1. ABSTRACT

Technetium is “trapped” on an Eichrom TEVA® resin to preconcentrate this analyte and to remove potential interferences. To do this from a biological sample, the sample is first treated with ammonia to stabilize the analyte, dried at 65°C (<75°C) to remove water and limit analyte loss, and finally ashed at 550°C to remove organic matter. The ash is then treated with 8 N nitric acid (HNO₃) and hydrogen peroxide to oxidize and extract the ⁹⁹Tc. The leachate is diluted to < 0.5 N HNO₃ and the ⁹⁹Tc concentrated on an Eichrom TEVA® resin. The ⁹⁹Tc is eluted with 8 N HNO₃ and the ⁹⁹Tc determined by inductively coupled plasma mass spectrometry (ICPMS). Rhenium (Re) is used as a recovery (yield) standard because it is not radioactive, has been shown to behave chemically similar to Tc and can be determined simultaneously with the ⁹⁹Tc.

2. APPLICABILITY

This procedure describes the basic steps necessary to determine Technetium-99 (⁹⁹Tc) in biological matrices including plant and animal tissue.

This method is a relatively simple, effective and, depending upon the initial sample size, sensitive method to determine trace ⁹⁹Tc in biological matrices. The method can also be used with other types of samples including waters and soils, but the recoveries have not been specifically verified. The method avoids some if the interferences encountered in standard radiochemical counting methods and eliminates the need to perform a separate count for a radioactive Tc tracer.

3. DISCUSSION

The Eichrom TEVA® resin is a liquid stationary phase consisting of a quaternary amine on a solid support. Technetium is “trapped” on an Eichrom TEVA® resin to preconcentrate this analyte and to remove potential interferences. To do this from a biological sample, the sample is first treated with ammonia to stabilize the analyte, dried at 65°C (<75°C) to remove water and limit analyte loss, and finally ashed at 550°C to remove organic matter. The ash is then treated with 8 N nitric acid (HNO₃) and hydrogen peroxide to oxidize and extract the ⁹⁹Tc. The leachate is diluted to < 0.5 N HNO₃ and the ⁹⁹Tc concentrated on an Eichrom TEVA® resin. The column is rinsed with 1 N HNO₃ to remove interferences. The ⁹⁹Tc is then eluted with 8 N HNO₃ and the ⁹⁹Tc determined by inductively coupled plasma mass spectrometry (ICPMS). Rhenium (Re) is spiked onto the samples in the very first step and is used as a recovery (yield) standard because it is not radioactive, has been shown to behave chemically similar to Tc and can be determined simultaneously with the ⁹⁹Tc.
DETERMINATION OF $^{99}\text{Tc}$ IN BIOLOGICAL SAMPLES USING TEVA® RESIN

The most probable interferences are molybdenum hydride ($^{99}\text{Mo}^1\text{H}$) and Ruthenium (Ru) at m/z 99. The relative abundances for Ru at m/z 99 and m/z 101 are 12.6% and 17%, respectively. The Ru interference can be corrected for by estimating the contribution of Ru at m/z 99 from the Ru response at m/z 101 (17% abundance) and subtracting it from the m/z 99 response. The probability of a significant interference by MoH is not likely but can be the possibility of having this interference can be determined by monitoring Mo at m/z 95 and/or Mo at m/z 98.

Tungsten hydrides ($^{184}\text{W}^1\text{H}$ and $^{186}\text{W}^1\text{H}$) and $^{187}\text{Os}$ may interfere with the Re measurements. Tungsten at m/z 182 can be monitored to assess the probability of a significant WH interference. An interelement correction for Os can be made using the Os response at m/z 189.

The W, Os, Mo and Ru interferences should be largely removed during the extraction and preconcentration of $^{99}\text{Tc}$ and Re on the TEVA® resin.

