

# Matrix of the Month

December, 2013:  
**Ochratoxin A**  
in Hot Wine Punch



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](mailto:info@LCTech.de)!

## Protocol

10 mL of hot wine punch are strongly mixed with 10 mL extraction solution (5 % NaHCO<sub>3</sub> mit 1 % PEG 8000) for 3 minutes.

The extract is filtrated by a fluted filter.

Then 10 mL of filtrated extract are diluted with 40 mL PBS-puffer (ph 7.2).  
In case of obvious turbidity, you have to centrifugate or filtrate the sample volume by a syringe filter (PVDF).

After the OtaCLEAN column has been opened let drop out the storage buffer.  
Now the diluted raw extract is applied onto the OtaCLEAN column.

A slight vacuum or overpressure could be applied in all steps, in which liquid is given onto the column. It is nevertheless very important, that the flow rate 2 mL/min is not exceeded.

Now let the liquid run through the column completely until there is no sample left on the column. If the column runs dry in this step, that is no problem. But prevent the column bed from drying completely.

To clean the column, the sample reservoir is rinsed with 10 mL water. This solution is applied onto the column. Residual water rests are to be removed by a slight vacuum or gas flow.

1 mL methanol is added onto the open column. Please wait until the methanol has reached the lower luer outlet of the column. The column is closed immediately and is incubated for 5 minutes, to completely break the analyt-antibody-binding. Once the column is reopened the eluates are collected and eluted with further 1 mL.

The eluate is adjusted to HPLC solvent conditions. It is then ready for analysis.

## HPLC Conditions

HPLC: Dionex Ultimate 3000 isocratic

Column oven: 40 °C

Separation column: EC125/3 Nucleosil 120-3 C-18 RP

Flow rate: 0.6 mL/min (40/55/5) (water/methanol/acetonitrile (v/v/v) + 0.1% acetic acid)

Fluorescence detection:

Excitation wavelength: 335 nm

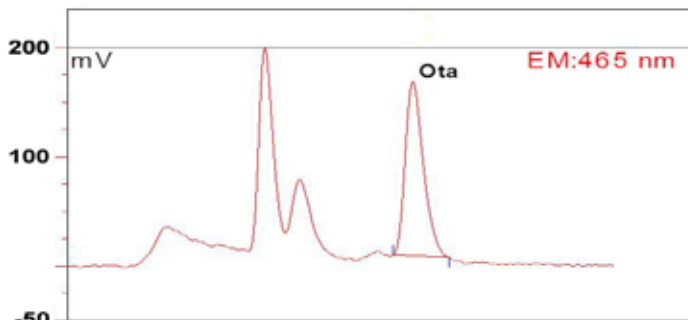
Emission wavelength: 465 nm

Recovery Rate

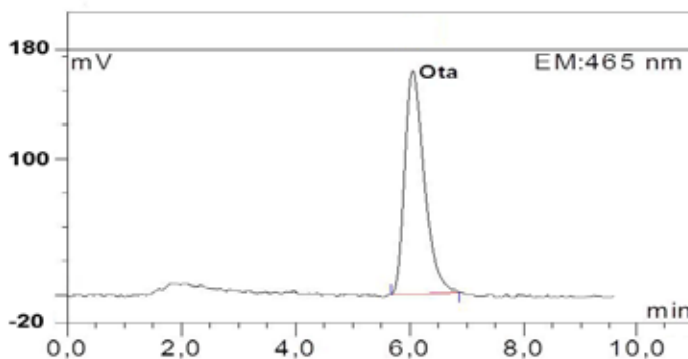
Content of Ochratoxin A in hot wine punch	
	Ochratoxin A
Standard*	100
Recovery rate** hot wine punch, spiked with 2 ppb	92

\* Standard is set = 100 % , \*\* corrected with non-spiked sample

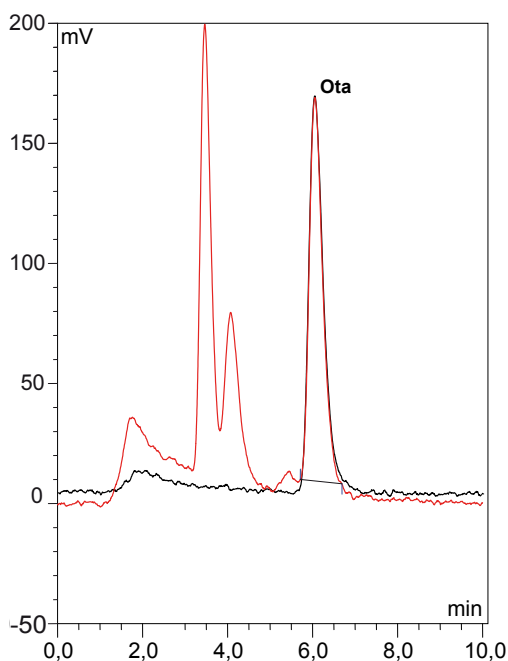
Chromatograms



Hot wine punch (OTA without post column derivatisation), spiked with 2 ppb



Standard, 2 ppb, (10 ng/2 mL)



Overlay of both chromatograms

This LCTech product was used:

OtaCLEAN,  
Immunoaffinity column  
for Ochratoxin A

P/N 10515

Do you have further questions?  
Please simply write an e-mail to [info@LCTech.de](mailto:info@LCTech.de)