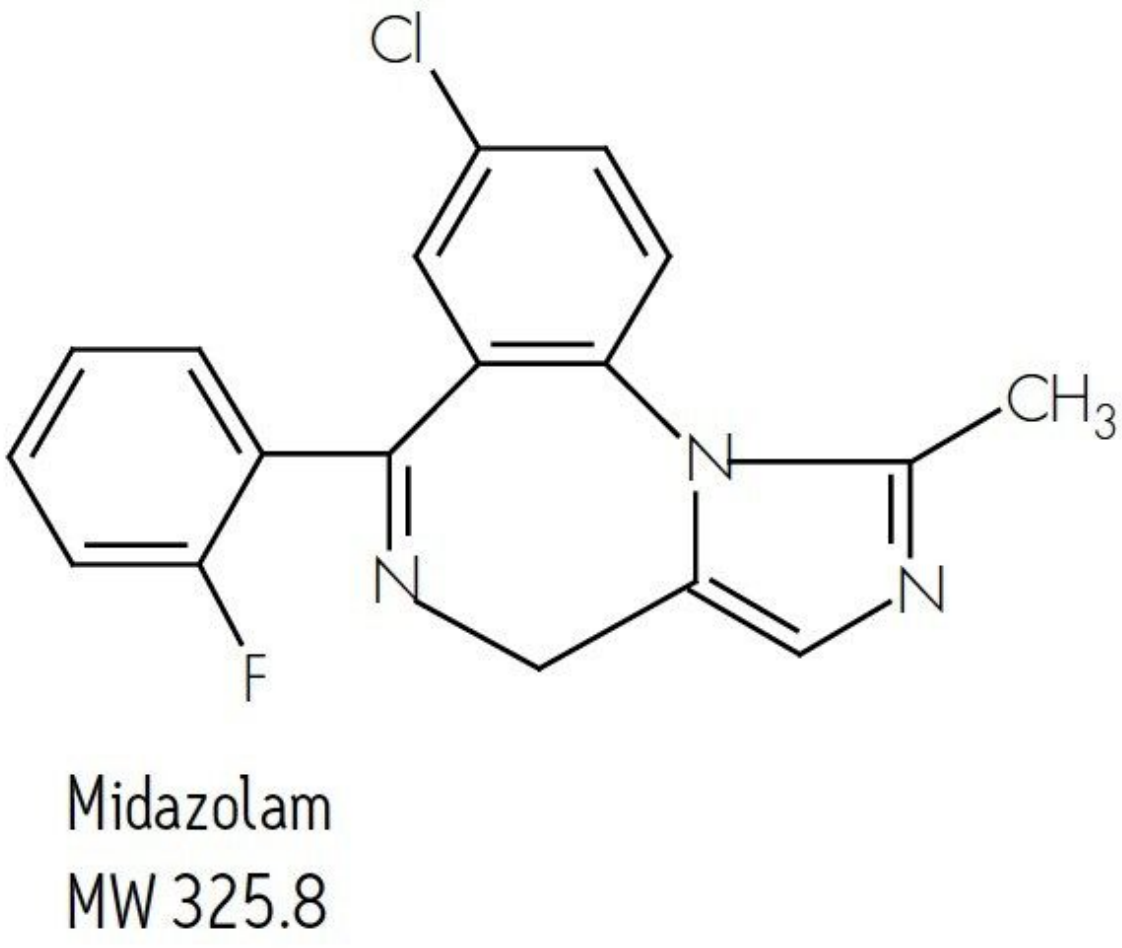
Abstract

This application brief demonstrates the analysis of midazolam on Oasis MCX.

Introduction

Midazolam is used to produce sleepiness or drowsiness and to relieve anxiety before surgery or certain procedures. In this example, the sample size was limited (less than 100 uL of plasma) and therefore the Oasis µElution plate was used.

Weak Base: pKa ~ 6.15 and therefore Oasis MCX was used.