With seaweed samples, a variation of this method gave >90% recovery of $^{99}\text{Tc}$ and Re and it was determined that Re was a good chemical recovery ("yield") standard for $^{99}\text{Tc}$ (Tagami 2003, Mas). A variation reported 80-90% recovery for biota and 50-70% for sediment. Recovery of $^{99}\text{Tc}$ from soils was also adequate using a slightly different variation of this method without the Re (Tagami 2000). It does appear that Re behaves similarly to Tc in the environment (Wakoff). In general, Tc and Re are trapped efficiently on the TEVA at very low HNO$_3$ concentrations. Poor chemical recoveries are due mostly to the actual sample matrix. Therefore, one should expect to correct for all bias with the Re recovery. In a recent test, Re and $^{99}\text{Tc}$ recoveries were 80+% with 10 g wet weight and somewhat less with samples as large as 25g. However with correction by the Re, the $^{99}\text{Tc}$ was 97±4% for all samples. Possible under estimation of $^{99}\text{Tc}$ may result from Re actually in the sample. Generally, Re is <1-2% of the Re tracer added to the sample.

4. SAFETY PRECAUTIONS

4.1 Chemical Handling

Handling of acids and bases and chemical vapors generated from these chemicals is a safety consideration in this procedure. Handle all chemicals will be handled per MCP-3635, "Chemical Hygiene Plan." Use proper personal protective equipment (PPE) per PRD-5121, "Personal Protective Equipment." At a minimum wear PPE consisting of safety glasses with side shields, nitrile gloves, and any additional PPE specified by the RWP, RCI or IH. Handle all the acids and bases in this procedure in a ventilation hood. Obtain more safety information on specific chemicals can be obtained, as needed, from MSDS sheets (available on the INEEL intranet), laboratory supervision, and industrial safety personnel. MSDS sheets can be found on the INEEL intranet.
4.2 **Hot surfaces**

**WARNING**
Thermal burns can result from skin contact with hot surfaces on the hot plates and heating blocks.

Avoid contact with hot surfaces on hot plates and heating blocks. Use care when removing beakers from hot plate. Use heat resistant gloves as appropriate, particularly when using the muffle furnace. Handle samples in hot crucibles with crucible tongs as appropriate.

4.3 **\(^{99}\text{Tc} - \text{Radioactive Materials and Sample Hazards}\)**

In general no "radioactive" samples are anticipated, as the major intent of this procedure is to determine \(^{99}\text{Tc}\) at near environmental levels in biological samples. However, broader application of the procedure might imply its use for samples determined to be radioactive. For radioactive samples, perform all work under an applicable Radiological Work Permit (RWP) for the area (see MCP-7, "Radiological Work Permit"). Perform all radiological work in a bench top work area, a radioactive fume hood, glove box, or hot cell as per the hazard index and the requirements and instructions listed on the RWP. Obtain RCT support as necessary when preparing \(^{99}\text{Tc}\) solutions or when performing sample spiking with \(^{99}\text{Tc}\).

4.4 **Waste Disposition**

Handle all waste generated from the performance of this method as directed by Waste Generator Services.

5. **APPARATUS AND REAGENTS**

5.1 **Apparatus**

5.1.1 Porcelain (or Pt or quartz) crucibles (50 mL)

5.1.2 Analytical balance, with at least 0.01 g readability, calibrated by the INEEL S&CL

5.1.3 15 mL graduated polyethylene vials

5.1.4 50 mL graduated polyethylene vials
5.1.5 Beakers, various sizes

5.1.6 Watch glasses, preferably Teflon®

5.1.7 Mechanical pipettors and associated tips, various sizes

5.1.8 250 mL polyethylene bottles

5.1.9 10 and 50 mL polyethylene syringes

5.1.10 0.45 and 1 μm Acrodisc CR syringe filters or equivalent

5.1.11 Oven

5.1.12 Furnace

5.1.13 Hot plate

5.1.14 TEVA® extraction columns or cartridges available from Eichrom Technologies, Inc. (Evanston, IL)

5.1.15 Inductively Coupled Plasma Mass Spectrometer

5.2 **Reagents**

5.2.1 Deionized water

5.2.2 Nitric Acid (15.7 N) – concentrated

5.2.3 Nitric Acid (1 N)

5.2.4 Nitric Acid (8 N)

5.2.5 Nitric Acid (0.1 N)

5.2.6 30% Hydrogen Peroxide

5.2.7 Rhenium stock solution (ideally $^{185}\text{Re}$) – 200 ng/mL in 0.1 N HNO₃

5.2.8 $^{99}\text{Tc}$ stock solution - 10 ng/mL in 0.1 N HNO₃ (0.17 nCi/mL = 170 pCi/mL = 6.29 Bq/mL = 377 DPM/mL) – 1-2 mL should be enough to run 10-20 batches

**NOTE:** $^{185}\text{Re}$ enriched standard is preferred in order to compensate for any natural Re in the sample via an isotope dilution determination of Re.

