4-NITROBIPHENYL

Measurements Research Branch

Analytical Method

Analyte:	4-Nitrobiphenyl	Method No:	P&CAM 273
Matrix:	Air	Range:	4-2200 µg/m³ in a 50-liter air sample
Procedure:	Collection on glass fiber filter and silica gel, elution with 2-propanol, analysis by gas chromatography	Precision:	8%
Date Issued:	8/1/78	Classification:	E (Proposed)
Date Revised:			

1. Synopsis

   1.1 A known volume of air is drawn through a three-stage sampler consisting of a high-efficiency glass fiber filter followed by two beds of silica gel to collect the 4-nitrobiphenyl aerosol and/or vapor.

   1.2 The filter and sorbent sections of the sampler are transferred to stoppered tubes and the 4-nitrobiphenyl is desorbed with 2-propanol.

   1.3 An aliquot of this solution is injected into a gas chromatograph.

   1.4 Peak areas are electronically determined and compared with a calibration curve obtained from injections of standard solutions of 4-nitrobiphenyl.

2. Range, Sensitivity and Detection Limit

   2.1 The analytical range is 0.2-110 µg/sample (4-2200 µg/m³ for a 50L air sample) when using 0.5 mL of desorbing solution and 1-µL injections into the gas chromatograph.

   2.2 The amount of 4-nitrobiphenyl vapor retained on the primary silica gel bed decreases as the relative humidity of the sampled air increases. In experiments in which atmospheres containing 700 µg/m³

of 4-nitrobiphenyl in air of 80% relative humidity and at 34 °C were sampled, less than 2% of the analyte appeared in the secondary silica bed when 45 L was sampled. At 50% relative humidity and 34 °C, 200 L of air containing 600 μg/m^3 was sampled and less than 0.3% was detected in the secondary silica bed.

2.3 The limit of detection is 0.05 μg/sample.

3. Interferences

3.1 In solutions containing 4-nitrobiphenyl at 1-2 ng/μL, the following compounds were found not to interfere when their concentrations were 1-3 ng/μL: 1-naphthol, 2-naphthol, 1-naphthylamine, 2-naphthylamine, 2-nitronaphthalene, 2-nitrobiphenyl, 3-nitrobiphenyl, and 3-aminobiphenyl.

3.2 Any compound sampled with 4-nitrobiphenyl and having the same retention time as the analyte will interfere with the analysis. This type of interference often can be eliminated by changing the operating conditions of the chromatograph, e.g., by temperature programming or by changing the stationary phase.

4. Precision and Accuracy

4.1 The precision of the overall method is 8% relative standard deviation for samples collected at 34 °C and 80% relative humidity. This value is based on 43 samples involving volumes of 4 to 12 liters and a sampling rate of 0.2 L/min. Concentrations sampled ranged from 120 to 600 μg/m^3.

4.2 Filters and silica gel spiked with 4-nitrobiphenyl and stored at ∿24 °C yielded recoveries $\geq$93% when desorbed within 28 days.

4.3 Analysis of the back-up silica gel section serves as a check on sample integrity. If more than 10% of the total recovered weight is found on this section, sample loss due to breakthrough may be indicated. Under these circumstances, the actual ambient concentration may be greater than the sample analysis indicates.

4.4 The accuracy of the method has not been determined.

5. Advantages and Disadvantages

5.1 The sampler is a small portable device containing no liquids. This is an advantage for sampling air in a worker's breathing zone without interfering with normal work activities.

5.2 Relative humidity has an effect on the collection efficiency of the absorbent. (Section 2.2)

5.3 At a flow rate of 0.2 L/min, the linear velocity through the 4-mm inlet of the holder is 26.5 cm/s. It is not known if this throat veolicty is sufficient for the capture of all particles of analytical importance.

5.4 This method has not been field tested.

6. Apparatus

6.1 Personal air sampling pump that can be maintained for four hours at a flow rate of 0.2 L/min with the sampler in line. Each pump should be calibrated with a representative sampler in line to minimize errors in volume measurement. A bubble flow meter or other suitable flow measuring device may be used.

