Supelco®
Analytical Products

Application Note

Improved Recoveries and Lower Background for the Analysis of PAHs in Olive Oil

Using SUPELCLEAN™ EZ-POP NP and an SLB®-5ms GC Column

A recent survey implemented by Supelco revealed that 58% of analytical chemists in the food industry find the analysis of compounds in fatty matrices to be highly problematic. Lipid interferences can produce elevated detection limits, contamination of LC and GC systems, and ultimately decrease instrument and column lifetime.

Current extraction and cleanup techniques for fatty samples may be time consuming, expensive, and/or provide insufficient background removal prior to analysis. For this reason, a new approach has been developed. The SUPELCLEAN™ EZ-POP NP, a dual-layer SPE cartridge containing Florisil® and Z-Sep/C18, was designed for the extraction of nonpolar analytes from oil matrices. By means of Lewis acid/base and hydrophobic interactions, fatty matrix interferences are preferentially retained by the cartridge while analytes of interest are eluted. In this way, lipid interferences are removed from the sample.

Experimental

The SUPELCLEAN™ EZ-POP NP and two competitor silica gel SPE cartridges were compared in terms of matrix removal and analyte recovery for the extraction of select polycyclic aromatic hydrocarbons (PAHs) from olive oil. Multiple replicates of both unspiked and spiked (20 ng/g with PAHs) oil samples were processed for each SPE cleanup technique. The cleanup procedures are summarized in **Tables 1** and **2**.

Table 1. Cleanup Procedure for Olive Oil using SUPELCLEAN™ EZ-POP NP

1.	Add 10 mL of acetone to the SUPELCLEAN™ EZ-POP NP cartridge(54341-U), and allow it to elute with gravity.
2.	Dry the cartridge at -10" Hg for 10 min.
3.	Accurately weigh 0.500 mL of oil directly onto the SPE cartridge.
4.	Add the internal standard directly to the oil on the cartridge.
5.	Allow the sample to penetrate the upper frit.
6.	Pulling vacuum, elute the analytes of interest with 15 mL of acetonitrile (rate of approximately 1 drop/sec.).
7.	Concentrate the samples to a final volume of 1 mL under nitrogen (5 psi) at 40 °C.

Table 2. Cleanup Procedure for Olive Oil using Silica Gel SPE Cartridges (Competitor A and B)[1*]

1.	Condition the SPE cartridges with 20 mL of hexane.
2.	Dilute 5 g of oil with hexane to a final volume of 10 mL. Load 1 mL onto the cartridge. Add the internal standard directly onto the cartridge.
3.	Wash the cartridge with 8 mL hexane:methylene chloride (70:30).
4.	Elute the analytes of interest with 8 mL hexane:methylene chloride (70:30).
5.	Concentrate the samples to a final volume of 1 mL under nitrogen (5 psi) at 40 °C.

* Gravity elution was used for the above steps.

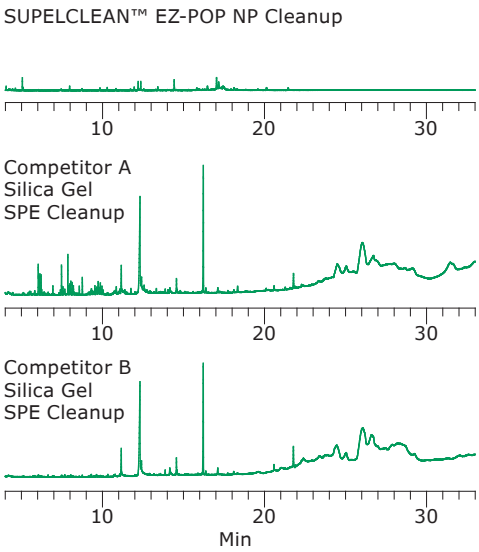
Extracts were then analyzed by GC-MS (SIM mode). Quantitation was performed against a 5-point calibration curve (1–20 ng/mL) prepared in unspiked olive oil extract with naphthalene-d8, fluoranthene-d_{10}, perylene-d_{12} internal standards, spiked at 10 ng/mL.

Results and Discussion

Matrix Removal

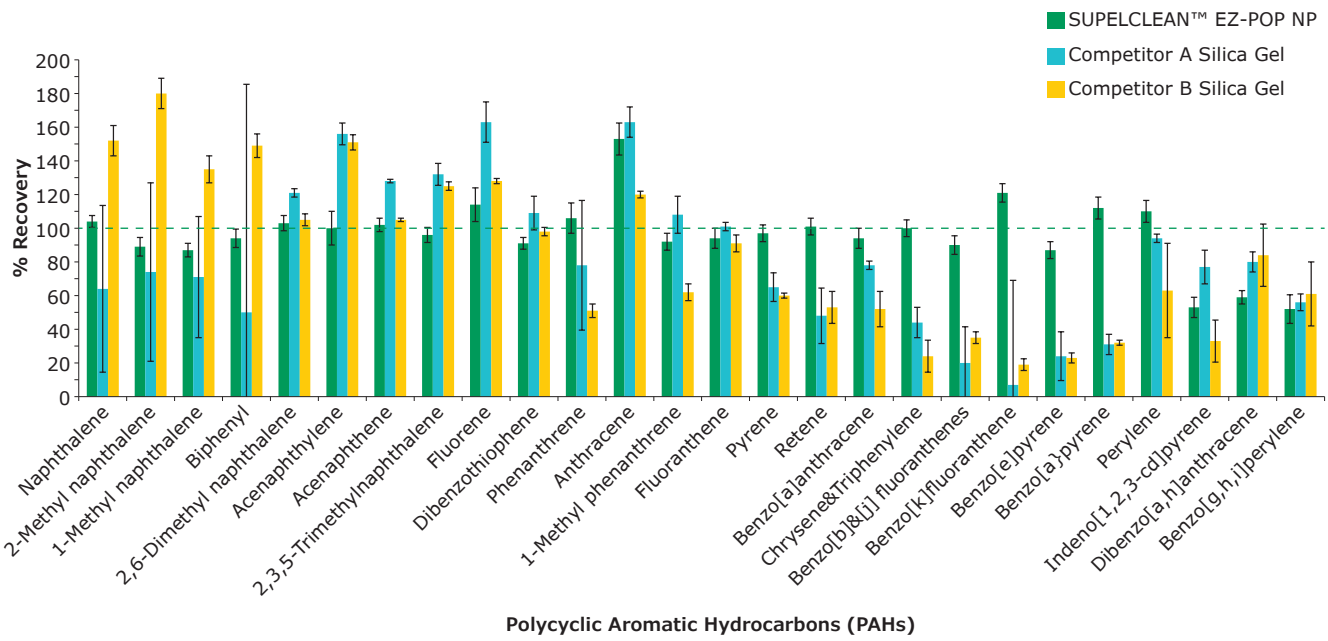
Figure 1 shows the GC-MS (full scan) chromatograms of the extracts after cleanup with SUPELCLEAN™ EZ-POP NP, Competitor A silica gel SPE, and Competitor B silica gel SPE, respectively. The chromatograms illustrate that the SUPELCLEAN™ EZ-POP NP cleanup produces much lower background than the silica gel SPE cleanup.

