LC-MS/MS Separation of MDMA and its Metabolites Using the Kinetex® 2.6 µm Biphenyl Column

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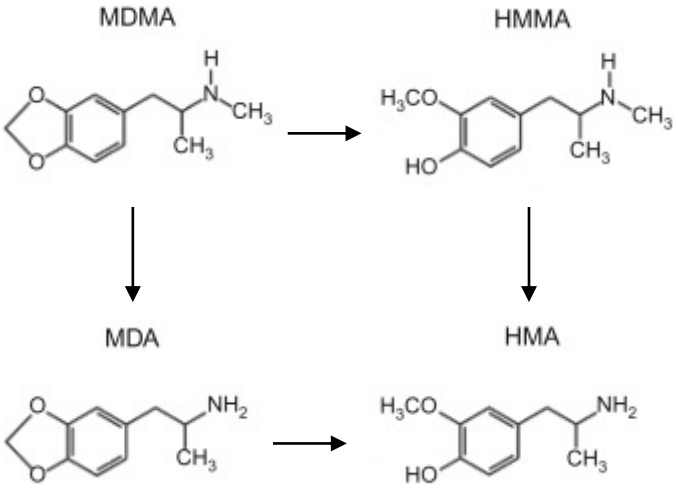
Overview

MDMA (3,4-Methylenedioxymethamphetamine) is a ring-substituted amphetamine analog with reinforcing psychoactive effects, in addition to neurotoxic potential toward central 5-HT neurons. It is metabolized into MDA (3,4-methylenedioxyamphetamine), HMMA (4-hydroxy-3-methoxymethamphetamine), and HMA (4-hydroxy-3-methoxyamphetamine) (**Figure 1**). Being able to successfully separate these drug substances is a useful tool for forensic labs. In this application note, we present an LC-MS/MS method to separate MDMA and its metabolites by utilizing the Kinetex 2.6 µm Biphenyl column.

The Kinetex Biphenyl column provided separation for MDMA and its metabolites through a mixture of pi-pi and polar interactions; and the higher efficiency provided by the core-shell particle morphology also results in narrow peaks and increased MS sensitivity (**Figure 2**).

Figure 1. MDMA Metabolism



LC-MS/MS Conditions

Column: Kinetex 2.6 µm Biphenyl
Dimension: 50 x 2.1 mm
Part No.: 00B-4622-AN
Mobile Phase: A: 0.1 % Formic Acid in Water
B: 0.1 % Formic Acid in Methanol
Gradient:

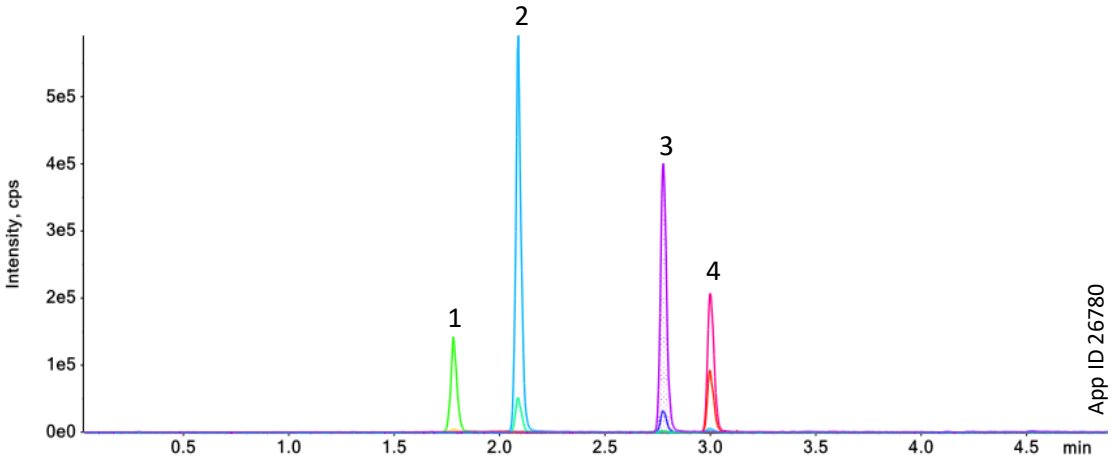
Time (min)	%B
0	5
4	95
5	95
5.1	5
7	5

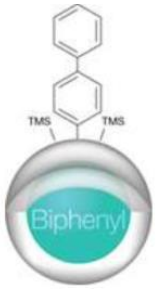
Flow Rate: 400 µL/min
Injection Volume: 1 µL
Temperature: 40 °C
LC System: Agilent® 1260 Infinity
Detection: MS/MS
Detector: SCIEX® 4500

MRM Transitions

Peak No.	Analyte	Q1 (m/z)	Q3 (m/z)
1	HMA-1	182	165
1	HMA-2	182	105
2	HMMA-1	196	165
2	HMMA-2	196	105
3	MDA-1	180	163.1
3	MDA-2	180	135
4	MDMA-1	194.1	163.2
4	MDMA-2	194.1	135.2

Figure 2. Kinetex® 2.6 µm Biphenyl





The Kinetex Biphenyl stationary phase offers multiple potential interactions to increase retention for polar aromatic compounds. These include pi-pi interactions and increased hydrogen bond accepting capacity.

Peak No.	Analyte	Retention Time (min)	Concentration (mg/mL)
1	HMA	1.78	100
2	HMMA	2.09	100
3	MDA	2.77	100
4	MDMA	3.01	100

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