

Melamine in Ice Cream Sample Preparation

1. Range of Detection

2,400-60,000 ng/mL (ppb). Samples with higher concentrations must be diluted further and re-analyzed.

2. Materials Required (Not Provided)

Microcentrifuge capable of spinning at 10,000-13,000 x g

Vortex mixer

Microcentrifuge tubes

Pipettes capable of delivering 100 and 900 μ L

Glass vials with Teflon lined caps

Methanol

ABRAXIS[®] Melamine ELISA Kit (PN 50005B)

3. 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.

4. Notes and Precautions

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

5. Procedure

- 5.1. Weigh 5 gm of ice cream samples to a glass vial and allow to thaw
- 5.2. Add 10 mL of distilled or deionized water and mix well by vortexing for 30 seconds
- 5.3. Add approximately 1 mL of sample (step 5.2) into a microcentrifuge tube
- 5.4. Centrifuge at 10,000-13,000 x g for 5 minutes. Sample should separate into 3 layers
- 5.5. Remove and discard the top (fat) layer
- 5.6. Carefully remove a portion of the middle layer
- 5.7. Dilute and aliquot of the middle layer 1:40 in 10% MeOH/20 mM PBS. For example, adding 25 μ L of the extracted milk with 975 μ L of 10% MeOH/20 mM PBS.
- 5.8. The sample is now ready to analyze according to the procedure described in the ABRAXIS[®] Melamine Kit package insert.

6. Evaluation of Results

Results obtained for ice cream samples prepared as described above must be multiplied by a factor of 120 to account for the sample dilution. 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 < 2,400 ppb Melamine detected. Results above the highest standard must be diluted and re-analyzed.

7. Performance Data

The sample preparation procedure detailed above was used with whole milk spiked with various amounts of Melamine. Average recoveries obtained with chocolate and vanilla ice cream samples were 102%.

8. For ordering or technical assistance contact

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