6.2 Sampler, equivalent to that shown in Figure 1. The sampler consists of three sections: a high-efficiency glass fiber filter, followed by 100-mg and 50-mg beds of silica gel. The first section, a 13-mm glass fiber filter (Spectrograde or Type A-E, Gelman Instrument Co., Ann Arbor, MI, or equivalent) is contained in a 13-mm filter holder (SX00-013-00 Millipore Corp., Bedford, MA, or equivalent). The sorbent tube is a 50-mm section of Pyrex glass, 6.4-mm o.d. x 4-mm i.d. flared on one end, containing the sections of silica gel (Grade 407, 20/45-mesh, 720-760 m^2/g, 0.737 g/cc; Coast Engineering Laboratories, Gardena, CA, or equivalent). The sorbent is held by 3.5-mm diameter, 100-mesh, stainless steel screens and 4-mm o.d. Teflon rings, as shown in Figure 1. To connect the two stages, the filter holder is pressed into the flared end of the sorbent tube. The pressure drop across the sampler at 0.2 L/min is 10 torr. Flexible vinyl caps are used to seal the ends of the sampler.

6.3 Gas chromatograph equipped with a flame ionization detector (FID) and injection port.

6.4 Glass column, 1.8-m x 2-mm i.d. x 6.4-mm o.d., packed with 3% OV-225 on 80/100 mesh Supelcoport (Supelco, Inc., Bellefonte, PA, or equivalent).

6.5 Potentiometric strip chart recorder.

6.6 Test tubes, 1-mL, fitted with polyethylene stoppers.

6.7 Glass syringes, 5- and 10-µL.

6.8 Pipettes of convenient volumes for the preparation of standard solutions.

6.9 Volumetric flasks of convenient volumes for the preparation of standard solutions.

6.10 Clinical centrifuge.

6.11 Analytical balance.

6.12 Test tube shaker, vortex type.

7. Reagents

Whenever possible, reagents used should be ACS reagent grade or better.

7.1 2-Propanol, UV grade, distilled in glass.

7.2 Acetone, distilled in glass.

7.3 Methyl alcohol.

7.4 Carbon tetrachloride.

7.5 4-Nitrobiphenyl.

8. Procedure

8.1 Cleaning of Equipment. All glassware used for the laboratory analysis is washed with detergent and rinsed with tap water, distilled water, and methyl alcohol.

8.2 Collection and Shipping of Samples

8.2.1 Immediately before sampling, the vinyl caps are removed from the ends of the sampler and saved for resealing after sampling.

8.2.2 The sorbent end of the sampler is connected to the pump with plastic or rubber tubing. The sampler is positioned vertically during sampling. Sampled air must not pass through any tubing before entering the sampler.

8.2.3 The atmosphere is sampled at a flow rate of 0.2 L/min for no longer than 4 hours. The flow rate and sampling time, or the volume of sampled air, must be measured as accurately as possible.

8.2.4 The temperature, pressure, and humidity of the atmosphere being sampled are measured and recorded, along with other pertinent data such as location, date, and time of sampling.

8.2.5 Blank samples are obtained by handling random collection devices in the same manner as in sampling, except that no air is drawn through them.

8.2.6 Samples shipped to the laboratory should be packed tightly to minimize breakage.

8.2.7 If samples of bulk material associated with the process under investigation are to be shipped to the laboratory, they should not be placed in the same container as the air samplers or blanks.

8.3 Analysis of Samples

8.3.1 Place the glass fiber filter and the 100-mg section of silica gel in a 1-mL tube or other suitable vial. Place the 50-mg section of silica gel in a separate test tube. Teflon rings and stainless steel screens associated with the sampler may be analyzed with the appropriate stages. Sampler caps can be discarded.

8.3.2 Add 0.5 mL of 2-propanol to each vial and cap it.

8.3.3 Allow the vials to stand for one hour, shaking them intermittently in a test tube vibrator. Centrifuge for 10 minutes.

