

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 900µL

Glass vials with Teflon lined caps

Methanol

ABRAXIS® 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 μL 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® 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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Date this Technical Bulletin is effective: 05/16/2024 Version: 01