Matrix of the Month

June 2019

Ochratoxin A in Palm Kernel Flour ~ 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

Palm Kernel Flour

Palm oil is global an important raw material in the food industry and for the production of energy. The oil is extracted from the pulp of the palm oil fruit. In additon to the palm oil, the less known palm kernel oil can also be produced from the kernel.

During the extraction process of palm kernel oil, the main by-product is palm kernel flour. The flour is mainly added to animal feed as it is rich in proteins and fibres. Palm kernel flour is mostly found in feed for ruminants, rabbits and poultry.

Mycotoxins may occur during the drying process or if palm kernel flour is stored incorrectly. High mycotoxin contamination in animal feed can lead to significant economic losses in livestock breeding as it has a strong impact on animal health.

Ochratoxin A in Food and Feed

Ochratoxin A is a naturally occurring mycotoxin produced by molds of the genera Aspergillus and Penicillium as primary contamination. The consumption of food and feed contaminated with moulds can lead to serious damage to human and animal health.

A maximum permitted level for mycotoxins has been set EU-wide. If the maximum level is pass, the food or feed may not be imported. LCTech has developed the immunoaffinity columns OtaCLEAN especially for the clean-up of ochratoxin A in food and feed. The column achieves very good recovery rates even with difficult matrices. Futhermore it has a high matrix tolerance and is able to bind Ochratoxin A specifically. It is suitable for both manual and automated processing.

Immunoaffinity column OtaCLEAN

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Processing Protocol

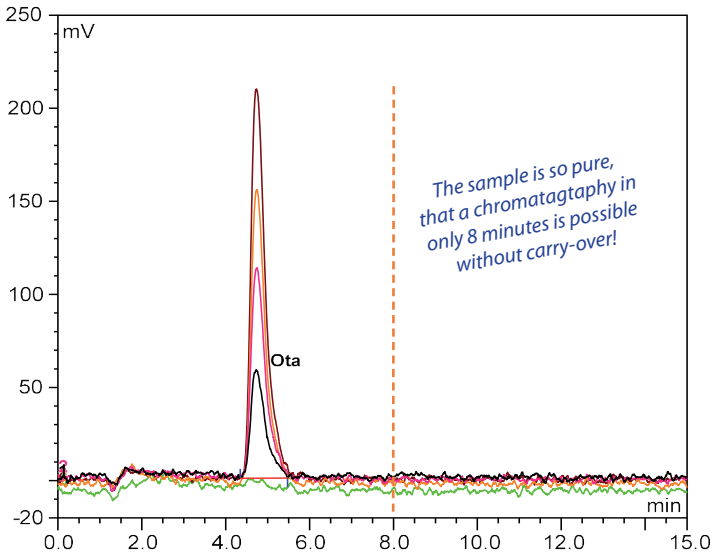
Homogenise 20 g of palm kernel flour and add 2 g of sodium chloride. Extract the sample with 100 mL methanol/ water (80/20 (v/v)) and 50 mL n-hexane in order to remove fat and oil.

For high extraction efficiencies, continue the extraction for at least 10 minutes. Filtrate the raw extract and dilute 2 mL with 12 mL PBS (contains 8 % Tween20). Load 14 mL of sample (represents 0.4 g matrix) onto an immunoaffinity column OtaCLEAN.

Wash the column with 10 mL deionized water to efficiently remove detergent residues. Place the wash solution in portions on the column. Elute the toxin with 2 mL methanol.

Keep in mind the the column bed is incubated with methanol for 5 minutes in order to ensure a fully denaturation of the antibodies and release of the toxin. At the end dilute the eluate for HPLC-conditions.

Chromatogram



Green = Palm kernel flour not spiked
Black = Palm kernel flour10 ppb, spiked
Red = Palm kernel flour20 ppb, spiked
Orange = Palm kernel flour30 ppb, spiked
Brown = Palm kernel flour 40 ppb,spiked

Conclusion

The chromatogram shows a baseline separated chromatography in less than 8 minutes without matrix interference. Thus the sample is so pure that a clear and fast analysis is possible without substantial impairments. The high reproducibility and the high toxin tolerance show the suitability of the OtaCLEAN column for food and feed analysis purpose.

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 (40/55/5 (v/v/v)) + 1 % Acetic acid
Fluorescence Detection:	without Derivatisation
Excitation Wavelength:	335 nm
Emission Wavelength:	465 nm

Recovery Rates
Content of Ochratoxin A in Palm Kernel Flour

Mycotoxin:	Ochratoxin A
Standard*	100
Recovery Rate*** Palm kernel flour, 10 ppb	86
Recovery Rate*** Palm kernel flour, 20 ppb	89
Recovery Rate*** Palm kernel flour, 30 ppb	80
Recovery Rate*** Palm kernel flour, 40 ppb	83

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

These LCTech Products were used:

OtaCLEAN
Immunoaffinity Columns 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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