Matrix of the Month

June 2018

Ochratoxin A in Beer
~ Manual and Automated ~

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

Beer

Soon, it is time again and fans throughout Germany will be cheering on the matches of the Football World Cup 2018. Whether at large public viewings with huge LED screens or at a cosy barbecue in the own garden - one thing most people would not want to miss: a delicious cool beer. The variety is enormous, as there are currently around 7,500 types of beer. Beer is obtained by fermentation of substances containing starch. According to the German Purity Law, issued by Duke Wilhelm IV of Bavaria on 24 April 1516, only malt, hops, water and yeast is allowed for production. In 2016, the Purity Law became 500 years old, making it the oldest food law regulation in force today.

However, beer is still a cereal product and therefore cannot always do justice to the term „pure“. The challenge is posed by toxic moulds that can develop during cultivation, storage, and processing of cereals leading to a loss in quality. For this reason, we have examined some types of beer for you this month. Chromatograms, recovery rates and a processing protocol are available on the following pages.

Immunoaffinity Columns OtaCLEAN for Ochratoxin A

In order to preserve the good quality of beer, LCTech supports laboratories worldwide in the field of sample clean-up in food and feed analysis. Especially for the clean-up of ochratoxin A we offer the immunoaffinity columns OtaCLEAN, a solution that convinces with very good recoveries even for difficult matrices. In addition to the convenient 3 mL format, they are also available in a 1 mL format and are suitable for manual and automated processing, e.g. with the robotic system FREESTYLE SPE.

Simply prepare the beer sample according to the processing protocol on the following page, equip the racks with the samples, configure the desired method in the software and press START - while the system is processing sample by sample, you can use the time gained for other important tasks in the laboratory.

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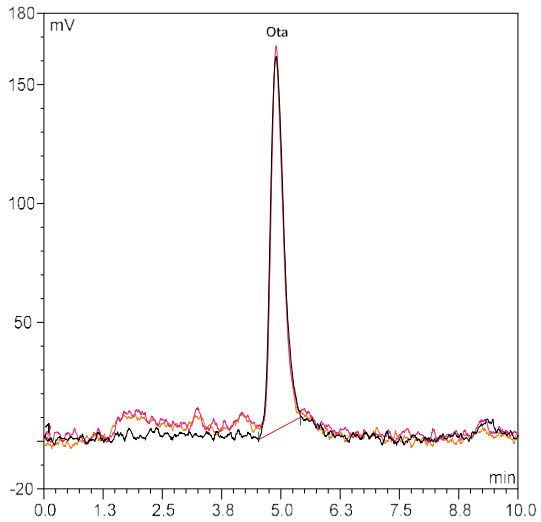
Manual Processing Protocol

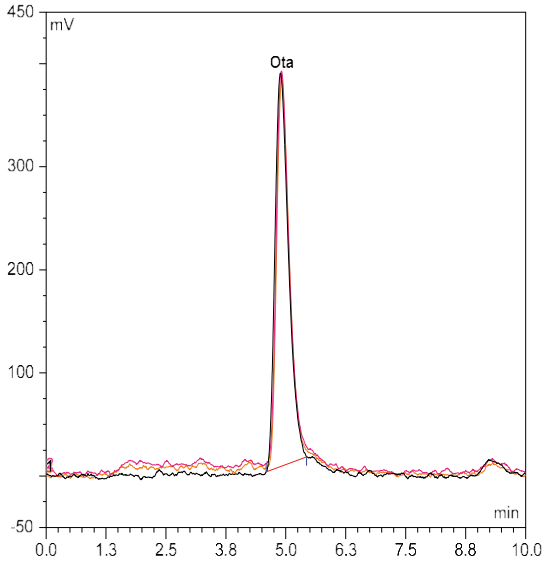
Degas 20 mL of Beer by sonication. Add 8 mL of 3 % $NaHCO_3$-solution and mix the sample material. Filtrate the raw extract and dilute 10 mL of it with 40 mL PBS. Load 50 mL (represents 7,14 g) of the sample onto a OtaCLEAN column. Wash the sample reservoir afterwards with 2 x 5 mL deionised water.

