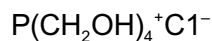


TETRAKIS(HYDROXYMETHYL)PHOSPHONIUM CHLORIDE

5046



MW: 190.58

CAS: 124-64-1

RTECS: TA2450000

METHOD: 5046, Issue 1

EVALUATION: PARTIAL

Issue 1: 15 March 2003

OSHA: None
 NIOSH: None
 ACGIH: None

PROPERTIES: hygroscopic, air sensitive solid; MP 151 °C; ionic; does not vaporize; soluble in water, methanol; insoluble in diethyl ether

SYNONYMS: tetramethylolphosphonium chloride, tetrahydroxymethylphosphonium chloride, THPC, Pyroset TKC, Proban CC, Retardol C, NCI-C55061, Tolcide THPC

SAMPLING		MEASUREMENT	
SAMPLER:	37-mm filter cassette (open-faced cassette containing two glass fiber filters coated with acidified 2,4-dinitrophenylhydrazine)	TECHNIQUE:	HPLC; UV DETECTION
FLOW RATE:	1.0 to 1.7 L/min	ANALYTE:	2,4-dinitrophenylhydrazone of formaldehyde
VOL-MIN:	1 L @ 6.5 mg/m ³	EXTRACTION:	4 mL acetonitrile; 60 °C, 1 hour
-MAX:	480 L @ 0.15 mg/m ³	INJECTION VOLUME:	20 µL
SHIPMENT:	Ship on ice (0 °C).	MOBILE PHASE:	45:55 acetonitrile:water @ 1.3 mL/min
SAMPLE STABILITY:	28 days @ 5 °C [1]	COLUMN:	3.9 x 150 mm stainless steel, packed with 5-µm C-18, Symmetry™ or equivalent
BLANKS:	2 to 10 field blanks per set 10 media blanks per set	DETECTOR:	UV @ 360 nm
ACCURACY		CALIBRATION:	DNPH-coated glass fiber filters fortified with THPC (media standards)
RANGE STUDIED:	Not studied	RANGE:	6.5 to 60 µg/sample (instrumental)[1]
BIAS:	Not determined	ESTIMATED LOD:	2 µg/sample
OVERALL PRECISION (Ŝ_{r,t}):	Not determined	PRECISION (S_r):	0.0585 for 8 to 31 µg per sample
ACCURACY:	Not determined		

APPLICABILITY: The theoretical working range is 0.43 to about 67 mg/m³ for a 15-L air sample. This method may be applicable to other tetrakis(hydroxymethyl)phosphonium salts, such as the sulfate. However, evaluation of this method has been limited to THPC.

INTERFERENCES: Formaldehyde gas is an interference. Concentrations of THPC can be corrected for formaldehyde gas by sampling for formaldehyde gas with NMAM 2016 [2]. Ozone has been observed to consume DNPH and to degrade the DNPH derivative of formaldehyde [3]. However, use of an ozone scrubber is not recommended because this scrubber may trap THPC aerosol. Ketones and other aldehydes can react with DNPH; the derivatives produced, however, are separated chromatographically from the analyte of interest.

OTHER METHODS: Other methods for THPC in air are unknown. A microanalytical method for tetrakis(hydroxymethyl) phosphonium salts in solution involves ion chromatography, post-column reaction with an acetylacetone reagent to form a formaldehyde derivative, and detection at 425 nm [4].

REAGENTS:

1. Formaldehyde-2,4-dinitrophenylhydrazone, (formaldehyde-DNPH), 99% pure, Supelco, Bellefonte, PA, or Aldrich Chemical Co., Milwaukee, WI.
2. Tetrakis(hydroxymethyl)phosphonium chloride (THPC),* 80% aqueous solution. Aldrich Chemical Co., Milwaukee, WI.
NOTE: Ascertain the correct percentage in solution by performing an assay [potentiometric titration of 0.22 g of 80% THPC solution (diluted with DI water and acidified with 10 mL of 25% sulfuric acid) with 0.1 M AgNO₃ and silver electrode] [1,8]. Density of 80% ± 2% aqueous solution: 1.30 to 1.40 g/mL @ 20 °C.
3. Acetonitrile,* high purity solvent for HPLC analysis, low carbonyl content.
NOTE: Carbonyl content of acetonitrile can be determined by passing 10 mL of the solvent through a commercially available cartridge containing silica gel coated with acidified DNPH and analyzing for formaldehyde-DNPH by HPLC. The concentration of formaldehyde-DNPH should be below the LOD. [2]
4. Water, deionized (DI water).
5. Calibration stock solution. Accurately weigh 10 mg of formaldehyde-DNPH in a 10-mL volumetric flask. Add acetonitrile to the 10-mL mark.
6. Fortification stock solution, 10 mg/mL. Add 140 µL of 80% solution of THPC to 14.86 g of DI water.

