



FERN

Uniting Federal, State and Local Laboratories for Food Emergency Response

SOP No: FERN-CHE.0003.00		Page 1 of 15
Title: LC/MS/MS Screen for the Presence of Melamine in swine and poultry tissues		
Revision: Original	Replaces: N/A	Effective: (5-07)
Authors/Point(s) of Contact: USDA, FSIS Western Laboratory: Emilio Esteban FERN Method Coordination Committee		

Note: This method has been approved for Emergency Response use and is posted following the FERN guidelines for urgent usage /level 1 validation. This validation level indicates that it meets minimal validation criteria. Emergency Response/Urgent Use methods will be removed from eLEXNET six months after the emergency has ended if there has been no validation data submitted to the Method Coordination Committee for consideration by a Technical Review Committee.

Contents

A.	INTRODUCTION	2
B.	EQUIPMENT	2
C.	REAGENTS AND SOLUTIONS	3
D.	STANDARDS	4
E.	SAMPLE PREPARATION	5
F.	ANALYTICAL PROCEDURE.....	5
G.	SCREENING CRITERIA	9
H.	SAFETY INFORMATION AND PRECAUTIONS.....	10
I.	QUALITY ASSURANCE PLAN	11
J.	WORKSHEET	13
K.	APPENDIX	15
L.	APPROVALS AND AUTHORITIES	15

Issuing Authority: Food Emergency Response Network (FERN)
Uncontrolled when printed



FERN

Uniting Federal, State and Local Laboratories for Food Emergency Response

SOP No: FERN-CHE.0003.00	Page 2 of 15
Title: LC/MS/MS Screen for the Presence of Melamine in swine and poultry tissues	
Revision: Original	Replaces: N/A
Effective: (5-07)	

A. INTRODUCTION

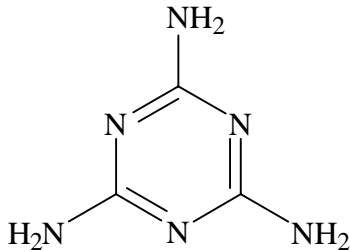
1. Theory

Melamine is extracted from muscle tissue with a mixture of acetonitrile and water. The extract is cleaned up by liquid/liquid extraction with methylene chloride followed by solid phase extraction (SPE). Melamine is eluted from the SPE cartridge with 5% ammonium hydroxide / methanol, evaporated to dryness, reconstituted with internal standard and 50% acetonitrile / water, and analyzed by LC/MS/MS.

2. Applicability

This method is applicable for screening of melamine in swine and poultry muscle at ≥ 50 ppb.

3. Structure



B. EQUIPMENT

Note: Equivalent equipment may be substituted for the following.

1. Apparatus

- Balance - analytical, 0.1 mg sensitivity, Model No. A120S, Sartorius.
- Balance - top loading, 0.01 g sensitivity, PJ3600 Delta Range, Mettler.
- Centrifuge - with 15 mL tube carriers, centrifuge: Model HN-S, rotor: Cat. No. 809, International Equipment Company.
- Centrifuge - with 50 mL tube carriers, centrifuge: Model PR-7000, rotor, Cat. No. 276, International Equipment Company.
- Centrifuge tubes - 50 mL, Pyrex® round-bottom with Teflon lined screw-cap, 29 mm, Cat. No. 21023-401, VWR.
- Centrifuge tubes - 15 mL, glass, disposable, screw top, Cat. No. 73785, Kimble.
- Culture tubes - glass, disposable, 15 mL, 16 x 100 mm, Cat. No. 73500 16100,

Issuing Authority: Food Emergency Response Network (FERN)
Uncontrolled when printed



FERN

Uniting Federal, State and Local Laboratories for Food Emergency Response

SOP No: FERN-CHE.0003.00	Page 3 of 15
Title: LC/MS/MS Screen for the Presence of Melamine in swine and poultry tissues	
Revision: Original	Replaces: N/A
Effective: (5-07)	

Kimble.

