

Separation of Trichlorobenzene from Sardines using Agilent EcoSpheres

Application Note

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Introduction

The analysis of pesticides and other contaminants in environmental samples is an important part of monitoring the condition of the environment. For example, determining trace levels of pesticides in livestock and crops is a crucial part of controlling the quality and safety of foodstuffs.

Most samples are produced for these testing procedures by macerating plant, soil and animal matter and then extracting the species of interest into an organic solvent ready for concentration and analysis by a suitable technique such as GC/MS. Due to the low concentrations of the contaminants and the presence of complex organic matrices, further separation steps are often required to purify the sample by the removal of extraneous material. Failure to remove these interfering compounds can affect the quality of the final analysis and increase analytical downtime.

There are many methods employed to clean up samples from environmental sources but gel permeation chromatography (GPC), also known as size exclusion chromatography (SEC), has proved one of the most popular. In this technique a size-based separation is employed to separate the species of interest, typically small molecules such as pesticides, from higher molecular weight matrices such as fats and lipids. GPC is a common and convenient technique for post extraction sample clean up as sample components are separated based on their molecular size in solution and not on any adsorption, hydrogen bonding or other physico-chemical interactions. Once isolated from the larger molecular weight interferences, the pesticides and priority pollutants can be collected for further analysis.

Sardine is a generic term for the young of several species of fish that are usually less than four inches long. A staple foodstuff in some parts of the world, sardines remain popular in part due to their healthy image as a brain food. As a wild-fished product, the danger of contamination of sardines from pesticides or other contaminants can be a concern.



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This note describes the separation of trichlorobenzene from a sample of macerated sardines as part of a quality control procedure.

Materials and Methods

A sample of macerated sardines was spiked with trichlorobenzene. To perform the analysis, a 300 x 7.5 mm glass column was packed with EcoSpheres, a soft gel material specifically designed for the clean up of samples from environmental sources. The EcoSpheres were swollen and packed in a 50:50 mixture of cyclohexane and ethyl acetate, solvents selected for their low toxicity.

Conditions

Column:	EcoSpheres Glass Column 300 x 25 mm (p/n PL1310-0054) packed with swollen EcoSpheres (p/n PL1460-4M03)
Eluent:	50:50 mixture of cyclohexane and ethyl acetate
Flow Rate:	5 mL/min
Inj Vol:	5 mL
System:	Modular system (dual piston pump with injection valve)
Data collection/analysis:	Agilent Cirrus GPC software and Agilent PL DataStream
Detector:	UV, 254nm

Results and Discussion

The chromatogram in Figure 1 shows that it is possible to separate the low molecular weight contaminant of interest from the high molecular weight lipid fraction of the macerated sardine. Collecting the region containing the trichlorobenzene from real samples allows the contaminant level to be assessed.

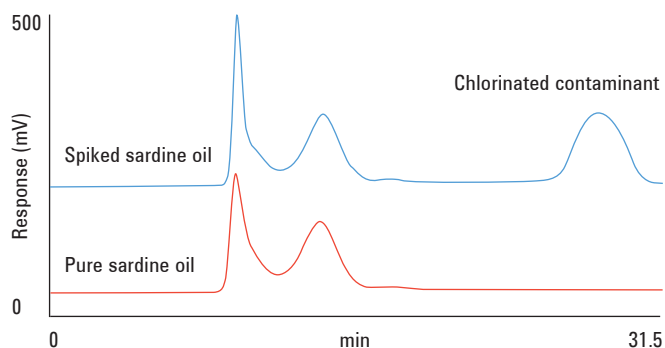


Figure 1. The presence of a trichlorobenzene contaminant in sardines revealed by EcoSpheres

Conclusion

EcoSpheres successfully revealed the existence of a chlorinated compound in a sample of fish oil, showing how the technology can be used to assess contaminant levels in environmental samples.

EcoSpheres is available as loose media for packing into any suitable glass column hardware. The media operate up to 300 psi (21 bar), and a packed 25 mm ID glass column can handle eluent flows up to 10 mL/min. As can be seen here, the microporous packing material gives good resolution with high loadings. EcoSpheres can be used with any isocratic LC system at the required flow rates. Its narrow particle size distribution delivers excellent peak shape and well-packed columns, and the low pore size excludes high molecular weight material, maximizing separation from low molecular weight additives.

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Published in UK, April 30, 2015

5991-5824EN



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