Figure 1. GC-MS Full Scan Chromatograms of Olive Oil Extract (same y axis)



column:	SLB®-5ms, 20 m x 0.18 mm I.D., 0.18 µm (28564-U)
oven:	60 °C (1 min.), 15 °C/min. to 250 °C, 8 °C/min. to 330 °C (7 min.)
inj. temp.:	300 °C
carrier gas:	helium, 1 mL/min constant flow
detector:	MS
MSD interface:	330 °C
injection:	1 µL, pulsed splitless (50 psi until 0.75 min, splitter open at 0.75 min.)
liner:	4 mm ID FocusLiner™ with taper and quartz wool

Figure 2. Analyte Recovery of PAHs from Olive Oil Extract (n=3)



Analyte Detection and Recovery

As illustrated in **Figure 2**, the SUPELCLEAN™ EZ-POP NP samples produced better overall analyte recoveries than those cleaned with the silica SPE cartridges. The reproducibility for all PAHs tested, exhibited by percent relative standard deviation (%RSD), fell within an acceptable range of less than 20% for the SUPELCLEAN™ EZ-POP NP cleaned samples. The SUPELCLEAN™ EZ-POP NP cleanup demonstrated improvements over silica gel cleanup in the following areas:

- Recoveries for the lighter PAHs (2–3 rings) in the SUPELCLEAN™ EZ-POP NP cleanup were not affected by matrix as they were for the silica gel samples.
- The heavier PAHs of 4–6 rings showed improved recoveries using SUPELCLEAN™ EZ-POP NP cartridge over the silica gel cartridges.
- The heavy background from the silica cleanup caused retention time shifts for the last six PAH peaks, making identification difficult. Also, the high level of late eluting background in the Competitor B silica sample prevented accurate quantitation of the last internal standard, perylene-d_{12}. On the contrary, there were no background complications associated with the SUPELCLEAN™ EZ-POP NP cleanup.

Hence, the SUPELCLEAN™ EZ-POP NP produced better overall recoveries and removed more problematic matrix interferences than the silica gel SPE cartridges, while maintaining good reproducibility.

Conclusion

This study compared the SUPELCLEAN™ EZ-POP NP to silica gel SPE for the extraction of PAHs from olive oil, a problematic fatty matrix. In terms of lipid removal, the SUPELCLEAN™ EZ-POP NP removed more unwanted background than silica gel SPE, greatly decreasing the matrix effects. The SUPELCLEAN™ EZ-POP NP produced better overall analyte recoveries than the silica gel SPE, with adequate reproducibility. Thus, the SUPELCLEAN™ EZ-POP NP provides suitable matrix removal for rugged GC-MS analysis of PAHs in olive oil.

Reference

1. Moret, S., Conte, L., *J. Sep. Sci.* 25, 96-100, (2002).

Featured and Related Products

Description	Qty.	Cat. No.
SUPELCLEAN™ EZ-POP NP SPE Cartridge	20	**54341-U**
Column		
SLB®-5ms Capillary GC Column 20 m x 0.18 mm I.D., 0.18 µm	1	**28564-U**
Reference Materials		
Polynuclear Aromatic Hydrocarbons Mix, *Trace*CERT® CRM 2000 µg/mL each component in methylene chloride: benzene (1:1)	ampule of 1 mL	**CRM48905**
Naphthalene-d_8 solution 2000 µg/mL in methylene chloride	ampule of 1 mL	**48715-U**
Fluoranthene-d_{10}	50 mg, 100 mg	**456292**
Perylene-d_{12} solution 2000 µg/mL in methylene chloride	ampule of 1 mL	**48081**
Solvent		
Acetonitrile, hypergrade for LC-MS LiChrosolv®	1 L, 2.5 L, 4 L	**1.00029**

Visit our gas chromatography page at
SigmaAldrich.com/GC

MilliporeSigma
400 Summit Drive
Burlington, MA 01803

To place an order or receive technical assistance

Order/Customer Service: **SigmaAldrich.com/order**
Technical Service: **SigmaAldrich.com/techservice**
Safety-related Information: **SigmaAldrich.com/safetycenter**

SigmaAldrich.com

MS_AN7746EN Ver. 1.0
35595
03/2021