



Ochratoxin A in red wine

Clean-up with *OtaCLEAN*



Red wine

The presence of ochratoxin A in wine has been monitored over several years and revealed a level of the toxin occurring, which required to set a regulation.

A level of 2 ppb (2 µg/L) has been established for the European Union, which are not exceeded in most cases.

In this matrix of the month we show the presence of OTA and the performance of mycotoxin clean-up for a good, reproducible analytical measurement by HPLC-FLD or LC-MS/MS for some specific wine varieties with different residual sweetness.

OtaCLEAN - IAC clean-up columns for the analysis of ochratoxin A

The *OtaCLEAN* column can find best contamination of OTA in red wine samples, which are more difficult to analyse than other matrices due to tannin and colouration. The concentration of the toxin by the antibody and the selective binding of the analyte allows a very accurate analysis not only of beverages and wine, but of many other difficult matrices.

With the combination of ***FREESTYLE SPE*** and the ***ThermELUTE™***, which injects the samples directly after clean-up, you achieve full automation of the analysis of aflatoxins B1, B2, G1, G2 as well as M1, zearalenone and ochratoxin A from the extract to the chromatogram.

Processing protocol

The wine samples are extracted with a 1% polyethylene glycol and 5% sodium hydrogen carbonate solution. Mix 10mL of wine with 10mL extraction solution as described above. After mixing for at least 3 minutes and subsequent filtration, 10 mL of the extract is diluted with PBS buffer to a final volume of 50 mL.

The sample is loaded onto the *OtaCLEAN* column at a maximum flow rate of 2 mL/min. Wash the Sample reservoir twice with 5mL deionised water and load it onto the column. This is used to wash the column bed as well. After emptying the column dry it with a flush of air.

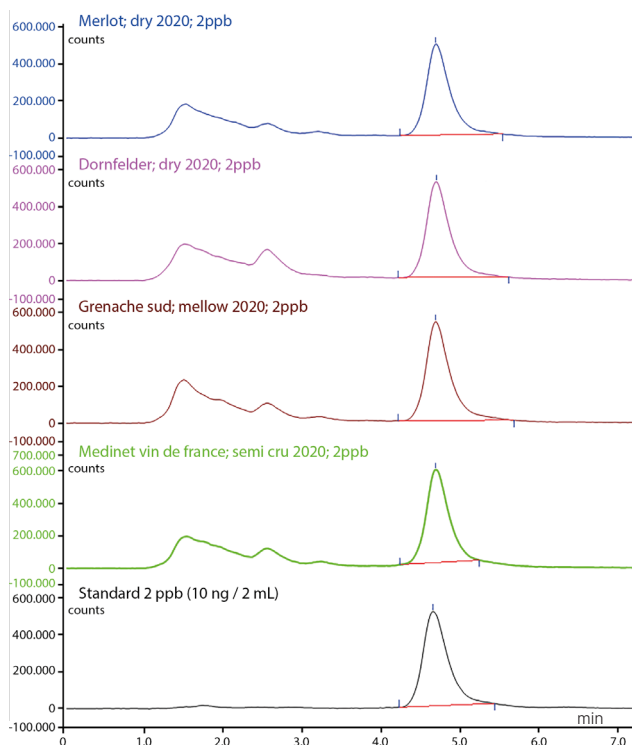
To elute the toxin, load 2 mL of Methanol and let it stand for 5 min for incubation. After elution the analytes are measured by HPLC fluorescence or by LC-MS/MS.



Fig 1.: *OtaCLEAN* automated on the *FREESTYLE* system



Chromatogram



This LCTech product was used:

10515 OtaCLEAN (25 pcs/box)

Conditions

HPLC	Isocratic
Column oven	40 °C
Separation column	RP EC 125/3 nucleosil 120-3 C18
Flow rate, Solvent	0.6 mL/min; HPLC water/methanol/ acetonitrile (40/55/5 (v/v/v) +1 % acetic acid)
Fluorescencedetection	Without derivatisation
Excitation wavelength	335 nm
Emission wavelength	465 nm

Recovery rates** in red wine

Analyt	Ochratoxin A
Standard *	100
Merlot 2020; 2 ppb	97
Merlot 2020; 5 ppb	99
Dornfelder 2020; 2 ppb	103
Dornfelder 2020; 5 ppb	104
Grenache Grand sud 2020 2 ppb	100
Grenache Grand sud 2020 5 ppb	96
Medinet 2020 2 ppb	103
Medinet 2020 5 ppb	95

* 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).

Conclusion

The clean-up of ochratoxin A by OtaCLEAN allows highest sample purity and concentration of analytes to meet the requirements of food and feed regulations.

The challenge in Wine alalysis, posed explicitly by some very sweet or coloured wine samples shows the high binding affinity of OtaCLEAN and the high matrix tolerance. The clean-up enables chromatography that is comparable to the one of the analytical standard and thus greatly simplifies data analysis and interpretation. The high matrix tolerance and the selectivity of the clean-up allows the use in wide range of matrices in food or feed analysis.

The presence of OTA in wine samples, as described in the literature, has increased the need to verify compliance with legal limits. In this case study, all wine samples contained quantifiable amounts of OTA between 0.05 and 0.13 ppb, which, however, did not exceed the legal limits.

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