* See SPECIAL PRECAUTIONS

EQUIPMENT:

1. Samplers, 37-mm, three-piece, plastic cassettes containing two glass fiber filters, a front filter and a backup filter, separated by 1 cm of space by means of a center cassette piece (spacer). Each filter is coated with 2 mg of acidified 2,4-dinitrophenylhydrazine (commercially available from SKC, Eighty Four, PA, as glutaraldehyde sampler, catalogue number 225-9003.*
2. Personal sampling pump, 1.0 to 1.7 L/min, with flexible connecting tubing.
3. Vials, 4-mL, glass, PTFE-lined rubber septa in caps for airtight seals.
4. Liquid chromatograph with UV detector, recorder, integrator, and column (p. 5046-1).
5. Syringes, 100-µL, 500-µL, and 10-mL.
6. Volumetric flasks, 25-mL, 10-mL.
7. Water bath, 60 °C ± 3 °C.
8. Syringe filters, PTFE membrane, 0.45-µm porosity.
9. Forceps.
10. Equipment for NMAM 2016, Issue 2, for formaldehyde gas [2].

SPECIAL PRECAUTIONS: On heating, THPC will decompose to form chlorine, ammonia, and oxides of phosphorus. Avoid high temperatures for THPC and contact with alkalis or strong oxidizing agents. THPC causes eye burns and moderate skin irritation. Dermal studies have shown that tetrakis(hydroxymethyl)phosphonium salts are promoters of skin cancer but not initiators. There was no evidence of carcinogenicity of THPC in rats and mice [4]. The liver is a target organ.

DNPH is a suspect carcinogen and is light sensitive [7]. Acetonitrile is toxic and is a fire hazard (flash point = 12.8 °C).

SAMPLING:

1. Calibrate each personal sampling pump with a representative sampler in line at the desired sampling rate.
2. Remove inlet cassette piece from sampler for open-faced sampling. Remove cap from outlet of sampler. Open-faced sampling is recommended because excessive quantities of THPC will exhaust the quantity of DNPH in a small area of the filter during closed-face sampling.
3. Attach sampler to the sampling pump with flexible tubing.
4. In a similar fashion, prepare DNPH-coated silica gel samples for air sampling of formaldehyde gas [2].
NOTE: A frit or filter can be added in the inlet to trap THPC aerosol and separate the THPC from the

formaldehyde gas. However, since the necessity of the frit or filter has not been determined, its use is optional.

5. Sample 1 to 480 liters of air at 1.0 to 1.7 L/min for THPC.
6. Conduct side-by-side air sampling for formaldehyde gas for the same period of time. Sample at 0.5 to 1.5 L/min for formaldehyde gas by NMAM 2016 [2].
7. Replace inlet cassette piece on the THPC sampler. Cap the outlet of the THPC sampler. Protect THPC samples from heat.
8. Place end caps onto the formaldehyde sampler and seal sampler in an envelope. Protect formaldehyde samples from heat.
9. Ship THPC and formaldehyde samples in separate containers on ice (0 °C).

SAMPLE PREPARATION:

10. Fold the front and back DNPH-coated filters using 2 pairs of forceps and place into separate 4-mL vials. Avoid finger contact.
11. Add 4.00 mL of carbonyl-free acetonitrile to each vial. Cap the vials tightly.
12. Place vials in water bath at 60 °C for 1 hour.
13. Remove vials from water bath and allow to cool to room temperature.
14. Filter the sample solutions with syringe filters. Collect filtrates in separate autosampler vials. Cap the autosampler vials.

NOTE: Follow NMAM 2016, Issue 2, for the analysis of formaldehyde gas samples [2].