5.2.9 Indium stock solution – 100 ng/mL in 0.1 N HNO₃

5.2.10 Ammonia solution (20% NH₃)
5.2.11 ICP-MS Calibration Standards: Prepare 50-mL volumes in 0.1 N HNO₃ including a blank and four different concentration levels for $^{99}$Tc and Re. Standards should be prepared fresh at least on a monthly basis. Examples are given below.

5.2.11.1 Std0 = 1 ng/mL In

5.2.11.2 Std1 = 1 ng/mL In, 0.5 ng/mL Mo, 0.5 ng/mL Os and 0.5 ng/mL Ru.

**NOTE:** Std1 can be used to standardize the instrument to determine the interference concentration levels or simply to verify that the "interelement corrections" from 7.4.2 are good.

5.2.11.3 Std2 = 1 ng/mL In, 1 ng/mL Re, 100 pg/mL $^{99}$Tc (1.7 pCi/mL = 6.29x10⁻² Bq/mL = 3.77 dpm/mL).

5.2.11.4 Std3 = 1 ng/mL In, 2 ng/mL Re, 200 pg/mL $^{99}$Tc (3.4 pCi/mL = 0.126 Bq/mL = 7.55 dpm/mL).

5.2.11.5 Std4 = 1 ng/mL In, 3 ng/mL Re, 300 pg/mL $^{99}$Tc (5.1 pCi/mL = 0.189 Bq/mL = 11.3 dpm/mL).

5.2.11.6 Std5 = 1 ng/mL In, 4 ng/mL Re, 400 pg/mL $^{99}$Tc (6.8 pCi/mL = 0.252 Bq/mL = 15.1 dpm/mL).

5.2.12 ICP-MS Calibration Verification Standard, 1 ng/mL In, 2 ng/mL Re, and 0.1 ng/mL $^{99}$Tc: Prepare 50-mL volume in 0.1 N HNO₃ from independent stock solutions if possible. Standards should be prepared fresh at least on a monthly basis.

6. **SAMPLE HANDLING**

6.1 Biological samples

Should remain frozen or refrigerated until use.

7. **PROCEDURES**

7.1 Sample Preparation

7.1.1 Record crucible mass.

7.1.2 Weigh wet sample into the tared crucible (<25 g wet weight) and record the sample mass.

7.1.3 Using Re as a recovery standard add 100 µL of a 200 ng/mL stock to every sample (i.e. 20 ng of Re-would prefer to have $^{185}$Re enriched
DETERMINATION OF $^{99}$Tc IN BIOLOGICAL SAMPLES USING TEVA® RESIN

standard to compensate for any natural Re in the sample via an isotope dilution determination of Re).

**NOTE:** Re needs to be added to every sample at this point in order to correct for losses that occur during the procedure.

7.1.4 For $^{99}$Tc-spiked samples add 100 µL of a 10 ng/mL standard (corresponds to addition of 1 ng or 17 pCi of $^{99}$Tc).

**NOTE:** At least one blank sample and one duplicate sample (if available) or a set of duplicates (if available) is spiked with $^{99}$Tc for each batch of samples prepared. Simple fortified blanks will not work (i.e. no sample matrix) as losses of $^{99}$Tc and Re are excessive.

7.1.5 Add 10 mL or enough to wet the sample of 20% ammonia solution as an analyte retention/ashing aid and mix with sample.

7.1.6 Dry at $<$75°C for 24 hr or until mass is stable.

**NOTE:** If time is an issue, the drying temperature can go as high as 110°C with little or no effect (Tagami 2003).

7.1.7 Cool and record mass of the crucible with the now dried sample.

7.1.8 Place sample into an oven/furnace at $<$250°C and raise the temperature to 550°C and “ash” for 3 hr.

7.1.9 Cool and record mass of the crucible with the ashed sample.

7.1.10 Add 10 mL of 8 N HNO₃, 2.5 mL of 30% H₂O₂ directly to the crucible and heat at $<$75°C for 3 hr under reflux conditions (i.e. covered with watchglass).

