

Aflatoxins in hazelnuts

Cleaned-up with *AflaCLEAN SMART*Automated with *FREESTYLE ThermELUTE™*



Hazelnuts

In January, we referred on the enormous number of complaints for nuts and nut products due to mycotoxins. To ensure the highest possible testing capacity for laboratories, we go into more detail about automating sample preparation for mycotoxins with our FREESTYLE ThermELUTE $^{\text{TM}}$ system.

FREESTYLE ThermELUTE™

From filtered and diluted crude extract to chromatogram without manual intermediate steps: performed by FREESTYLE ThermELUTE™ combination with HPLC. The FREESTYLE BASIC is equipped with an SPE- and a ThermELUTE™-module. This combination enables fully automated and very fast processing of the convenient SMART immunoaffinity columns. Elution is performed directly from the immunoaffinity column into the HPLC.

FREESTYLE ThermELUTE™ is unique

- Great reduction in workload with simultaneous increase in analysis numbers and greater measurement sensitivity (ppt instead of ppb)
- Measurement and monitoring even in the range of the required MRL for baby food
- Any type of food and feed without restrictions for matrices - individual sample volumes definable

FREESTYLE processes the complete sample

- Loading of a sample aliquot, washing and transfer to heating block
- Transfer large volume aqueous elution directly into HPLC
- Automation is already useful from 10 samples per day
- Matrix-independent increase in measurement sensitivity





Full automation in mycotoxin analysis

Automated processing with AflaCLEAN

Extract 20 g of homogenized matrix (hazelnut) with 100 mL of 80/20 (v/v) methanol/water. For defatting, add an additional 50 mL of n-hexane for at least 5 minutes. After a supporting centrifugation at 3.000 xg for another 5 minutes, you can use the methanolic sample (lower phase) for further processing. Dilute 7 mL of the matrix equivalents with 43 mL of PBS buffer. Depending on the limit of quantitation, load 1-10 mL of the diluted sample onto the AflaCLEAN SMART column in

the FREESTYLE ThermELUTE™. Through process control, the AflaCLEAN SMART immunoaffinity column is automatically washed with 2 mL of deionized water and after thermal denaturation, the analyte, salt-free, is quantitatively transferred to the sample loop in the AflaELUATOR. Synchronized processing allows direct injection into the HPLC and immediate analysis of the sample. Crosscontamination from sample to sample is eliminated by using sample-specific columns and consistent cleaning of the system (see Figure 1).





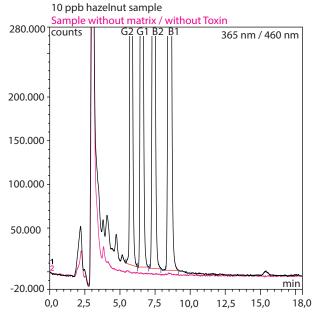


Fig. 1: Cross-contamination test - no carry-over

Conditions				
HPLC / UPLC	isocratic			
Column oven	35 °C			
Separation column	P/N 10522			
Flowrate, Solvent	1.2 mL/min; LC-solvent water/ methanol/acetonitril (60/30/15 (v/v/v))			
Fluorescence detection	UVE (photochemical)			
Excitation wavelength	365 nm			
Emission wavelength	460 nm			

Recovery rates**					
Aflatoxin	B1	В2	G1	G2	
Standard*	100	100	100	100	
0,025 ppb	107	108	99	93	
0,125 ppb	107	104	97	98	
1,25 ppb	105	106	92	101	
2,5 ppb	104	107	97	104	

^{*} Standard was set = 100%

The recoveries are 3 measurements each, and the measurement errors are within the column specifications.

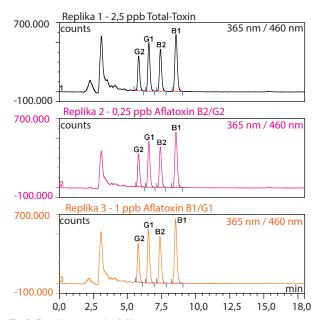


Fig.2: Excellent reproducibility -3 identical samples were processed and show a reproducible analysis for the aflatoxins B/G with high measurement sensitivity

Conclusion

The FREESTYLE ThermELUTE™ is the most innovative fully automated system for the processing, clean-up and complete analysis of mycotoxins, such as aflatoxins B/G or aflatoxin M1, but also ochratoxin A or zear-alenone.

The technology makes it possible to achieve measuring ranges far below the European regulations for baby food even with an HPLC and allows a massive reduction in sample volume in the food and feed sector. With a slight adjustment of the volumes, the extraction and sample dilution protocols, for the standard 3 mL AflaCLEAN or OtaCLEAN columns can be used. In addition, salt-free aqueous injection offers the advantage of long analytical column life.

Measurement sensitivity, automation in sample processing and reproducibility are characteristic features of the system in addition to the high-quality and robust processing.

These LCTech products were used:

FREESTYLE ThermELUTE™
12863 AflaCLEAN SMART (1000 pcs/unit)
12862 AflaCLEAN SMART (100 pcs/unit)
10519 UVE
10522 HPLC column for mycotoxins
13893 AflaELUATOR

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



^{**} Corrected with non-spiked sample / The results are in accordance with the performance specifications of EC 401 / 2006 (section 4.3.1).