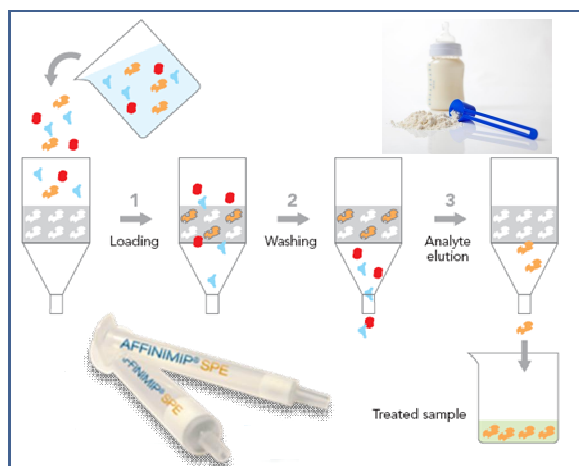


Selective Solid Phase Extraction of Bisphenol A from Powdered Infant Formula at Low Concentrations Using Molecularly Imprinted Polymers



Background

Bisphenol A (or BPA) is a molecule widely used in the food packaging industry. The migration of this endocrine disruptor compound from the packaging to food is the main source of consumers' exposure to BPA. Its consumption is critical for babies. So, a European directive prohibits the use of BPA to manufacture infant feeding bottles (Directive 2011/8/EU of 28 January 2011).

So, BPA is a topical issue with a worldwide regulation going to still lower concentrations of BPA allowed in food. So, highly sensitive and reliable detection methods are required for routine analysis of BPA in food samples, particularly for baby food.

In a previous application note, we described a protocol enabling the determination of very low concentration of BPA in liquid infant formula. This application note describes the analysis of very low concentration of BPA in powdered infant formula using AFFINIMIP® SPE Bisphenols cartridge.

We demonstrate in this application note that a reliable quantification of Bisphenol A at low concentrations (2 and 4 µg/L) using fluorescence detector is possible. Therefore, the use of AFFINIMIP® SPE Bisphenols enables to eliminate the tedious derivatization step required by gas chromatography.

This method is also perfectly suitable for clean-up before GC-MS/MS or LC-MS/MS.

Results

High analyte recovery and good reproducibility

Concentration of BPA in reconstituted milk (µg/L)	Mean concentration (µg/L)	Recoveries %	RSD _r %
2.1	2.3 (n=5)	108	8.7
4.3	4.0 (n=4)	95	3.7

Table 1. Recovery of Bisphenol A spiked at different concentrations after AFFINIMIP® SPE Bisphenols clean-up of 40mL of loading solution (equivalent to 10mL of reconstituted infant milk) and relative standard deviation calculated from results generated under **repeatability conditions**.

Perfect cleanup at very low concentrations

The preparation of a fluid loading solution enables the analysis of a large amount of reconstituted infant milk (equivalent to 10mL). It produces a good clean-up and a high enrichment level, thus enabling the determination of very low amount of Bisphenol A with a simple protocol.

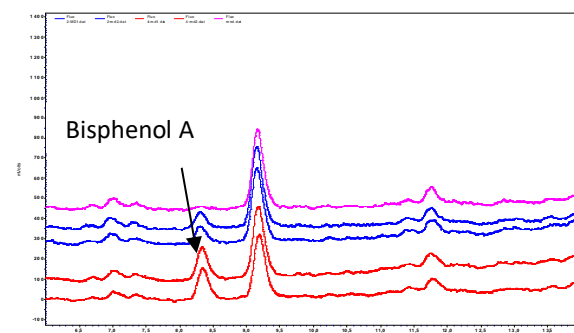


Figure 1. Chromatograms obtained after clean-up with AFFINIMIP® SPE Bisphenols of equivalent at 10mL of Infant Formula spiked with Bisphenol A at 4.3µg/L (tested twice, red) or at 2.1µg/L (tested twice, blue) or not spiked (pink).

Experimental conditions

Materials

All reagents and chemicals were ACS grade quality or better. Bisphenol A was obtained from Alfa Aesar. Powdered Infant milk was purchased at a supermarket.

Preparation of the loading solution

Infant milk was reconstituted according to the manufacturer's instruction (4.4 g in 30 mL of water). The reconstituted solutions were then spiked with Bisphenol A at 2.1 and 4.3 µg/L. It is equivalent to respectively 14.4 and 29.4 ng of BPA / g of powdered milk.

The reconstituted milk was warmed up at about 50°C during 20 seconds using microwaves. Then 20 mL of acetonitrile were added to the 20 mL of warm milk.

The solution was centrifuged at 4000 rpm during 10 minutes and the supernatant was collected and filtered on filter paper (4-7µm). This extract was diluted 1:1 with water.

This fluid solution was used as the loading solution.

Solid phase extraction (SPE) protocol

The SPE procedure used a 3mL **AFFINIMIP® SPE Bisphenols** cartridge. The details of each step are as follow:

- Condition the SPE cartridge with 5mL of Methanol-2% acetic acid, 5mL Acetonitrile (ACN), then with 5mL of deionized Water
- Load up to 40mL of the loading solution
- Wash the cartridge with 10mL of deionized Water
- Wash the cartridge with 6mL of deionized Water /Acetonitrile (60/40, v/v)
- Dry 30 seconds
- Elute Bisphenol A with 3mL of Methanol

The elution fraction was then evaporated and dissolved in the mobile phase.

Analysis

HPLC was performed on a ThermoFinnigan Spectra System with a Thermo Hypersil Gold C18 column (150mm x 4.6mm). Separation was carried out using a gradient at a flow rate of 1mL/min. The detection system was a Jasco FP-2020 with Fluorescence detector set to excitation/emission wavelengths of 230 and 315nm, respectively. The injection volume was 50µL.

	Time (min)	% Water	% ACN
Mobile Phase	0	65	35
	2	65	35
	12	50	50
	20	50	50
	20.5	65	35
	40	65	35

Product references

• **AFFINIMIP® SPE Bisphenols**

- FS106-02 for 25 polypropylene cartridges 3mL
- FS106-02G for 25 glass cartridges 6mL
- FS106-03 for 50 polypropylene cartridges 3mL
- FS106-03G for 50 glass cartridges 6mL

Related matrices: canned foods, infant milk, beer, water...