



March 2017

Ochratoxin A in Chili Fully Automated Processing via FREESTYLE ThermELUTE™

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

Sample Preparation

MYCOTOXINS

Chili

Chilis are the hot relatives of paprika. Worldwide there are over a hundred varieties of chili with different levels of spiciness. Spices enhances our food. They often are imported, mostly from Asia. However, they not always meet our quality requirements. By drying and storage of food, mycotoxins can be arised. Therefore, aflatoxins and ochratoxin A repeatedly lead to rejections of the food at the controls of the EU.

Fully Automated Processing via FREESTYLE ThermELUTE™ and OtaCLEAN SMART

The challenge many laboratories have to face nowadays is to be able to process many samples as quickly as possible. To facilitate this task, LCTech has developed the OtaCLEAN SMART immunoaffinity column for the analysis of ochratoxin A in food. Through miniaturization of the column, not only the processing time of the sample is dramatically reduced, but also the required amounts of sample and solvent, while the high performance of the columns remains the same. Like the other LCTech SMART columns, the operating mechanism is based on the principle of immunoaffinity chromatography with increased flow rates and a customized, small elution volume in order to shorten the processing time.

The robotic system FREESTYLE ThermELUTE™ in combination with the immunoaffinity columns OtaCLEAN SMART and any HPLC system enables a fully automated mycotoxin analysis with such sensitive results, that you can be sure always to measure even in the lowest ppt-range. By means of a unique technique, the comprehensive automation „from raw extract to chromatogram“ is realised without any manual intervention for processing around the clock.

The Highlights:

- High sample throughput up to 500 samples / week
- Automation for sample processing round the clock, even at weekends
- Incredible sensitivity in the lower ppt-region
- Very good recovery rates for all matrices
- Reproducible results



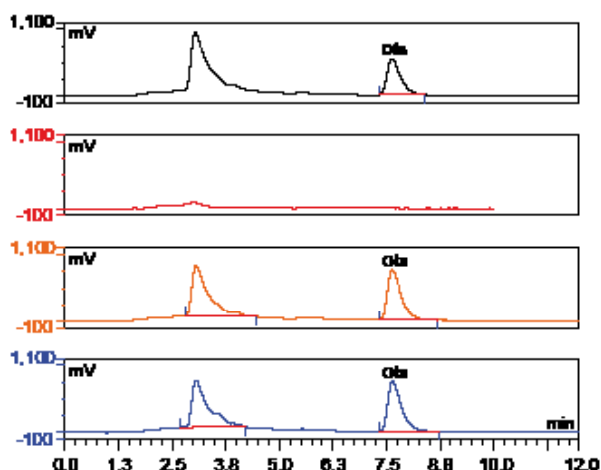
Protocol of Automated Processing

Homogenise 10 g chili and add 2 g of sodium chloride. Extract the mixture with 100 mL methanol/water (80/20 (v/v)) and 50 mL n-hexane, to remove fat and essential oils. The extraction should be performed for 20 minutes, in order to avoid lower extraction effects. Filtrate the raw extract and dilute 2 mL with 12 mL PBS (contains 8 % Tween20).

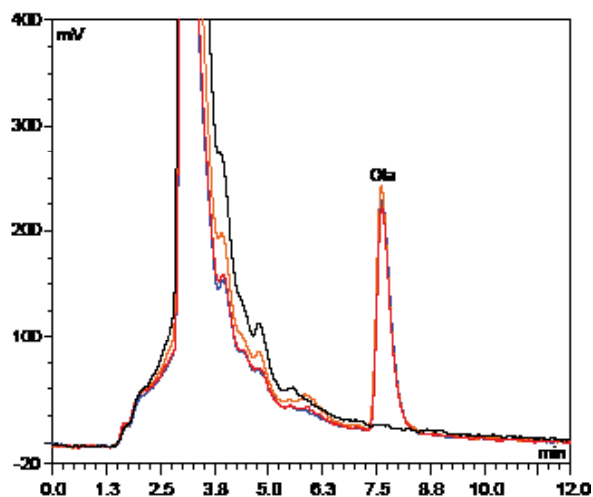
The robotic system FREESTYLE ThermELUTE™ loads the sample fully automated onto the immunoaffinity column OtaCLEAN SMART. Afterwards the system washes the column with 2 mL of diluted phosphate buffer (0.002 % PBS).

The column will be thermal eluted via ThermELUTE™ technology and directly injected into the HPLC sample loop. As result you achieve very good and high resolution chromatograms.

Chromatograms



Black: Chili (A), not spiked; Red: Blind sample (control of cross-contamination); Orange & blue: Chili (A), 10 ppb spiked



Overlay of chromatograms
Black: Chili (B), not spiked; Red, orange & blue: 10 ppb spiked Chili (B), individually processed, purified and injected

HPLC-Conditions (Ochratoxin A)

HPLC:	isocratic
Column Oven:	40 °C
Separation Column:	RP EC 125/3 nucleosil 120-3 C18
Flow Rate:	0.6 mL/min
Eluent:	HPLC-water/methanol/ acetonitrile + 1 % acetic acid (40/55/5 (v/v/v))
Fluorescence Detection:	without derivatisation
Excitation Wavelength:	335 nm
Emission Wavelength:	465 nm

Recovery Rates

Content of Ochratoxin A in Chili

Mycotoxin	Ochratoxin A
Standard*	100
Recovery Rates** Chili (A), 10 ppb	93
Recovery Rates** Chili (B), 10 ppb	87

*Standard is set = 100 %, **Corrected with non-spiked sample/
The results correspond to the performance specifications of EC 401/2006 (Section 4.3.1)



FREESTYLE ThermELUTE™
with SPE module for fully
automated sample prepara-
tion in mycotoxin analysis

These LCTech products were used:

OtaCLEAN SMART,
Immunoaffinity Columns for Ochratoxin A
P/N 13346 / 13351

FREESTYLE ThermELUTE™, Robotic System
for Automated Sample Preparation and Analysis
P/N 12663 / 12668 / 13691