- h. Graduated cylinders - class A, 25 mL, 100, 500 mL.
- i. Homogenizer - Omni 7H, Omni International.
- j. Micropipettors - Adjustable, 100 µL to 10 mL, Eppendorf.
- k. Monoject tuberculin syringe, sterile - 1 mL (VWR Part No.: MJ8881-501178)
- l. Nitrogen evaporator - N-Evap, Model No. 111, Organomation Associates Inc.
- m. Pasteur pipettes - glass, disposable, 9 inch, Cat. No. 14672-380, VWR.
- n. Repipettors - bottle top dispensers, 0 – 25 mL and 1 – 10 mL Dispensettes, Brinkmann.
- o. Shaker - horizontal flatbed, two speed, Cat. No. 511105, Eberbach.
- p. Solid Phase Extraction columns - Strata X-C, 60 mg, 3mL, Phenomenex.
- q. Spatula - stainless steel.
- r. Squirt Bottle – Teflon, Cat. No. 8-250, Nalgene.
- s. Syringe - 3 mL, disposable, Cat. No. BD301077, Becton Dickinson.
- t. Syringe Filters - Millex, HV, 0.45 µm, Cat. No. SLHVR04NL, Millipore.
- u. Test tube racks - for 15 and 50 mL tubes.
- v. Vacuum manifold - for solid phase extraction, Cat. No. 5-7030, Supelco.
- w. Volumetric flask - class A, 50 mL, 1L.
- x. Vortex mixer - variable speed, Cat. No. S8223-1, American Scientific Products.

2. Instrumentation

- a. HPLC - Waters Alliance 2695.
- b. Mass Spectrometer - Waters Micromass Quattro micro API.
- c. Column - Phenomenex Synergi™ Polar RP (150 x 4.6mm; 4 µm particle size).

C. REAGENTS AND SOLUTIONS

Note: Equivalent reagents and solutions may be substituted for the following.

1. Reagents

- a. Acetic Acid - MS grade, Cat. No. 49199 Fluka / Sigma Aldrich.
- b. Acetonitrile (ACN) - HPLC grade, Cat. No. 015-4, Burdick & Jackson.
- c. Ammonium Acetate - MS grade, Cat. No. 73594 Fluka / Sigma Aldrich.

Issuing Authority: Food Emergency Response Network (FERN)
Uncontrolled when printed



FERN

Uniting Federal, State and Local Laboratories for Food Emergency Response

SOP No: FERN-CHE.0003.00	Page 4 of 15
Title: LC/MS/MS Screen for the Presence of Melamine in swine and poultry tissues	
Revision: Original	Replaces: N/A
Effective: (5-07)	

- d. Ammonium Hydroxide (NH₄OH) - 30%, Cat. No. 9721-33, J.T. Baker.
- e. Hydrochloric acid (HCl) - Concentrated, Cat. No. 9535-05, J.T. Baker.
- f. Methanol - HPLC grade, Cat. No. 230-4, Burdick & Jackson.
- g. Methylene Chloride - HPLC grade, Cat. No. D151-4, Fisher.
- h. 18 MΩ deionized (DI) H₂O - from Millipore Rx system.

2. Solutions

- a. ACN:H₂O (1:1, v/v):
Mix 500 mL of ACN with 500 mL of DI water.
- b. Rinse Solution:
Fill a jug with about half ACN and half water. No measurement required.
- c. 1N HCl:
Measure 83 mL of concentrated HCl using a 100 mL graduated cylinder. Add to a 1L volumetric flask. Bring to volume with DI water.
- d. 0.1N HCl:
Pipette 8.33 mL of concentrated HCl into a 1L volumetric flask. Bring to volume with DI water.
- e. 5% Ammonium hydroxide in methanol:
Using class A graduated cylinders, measure 25 mL ammonium hydroxide and 475 mL methanol. Combine in a glass bottle.
- f. Mobile Phase A (25 mM ammonium acetate / 25 mM acetic acid in water):
Weigh 1.93 g of ammonium acetate and add to a 1L volumetric flask. Add 1430 μL of acetic acid. Bring to 1L with DI water.

D. STANDARDS

Note: Equivalent standards and solutions may be substituted for the following.