CALIBRATION AND QUALITY CONTROL:

15. Calibrate daily with at least six working (media) standards over the range of interest.
 - a. Prepare a series of aqueous THPC solutions for fortification of blank DNPH-coated filters (media blanks) by dilution of THPC fortification stock solution (item 6) with DI water. Suggested concentrations of THPC in solution include 400 and 800 µg/mL. Media blanks and coated filters for actual air sampling should be from the same lot and should be handled and stored in the same manner.
 - b. Place folded filters in 4-mL vials and fortify with known quantities of THPC solution in the range of 20 to 80 µL.
 - c. Prepare the media standards (steps 11-14) and analyze (steps 17-19).
 - d. Prepare a calibration graph (peak area or height vs. µg THPC per sample).
16. Determine percent yield of formaldehyde from THPC in the calibration range (step 15) at least once.
 - a. Calculate the concentration of formaldehyde-DNPH derivative in the calibration stock solution (about 1000 µg/mL). Calculate the equivalent concentration of free formaldehyde in solution by multiplying the derivative concentration by 0.143 (molecular weight of formaldehyde divided by the molecular weight of formaldehyde-DNPH).
 - b. Prepare a series of dilutions [working standards (liquid standards)] from 0.02 to about 1 µg/mL.
 - c. Prepare six samples at the same level by fortifying six DNPH-coated filters with a known quantity of THPC (step 15).
 - d. Analyze samples (steps 17-19).
 - e. Calculate the molar quantity of formaldehyde found in each sample.
 - f. Calculate the molar quantity of THPC applied to each DNPH-coated filter.
 - g. Consider that one mole of THPC releases one mole of formaldehyde in the presence of DNPH. Calculate the percent yield of formaldehyde (yield should be 100%).

MEASUREMENT:

17. Set the liquid chromatograph according to manufacturer's recommendations and to conditions given on page 5046-1.
18. Inject a 20-µL aliquot of filtered sample solution.
19. Measure peak height or peak area. If sample peak is larger than the largest standard peak, dilute an aliquot of the remaining sample solution, reanalyze, and apply appropriate dilution factor in the calculations.
20. To ensure validity of the samples, identify those samples which contain more than 1000 µg of THPC per

sample. The capacity of the samplers may have been exceeded for these samples, and collection of smaller samples would be warranted.

CALCULATIONS:

21. Determine mass, μg , of THPC found on the front filter, W_F , on the backup filter, W_B , and on the average media blank, B , from the calibration graph.
22. Calculate concentration, C , of THPC in the air volume sample, V (L). This air concentration of THPC is uncorrected for formaldehyde gas present at the sampling site (if any).

$$C = \frac{W_F + W_B - 2B}{V}, \text{mg} / \text{m}^3$$

NOTE: $\mu\text{g}/\text{L} = \text{mg}/\text{m}^3$

23. Calculate the air concentration or average air concentration for formaldehyde gas, C_{form} , in mg/m^3 in side-by-side samples collected according to NMAM 2016 [2] (see steps 4 and 6 of NMAM 5046).
24. Calculate the corrected concentration, C' , of THPC (corrected for the concentration of formaldehyde gas in the air). The value of 6.35 is found by dividing the molecular weight of THPC by the molecular weight of formaldehyde.

$$C' = C - 6.35(C_{\text{form}}), \text{mg} / \text{m}^3$$

EVALUATION OF METHOD:

THPC aerosol can be determined by measuring the formaldehyde which is released from THPC in the presence of DNPH and heat. However, formaldehyde gas, if present in the air, will be an interference in the measurement of THPC. This method attempts to measure THPC aerosol by correcting for the presence of formaldehyde gas in the air. The samplers for formaldehyde gas used in NMAM 2016 [2] conceptually would have frits or filters in the inlets to trap THPC aerosol and allow formaldehyde gas to pass through to the beds of DNPH-coated silica gel. However, since the necessity of the frit or filter has not been determined, use of frits or filters is optional. The degree of interference from THPC aerosol in measuring formaldehyde gas by NMAM 2016 has not been determined. This method has not been tested by air sampling for THPC aerosol or by air sampling for formaldehyde gas in a THPC aerosol environment.

This method was evaluated using 37-mm DNPH-coated glass fiber filters in three-piece cassettes and aqueous tetrakis(hydroxymethyl)phosphonium chloride. Average recoveries of 8-, 14-, 20- and 31- μg quantities of THPC from 37-mm DNPH-coated glass fiber filters ranged from 87% to 104% (23 samples, pooled RSD = 5.85%) [1]. Samples of THPC on DNPH-coated filters were stable during storage at 5 °C in the dark; average recoveries of 15.0- μg quantities of THPC from DNPH-coated filters ranged from 108% to 131% after 7 to 28 days of storage at 5 °C ($n = 21$ samples, RSD = 2.7% to 5.4%) [1].

