

Determination of Urinary Mycotoxin Biomarkers Using a Sensitive Online Solid Phase Extraction-UHPLC-MS/MS Method

Jessica Schmidt, Benedikt Cramer, Paul C. Turner, Rebecca J. Stoltzfus, Jean H. Humphrey, Laura E. Smith and Hans-Ulrich Humpf

Table S1. sMRM parameters of the validated online SPE-UHPLC-MS/MS method.

Analyte	Q1 mass [Da] ^a	Q3 mass [Da] ^a	Expected Retention Time [min]	Window [sec]	DP [V] ^b	CE [V] ^c	CXP [V] ^d
AFM ₁	329.1 [M+H] ⁺	229.0	5.7		41	53	15
	329.1 [M+H] ⁺	273.0	5.7		41	33	15
ALT	293.1 [M+H] ⁺	239.0	6.1		30	27	12
	293.1 [M+H] ⁺	139.0	6.1		30	84	12
AME	271.0 [M-H] ⁻	213.1	8.5		-44	-45	-20
	271.0 [M-H] ⁻	211.1	8.5		-44	-50	-20
AOH	257.0 [M-H] ⁻	147.1	7.0		-80	-38	-11
	257.0 [M-H] ⁻	157.1	7.0		-80	-36	-11
CIT	251.1 [M+H] ⁺	215.0	6.9	60	75	35	13
	251.1 [M+H] ⁺	205.0	6.9	60	75	36	13
DH-CIT	267.1 [M+H] ⁺	203.1	6.6	45	51	39	14
	267.1 [M+H] ⁺	231.1	6.6	45	51	32	14
FB ₁	722.4 [M+H] ⁺	334.2	6.4		41	55	18
	722.4 [M+H] ⁺	352.2	6.4		41	51	20
OTA	404.1 [M+H] ⁺	221.0	8.5		35	48	11
	404.1 [M+H] ⁺	102.0	8.5		35	88	11
	404.1 [M+H] ⁺	239.0	8.5		35	31	11
ZEN	317.2 [M-H] ⁻	175.0	8.6		-140	-32	-11
	317.2 [M-H] ⁻	131.0	8.6		-140	-38	-11
α -/ β -ZEL	319.2 [M-H] ⁻	174.1	7.5	60	-160	-34	-15
	319.2 [M-H] ⁻	160.1	7.5	60	-160	-39	-15
¹³ C ₁₇ -AFM ₁	346.3 [M+H] ⁺	242.2	5.7		41	53	15
	346.3 [M+H] ⁺	288.1	5.7		41	33	15
¹³ C ₃ -CIT	254.1 [M+H] ⁺	218.0	6.9	45	75	35	13
	254.1 [M+H] ⁺	208.0	6.9	45	75	36	13
¹³ C ₃ -DH-CIT	270.1 [M+H] ⁺	206.1	6.6		54	39	14
	270.1 [M+H] ⁺	234.1	6.6		54	32	14
¹³ C ₃₄ -FB ₁	756.1 [M+H] ⁺	356.3	6.4		41	55	18
	756.1 [M+H] ⁺	374.3	6.4		41	51	20
<i>d</i> ₅ -OTA	409.1 [M+H] ⁺	221.0	8.5		35	48	11
	409.1 [M+H] ⁺	102.0	8.5		35	88	11
	409.1 [M+H] ⁺	239.0	8.5		35	31	11
<i>d</i> ₂ -ZEN	319.2 [M-H] ⁻	177.0	8.6		-140	-32	-11
	319.2 [M-H] ⁻	133.0	8.6		-140	-38	-11

^a Quantifier above, qualifier below; ^b declustering potential; ^c collision energy; ^d collision cell exit potential

Table S2. Accuracy and relative standard deviations determined during intra- and interday repeatability at the second highest calibration level.

Analyte	Intraday ^a		Interday ^b	
	Accuracy [%]	RSD ^c [%]	Accuracy [%]	RSD ^c [%]
AFM ₁	103	3	101	4
ALT	99	1	97	3
AME	134	5	117	15
AOH	110	3	104	5
CIT	103	2	102	4
DH-CIT	103	5	103	6
FB ₁	117	8	128	10
OTA	106	4	105	5
ZEN	117	5	116	4
α-ZEL	115	6	115	6
β-ZEL	103	7	110	10

^a *n* = 12; ^b *n* = 4 on three different days; ^c relative standard deviation.

Table S3. Concentrations of the analytes and stable isotope-labelled standards in the calibration solutions.

Analyte	Concentration Levels in Calibration Solutions [ng/mL]								Int. Standard	1–8
	1	2	3	4	5	6	7	8		
AFM ₁	0.025	0.063	0.13	0.19	0.25	0.63	1.3	2.5	¹³ C ₁₇ -AFM ₁	0.20
ALT	0.70	1.8	3.5	5.3	7.0	18	35	70	—	—
AME	0.040	0.10	0.20	0.30	0.40	1.0	2.0	4.0	—	—
AOH	0.90	2.3	4.5	6.8	9.0	23	45	90	—	—
CIT	0.30	0.75	1.5	2.3	3.0	7.5	15	30	¹³ C ₃ -CIT	3.0
DH-CIT	0.20	0.50	1.0	1.5	2.0	5.0	10	20	¹³ C ₃ -DH-CIT	2.0
FB ₁	0.014	0.035	0.070	0.11	0.14	0.35	0.70	1.4	¹³ C ₃₄ -FB ₁	0.14
OTA	0.012	0.030	0.060	0.090	0.12	0.30	0.60	1.2	<i>d</i> ₅ -OTA	0.12
ZEN	0.035	0.088	0.18	0.26	0.35	0.88	1.8	3.5	<i>d</i> ₂ -ZEN	0.35
α-ZEL	0.15	0.38	0.75	1.1	1.5	3.8	7.5	15	<i>d</i> ₂ -ZEN	0.35
β-ZEL	0.15	0.38	0.75	1.1	1.5	3.8	7.5	15	<i>d</i> ₂ -ZEN	0.35