1. Source

- a. Melamine - Cat. No. M2659, Aldrich.
- b. ¹⁵N₃ Melamine (for internal standard) - Cat. No. IN 5451, ICON Services.
Cat. No. 592889, Aldrich. (no longer available from either source 5/17/07)

2. Preparation

- a. Melamine stock standard (100 μg/mL):

Issuing Authority: Food Emergency Response Network (FERN)
Uncontrolled when printed



FERN

Uniting Federal, State and Local Laboratories for Food Emergency Response

SOP No: FERN-CHE.0003.00	Page 5 of 15
Title: LC/MS/MS Screen for the Presence of Melamine in swine and poultry tissues	
Revision: Original	Replaces: N/A
Effective: (5-07)	

Weigh 5 mg of melamine into a 50 mL volumetric flask. Fill flask about half way with 50% ACN:H₂O. Sonicate to dissolve. Bring to volume with ACN:H₂O (1:1, v/v).

- b. Melamine working standard (1 µg/mL):

Pipette 500 µL of the melamine stock solution into a 50 mL volumetric flask. Bring to volume with ACN:H₂O (1:1, v/v).

- c. ¹⁵N₃ Melamine internal standard (IS) (1 µg/mL):

Prepare 1 µg/mL solution of ¹⁵N₃ Melamine in ACN:H₂O (1:1, v/v).

- d. Calibration curve standards:

Concentration	Tissue equivalent	Volume melamine working std. (1 µg/mL)	Volume IS (1 µg/mL)	Volume ACN:H ₂ O (1:1, v/v)
0 ng/mL	0 ppb	0 µL	500 µL	500 µL
125 ng/mL	25 ppb	125 µL	500 µL	375 µL
250 ng/mL	50 ppb	250 µL	500 µL	250 µL
500 ng/mL	100 ppb	500 µL	500 µL	0 µL

3. Storage and Stability

Store refrigerated at 2 - 8 °C. Stability has not been tested.

E. SAMPLE PREPARATION

After removing excessive fat from muscle sample, cut it into smaller pieces and homogenize with a mechanical food processor. Transfer homogenized sample into plastic bags and store in a freezer at ≤ -20°C. Let the sample partially thaw prior to analysis.

F. ANALYTICAL PROCEDURE

Note: avoid plastic - it may leach melamine and contaminate samples.

Melamine is very sticky. Rinse all re-usable glassware with acetonitrile/water rinse solution before use.

1. Meat Sample Preparation

- a. Weigh 5.00 ± 0.05 g of sample into a 50 mL glass centrifuge tube. Also weigh two known blank tissues for use as positive and negative controls.

Issuing Authority: Food Emergency Response Network (FERN)
Uncontrolled when printed



FERN

Uniting Federal, State and Local Laboratories for Food Emergency Response

SOP No: FERN-CHE.0003.00	Page 6 of 15
Title: LC/MS/MS Screen for the Presence of Melamine in swine and poultry tissues	
Revision: Original	Replaces: N/A
Effective: (5-07)	

- b. Fortify a blank tissue with 250 μ L of 1 μ g/mL melamine working standard for use as a 50 ppb positive control.
- c. Add 25 mL of ACN:H₂O (1:1, v/v).
- d. Tissuemize to homogenize at medium to high speed for 30 seconds to 1 minute. Between each sample, clean the homogenizer probe 5 times with rinse solution. Fill a 50 mL glass tube with sufficient volume to clean a probe (~ 40 - 45 mL). Put the probe in the tube and tissuemize on high speed for about 5 seconds. Repeat 4 more times, each time with a fresh volume of rinse solution.
- e. Centrifuge at 3500 g for 10 minutes, or sufficient to pack tissue.
- f. Remove 5 mL aliquot of the extract into a glass 50 mL glass centrifuge tube.
- g. Add 110 μ L of 1 N HCl and vortex to mix.
- h. Add 10 mL of methylene chloride.
- i. Shake for 2 minutes.
- j. Centrifuge at 3500 g for 10 minutes, or sufficient to separate layers.
- k. Remove the aqueous layer (top) into a disposable, screw cap, glass centrifuge tube.
- l. Add 2.5 mL of water to the original 50 mL tube.
- m. Shake for 2 minutes.
- n. Centrifuge at 3500 g for 10 minutes, or sufficient to separate layers.
- o. Carefully remove the top layer; combine aqueous layers.
- p. Cap and vortex for 10 seconds.
- q. Centrifuge at 3500 g for 10 minutes if necessary.
- r. Thoroughly clean SPE manifold and fittings with rinse solution.
- s. Pre-wash the SPE column with 5 mL of methanol followed by 5 mL of water (vacuum optional).
- t. Load the sample onto the column (gravity)
- u. Wash the column with 2 mL of 0.1N HCl, followed by 1 mL of methanol, discard washes. (gravity)
- v. Aspirate the column for 1 minute. (vacuum)
- w. Elute melamine with 5 mL of 5% ammonium hydroxide/methanol into a clean, glass, disposable tube. (gravity)
- x. Thoroughly clean the N-evap tips by spraying with rinse solution from a Teflon squirt bottle and collecting the rinse in a beaker for proper disposal.

