



December 2016

Aflatoxin B/G in Brazil Nut Fully Automated via FREESTYLE ThermELUTE™

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

Brazil Nuts: A Must in Winter Season

Especially in the months October till December nuts are very popular. Brazil nuts have a very strongly aromatic taste and are full of positive ingredients like zinc, iron or vitamin E. Aflatoxins often damage nuts due to incorrect or too long storage. Aflatoxins are toxins, which are not immediately recognisable by smell, taste or with the naked eye. Particular for the import from third countries the implementing regulation of the European commission, (EG) Nr. 1152/2009 set intensive sampling and investigations in order to control the high risk of aflatoxin contamination.

Mycotoxin Analysis via FREESTYLE ThermELUTE™

The robotic system FREESTYLE ThermELUTE™ facilitates fully automated mycotoxin analysis in combination with any HPLC system. It achieves such sensitive results that the user can be sure to measure below the European limits. With the unique technique of thermal denaturation the comprehensive automation „from raw extract to chromatogram“ is realised without any manual working steps.

The system achieves the high sample throughput of 500 samples/week by using the SMART immunoaffinity columns with a size of only 3.5 cm. Through miniaturisation of the overall process, not only the processing time of the sample is dramatically reduced, but also the required amounts of sample and solvent. Each sample is processed with a SMART column, which grants top performance for each sample and reliably prevents cross-contamination.



ThermELUTE™
gripper injects
SMART-column

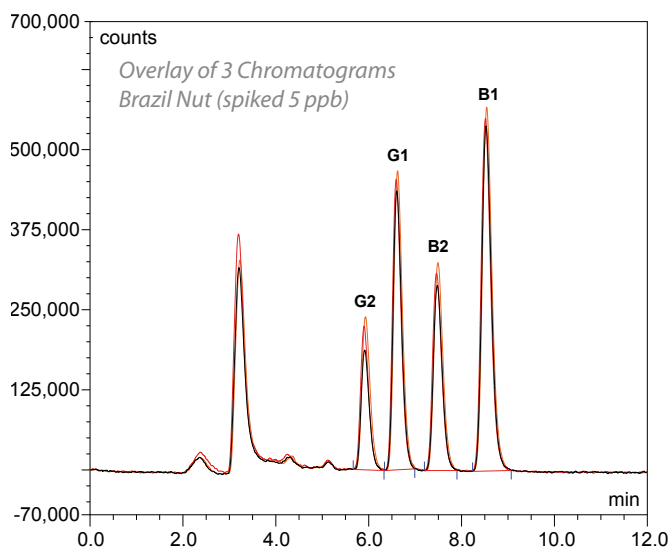
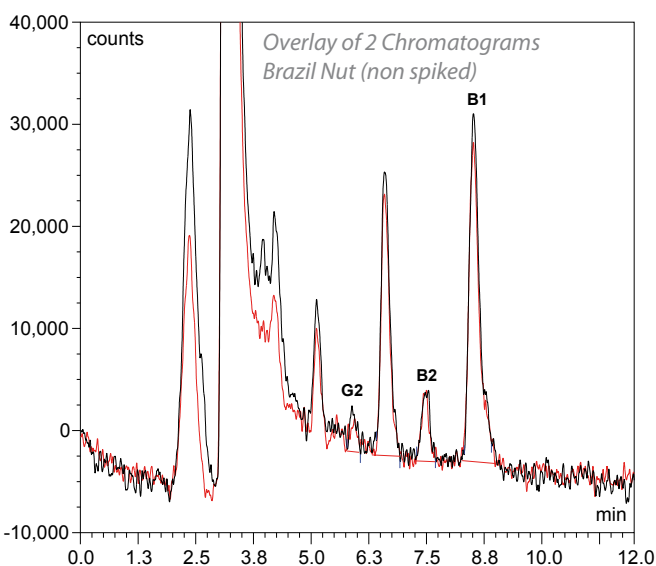
... easy and reliable sample preparation around the clock!

Protocol of Manual Processing

Homogenise 10 g of brazil nuts and add 2 g of sodium chloride. Extract the sample with 100 mL (80/20 (methanol/water (v/v))) and dilute with 50 mL n-hexane in order to remove fat and oils. Filtrate the sample and dilute 2 mL with 12 mL PBS buffer (pH value 7.2) which contains 8% Tween20. Depending to the detector sensitivity you can dilute the extract further (in this case 1:4).

FREESTYLE ThermELUTE™ loads the extracted, diluted and filtrated sample automatically onto the high-performance AflaCLEAN SMART column. Due to the high sensitivity of the FREESTYLE ThermELUTE™ the amount of matrix can be either drastically reduced (down to 0.02 g) or it can be achieved a very high measure sensitivity until the range of ppt.

With a applied flow rate of 1.5 mL the column will be washed with 2 mL deionized water. Afterwards the column will be thermal eluted by ThermELUTE™ technology and on-line injected directly into the HPLC. The results are excellent, high-resolution chromatograms within 10 minutes measuring time.



HPLC-Conditions (Aflatoxins B/G)

HPLC:	isocratic
Column Oven:	36 °C
Separation Column:	RP C-18 (P/N 10522)
Flow Rate:	1.2 mL/min
Eluent:	HPLC-water/methanol/ acetonitrile (60/30/15 (v/v/v))
Fluorescence Detection:	UV-Derivatisation with UVE Photochemical Reactor
Excitation Wavelength:	365 nm
Emission Wavelength:	460 nm

Recovery Rates

Content of Aflatoxins B/G in Brazil Nuts

Mycotoxin	B1	B2	G1	G2
Standard*	100	100	100	100
Recovery Rate** Brazil Nuts, 5 ppb	92	95	99	90

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



AflaCLEAN SMART Columns with Luer Tip

These LCTech products were used:

AflaCLEAN SMART,
Immunoaffinity Columns for Aflatoxins B/G
P/N 12862 / 12863

UVE Photochemical Reactor
P/N 10519

FREESTYLE ThermELUTE™, Robotic System
for fully Automated Sample Preparation
P/N 12663 / 13317