Elute the toxin with 2 mL methanol of the column. Keep in mind that the column bed is incubated with methanol for 5 minutes in order to ensure a fully denaturation of the antibodies and release of toxins.

Chromatogram



Black: *Standard represents 0.4 ppb*
Red: *Dark Beer, 0.4 ppb*
Orange: *Dark Beer, 0.4 ppb*



Black: *Standard represents 1 ppb*
Red: *Dark Beer, 1 ppb*
Orange: *Dark Beer, 1 ppb*

HPLC-Conditions
(Ochratoxin A)

Mycotoxin:	**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 % (40/55/5 (v/v/v))
Fluorescence Detection:	without Derivatization
Excitation Wavelength:	335 nm
Emission Wavelength:	465 nm

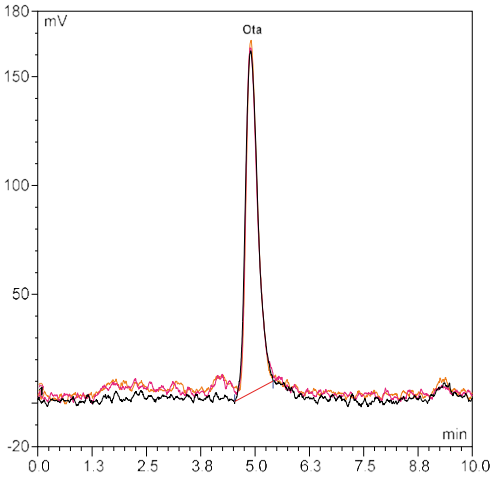
Recovery Rates
Content of Ochratoxin A in Beer

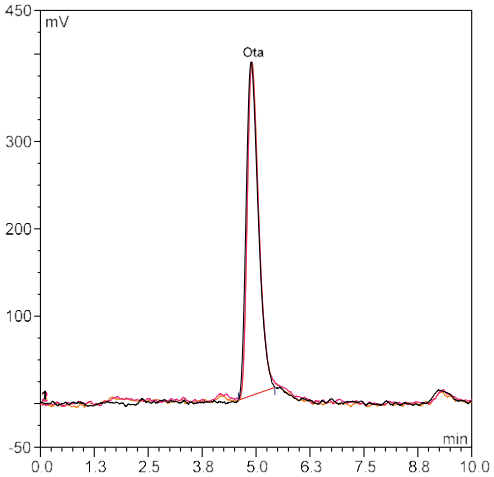
Mycotoxin	**Ochratoxin A**
Standard*	100
Recovery Rate** Dark Beer, 0.4 ppb	93
Recovery Rate** Dark Beer, 1 ppb	94
Recovery Rate** Light Beer, 0.4 ppb	96
Recovery Rate** Light Beer, 1 ppb	95
Recovery Rate** Bock Beer, 0.4 ppb	93
Recovery Rate** Bock Beer, 1 ppb	91
Recovery Rate** Wheat Beer, 0.4 ppb	93
Recovery Rate** Wheat Beer, 1 ppt	97