7.1.11 Cool, decant and filter with a syringe through a 0.45 µm Acrodisc CR filter into a 250 mL polypropylene bottle.

7.1.11.1 IF filtering is difficult, THEN stack a 1-µm Acrodisc CR filter with the 0.45-µm Acrodisc CR filter for ease of filtering.

7.1.11.2 IF filtering of plant samples is difficult due to the presence of micro particulates, THEN use vacuum filter units employing 0.45-µm pore size filter membranes as necessary.

7.1.12 Rinse remaining solids at least 2 times with aliquots of deionized water, decanting and filtering the solution through the 0.45 µm Acrodisc CR filter into the 250 mL polypropylene bottle.

7.1.13 Dilute to $>$200 mL with deionized water.
7.2 **Eichrom TEVA Cartridge/Column Preparation**

7.2.1 Precondition with 5 mL of 8 N HNO₃ and discard the solution.

7.2.2 Follow with 10 mL of 0.1 N HNO₃ and discard the solution.

7.3 **Eichrom TEVA Column Separation and final dilution**

7.3.1 Load sample from 7.1.13 onto the TEVA column/cartridge at an average flow rate of between 1 and 1.5 mL/min.

**NOTE:** The ⁹⁹Tc and Re will be retained on the column/cartridge.

7.3.1.1 If necessary to achieve a reasonable flow rate then use a pump or the vacuum box to achieve this flow rate with the cartridges or substitute 2 mL columns with larger particles for sufficient gravity flow.

7.3.1.2 Discard the liquid.

7.3.2 Wash (to remove Mo, Ru and W) with 20 mL of 1 N HNO₃. The analytes are retained on the column, so the liquid coming through the column is discarded.

**NOTE:** Larger concentrations of HNO₃ may preclude some of the ⁹⁹Tc and Re with the contaminants.

7.3.3 Elute the ⁹⁹Tc and Re with 5+ mL of 8 N HNO₃ into a 15 mL graduated polyethylene tube or directly into the beaker to be used for evaporation.

7.3.4 Spike with 100 µL of 100 ng In/mL to be used as an internal standard.

7.3.5 Evaporate to near dryness at <75°C on a hot plate (because 8 N HNO₃ should not be aspirated directly into the ICP-MS).

**NOTE:** Keeping the temperature <75°C will minimize the probability of any losses or differential losses due to the volatility of Tc and Re as HTcO₄ and HReO₄.

7.3.6 Dilute to with 0.1 N HNO₃, transfer to a 15 mL graduated polyethylene tube and dilute to 10 mL with 0.1 N HNO₃. The resulting solution should be 0.1+ N HNO₃ (i.e. ≈ 1% HNO₃).

7.4 **ICP-MS Instrument Setup**

7.4.1 Use the following masses ⁹⁵Mo (15.9%), ⁹⁸Mo (24.1%, 1.9% Ru), ⁹⁹Tc, ¹⁰¹Ru (17%), ¹⁰²Ru (31.6%, optional line to confirm Ru interference potential), ¹¹⁷In (95.7%), ¹¹⁸Sn (24.2%), ¹⁸⁷Re (37.4%), ¹⁸³Re (62.6%), ¹⁸²W (26.3%) and ¹⁸⁹Os (16.1).
7.4.2 Interelement corrections include

- $^{99}$Tc = $^{99}$Tc – 0.747 * $^{101}$Ru
- $^{115}$In = $^{115}$In – 0.0140 * $^{118}$Sn
- $^{187}$Re = $^{187}$Re – 0.0994 * $^{189}$Os
- $^{98}$Mo = $^{98}$Mo – 0.112 * $^{101}$Ru
- $^{103}$Ru = $^{103}$Ru – 0.0448 * $^{105}$Pd (optional)

7.4.3 Tune ICP-MS using manufacturer specification.

7.4.4 Make three replicate determinations per analysis with a total acquisition time of three minutes per sample, assuming an uptake rate of ≤1 mL/minute.

