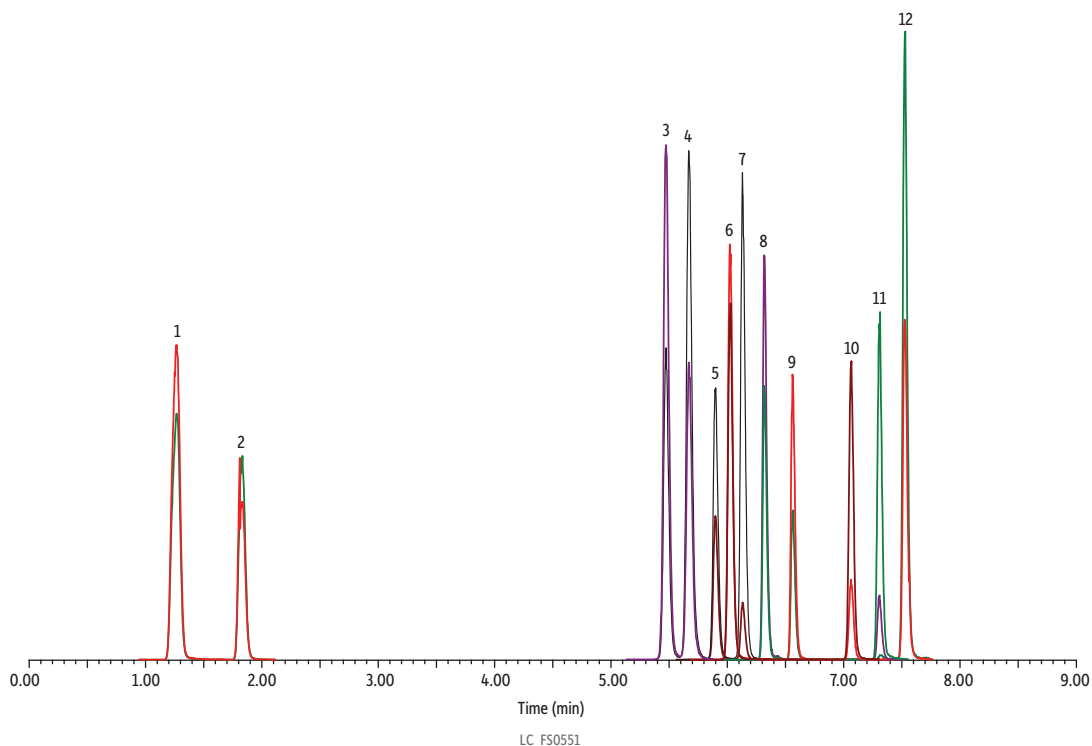


Ergot Alkaloid Epimers in Fortified Flour Sample on Raptor Biphenyl by LC-MS/MS



Peaks	tr (min)	Precursor Ion	Product Ion 1	Product Ion 2
1. Ergometrine	1.27	326.2	223.2	208.1
2. Ergometrinine	1.83	326.2	223.2	208.1
3. Ergosine	5.47	548.4	208.1	223.2
4. Ergosinine	5.67	548.4	208.1	223.2
5. Ergotamine	5.90	582.4	223.2	268.2
6. Ergocornine	6.03	562.4	268.2	223.2
7. Ergotaminine	6.13	582.4	223.2	268.2
8. Ergocryptine	6.32	576.4	268.2	223.2
9. Ergocristine	6.56	610.4	223.2	592.4
10. Ergocorninine	7.07	562.4	268.2	223.2
11. Ergocryptinine	7.31	576.4	268.2	223.2
12. Ergocristinine	7.53	610.4	223.2	592.4

Column Raptor Biphenyl (cat.# 9309A12)
Dimensions: 100 mm x 2.1 mm ID
Particle Size: 2.7 µm
Pore Size: 90 Å
Guard Column: Raptor Biphenyl EXP guard column cartridge 5 mm, 2.1 mm ID, 2.7 µm (cat.# 9309A0252)
Temp.: 60 °C

Standard/Sample
Diluent: 50:50 Water:methanol
Conc.: 6.25 ng/mL final concentration after sample preparation
Inj. Vol.: 5 µL

Mobile Phase
A: Water, 0.05% formic acid
B: Methanol, 0.05% formic acid

Time (min)	Flow (mL/min)	%A	%B
0.00	0.4	75	25
5.00	0.4	50	50
9.00	0.4	0	100
9.01	0.4	75	25
11.0	0.4	75	25

Detector MS/MS
Ion Mode: ESI+
Mode: MRM
Instrument UHPLC

Sample Preparation A blended flour was prepared by mixing white rice flour (75%); brown rice flour (5%); millet flour (5%); oat flour (5%); all-purpose wheat flour (5%); and all-purpose, gluten-free flour (5%). Two grams of the flour sample were weighed into a 50-mL polypropylene centrifuge tube (cat.# 25846) and fortified at 50 µg/kg for all analytes with a stock standard solution. After sitting at room temperature for 10 minutes, 16 mL of extraction solution (80:20 acetonitrile:water) containing 0.5% formic acid were added, and the tube was stirred to create a homogenous suspension. The extraction was carried out by shaking horizontally on a digital pulse mixer (Glas-Col LLC, Terre Haute, IN) at 800 rpm for 20 minutes. After centrifuging for 5 minutes at 4000 rpm, 1 mL of extract was evaporated to dryness at 45 °C under a gentle stream of nitrogen. The dried extract was reconstituted with 1 mL of 50:50 water:methanol solution, and a 0.4 mL aliquot was transferred to and filtered using a Thomson SINGLE StEP filter vial with a 0.2 µm PTFE filter (cat.# 25874). Five µL of the filtered solution was injected for the LC-MS/MS analysis.