Experimental

Test Conditions

Oasis MCX 96-Well µElution Plate

Condition:	200 µL MeOH
Equilibrate:	200 µL H_2O
Load:	100 µL sample (50 µL plasma diluted 1:1 with 4% H_3PO_4)
Wash 1:	200 µL 2% HCOOH
Wash 2:	200 µL MeOH
Elute:	50 µL (25 µL x 2) 5% NH_4OH in MeOH
Dilute:	100 µL H_2O or 100 µL 2% FA in H_2O to neutralize
Inject:	10 µL
Column:	SunFire C_{18} 2.1 x 20 mm IS, 3.5 µm
Mobile phase A:	10 mM $CH_3COO\text{-}NH_4$+, pH 5.5
Mobile phase B:	MeOH with 10 mM $CH_3COO\text{-}NH_4$+, pH 5.5
Flow rate:	0.4 mL /min
Instrument:	2777 Sample Manager and 1525µ Binary HPLC Pump

Gradient

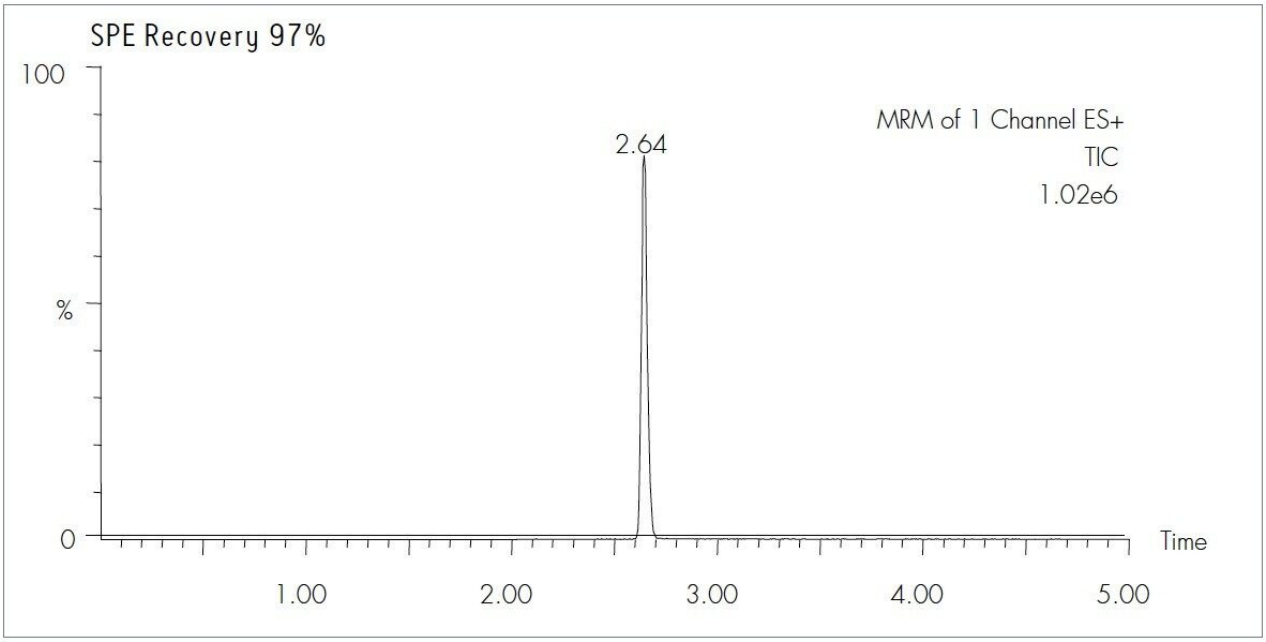
Time(min)	Profile

	%A	%B
0.0	95	5
3.0	5	95
4.0	5	95
4.1	95	5
5.0	95	5

Quattro Premier

ESI- source temp:	150 °C
Desolvation temp:	350 °C
Cone gas flow:	50 L /Hr
Desolvation gas flow:	600 L /Hr
Collision cell:	$2.2e^{-3}$ bar (Ar gas)
Midazolam	MRM Transition
	m/z 326.2 → 291.2

Results and Discussion



WA60087, June 2007