Mycotoxins: Sample Preparation and Analysis

Matrix of the Month

April, 2014:
Ochratoxin A
in Peanut Puffs



Do you have a special matrix that we should test for mycotoxins? Please let us know and write an e-mail to info@LCTech.de!

Protocol

20 g homogenised peanut puffs are mixed with 2 g sodium chloride and extracted by thoroughly mixing with 100 mL 80/20 (methanol/water (v/v)) and 50 mL n-hexane (5-10 min). The extract is filtrated and centrifuged for 10 min at 2000 x g to support phase separation of aqueous and n-hexane phase. 2 mL of the aqueous phase (lower phase) are diluted with 12 mL PBS buffer containing 8% Tween20.

The extract is applied to the immunoaffinity column (Afla-OtaCLEAN or OtaCLEAN), afterwards the sample reservoir is rinsed with 10 mL of deionised water. This washing solution is also applied to the immunoaffinity column.

The column is dried and eluted with methanol (2 $\rm mL$). The methanol should incubate on the column bed for 5 minutes to ensure that the antibody is entirely denaturated.

The eluate is diluted to HPLC conditions or concentrated according to HPLC sensitivity.

HPLC Conditions

Ochratoxin A

HPLC: Dionex Ultimate 3000 isocratic

Column oven: 40 °C

Separation column: EC125/3 Nucleosil 120-3 C-18 RP (e.g. 10522)

Flow rate: 0.6 mL/min, water/methanol/acetonitrile (40/55/5 (v/v/v)) + 1 % acetic acid

Fluorescence detection: Excitation wavelength: 335 nm
Emission wavelength: 465 nm

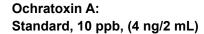
Recovery Rates

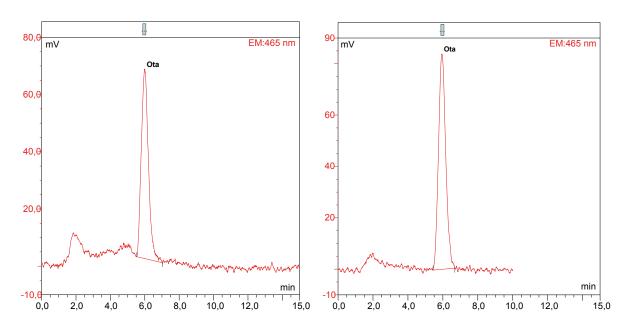
Content of Ochratoxin A in peanut puffs	
	Ochratoxin A
Standard*	100
Recovery rate** peanut puffs, spiked with 10 ppb	88

^{*} Standard is set = 100 %, ** corrected with non-spiked sample

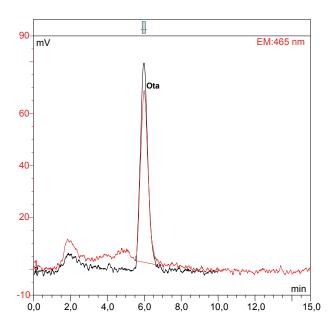


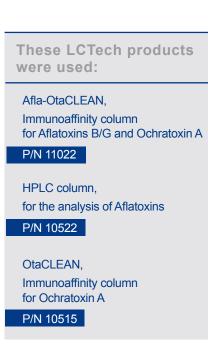
Ochratoxin A: Peanut Puffs, 10 ppb





Ochratoxin A:
Overlay of both chromatograms





Do you have further questions?
Please simple write an e-mail to info@LCTech.de!

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