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

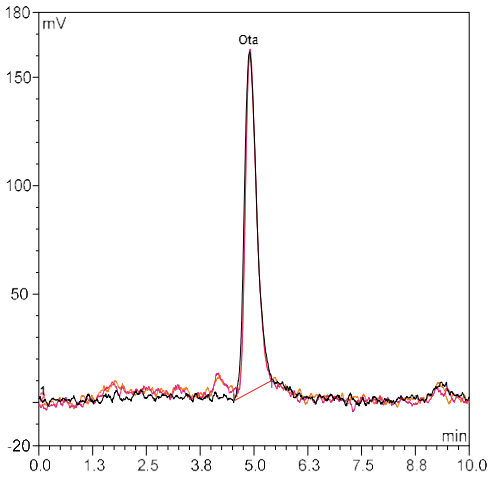
More chomotograms on page 3

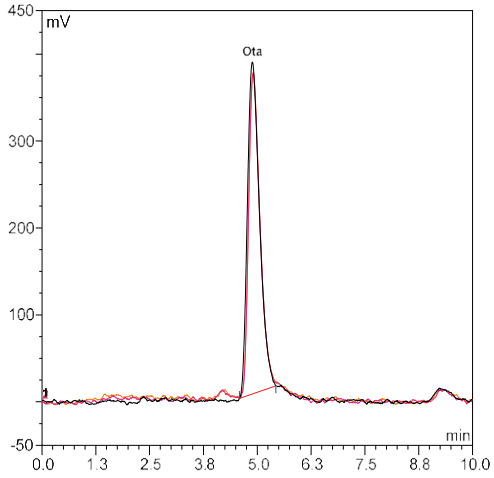
Matrix
of the Month



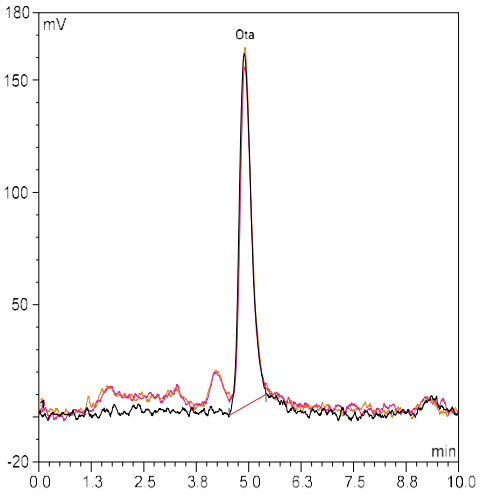


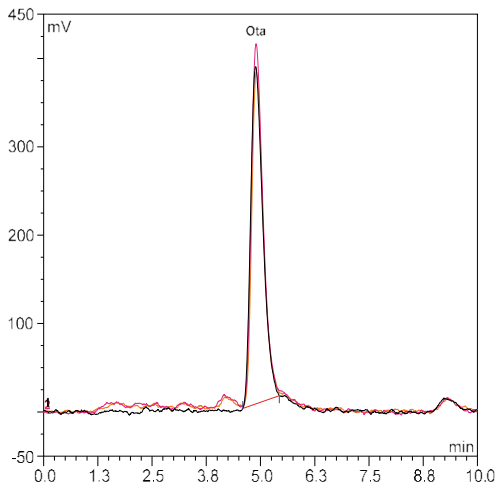
Black: *Standard represents 0.4 ppb (left); 1 ppb (right)*
Red: *Light Beer, 0.4 ppb (left); 1 ppb (right)*
Orange: *Light Beer, 0.4 ppb (left); 1 ppb (right)*





Black: *Standard represents 0.4 ppb (left); 1 ppb (right)*
Red: *Bock Beer, 0.4 ppb (left); 1 ppb (right)*
Orange: *Bock Beer, 0.4 ppb (left); 1 ppb (right)*





Black: *Standard represents 0.4 ppb (left); 1 ppb (right)*
Red: *Wheat Beer, 0.4 ppb (left); 1 ppb (right)*
Orange: *Wheat Beer, 0.4 ppb (left); 1 ppb (right)*

LCTech immunoaffinity columns for excellent chromatographic results with very high recovery rates at high loading capacities.

These LCTech products were used:

OtaCLEAN,
Immunoaffinity Column for Ochratoxin A
P/N 10515 / 11535

HPLC Separation Column RP C-18
P/N 10522

FREESTYLE SPE, Robotic System
for Automated Sample Clean-up
P/N 12663 / 12668

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