

# Aflatoxin B/G in chili/paprica Cleaned-up with *AflaCLEAN™ SMART*



# AflaCLEAN<sup>™</sup> SMART - IAC clean-up column for the analysis of aflatoxin B/G

Despite their small size, the immunoaffinity columns of only 3 cm convince with a high loading capacity, high matrix compatibility, lower price, reduced solvent consumption and shorter processing times.

In the following use case, only 7 minutes are required for sample processing. Less than 2 minutes for column loading and washing + 5 minutes for the elution process.

## **Processing protocol**

# Contamination risk of Chili and paprica

The risk of contamination with aflatoxins and ochratoxin A is very high in chili and paprica due to the *drying processes*.

Intensive controls on imported material have been able to ensure the safety of chili and paprica products for the food sector, but samples containing aflatoxins and ochratoxin A far above regulatory limits are repeatedly found. (*Rajendran et al. 2021; Enamullah et al.2022*)

Consistent and continuous analysis of chili and paprica is therefore necessary to minimize the hazards of mycotoxin exposure by these products and to ensure food quality.



Mix 20 g homogenized sample material with 2 g sodium chloride. For extraction use 100 mL methanol/water (80/20 (v/v)). During the extraction process, add 50 mL of n-hexane to efficiently remove fats and oils. An extraction of at least 15 minutes is recommended.

Filter the crude extract and then centrifuge at 3000 xg for 5 min to achieve optimal separation of the methanolic lower phase from the n-hexane phase. Mix 2 mL of the lower methanolic phase with 12 mL of PBS buffer containing 8% Tween20. Load 2.8 mL onto the AflaCLEAN<sup>™</sup> SMART immunoaffinity column at a flow rate of 3 mL/min. Rinse the sample reservoir with 2 mL of deionized water and load the wash solution onto the IAC column at the same flow rate.

After washing the column bed, dry the column with an air stream. Add 0.4 mL of methanol onto the column and let it incubate. Ensure that it is allowed to soak in the column bed for at least 5 min to ensure complete denaturation and thus elution of the toxin. Samples can be analyzed when diluted to the HPLC solvent conditions.

### APPLICATION NOTE AN1167





Linear (Aflatoxin B2) ..... Linear (Aflatoxin B1)

The linearity shown in the *figure* demonstrates the consistently good recoveries for matrices with different toxin loads.

··· Linear (Aflatoxin G2) ······ Linear (Aflatoxin G1)

The high recoveries demonstrate the suitability of the AflaCLEAN<sup>™</sup> SMART columns for the analysis of chili and paprica samples using the extraction and sample dilution protocol presented here.



Overlaid chromatograms of individual chili samples, naturally contaminated, spiked and without toxin findings.

The highly selective clean-up of the AflaCLEAN<sup>™</sup> SMART column enables reproducibale baseline-separated chromatography in less than 10 minutes for all 4 aflatoxins. Possible interferences are completely removed from the analytical chromatography.

#### These LCTech products were used:

12862	AflaCLEAN™	SMART	(100 pcs/box)
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- 10519 UVE (Photochemical derivatization)
- HPLC column for mycotoxins

Recovery rates						
Aflatoxin	B1	B2	G1	G2		
Standard*	100	100	100	100		
Recovery rates ** 20ppb						
Chili habanero	98	91	94	86		
Chili cayenne	91	90	92	84		
Chili jalapeño	90	88	95	84		
Hot paprica	99	95	93	91		
Sweet paprica	98	100	91	95		
Smoked paprica	93	91	93	81		
Recovery rates ** 10ppb						
Chili habanero	99	98	96	97		
Chili cayenne	92	95	100	88		
Chili jalapeño	92	95	100	88		
Hot paprica	99	100	91	89		
Sweet paprica	100	98	96	96		
Smoked paprica	97	95	95	86		

\* Standard was set = 100% set

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

Conditions				
HPLC	Isocratic			
Column oven	35 °C			
Separation column	PN 10522			
Flow rate, Running medium	1.2 mL/min (60/30/15) (HPLC water/methanol/acetonitrile)			
Fluorescencedetection	Derivatization by UVE (photo- chemical) (PN 10519)			
Excitation wavelength	365 nm			
Emmission wavelength	460 nm			

## Conclusion

Fast, efficient sample clean-up for HPLC- FLD or LC-MS/MS analysis of spices (here chili and paprica) using a toxin / analyte specific clean-up column.

With a SMART sample preparation you are guaranteed easy & fast processing, high efficiency and reproducible performance.

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



LCTech GmbH Daimlerstr. 4

84419 Obertaufkirchen, Germany Tel. +49 8082 2717-0

info@LCTech.de www.LCTech.de

Illustrations and

Aflatoxin B1

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