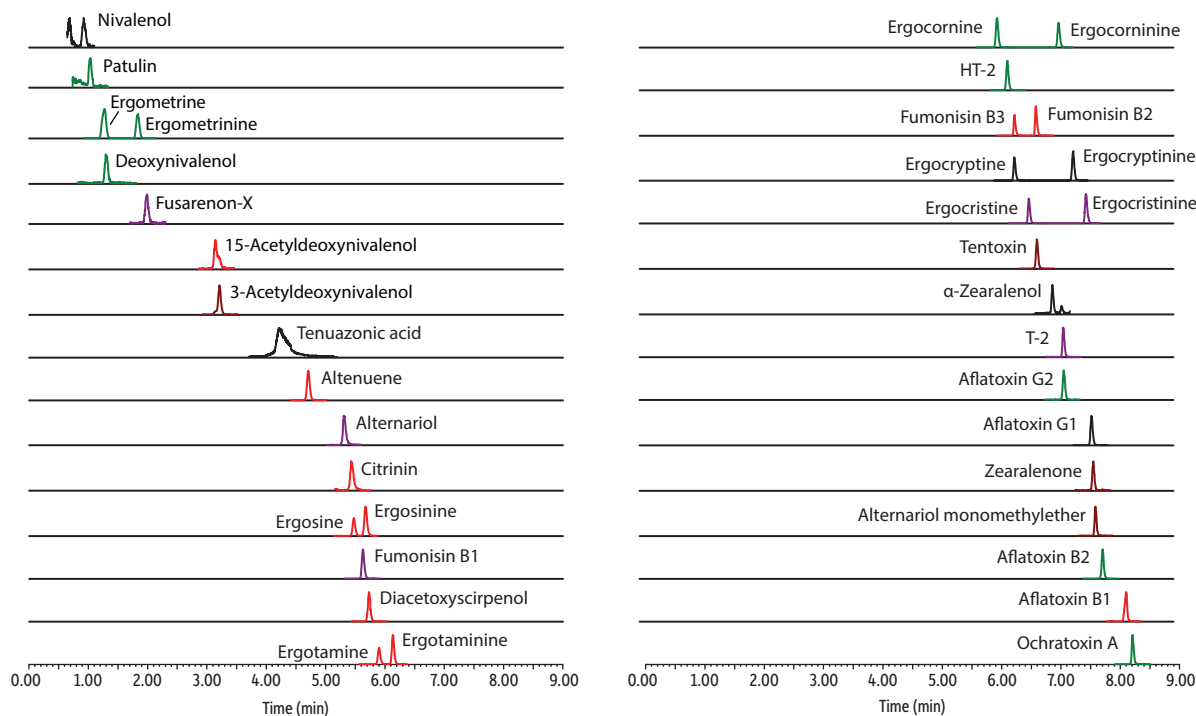


Alternaria Toxins, Ergot Alkaloid Epimers, and Other Major Mycotoxins in Fortified Flour Sample on Raptor Biphenyl by LC-MS/MS

- Simultaneous determination of alternaria toxins and ergot alkaloids with acidic LC condition.
- Suitable chromatographic separation of ergot alkaloid epimers.
- Short run time for multi-mycotoxin analysis.



LC_FS0550

Peaks	tr (min)	Precursor Ion	Product Ion 1	Product Ion 2
1. Nivalenol	0.92	295.1	137.1	91.0
2. Patulin	1.03	155.0	99.0	81.0
3. Ergometrine	1.27	326.2	223.2	208.1
4. Deoxynivalenol	1.30	297.2	231.0	249.0
5. Ergometrinine	1.83	326.2	223.2	208.1
6. Fusarenon-X	1.98	355.1	137.1	247.1
7. 15-Acetyldeoxynivalenol	3.14	339.2	137.1	321.2
8. 3-Acetyldeoxynivalenol	3.21	339.2	213.1	231.1
9. Tenuazonic acid	4.22	198.1	125.0	153.1
10. Altenuene	4.70	293.2	257.1	275.2
11. Alternariol	5.30	259.0	185.1	130.0
12. Citrinin	5.43	251.2	233.1	205.1
13. Ergosine	5.47	548.4	208.1	223.2
14. Fumonisin B1	5.63	722.5	352.3	334.2
15. Ergosinine	5.67	548.4	208.1	223.2
16. Diacetoxyscirpenol	5.73	384.2	247.1	307.2
17. Ergotamine	5.90	582.4	223.2	268.2
18. Ergocornine	6.03	562.4	268.2	223.2
19. Ergotaminine	6.13	582.4	223.2	268.2

Peaks	tr (min)	Precursor Ion	Product Ion 1	Product Ion 2
20. HT-2	6.20	447.2	345.1	285.1
21. Fumonisin B3	6.32	706.4	336.2	318.3
22. Ergocryptine	6.32	576.4	268.2	223.2
23. Ergocristine	6.56	610.4	223.2	592.4
24. Fumonisin B2	6.68	706.4	336.2	318.3
25. Tentoxin	6.70	415.2	312.2	302.2
26. alpha-Zearalenol	6.96	303.1	285.1	175.0
27. Ergocorninine	7.07	562.4	268.2	223.2
28. T-2	7.14	489.2	387.1	245.1
29. Aflatoxin G2	7.15	331.2	189.0	313.0
30. Ergocryptinine	7.31	576.4	268.2	223.2
31. Ergocristinine	7.53	610.4	223.2	592.4
32. Aflatoxin G1	7.62	329.1	199.7	243.0
33. Zearalenone	7.65	319.2	283.1	187.0
34. Alternariol monomethylether	7.69	273.0	199.1	128.0
35. Aflatoxin B2	7.81	315.1	287.0	259.0
36. Aflatoxin B1	8.20	313.2	241.1	284.9
37. Ochratoxin A	8.31	404.1	239.0	358.0

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 Aflatoxins standard (cat.# 34121)
 Ochratoxin A standard (cat.# 34122)
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.

Notes The chromatogram shows peaks with the MS transition of product ion 1.
 Note that method development work demonstrated that whenever a new column is installed it must be rinsed and maintained under mobile phase overnight to ensure an acceptable peak shape for tenuazonic acid.