Aflatoxins B/G und Ochratoxin A in soy flakes

manual and automated

Soy flakes

As a protein-rich raw material, the cultivation of the soybean is becoming increasingly important worldwide. The soybean is a crop that belongs to the pulses family. As we know today, it has been cultivated by humans for a long time. There is already evidence of soy cultivation in north-eastern China between 1700 and 1100 BC. Soy, and also soy flakes, contain a high proportion of unsaturated fatty acids as well as a lot of protein. The special thing about this protein is that it resembles animal protein despite its plant origin. The soy flakes are made from the fruit of the soybean, which has previously undergone a gentle processing process to remove the shell, bitter substances and the so-called trypsin inhibitor, which can disrupt the human digestive system. Processed in this way, soy flakes are easily digestible, wholesome and edible in a variety of ways.

Immunoaffinity columns for aflatoxins B1, B2, G1 and G2

Just like OtaCLEAN and Afla-OtaCLEAN, the AflaCLEAN columns are available in the practical 3 mL polypropylene format. The column bed consists of a soft gel with a storage buffer on top containing a preservative. The loading capacity is 150 ng aflatoxin B1 absolute. The columns can be stored at room temperature or 4 °C to 30 °C for 24 months (from date of manufacture). The AflaCLEAN columns are suitable for manual as well as automated processing, e.g. with the LCTech robotic system FREESTYLE SPE.

For more detailed information on our immunoaffinity columns, visit our website.

Advantages of the AflaCLEAN column at a glance:

- 3 mL format for homogeneous running speed and optimal clean-up
- Outstanding shelf life up to 24 months at room temperature
- Loading capacity: 150 ng aflatoxin B1
- Recoveries:
  B1 > 90 %, B2 > 80 %, G1 > 90 %, G2 > 60 %

Processing protocol

Extract 20 g soy flakes with 2 g sodium chloride in 100 mL methanol/water (80/20 (v/v)) for 30 minutes. Add 50 mL n-hexane during the 30 min extraction to extract fats and oils. Filter the crude extract, if there is no clear phase separation after filtration, centrifugation at 3000 x g for 10 minutes is helpful.

Dilute 2 mL of the methanolic (lower) phase with 12 mL of PBS buffer containing 8 % Tween20, which reduces non-specific binding of matrix components to the IAC column and prevents precipitation of matrix components after dilution with PBS buffer.

Processing protocol continued →

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Processing protocol / 2

Load 14 mL onto an immunoaffinity column (AflaCLEAN, Afla-OtaCLEAN or OtaCLEAN, this corresponds to a matrix amount of 0.4 g. The sample sample reservoir is rinsed with 5 mL deionised water and the rinsing solution is loaded onto the immunoaffinity column. The column is washed again with 5 mL deionised water.
After the wash solution has passed through the column, the column is dried by a flush of air and eluted using 2 mL methanol. The methanol should be allowed to soak in the column bed for at least 5 minutes to complete denaturation of the antibodies and thus elution of the toxins. The eluate is diluted to hplc solvent concentration or injected directly in small volumes. The samples can be analysed by HPLC fluorescence or by LC-MS/MS.

Conclusion

Soy samples, in this case soy flakes, can be analysed for the major storage fungal toxins using the products AflaCLEAN and OtaCLEAN, but also using the combination column Afla-OtaCLEAN. Selective clean-up using the AflaCLEAN and Afla-OtaCLEAN immunoaffinity columns allows sensitive, selective clean-up of aflatoxins with very good recovery rates and excellent chromatographic results.
The purified samples can be analysed by HPLC fluorescence and photochemical derivatisation or by LC-MS/MS. Ochratoxin A, which can also be found as a highly regulated toxin in soy, can also be highly selectively purified using the OtaCLEAN and the Afla-OtaCLEAN to determine and control the low tolerated toxin levels allowed in soy matrices.
These products allow sensitive mycotoxin analysis by HPLC fluorescence or LC-MS/MS and can be applied on various matrices.

Recovery rates Aflatoxins**

Aflatoxin	B1	B2	G1	G2
Standard*	100	100	100	100
Soy flakes AflaCLEAN 10 ppb	102	94	98	95
Soy flakes Afla-OtaCLEAN 10 ppb	103	93	101	86

Recovery rates Ochratoxin**

	Ochratoxin A
Standard*	100
Soy flakes OtaCLEAN 10 ppb	88
Soy flakes Afla-OtaCLEAN 10 ppb	89

* Standard was set = 100% ,
** Corrected with non-spiked sample / The results are in accordance with the performance specifications of EC 401 / 2006 (section 4.3.1).

Conditions

	Aflatoxin	Ochratoxin A
HPLC	isocratic	isocratic
Column oven	36 °C	40 °C
Separation column	RP C18 PN 10522	RP EC 125/3 nucleosil 120-3 C18
Flowrate, Solvent	1.2 mL/min; HPLC-Water/ Methanol/Acetonitrile (60/30/15 (v/v/v))	0.6 mL/min; HPLC-Water/Methanol/Acetonitrile (40/55/5 (v/v/v)+1% Essigsäure)
Derivatisierung	Photochemical by means UVE	without derivatisation
Fluorescence detection		
Excitation wavelength	365 nm	335 nm
Emission wavelength	460 nm	465 nm

These LCTech products were used:

10514	AflaCLEAN
11022	Afla-OtaCLEAN
10515	OtaCLEAN
10519	UVE
10522	HPLC-column
10750	Pre-column holder
10523	Pre-column (Guard)

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

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