Issuing Authority: Food Emergency Response Network (FERN)
Uncontrolled when printed



FERN

Uniting Federal, State and Local Laboratories for Food Emergency Response

SOP No: FERN-CHE.0003.00	Page 7 of 15
Title: LC/MS/MS Screen for the Presence of Melamine in swine and poultry tissues	
Revision: Original	Replaces: N/A
Effective: (5-07)	

- y. Evaporate the eluant to dryness using an N-evap set to 50 - 60 °C.
- z. Add 100 µL of ACN:H₂O (1:1, v/v), and 100 µL of IS (¹⁵N₃ Melamine, 1 µg/mL).
- aa. Vortex the sample and sonicate for at least one minute to re-dissolve the residue.
- bb. Filter through a 0.45 µm HV filter into an autosampler vial. Transfer the filtered extract into a vial insert using a glass Pasteur pipette. Put the insert into the original vial, cap, and submit for LC-MS/MS analysis.

2. Instrumental Settings

Note: The following instrument parameters may be optimized.

a. HPLC Conditions

- i. Mobile Phase A: 25 mM Ammonium Acetate / 25 mM Acetic Acid in H₂O
- ii. Mobile Phase B: Acetonitrile
- iii. Mobile Phase C: Methanol
- iv. Flow Rate: 0.5 mL/min
- v. Gradient:

Time (min)	%A	%B	%C
0.0	80	15	5
5.0	30	65	5
5.1	5	90	5
7.0	5	90	5
7.1	80	15	5
12.0	80	15	5

- vi. Injection Volume: 50 µL
- vii. Column Temperature: 25°C

b. Interface Conditions:

- i. Polarity: Positive
- ii. Ion Source: Electro Spray
- iii. Source Temperature : 125
- iv. Desolatiion Temperature : 450
- v. Cone Gas Flow : 20 L/hour

Issuing Authority: Food Emergency Response Network (FERN)
Uncontrolled when printed



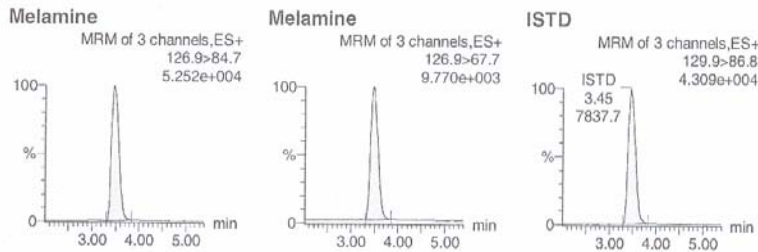
FERN

Uniting Federal, State and Local Laboratories for Food Emergency Response

SOP No: FERN-CHE.0003.00		Page 8 of 15
Title: LC/MS/MS Screen for the Presence of Melamine in swine and poultry tissues		
Revision: Original	Replaces: N/A	Effective: (5-07)

