Aflatoxin B/G and Ochratoxin A in Dates
Cleaned-up with *Afla-OtaCLEAN*

The Date - “Gold of the Orient“

The “real date palm“ is a plant species from the palm family. It is an ancient oriental cultivated plant whose human use can be traced back to the year 6000 BC.

Dates are rich in sugar, have a high calorie content and grow mainly in warmer regions of the earth, so they are also known as “bread of the desert“. In dried fruit and especially in dried dates, aflatoxins B/G and ochratoxin A can be formed due to incorrect storage conditions and during the drying process. Since these toxins can be highly harmful to human and animal health, tests are carried out regularly. In order to test extracts (e. g. from dates), for both aflatoxins B/G and ochratoxin A in just one step, LCTech has developed the combined immunoaffinity column Afla-OtaCLEAN, which can be used in combination with the EluVac Vacuum Manifold, among others.

Parallel Clean-up - EluVac Vacuum Manifold

Aflatoxin B/G and ochratoxin A are the two most regulated mycotoxins and are formed by molds of the genus Aspergillus and Penicillum. Since mycotoxin clean-up is very important today, LCTech has developed the EluVac Vacuum Manifold as a way to efficiently clean-up more samples in combination with the Afla-OtaCLEAN immunoaffinity column and to speed up this way the clean-up process.

More samples in less time and flexibly adaptable. The EluVac Vacuum Manifold allows the simultaneous clean-up of up to 20 samples, which means that the sample throughput in your laboratory can be increased enormously with only little effort. The EluVac can also be easily and quickly adapted to other SPE - applications in environmental, food and feed analysis.

Processing Protocol

Homogenise 10 g dates with 1 g sodium chloride and extract through 50 mL methanol/water (80/20 (v/v)) and 25 mL of n-hexane to defat and remove essential oils. Run the extraction for at least 30 min. to achieve maximum extraction efficiency.
Subsequently, filtrate the raw extract and dilute 10.5 mL of the n-hexane free phase with 64.5 mL of PBS. In case of precipitation, filtrate the sample through a glass fibre filter to prevent clogging or coelution of matrix components from the column.
Load 50 mL of the diluted sample onto an immunoaffinity column Afla-OtaCLEAN to quantitatively bind the aflatoxins and the ochratoxin A.

Wash the column with 10 mL of deionised water. Use the wash solution to first rinse the sample reservoir and then the Afla-OtaCLEAN column. Next step is to dry the column with a short stream of air.
Then elute the column with 2 mL of methanol. Make sure that the methanol incubates within the column bed for at least 5 min. to ensure complete denaturation of the antibodies.
Simplify the process of sample clean-up by using the EluVac Vacuum Manifold for manual processing. Process up to 20 columns in parallel at individual column flow rates and achieve a huge increase in your sample throughput.

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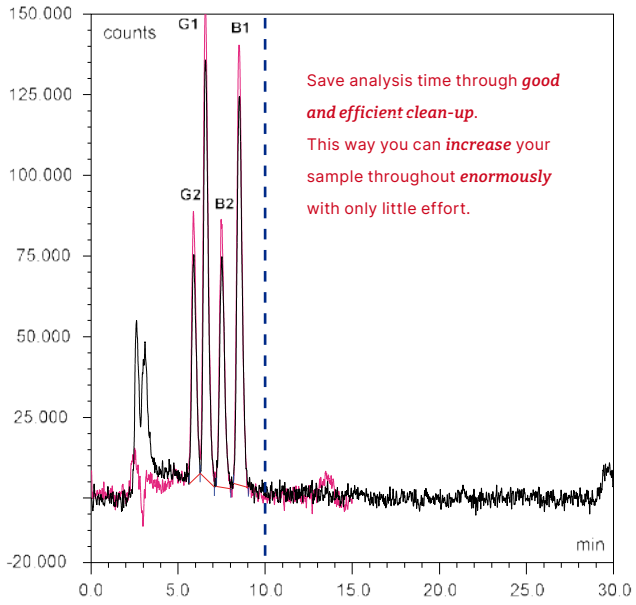
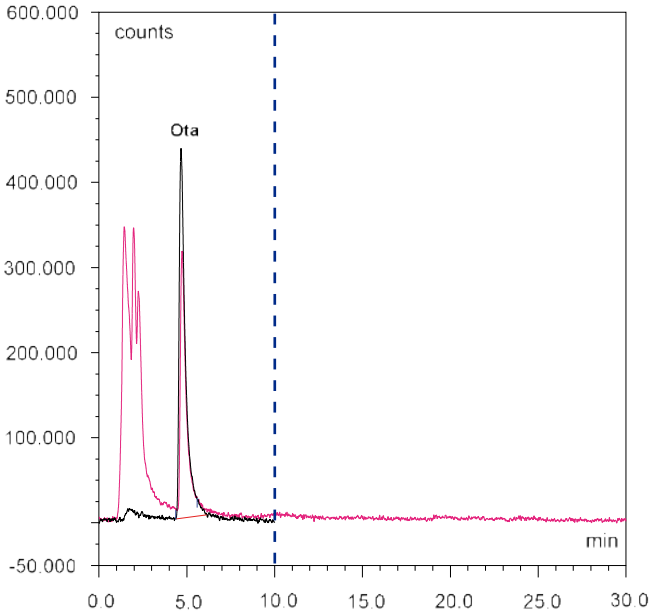
Chromatograms



Red: Standard Aflatoxins 14 ng / 2 mL (equivalent to 10 ppb)
Black: Dates, 10 ppb, cleaned-up with Afla-OtaCLEAN



Red: Dates, 10 ppb cleaned-up with Afla-OtaCLEAN
Black: Standard 14 ng/2 mL Ochratoxin A (equivalent to 10 ppb)

Conditions	
Aflatoxin B/G	
HPLC	isocratic
Column Oven	36 °C
Separation Column	RP C-18 (P/N 10522)
Flow Rate, Eluent	1.2 mL/min; HPLC-water/methanol/ acetonitrile (60/30/15 (v/v/v))
Fluorescence Detection	with derivatisation (UVE/photochemical)
Excitation Wavelength	365 nm
Emission Wavelength	460 nm
Ochratoxin A	
HPLC	isocratic
Column Oven	40 °C
Separation Column	RP EC 125/3 nucleosil 120-3 C18
Flow Rate, Eluent	0.6 mL/min; HPLC-water/methanol/acetonitrile (40/55/5 (v/v/v) + 1 % acetic acid)
Fluorescence Detection	without derivatisation
Excitation Wavelength	335 nm
Emission Wavelength	465 nm

Recovery Rates**

Aflatoxin	B1	B2	G1	G2
Standard*	100	100	100	100
Recovery Rates** Dates, 10 ppb	98	98	98	94
Mycotoxin	**Ochratoxin A**			
Standard*	100			
Recovery Rates** Dates, 10 ppb	82			

*Standard was set = 100%, **Corrected with non-spiked sample / The results are in accordance with the performance specifications of the EC 401 / 2006 (section 4.3.1).

These LCTech products were used:

11022 / 11771	Afla-OtaCLEAN Immunoaffinity Columns for Aflatoxin B/G and Ochratoxin A
12415	EluVac Vacuum Manifold Set
10519	UVE Photochemical Reactor
10522	HPLC Separation Column RP C-18

EluVac Vacuum Manifold with Immunoaffinity Columns *Afla-OtaCLEAN*

Do you have a special request as to which matrix we should test for you? Contact us by e-mal at: info@lctech.de

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