7.4.5 Analysis steps

7.4.5.1 Calibrate using the standards in 7.4.4.1-7.4.4.6 and verify that the $R^2$ for $^{99}$Tc and Re is greater than 0.99.

**NOTE:** Std1 can be used to standardize the instrument to determine the interference concentration levels or simply to verify that the “interelement corrections” from 7.4.2 are good.

7.4.5.2 Analyze the blank (Std0).

7.4.5.3 Analyze the calibration verification solution.

7.4.5.4 IF the blank and calibration verification are not acceptable (e.g. blank <1 pg/mL and calibration verification ±10%), THEN repeat 7.4.5.1 through 7.4.5.3.

7.4.5.5 IF the blank and calibration verification are acceptable (e.g. blank <1 pg/mL and calibration verification ±10%), THEN run five samples.

7.4.5.6 Repeat 7.4.5.2 through 7.4.5.5 until all samples have been analyzed ending the analysis sequence with a successful blank and calibration verification.

7.4.6 Perform instrument shutdown, maintenance and troubleshooting, as necessary, per guidance in the ICPMS vendor-supplied literature.
8. QUALITY CONTROL REQUIREMENTS

8.1 Since this is a destructive analysis, the actions to be taken should a QC sample result fall outside of the expected range may be limited. Repeating the instrumental analysis may be possible if enough solution remains, but repeating the sample preparation may not be possible due to limited sample availability. Specific actions must be discussed with and agreed to by the requestor.

8.2 At a minimum prepare one ⁹⁹Tc spiked sample preparation for every 20 samples if there is enough sample. As previously noted, the ⁹⁹Tc recovery can vary widely however, after correction with the Re recovery the ⁹⁹Tc should be in the range of 100±20% or other value specified by the requestor.

8.3 At a minimum prepare one duplicate sample preparation for every 20 samples if there is enough sample to do so. Sample homogeneity and concentration level may affect the results, however in general, duplicate samples should be within ±20% or a values specified by the requestor.

8.4 At a minimum prepare one blank sample preparation (i.e. a blank reference material) for every 20 samples. These should be non-detects.

NOTE: Simple reagent blanks generally exhibit high losses of Tc and Re so the blank reference material should be some type of material closely related to the sample material.

8.5 At a minimum prepare one ⁹⁹Tc spiked blank sample preparation (i.e. spike a blank reference material) for every 20 samples. The ⁹⁹Tc recovery can vary widely however, after correction with the Re recovery should be in the range of 100±20%.

NOTE: Simple reagent blanks generally exhibit high losses of Tc and Re so the blank reference material should be some type of material closely related to the sample material.
9. **CALCULATIONS**

9.1 **Useful conversions and factors:**

- $^{99}\text{Tc}$ activity = 0.017 Ci/g
- 1 Bq = 2.7x10^{-11} Ci = 27 pCi.
- Dry weight % = %DW = 100*W_{dry}/W_{wet}
- Ash content % = %DW * W_{ash}/W_{dry}

9.2 **% Re recovery**

9.2.1 If using natural Re then \% Re recovery = 100\% * C_{diluted \text{ solution}}/(2 \text{ ng/mL})

9.2.2 If using isotope dilution with $^{185}$Re enriched standard, then for natural Re

\[ C_X = 186.21 \times \left( \frac{C_S V_S}{V_X} \right) \left( \frac{A_S - R_M B_S}{R_M B_X - A_X} \right) \]

where:

- $C_X$ is the concentration of natural Re in the diluted sample in ng/mL.
- $C_S$ is the concentration of Re in nMol/mL of the stock Re spiking solution.
- $A_X$ and $B_X$ are the natural atom fractions of $^{185}$Re and $^{187}$Re in the sample, respectively.
- $A_S$ and $B_S$ are the atom fractions of $^{185}$Re and $^{187}$Re in the enriched standard, respectively.
- $R_M$ is the measured ratio of the spiked sample.
- $V_X$ is the final dilution of the sample (10 mL).
- $V_S$ is the volume of the standard added (0.1 mL).