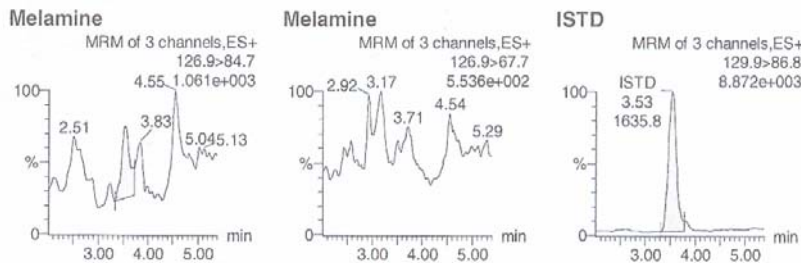
- vi. Desolvation Gas Flow : 900 L/hour
- c. Multiple Reaction Monitoring (MRM) Parameters:
 - i. Resolution Q1 & Q3 Unit
 - ii. Cone Voltage : 35V
 - iii. Dwell Time : 0.2 sec
 - iv. Melamine Transitions:
 - (a) m/z 127→ 85 27eV collision voltage (quant. Ion)
 - (b) m/z 127→ 68 17eV collision voltage
 - v. ¹⁵N₃ Melamine Transition:
 - (a) m/z 130→ 87 17eV collision voltage
- 3. Sample Chromatograms – Examples of typical chromatograms found with this method are shown below.

a. 50 ppb External Standard



#	Cmpd	Trace	RT	Peak Area	Sec.Trace	Sec.Area	Ion Ratio	Pred.Ratio	ppb	%Rec	S/N	Sec. S/N
1	Melamine	126.9>84.7	3.45	9538	126.9>67.7	1775	5.37	5.37	50.3	100.7	1856	759
2	ISTD	129.9>86.8	3.45	7838				1.00	632.6	126.5	1512	

b. Blank Poultry Muscle



#	Cmpd	Trace	RT	Peak Area	Sec.Trace	Sec.Area	Ion Ratio	Pred.Ratio	ppb	%Rec	S/N	Sec. S/N
1	Melamine	126.9>84.7	3.53	107	126.9>67.7	0	0.00	5.37	2.4		10	0
2	ISTD	129.9>86.8	3.53	1636				1.00	132.0	26.4	210	

Issuing Authority: Food Emergency Response Network (FERN)
Uncontrolled when printed

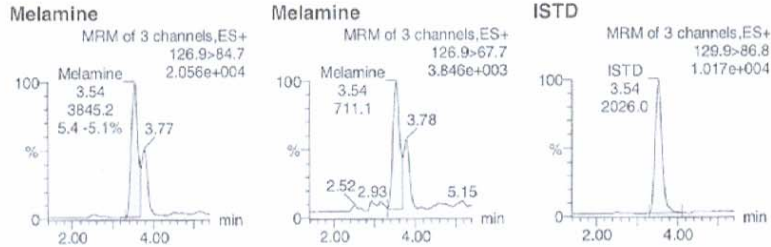


FERN

Uniting Federal, State and Local Laboratories for Food Emergency Response

SOP No: FERN-CHE.0003.00	Page 9 of 15
Title: LC/MS/MS Screen for the Presence of Melamine in swine and poultry tissues	
Revision: Original	Replaces: N/A
Effective: (5-07)	

c. 50 ppb Poultry Muscle Recovery



#	Cmpd	Trace	RT	Peak Area	Sec.Trace	Sec.Area	Ion Ratio	Pred.Ratio	ppb	%Rec	S/N	Sec. S/N
1	Melamine	126.9>84.7	3.54	3845	126.9>67.7	711	5.41	5.70	77.8	344	90	
2	ISTD	129.9>86.8	3.54	2026				1.00	324.5	64.9	333	

G. SCREENING CRITERIA

1. The retention time must match that of an external standard within 5%.
2. The melamine product ion abundance ratios must match that of an external standard within 20% relative.

Example calculation:

$$\text{Melamine ion ratio} = (\text{area of } m/z \text{ 127 to 85 peak}) \div (\text{area of } m/z \text{ 127 to 68 peak})$$

Note: Most mass spectrometers calculate the reciprocal of the above and state it as a percentage, which is equally valid.

$$\text{Passing ion ratio of a sample} = \text{ion ratio of a standard} \pm (0.2 * \text{ion ratio of a standard})$$

3. The solvent blank, ACN:H₂O (1:1, v/v), injected after the standard curve must be negative for melamine (no peak within the retention time window with passing ion ratio), demonstrating no carryover.
4. The positive tissue control must be positive for melamine (meeting retention time and ion ratio criteria).
5. The negative tissue control must not contain melamine at levels higher than 10% of the positive tissue control. Melamine peaks meeting the retention time and ion ratio criteria must be compared to the positive tissue control.

