



April 2019

## Aflatoxin B/G and Ochratoxin A in Juniper Berry ~ 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](mailto:mycotoxins@LCTech.de)

### Sample Preparation

MYCOTOXINS

#### Juniper Berry

Traditional medicinal plant or poisonous berry? The juniper plant is provided with many myths. The antique Egyptians and Greeks used the plant already as remedy, on the other hand the people in the Middle Ages used the berry as a magic agent. Today, the juniper is no longer a magic remedy, but it is applied as a medicinal plant and spice.

The juniper berries are often called healing plants, because of the proportion of tannins, essential oils and saponin, that in its special combination causes a diuretic effect.

The berry is also used as a spice in the kitchen and in the production of gin. The plant is only poisonous if it is overdosed and can therefore trigger kidney pain or tachycardia.

It is mostly applied in dried form. As a result of incorrect storage, unwanted mould formation can occur, resulting in an excessively high mycotoxin concentration, which reduces the quality of the product and can cause damage to health.

#### Two in One – Combined Immunoaffinity Column Afla-OtaCLEAN

Aflatoxin B/G and Ochratoxin A are often found together in many foods and feed, also in the juniper berry. To purify both toxin groups you usually need two individual columns.

LCTech simplified this processes with the development of the two-in-one immunoaffinity column Afla-Ota-CLEAN. It offers the option to clean-up the Aflatoxine B/G und Ochratoxin A in one step of a procedure. This saves half of the time and at the same time the extract can be tested for several mycotoxins.

The Afla-OtaCLEAN columns are also suitable for difficult matrices to purify both toxin groups in one operation. In this way, time and money can be saved and the product quality can be significantly improved, especially in the area of spices.



Immunoaffinity columns Afla-OtaCLEAN

## Processing Protocol

Homogenise 10 g of juniper berries and add 1 g sodium chloride. Extract the sample through 50 mL methanol/water (80/20 (v/v)) and add 25 mL n-hexane in order to remove fat for at least 10 minutes.

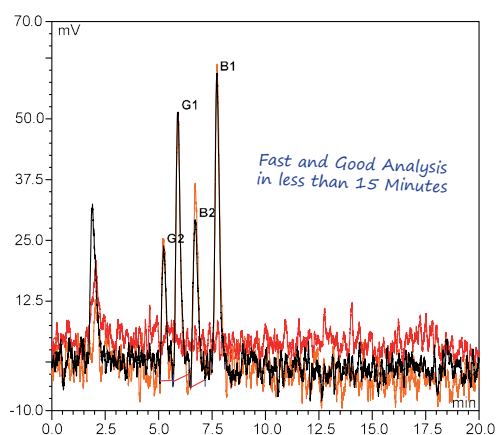
Filtrate the raw extract and dilute 2 mL of the n-hexane free phase with 12 mL PBS (contains 8 % Tween20).

Load 14 mL of the sample (represents 0.4 g of matrix) onto the respective Afla-OtaCLEAN immunoaffinity column.

Wash the column with 10 mL deionized water and dry it afterwards by flushing air through it.

Elute the toxin with 2 mL methanol. Keep in mind that the column bed is incubated with methanol for 5 minutes to ensure a fully denaturation of the antibodies and release of toxin.

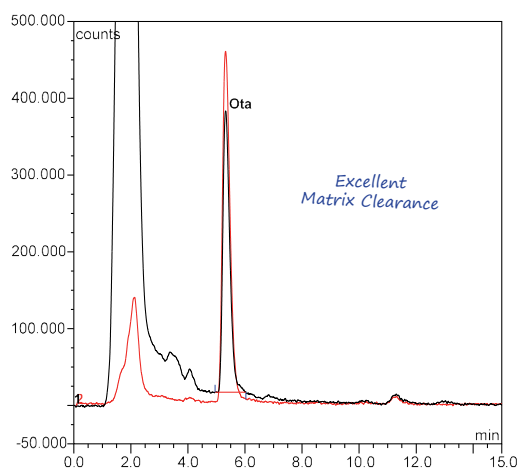
## Chromatograms



**Black:** Standard 10 ppb (4 ng/ 2 mL)

**Red:** Juniper berry not spiked, cleaned-up with Afla-OtaCLEAN

**Orange:** Juniper berry 10 ppb cleaned-up with Afla-OtaCLEAN



**Black:** Juniper berry spiked with 10 ppb

**Red:** Ochratoxin A Standard 10 ppb (4 ng/2 mL)

## HPLC-Conditions

(Aflatoxin B/G / Ochratoxin A)

Mycotoxine:	Aflatoxin B/G	Ochratoxin A
HPLC:	isocratic	isocratic
Column Oven:	36 °C	40 °C
Separation Column:	RP C-18 (P/N 10522)	RP C-18 (P/N 10522)
Flow Rate:	1.2 mL/min	0.6 mL/min
Eluent:	HPLC-Water/ Methanol/Acetonitrile (60/30/15 (v/v/v))	HPLC-Water/ Methanol/Acetonitrile (40/55/5 (v/v/v)) + 1 % Acetic Acid
Fluorescence Detection:	Derivatisation with UVE Photochemical Reactor	Without Derivatisation
Excitation Wavelength:	365 nm	335 nm
Emission Wavelength:	460 nm	465 nm

## Recovery Rates

Content of Aflatoxin B/G and Ochratoxin A in Juniper Berry

Mycotoxine	B1	B2	G1	G2	OTA
Standard*	100	100	100	100	100
Recovery Rate** Juniper Berry, 10 ppb (Afla-OtaCLEAN)	93	87	98	91	82

\*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:

Afla-OtaCLEAN Immunoaffinity Columns for  
Ochratoxin A and Aflatoxin B/G  
P/N 11022 / 11771

HPLC Separation Column RP C-18  
P/N 10522

UVE Photochemical Reactor  
P/N 10519