After solving for $C_X$, the contribution of the natural Re at m/z 185 is subtracted from the response at m/z 185 to determine the concentration of the $^{185}$Re enriched spike from a calibration curve. Once the concentration of the Re spike has been determined, use equation 9.2.1.
9.3 For $^{99}\text{Tc}$

\[ pg^{99}\text{Tc} / g = \frac{C_{\text{Tc-wet}} \cdot V_X}{W_{\text{wet}}} \]

\[ pCi^{99}\text{Tc} / g = A_{\text{Tc-wet}} = \frac{C_{\text{Tc}} \cdot V_X}{W_{\text{wet}}} \cdot 0.0170 \]

\[ pCi^{99}\text{Tc} / g = A_{\text{Tc-wet-corrected}} = \frac{A_{\text{Tc-wet}}}{\% \text{Re}_{\text{rec}}} \]

where:

- $C_{\text{Tc-wet}}$ is the dry weight $^{99}$Tc concentration.
- $A_{\text{Tc-wet}}$ is the dry weight $^{99}$Tc activity.
- $C_{\text{Tc}}$ is the $^{99}$Tc concentration in the final dilution (pg/mL).
- $V_X$ is the final dilution volume in mL (10 mL).
- $W_{\text{wet}}$ is the dry weight used in the analysis.
- 0.0170 is the activity of $^{99}$Tc in pCi/pg.

Values are adjusted for "yield" by dividing by the % Re recovery.

9.4 Total uncertainty

Total uncertainty is a term that is inclusive of all of the sources of error in the analysis. For the determination of $^{99}$Tc in this procedure, this will include the uncertainty of the 3 determinations of the $^{99}$Tc and Re intensities and the uncertainty associated with the instrument variability over time.

\[ \text{Total Uncert.} = \left[^{99}\text{Tc}_{\text{corrected}} \right] \cdot \sqrt{RSD_{^{99}\text{Tc}}^2 + RSD_{\text{Re}}^2 + RSD_{\text{Calibration Checks}}^2} \]

Where RSD is the relative standard deviation of the various measurements and

\[ RSD = \frac{s}{\bar{x}} \]

9.5 Detection limits

Detection limits will be dependant upon the Ru in the sample which adversely effects the precision of the $^{99}$Tc measurement at m/z 99. Therefore, the detection
limits are determined on a sample by sample basis by multiplying the $^{99}$Tc 
standard deviation or total uncertainty from the 3 replicates by the single-sided 
Student t value at $p=0.01$ for 2 degrees of freedom or 6.965.

10. **RECORDS**

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<th>Uniform File Code</th>
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<td>ENV5-d</td>
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11. **REFERENCES**


11.6 MCP-7, "Radiological Work Permit"

11.7 MCP-3635, "Chemical Hygiene Plan"

11.8 PRD-5121, "Personnal Protective Equipment"

11.9 MCP-3562, "Hazard Identification, Analysis and Control of Operational Activities"
12. METHOD HISTORY

12.1 Revision History

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<thead>
<tr>
<th>Revision</th>
<th>Author(s)</th>
<th>Date</th>
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<tbody>
<tr>
<td>0</td>
<td>William F. Bauer</td>
<td>November 2004</td>
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12.2 Current Revision Summary

New method

13. APPROVALS

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## APPENDIX A
### Procedure Basis

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<tr>
<th>Step</th>
<th>Basis/Summary</th>
<th>Source</th>
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<tr>
<td>7.1 thru 7.3</td>
<td>Wear nitrile glove and safety glasses/goggles as appropriate</td>
<td>JSA ACMM3705,</td>
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<tr>
<td>7.1.4 and 7.4.4</td>
<td>Perform spiking of samples with $^{99}$Tc and preparation of standards containing $^{99}$Tc with RCT support as appropriate</td>
<td>JSA ACMM3705,</td>
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<tr>
<td>7.1.8</td>
<td>Placing samples into 250°C oven wear heat resistant gloves and use tongs.</td>
<td>JSA ACMM3705,</td>
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<td>7.1.5 thru 7.3.5</td>
<td>Work generally performed in ventilation hood as appropriate to minimize exposure to vapors</td>
<td>JSA ACMM3705,</td>
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