The Internal standard is used, the melamine quantification peak area (m/z 127 to 85) divided by the internal standard peak area (m/z 130 to 87) must be less than or equal to

Issuing Authority: Food Emergency Response Network (FERN)
Uncontrolled when printed



FERN

Uniting Federal, State and Local Laboratories for Food Emergency Response

SOP No: FERN-CHE.0003.00		Page 10 of 15
Title: LC/MS/MS Screen for the Presence of Melamine in swine and poultry tissues		
Revision: Original	Replaces: N/A	Effective: (5-07)

10% of that of the positive tissue control.

- For a sample to be screen positive, it must be found to contain melamine at or above the 50ppb minimum proficiency level of this analytical method.

The internal standard is used, the melamine quantification peak area (m/z 127 to 85) divided by the internal standard peak area (m/z 130 to 87) must be greater than or equal to that of the positive tissue control.

H. SAFETY INFORMATION AND PRECAUTIONS

- Required Personal Protective Equipment (PPE) - safety glasses, disposable gloves, lab coats.
- Hazards

<i>Reagents</i>	<i>Hazard</i>	<i>Recommended Safe Procedures</i>
Acetonitrile, Methanol	Flammable, poisonous; inhalation will cause headache, fatigue, nausea	Wear gloves and work in the hood. Use protective eyewear. Avoid contact with skin, eyes.
Methylene Chloride	Flammable, poisonous, carcinogenic. Inhalation may cause dizziness, sleepiness, nausea, and respiratory problems. In large dosages can cause unconsciousness or death.	Wear neoprene gloves when pouring or working with large volumes. Methylene chloride will eat through nitrile gloves! Work in the hood. Use protective eyewear. Avoid contact with skin, eyes.
Hydrochloric acid, Ammonium hydroxide	Corrosive, burns	Wear PPE, avoid skin contact, work in a fume hood.

- Disposal Procedures

<i>Procedure Step</i>	<i>Hazard</i>	<i>Recommended Safe Procedures</i>
Acetonitrile, Methanol	Flammable, poisonous; inhalation will cause headache, fatigue, nausea	Store waste in a tightly sealed container away from non-compatibles in a cool, well ventilated, flammable liquid storage area/cabinet for disposal in accordance with local, state and federal regulations.

Issuing Authority: Food Emergency Response Network (FERN)
Uncontrolled when printed



FERN

Uniting Federal, State and Local Laboratories for Food Emergency Response

SOP No: FERN-CHE.0003.00		Page 11 of 15
Title: LC/MS/MS Screen for the Presence of Melamine in swine and poultry tissues		
Revision: Original	Replaces: N/A	Effective: (5-07)

Methylene Chloride	Flammable, poisonous, carcinogenic. Inhalation may cause dizziness, sleepiness, nausea, and respiratory problems. In large dosages can cause unconsciousness or death.	Store waste in a tightly sealed container away from non-compatibles in a cool, well ventilated, flammable liquid storage area/cabinet for disposal in accordance with local, state and federal regulations.
Hydrochloric acid, Ammonium hydroxide	Corrosive, burns	Collect waste and store in a tightly sealed container. Store away from non-compatibles in a cool, well ventilated, acid liquid storage area/cabinet for disposal in accordance with local, state, and federal regulations, or neutralize and dispose in accordance with local, state and federal regulations.

I. QUALITY ASSURANCE PLAN

1. Performance Standard

Positive control is positive for melamine.
 Negative control is negative for melamine.

2. Critical Control Points and Specifications

<i>Record</i>	<i>Acceptable Control</i>
a. Weight of muscle sample	5.00 ± 0.05 g

3. Readiness To Perform

a. Familiarization

i. Phase I: Standards - Duplicate standard curve (D.2.d.) on each of 3 days, which will include the following:

- (a) 0 ppb
- (b) 25 ppb
- (c) 50 ppb
- (d) 100 ppb

ii. Phase II: Analyst Fortified samples - 3 replicates at 0ppb and 3 replicates

Issuing Authority: Food Emergency Response Network (FERN)
 Uncontrolled when printed



FERN

Uniting Federal, State and Local Laboratories for Food Emergency Response

SOP No: FERN-CHE.0003.00	Page 12 of 15
Title: LC/MS/MS Screen for the Presence of Melamine in swine and poultry tissues	
Revision: Original	Replaces: N/A
Effective: (5-07)	

at 50 ppb over a period of 3 different days.

