Affinisep

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

Introduction

Bisphenol A (or BPA) (see Figure 1) is a molecule widely used in industry for the synthesis of polycarbonate plastics and epoxy resins. Polycarbonate plastics are used to make a variety of common products including baby and water bottles. Epoxy resins are used as coatings on the inside of almost all food and beverage cans.

Figure 1. Chemical structure of Bisphenol A

Bisphenol A is also an endocrine disruptor, which can mimic the body’s own hormones and may lead to negative health effects. The migration of BPA from the packaging to food is the main source of consumers’ exposure to BPA. So, the European commission has defined a specific migration limit at a maximum level of 0.6 mg of BPA/kg of food (Directive 2011/8/EU of 28 January 2011). In addition, the directive prohibits the use of BPA to manufacture infant feeding bottles.

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.

For such concentrations, a clean-up step is crucial in order to improve the sensitivity, the reliability and the specificity before analysis.

Affinisep has developed a new class of intelligent polymers based on molecularly imprinted polymers (AFFINIMIP®) specific to Bisphenol A used as a powerful technique for clean-up and pre-concentration.

AFFINIMIP® SPE Bisphenols cartridge is a simple, fast, sensitive and selective tool for the extraction of Bisphenol A from milk samples with a high recovery yield (> 80%), a good repeatability and reproducibility.

We demonstrate in this application note that a reliable quantification of Bisphenol A at low concentrations (1 and 2µg/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.

Experimental conditions

Materials

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

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 15mL of Infant Formula
- 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 SPE procedure lasted approximately 50 minutes.

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

Results

Table 1. Recovery of Bisphenol A spiked at different concentrations after AFFINIMIP® SPE Bisphenols clean-up of 15mL of Infant Formula and relative standard deviation calculated from results generated under repeatability conditions (n=3).

<table>
<thead>
<tr>
<th>C° (µg/L)</th>
<th>Mean (µg/L)</th>
<th>Recoveries %</th>
<th>% RSD</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.0</td>
<td>0.9</td>
<td>88.4</td>
<td>1.5</td>
</tr>
<tr>
<td>2.0</td>
<td>1.7</td>
<td>85.7</td>
<td>2.7</td>
</tr>
</tbody>
</table>

Table 2. Recovery of Bisphenol A spiked at different concentrations after AFFINIMIP® SPE Bisphenols clean-up of 15mL of Infant Formula and relative standard deviation calculated from results generated under reproducibility conditions.

<table>
<thead>
<tr>
<th>C° (µg/L)</th>
<th>Mean (µg/L)</th>
<th>Recoveries %</th>
<th>% RSD</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.0</td>
<td>0.8</td>
<td>84.4</td>
<td>7.4</td>
</tr>
<tr>
<td>2.0</td>
<td>1.7</td>
<td>85.8</td>
<td>5.3</td>
</tr>
</tbody>
</table>

Figure 2. Chromatograms obtained after clean-up with AFFINIMIP® SPE Bisphenols of 15mL of Infant Formula spiked with Bisphenol A at 2µg/L (tested twice, blue) or at 1µg/L (tested twice, red) or not spiked (pink).

Figure 3. Chromatograms of Infant Formula containing 1µg/L of Bisphenol A before clean-up (Red) and after clean-up (Blue) with AFFINIMIP® SPE Bisphenols.

Product reference

- **AFFINIMIP® SPE Bisphenols**
  - Catalog number: FS106-02 for 25 cartridges
  - FS106-02G for 25 Glass cartridges
  - FS106-03 for 50 cartridges
  - FS106-03G for 50 Glass cartridges

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