$C_{21}H_{22}N_2O_2$ MW: 334.42 CAS: 57-24-9 RTECS: WL2275000

METHOD: 5016, Issue 2 **EVALUATION: FULL** **Issue 1: 15 May 1985**
Issue 2: 15 August 1994

OSHA : 0.15 mg/m^3
NIOSH: 0.15 mg/m^3/10 h; Group I Pesticide
ACGIH: 0.15 mg/m^3

PROPERTIES: solid; MP 268 °C; VP not significant

SYNONYMS: strychnidin-10-one

SAMPLING

SAMPLER: FILTER (glass fiber, 37-mm)

FLOW RATE: 1 to 3 L/min

VOL-MIN: 70 L @ 0.15 mg/m^3
-MAX: 1000 L

SHIPMENT: routine

SAMPLE STABILITY: at least 7 days @ 25 °C [1]

BLANKS: 2 to 10 field blanks per set

ACCURACY

RANGE STUDIED: 0.073 to 0.34 mg/m^3 [3] (180-L samples)

BIAS: 4.4%

OVERALL PRECISION ($\hat{S}_{rT}$): 0.059 [1]

ACCURACY: ± 14.6%

MEASUREMENT

TECHNIQUE: HPLC, UV DETECTION

ANALYTE: strychnine

DESORPTION: 5 mL mobile phase

INJECTION VOLUME: 20 µL

COLUMN: 25 cm x 4.2-mm ID, packed with µBondpack C18, 10-µm particle size

MOBILE PHASE: aqueous 1-heptane sulfonic acid + CH_3CN; pH 3.5; 1 mL/min @ ambient temperature

DETECTOR: UV absorption @ 254 nm

CALIBRATION: solutions of strychnine in mobile phase

RANGE: 10 to 70 µg per sample

ESTIMATED LOD: 0.8 µg per sample [1,2]

PRECISION ($\hat{S}_r$): 0.042 @ 13 to 62 µg per sample [1]

APPLICABILITY: The working range is 0.05 to 10 mg/m^3 for a 200-L air sample.

INTERFERENCES: None known.

OTHER METHODS: This revises Method S302 [2].

REAGENTS:

1. Strychnine, 98%.*
2. Acetonitrile, chromatographic quality.
3. Water, chromatographic quality.
4. Acetonitrile:water, 1:1 (v/v). Mix equal volumes of acetonitrile and distilled water.
5. Acetic acid, glacial.*
6. Acetic acid, 0.01 N. Dilute 0.6 mL glacial acetic acid to 1 L with distilled water.
7. 1-Heptanesulfonic acid sodium salt, 95%.
8. Calibration stock solution, 1 mg/mL strychnine in 0.01 N acetic acid. Use an ultrasonic bath to aid dissolution.
9. Mobile phase: Dissolve 1.1014 g 1-heptanesulfonic acid sodium salt in 980 mL. 1:1 acetonitrile:water. Adjust pH to 3.5 with glacial acetic acid. Dilute to 1 L with 1:1 acetonitrile:water. Filter through 0.45-µm PTFE filter and refrigerate. Prepare fresh weekly and if cloudiness appears.

* See SPECIAL PRECAUTIONS.

EQUIPMENT:

1. Sampler: glass fiber filter, 37-mm diameter, in cassette filter holder.
2. Personal sampling pump, 1 to 3 L/min, with flexible connecting tubing.
3. HPLC, UV absorption detector at 254 nm, recorder, integrator and column (page 5016-1).
4. Syringe filter, polypropylene housing, nylon membrane, 0.2-µm pore size.
5. Syringe, 20-µL.
6. Micropipets or syringes, 10- to 100-µL.
7. Jars, glass, squat-form ointment, with PTFE-lined screw caps.
8. Volumetric flasks, glass, 10-mL.
9. Pipet, 5-mL delivery, with pipet bulb.
10. Tweezers.
11. Ultrasonic bath.

SPECIAL PRECAUTIONS: Strychnine is a potent convulsant. Use impervious clothing, gloves and face shields when handling it [3,4].