NOTE: Phase I and Phase II may be performed concurrently.

- iii. Phase III: Check samples for analyst accreditation.
 - (a) 8 blind check samples: at least one of which must be negative, others shall be fortified at 50 ppb.
 - (b) Report analytical findings to supervisor / Quality Assurance Manager (QAM).
 - (c) Letter from QAM is required to commence official analysis.
- b. Acceptability criteria.
Refer to I. 1.

4. Intralaboratory Check Samples

- a. System, minimum contents.
 - i. Frequency: One per week per analyst when samples are analyzed.
 - ii. Records are to be maintained for review.
- b. Acceptability criteria.
Refer to I. 1.
If unacceptable values are obtained, then:
 - i. Stop all official analyses by that analyst.
 - ii. Take corrective action.

5. Sample Acceptability and Stability

- a. Matrix: swine and poultry muscle
- b. Sample size: approximately 500 g
- c. Condition upon receipt: cold, not spoiled
- d. Sample storage:
 - i. Time: 2 months
 - ii. Condition: frozen < -10°C

6. Sample Set

- a. Negative control (tissue blank).
- b. Positive control (tissue blank fortified with 50 ppb melamine).
- c. Test samples to be analyzed.

Issuing Authority: Food Emergency Response Network (FERN)
Uncontrolled when printed



FERN

Uniting Federal, State and Local Laboratories for Food Emergency Response

SOP No: FERN-CHE.0003.00	Page 13 of 15
Title: LC/MS/MS Screen for the Presence of Melamine in swine and poultry tissues	
Revision: Original	Replaces: N/A
Effective: (5-07)	

- 7. Sensitivity
 - a. Minimum proficiency level (MPL): 50 ppb.

J. WORKSHEET

Following worksheet is an example.



FERN

Uniting Federal, State and Local Laboratories for Food Emergency Response

SOP No: FERN-CHE.0003.00	Page 14 of 15
Title: LC/MS/MS Screen for the Presence of Melamine in swine and poultry tissues	
Revision: Original	Replaces: N/A
Effective: (5-07)	

Start Date	
End Date	
Notebook	
Data Folder	

Analyst	
Peer Review	
Supervisor	

Standards	ID
0 x	
1/2 x	
X	
2x	
fortification	
internal standard	

Reagents	ID	DISP/MIPP	Volume
ACN:H2O 1:1 v/v			
1N HCl			
methylene chloride			
Water	NA		
methanol			
0.1 N HCl			
5% NH ₄ OH/methanol			
aq. mobile phase		NA	NA
ACN mobile phase		NA	NA
MeOH mobile phase		NA	NA

Equipment	ID
balance	
tissuemizer	
shaker	
centrifuge	
micropipette	
N-evap	
LC/MS/MS	

Comments

Sample	Tissue	Weight	Ion Ratio	RT	Ppb	Spike	Recovery
		4.95 - 5.05 g		min.			
1							
2							
3							
4							
5							
6							
7							
8							
9							
10							
11							
12							

Issuing Authority: Food Emergency Response Network (FERN)
 Uncontrolled when printed



FERN

Uniting Federal, State and Local Laboratories for Food Emergency Response

SOP No: FERN-CHE.0003.00	Page 15 of 15
Title: LC/MS/MS Screen for the Presence of Melamine in swine and poultry tissues	
Revision: Original	Replaces: N/A
Effective: (5-07)	

K. APPENDIX

Reference: LC-MS/MS Method for the Analysis of Melamine in Porcine Meat Tissue (Not for Publication), California Animal Health and Food Safety Laboratory System, University of California, Davis, April 30, 2007.

L. Approvals and Authorities

Issuing Authority: Food Emergency Response Network (FERN)
Uncontrolled when printed

For the most current copy, check the Master Document List in eLEXNET

FERN SOP Template, Rev.12-01-06