

**FORENSIC TOXICOLOGY LABORATORY
OFFICE OF CHIEF MEDICAL EXAMINER
CITY OF NEW YORK**

HEAVY METALS

**ANTIMONY, ARSENIC, BISMUTH, MERCURY
by
REINSCH TEST**

PRINCIPLE

The classical Reinsch test is an adequate presumptive procedure to indicate whether or not heavy metals are present in biological samples. The procedure is based on the fact that copper will be displaced by elements below it in the electrochemical series. Antimony, arsenic, bismuth and mercury will deposit on a copper wire or copper strip and the resulting discolored copper is an indication of a positive result. This screening test is rapid, sensitive, reliable, and can be used on biological fluids or tissue homogenates without extensive preliminary treatment. The test is sensitive to approximately 0.5 mg/L of arsenic, 1.0 mg/L of antimony and bismuth and 2.5 mg/L of mercury.

SAFETY

The handling of all reagents, samples and equipment is performed within the guidelines which are detailed in the safety manual.

REAGENTS

All chemicals should be analytical reagent grade.

1. Hydrochloric acid, concentrated
2. Nitric acid, concentrated
3. Copper wire. Form approximately 6 inches into a spiral for $\frac{1}{2}$ the length.
4. Mercury stock solution, 1000 mg/L

Dissolve 1.354 g of mercuric chloride in 1000 mL of distilled water. Add 2.0 mL hydrochloric acid.

5. Mercury reference solution, 10 mg/L

Dilute 1.0 mL of the mercury stock solution to 100 mL in a volumetric flask with distilled water.

6. Distilled or deionized water.
7. Arsenic stock solution, 1000 mg/L

Dissolve 1.32 g arsenic trioxide in 10.0 mL of 10N sodium hydroxide and add 500 mL of water. Neutralize the solution with concentrated hydrochloric acid, and then dilute it to 1L with water.

8. Arsenic reference solution, 10 mg/L

Dilute 1.0 mL of the arsenic stock solution to 100 mL in a volumetric flask with distilled water.

9. Potassium cyanide, 10 g/100 mL

Dissolve 10 g of potassium cyanide in water and dilute to 100 mL.

SAMPLE PREPARATION

1. Urine, 20 mL. Record dilution if sample volume is less than 20 mL.
2. Blood, 1.0 mL diluted to 20 mL with water.
3. Tissues, 5.0 mL of homogenate (1:5), diluted to 20 mL.
4. Gastric, 5.0 mL of a 1:10 dilution, diluted to 20 mL.
5. Miscellaneous samples (vomitus, powders, tablets, residues). Dissolve or suspend in 20 mL water.

PROCEDURE

1. Wash the copper wire spirals with concentrated nitric acid and dry.
2. Place 20 mL of sample as prepared above into a 50 mL test tube labeled with the contents.
3. Prepare matrix matched blanks and controls from negative matrix. Use water for urine and gastric. Place into 50 mL test tubes. Prepare a suitable negative matrix for all samples tested.
4. Pipet 5 mL of 10 mg/L arsenic reference solution (positive arsenic control) into a 50 mL test tube. Into another 50 mL test tube pipet 10 mL of the 10 mg/L mercury reference solution (positive mercury control). Dilute both to 20 mL with negative matrix. Prepare positive controls in each matrix used.
5. Add 5.0 mL of concentrated hydrochloric acid to each tube. Mix by Vortex.
6. Add the freshly washed copper wire (shaped into a spiral) into each tube.
7. Heat the solution gently for about one hour under the fume hood.
8. Remove the copper wire from the test tube, wash it with water and dry it on a piece of white paper.

INTERPRETATION

A silver deposit indicates the presence of mercury; shiny black deposit indicates the presence of bismuth; dull black deposit indicates the presence of arsenic; dark purple deposit indicates the presence of antimony.

LIMIT OF DETECTION

Limit of detection for arsenic in urine or gastric is 1 µg/mL, in blood, 6 µg/mL, and in liver, 2 µg/g liver.

Limit of detection for mercury in blood, urine, gastric, blood and liver is 100 µg/mL.

INTERFERING SUBSTANCES

Dark deposits may indicate selenium and tellurium, whereas a speckled discoloration indicates the presence of high concentrations of sulphur. Parallel analysis of all reagents may be necessary to rule out false positive results due to the reagents. Concentrations of heavy metals normally present in urine are too low to be detected in samples of the size suggested for the test described above. A positive test should be confirmed with a more precise quantitative procedure.

ACCEPTANCE CRITERIA

1. Only specimens that have been analyzed with successful controls can be reported.
2. Negative control must not leave a deposit on the copper strip or wire.
3. Positive control must produce a deposit on the copper strip or wire.

REPORTING

1. Samples which do not leave a deposit on the copper strip or wire will be reported as “antimony, arsenic, bismuth and mercury not detected”.
2. Samples which produce a deposit on the copper strip or wire will be reported as “antimony detected”, “arsenic detected”, “bismuth detected” or “mercury detected” depending on the appearance of the deposit.

Note: To confirm that the dull black deposit is due to arsenic, place the coated copper into 2.0 mL of the potassium cyanide solution. If the black deposit is due to arsenic, it will dissolve; if due to bismuth or antimony, the black deposit will not be removed.

Note: The final toxicology report will indicate that positive results are unconfirmed and that confirmation is available upon request.

REFERENCES

Sidney Kaye, ed., *Handbook of Emergency Toxicology*, Charles C. Thomas, Springfield, Illinois, 1988.

Irving Sunshine, ed., *Handbook of Analytical Toxicology*, CRC Press, Cleveland, Ohio, 1969.