

## **Melamine in Cookies Sample Preparation**

### **1. Intended Use**

For the detection of Melamine in cookies. For other matrices, please see the appropriate application bulletin.

### **2. Range of Detection**

2,000-50,000 ng/mL (ppb). Samples with higher concentrations must be diluted further and re-analyzed.

### **3. Materials Required (Not Provided)**

Blender

Overhead tube rotator

Vortex mixer

Pipettes capable of delivering 100 and 900 $\mu$ L

Glass vials with Teflon lined caps

Methanol

ABRAXIS<sup>®</sup> Melamine ELISA Kit (PN 50005B)

### **4. Preparation of Solutions**

10% MeOH/20 mM Phosphate Buffered Saline (PBS), pH 7.4

To 800 mL of distilled or deionized water, add: Sodium phosphate dibasic anhydrous 2.277 g; Sodium phosphate monobasic monohydrate 0.548 g; Sodium chloride 18.0 g, add 100 mL of methanol and then bring to 1 L with distilled or deionized water, pH 7.2-7.4.

### **5. Notes and Precautions**

To eliminate matrix interference from cookies (fat, etc.) for the presence of Melamine, samples must be diluted in 10% MeOH/20 mM PBS.

### **6. Procedure**

- 6.1. 20 mL of distilled or deionized water is added to 10 g of cookie sample and homogenized for 10 minutes using a blender.
- 6.2. Pipette 1 mL of the homogenized sample into a plastic tube.
- 6.3. Centrifuge for 10 minutes at 2500 +/- 200 g.
- 6.4. Remove and discard the top (fat) layer.
- 6.5. Carefully remove a portion of the middle layer
- 6.6. Dilute and aliquot of the middle layer 1:50 in 10% MeOH/20 mM PBS. For example, adding 40  $\mu$ L of the cookie extract (step 4.5) with 1.96 mL of 10% MeOH/20 mM PBS.
- 6.7. The sample is now ready to analyze according to the procedure described in the ABRAXIS<sup>®</sup> Melamine Kit package insert.

### **7. Evaluation of Results**

Results obtained for cookie samples prepared as described above must be multiplied by a factor of 100 to account for the sample dilution (this is the dilution factor introduced by the extraction/dilution

procedure). Only use results within the analytical range of the assay (20-500 ppb). Results lower than the lowest standard (20 ppb) should not be multiplied by a dilution factor and should not be reported as negative, but should be reported as < 2,000 ppb Melamine detected (using the dilution factor). Results above the highest standard must be diluted and re-analyzed. If further dilutions are performed on the samples, this dilution factor needs to be multiplied by the sample value to obtain the final Melamine concentration on the sample.

## **8. Performance Data**

The sample preparation procedure detailed above was used with cookies spiked with various amounts of Melamine. Average recoveries were 110 %.

## **9. For ordering or technical assistance contact**

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