Avoid skin contact with acetic acid as it may cause severe burns.

SAMPLING:

1. Calibrate each personal sampling pump with a representative sampler in line.
2. Sample at an accurately known flow rate between 1 and 3 L/min for a total sample size of 70 to 1000 L.

SAMPLE PREPARATION:

3. Transfer the filter carefully using tweezers to an ointment jar.
4. Add 5.0 mL mobile phase. Seal and gently swirl the jar to wet the filter.
5. Filter the sample solution through a syringe filter.

CALIBRATION AND QUALITY CONTROL:

6. Calibrate daily with at least six working standards over the range 1 to 70 µg strychnine per sample (0.2 to 14 µg/mL).
   a. Add known amounts of calibration stock solution to mobile phase in 10-mL volumetric flasks and dilute to the mark.
   b. Analyze together with the samples and blanks (steps 9, 10, and 11).
   c. Prepare calibration graph (peak area vs. µg strychnine).
7. Determine recovery (R) at least once for each lot of filters used for sampling in the calibration range. Prepare three filters at each of five levels plus three media blanks.

a. Deposit a known amount of calibration stock solution onto the filter. Allow filters to air dry.
b. Store samples overnight in ointment jars.
c. Prepare for analysis (steps 3 through 5) and analyze together with working standards (steps 9 through 11).
d. Prepare a graph of R vs. µg strychnine recovered.

8. Analyze three quality control blind spikes and three analyst spikes to ensure that the calibration graph and R graph are in control

MEASUREMENT:

9. Set HPLC system according to manufacturer's recommendations and to the conditions given on page 5016-1.
10. Inject sample aliquot using syringe, fixed volume sample loop or autosampler.
11. Measure peak area.

CALCULATIONS:

12. Determine the mass, µg (corrected for R) of strychnine found in the sample (W) and in the average media blank (B) from the calibration graph.
13. Calculate concentration, C, of strychnine in the air volume sampled, V (L):

$$C = \frac{W - B}{V}, \text{ mg/m}^3.$$

EVALUATION OF METHOD:

Method S302 was issued on February 17, 1978 [2], and evaluated over the range 0.073 to 0.34 mg/m^3 using a 180-L sample [1,5]. Overall precision, $\hat{S}_{rT}$, was 0.059 with an average recovery of 100.9%, representing a non-significant bias. The concentration of strychnine was independently verified by analysis of filter samples with a UV spectrophotometer at 254 nm. Recovery was 0.98 in the range of 13.5 to 54.1 µg per sample. Collection efficiency was determined to be at least 98.8% for 180-L sample at a concentration to be 0.15 µg/mL. Average recovery for one-day old samples was 105% vs. 98.8% for seven-day old samples stored at room temperature.

Since the filter samples for the recovery and stability studies were prepared with strychnine acetate rather than the free base, the recovery of the free base and the stability of such samples is unknown.

REFERENCES:

[1] Back-up Data Report for Strychnine, NIOSH Contract No. 210-76-0123, available as "Ten NIOSH analytical Methods," Order Nol PB288-629 from NTIS, Springfield, VA 22161.
[2] NIOSH Manual of Analytical Methods, 2nd ed., Vol. 5, S302, U.S. Department of Health, Education, and Welfare, Publ. (NIOSH) 79-141 (1979).
[3] Criteria for a Recommended Standard...Occupational Exposure During the Manufacture and Formulation of Pesticides, U.S. Department of Health, Education, and Welfare, Publ. (NIOSH) 78-174 (1978), available as PB81-227001 from NTIS, Springfield, VA 22161.
[4] NIOSH/OSHA Occupational Health Guidelines for Chemical Hazards, U.S. Department of health and Human Services, Publ. (NIOSH) 81-123 (1981), available as GPO Stock #017-033-00337-8 from Superintendent of Documents, Washington, D.C. 20402.
[5] NIOSH Research Report - Development and Validation of Methods for Sampling and analysis of Workplace and Toxic Substances, U.S. Department of Health and Human Services, Publ. (NIOSH) 80-133 (1980).

METHOD REVISED BY:

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