Matrix of the Month

March 2021

Aflatoxin B/G in Sesame
~ 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: info@LCTech.de

Sample Preparation

MYCOTOXINS

Sesame

Sesame is considered one of the oldest oils in the world and was already cultivated in 300 BC. Sesame originally came from South Asia. Today, the oil plant is cultivated in many tropical and subtropical countries around the world.

Sesame seeds come in different varieties - white, brown and black. These differ not only in taste, but also in the composition of the nutrients. As the fairy tale saying „open sesame" reveals, the seed of the hard capsule fruit is rich in nutrients. Unfortunately, sesame is nowadays often contaminated by mycotoxins and pesticides (ethylene oxide), which are supposed to prevent the growth of moulds and germs. For this reason, strict legal regulations apply throughout the EU for the maximum permissible content.

Automated Mycotoxin Clean-up with FREESTYLE SPE

Mycotoxin analysis is essential in the food and feed sector. LCTech has developed the FREESTYLE SPE to support you in your daily routine tasks in the laboratory and to save time.

Achieve very good recoveries and reproducible results. You can directly transfer any manual SPE method with the already proven system. Simply follow the prepared processing steps described on the following page. Then position the sample in the FREESTYLE SPE, parameterise the method in the software with a few mouse clicks and start the system - done.

Robotic System FREESTYLE

Matrix of the Month

Processing Protocol

Homogenise 20 g of sesame seeds with 2 g of sodium chloride and extract through 100 mL methanol/water (80/20 (v/v)) and 50 mL of n-hexane to remove essential oils.

Run the extraction for at least 30 minutes to achieve the maximum extraction efficiencies. Filter the raw extract and centrifuge at 3000xg for 5 minutes to support phase separation between the n-hexane and the methanolic phase.

Carefully remove 10.5 mL from the methanolic phase (lower liquid phase) and dilute with 64.5 mL PBS. If precipitation and turbidity occur during dilution of the raw extract, filter the sample using a Whatman GF/A filter.

Load a maximum of 50 mL of the diluted sample (corresponds to 1.4 g matrix equivalents) onto an AflaCLEAN immunoaffinity column. Wash the reservoir with 2 x 5 mL deionised water and load the rinsing solution onto the column as well. Then dry the column with a short stream of air.

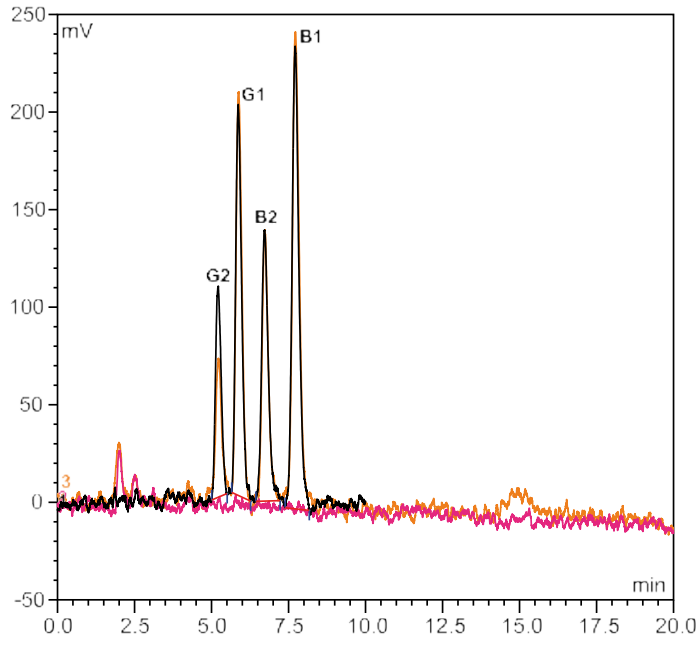
Pipette the eluent (corresponds to 2 mL methanol) onto the column bed. Make sure that the methanol is allowed to act in the column bed for 5 minutes to ensure complete denaturation of the antibodies and thus release of the toxins.

The collected eluate can be diluted or directly analysed in small volumes in LC by fluorescence and post-column derivatisation or by LC-MS/MS.

HPLC-Conditions
Aflatoxin B/G

HPLC:	isocratic
Column Oven:	38 °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)
Flourescence Detection:	With photochemical reactor UVE
Excitation Wavelength:	335 nm 365 nm
Emission Wavelength:	460 nm

Chromatogram



Black: *14 ng/2ml Aflatoxin Mix (corresponds to 10ppb total aflatoxin)*
Red: *Sesame not spiked*
Orange: *Sesame spiked with 10 ppb aflatoxin (10 µg/kg)*

Recovery Rates
Content of Aflatoxins B1, B2, G1 und G2 in sesame

Aflatoxins	B1	B2	G1	G2
Standard*	100	100	100	100
Recovery Rate** Sesame, 10 ppb	95	92	97	79

* 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:

AflaCLEAN Immunoaffinity Columns for Aflatoxin B/G
P/N 10514 / 11721

FREESTYLE SPE, Robotic System for Automated Sample Preparation
P/N 12663 / 12668

UVE Photochemical Reactor
P/N 10519

HPLC Separation Column RP C-18
P/N 10522

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