The limit of detection and lower limit of quantitation of THPC were 2 and 6.5 μg per filter, respectively. These limits were found by least squares calculations based on six DNPH-coated filters fortified with known quantities of THPC.

The possibility of migration of formaldehyde from the front filter to the backup filter during storage at 5 °C in sealed three-piece cassettes was investigated. Front filters in six three-piece cassettes were fortified with 31.4-µg quantities of THPC, and the cassettes were sealed. The six cassettes were stored for 10 days at 5 °C in the dark and then front and backup filters were analyzed. Average recoveries of THPC from front and backup filters were 102% and 11%, respectively (RSDs were 5.7% and 23%, respectively). The average total recovery of THPC from front and backup filters was 113% (RSD = 5.0%). These data provide evidence that (a) formaldehyde from THPC can indeed migrate from one DNPH-coated filter to another and (b) one needs to analyze both the front and backup DNPH-coated filters for THPC in case there is migration.

Yields of formaldehyde from THPC were determined in order to study the stoichiometry of the breakdown of THPC. DNPH-coated glass fiber filters in 4-mL vials were fortified with known quantities of THPC at four levels (n = 23 samples). The samples were treated with acetonitrile and heat by the usual procedure and analyzed with liquid standards (standard solutions of formaldehyde-DNPH in acetonitrile). Theoretical quantities of formaldehyde were calculated on the basis that one mole of THPC releases one mole of formaldehyde in the presence of DNPH and heat. The average of the four average yields of formaldehyde was 100.3%.

The upper limit for collection of THPC on a filter is calculated at 1 mg. Since at 100% yield one mole of THPC releases one mole of formaldehyde in the presence of DNPH and heat, 1 mg of THPC would release 158 µg of formaldehyde, a quantity which theoretically would react with 1.04 mg of DNPH. One DNPH-coated filter contains 2 mg of DNPH, a quantity which allows for the presence of excess DNPH when 1 mg of THPC is collected.

REFERENCES:

- [1] Tucker SP, Striley CA [2002]. Tetrakis(hydroxymethyl)phosphonium Chloride Backup Data Report for Method 5046. National Institute for Occupational Safety and Health, DART, Cincinnati, OH. Unpublished report for evaluation of method.
- [2] NIOSH [1994]. Formaldehyde by HPLC, method 2016, issue 2, NIOSH Manual of Analytical Methods, 4th ed., Eller PM and Cassinelli ME (eds.), U.S. Department of Health and Human Services, Public Health Service, Centers for Disease Control and Prevention, National Institute for Occupational Safety and Health, Cincinnati, OH. DHHS (NIOSH) Publication No. 94-113.
- [3] Kleindienst TE, Corse EW, and Blanchard FT [1998]. Evaluation of the Performance of DNPH-Coated Silica Gel and C₁₈ Cartridges in the Measurement of Formaldehyde in the Presence and Absence of Ozone. *Environmental Science & Technology*, 32(1), 124-130.
- [4] World Health Organization [2000]. Environmental Health Criteria 218, International Programme on Chemical Safety, International Labour Organization, Geneva, <http://www.inchem.org/documents/ehc/ehc/ehc218.html>.
- [5] Elizy SE Jr. and Connick WJ Jr. [1973]. Gasometric Analysis of THPC., *American Dyestuff Reporter*, 62 (6), pp. 47 and 50.
- [6] Loewengart G and Van Duuren BL [1977]. Evaluation of Chemical Flame Retardants for Carcinogenic Potential, *Journal of Toxicology and Environmental Health*, 2, 539-546.
- [7] Otson R and Fellin P [1988]. A Review of Techniques for Measurement of Airborne Aldehydes. *Science of the Total Environment*, 77 (2/3), 95-131.
- [8] Aldrich Chemical Co. [2000]. Potentiometric titration of tetrakis(hydroxymethyl)phosphonium chloride with silver nitrate and silver electrode, product 40,486-1, Lot 10715B1, Technical Service Department, Aldrich Chemical Co., Milwaukee, WI (August 18, 2000). Unpublished procedure by Fluka in Backup Data Report [1].

METHOD DEVELOPED BY:

Samuel P. Tucker, Ph.D., NIOSH/DART