April 2018
Aflatoxin M1 in Greek Yoghurt
~ 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

Greek Yoghurt

Greek yoghurt is especially characterised for its creamy consistency. Due to the special manufacturing process, this yoghurt contains fewer carbohydrates, but twice as much protein as other yoghurts and is therefore particularly popular for athletes. The very high fat content is no problem in this context, because Greek yoghurt has „good" fats. Of course, it should not be enjoyed in excess. In Greece, the yoghurt is traditionally refined with fruits, nuts and honey, and is also used for hearty dishes, such as tsatsiki.

Sample Clean-Up of Aflatoxin M1 in Food and Feed

Aflatoxin M1 is produced within the organism of animals from aflatoxin B1, after consumption of feed contaminated with Aflatoxin B1. Afterwards, you will find the toxin in dairy products, such as Greek yoghurt.

LCTech developed the AflaCLEAN M1 Select immunoaffinity column for sample preparation routine analysis using HPLC with fluorescence detection or LC-MS. It is especially designed for the clean-up of aflatoxin M1 in dairy products and achieves excellent recoveries. Like all LCTech columns, AflaCLEAN M1 Select is suitable for both manual and automated processing, e. g. with the LCTech FREESTYLE SPE robotic system.

Each manual SPE-method which has already established in your laboratory can be quick and easy transferred to the FREESTYLE system. Extract, filtrate and dilute the Greek yoghurt according to the protocol of manual processing. Equip the racks with the immunoaffinity columns, choose the method in the software and press the start button. Subsequently, you get reproducible and reliable results.

Manual Processing Protocol

Homogenise 20 g yoghurt with 100 mL water. For particularly good extraction efficiencies, continue the extraction for 30 minutes. Filtrate the raw extract and dilute 21 mL of it with 129 mL PBS.

Load 100 mL of the sample (represents 2.8 g matrix) onto a immunoaffinity column AflaCLEAN M1 Select. Wash the sample reservoir afterwards with 10 mL deionised water and load this solution also onto the IAC-column.

Dry the column by flushing air through it and elute the toxin afterwards with 2 mL of methanol. Keep in mind that the column bed is incubated with the methanol for at least 5 minutes in oder to ensure the complete denaturation of the antibodies.

Dilute the eluate to HPLC conditions and measure it afterwards via fluorescence or LC-MS.

Chromatogram
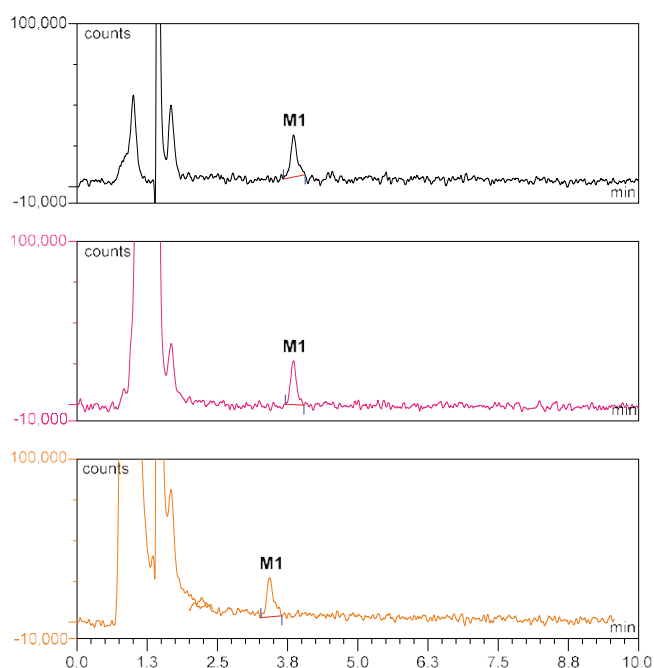
Black: Yoghurt spiked after extraction (160 ppt)
Red: Standard 160 ppt (0.448 ng / 2 mL)
Orange: Yoghurt spiked before extraction (160 ppt)

FREESTYLE SPE with EVAporation-module for automated sample preparation in mycotoxin-analysis

HPLC-Conditions
(Aflatoxin M1)

Mycotoxin:	Aflatoxin M1
HPLC:	isocratic
Column Oven:	36 °C
Separation Column:	RP C.18
Flow Rate:	1.2 mL/min
Eluent:	HPLC-water/methanol/ acetonitrile (60/30/15 (v/v/v))
Fluorescence Detection:	without derivatization
Excitation Wavelength:	365 nm
Emission Wavelength:	435 nm

Recovery Rates
Content of Aflatoxin M1 in Greek Yoghurt

Mycotoxin	Aflatoxin M1
Standard*	100
Recovery Rate** Greek Yoghurt, 160 ppt	96

*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:

AflaCLEAN M1 Select, Immunoaffinity Column for Aflatoxin M1
P/N 14202/ 14201

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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2