8.3.4 Gas Chromatographic Conditions

Column:	3% OV-225 on 80/100 mesh Supelcoport in 1.8-m x 2-mm i.d. x 6.4-mm o.d. glass tubing.
Column Temperature:	190 °C
Injector Temperature:	210 °C
Detector Temperature:	200 °C
Helium:	22 mL/min
Hydrogen:	85 mL/min
Air:	378 mL/min
Detector:	FID; Attenuation 1 x 2 at low concentrations
Injection Volume:	1.0 µL
Efficiency:	2500 theoretical plates
Capacity Ratio:	15.9

8.3.5 Injection. Flush the 5-µL syringe first with acetone (to insure its cleanliness and to wet the barrel and plunger) and then with the sample solution. Carefully draw 1 µL of sample solution into the syringe (to minimize the intake of particulate matter) and inject the aliquot into the gas chromatograph.

8.4 Efficiency of Desorption

8.4.1 Importance of Determination. The efficiency of desorption for 4-nitrobiphenyl can vary from one laboratory to another and also from one batch of silica gel or filters to another. Thus, it is necessary to determine the fraction of 4-nitrobiphenyl that can be recovered from these matrices. Since the efficiency of desorption may also vary with the amount of 4-nitrobiphenyl present, determinations should be made for at least two levels in the range of anticipated sample sizes. If the efficiencies of desorption for the filter and primary silica gel bed do not vary by more than 5%, these two primary stages may be analyzed together. If greater variances are found, it may be necessary to analyze the stages individually and make appropriate corrections (Section 10.3).

8.4.2 Procedure for Determining the Efficiency of Desorption. Place the filters and silica gel from unused samplers in 1-mL test tubes. Add to each a known amount of 4-nitrobiphenyl in 10 μL of carbon tetrachloride using the same microliter syringe for each. Five filter and silica gel samples are thus prepared at each of two different levels in the range of interest. Cap the tubes. Treat a parallel blank in the same manner (no analyte is added). Prepare at least three standards at each of the various levels by adding 10 μL (same syringe as above) of the 4-nitrobiphenyl solutions to 0.5 mL of desorbing solution. Analyze the samples and standards as described in Section 8.3. The efficiency of desorption is the average gas chromatographic peak area given by the samples, corrected for the blank if necessary, divided by the average area given by the standards.

9. Calibration and Standardization

9.1 For accuracy in the preparation of standards, it is recommended that one standard be prepared in a relatively large volume and at a high concentration. The initial standard is prepared by weighing a selected amount of 4-nitrobiphenyl (e.g., 125 mg) into a 250-mL volumetric flask and adding the desorbing solution to the calibration mark. Dilutions encompassing the concentration range of interest down to 0.3 ng/μL are then made from the stock solution. Solutions should be stored in a refrigerator when not in use.

CAUTION: 4-NITROBIPHENYL HAS BEEN IDENTIFIED AS A CARCINOGEN. APPROPRIATE PRECAUTIONS MUST BE TAKEN IN HANDLING THE COMPOUND TO AVOID PERSONNEL EXPOSURE AND AREA CONTAMINATION.

9.2 The standard solutions should be analyzed under the same gas chromatographic conditions and during the same time period as the samples. This will minimize inaccuracy due to day-to-day variations in the setting of instrumental conditions.

9.3 Calibration curves for 4-nitrobiphenyl are prepared by plotting the peak areas for the standards against their concentration in µg/0.5 mL.

10. Calculations

10.1 In the event that the blank produces a peak with the same retention time as the analyte, the analyst should determine the source of the interference and eliminate or compensate for it.

10.2 Read the weight (W_1) in µg of 4-nitrobiphenyl present in the sample from the calibration curve.

10.3 The corrected weight (W_2) is determined by

$$W_2 = \frac{W_1}{D}$$

where D is the efficiency of desorption for that sampler stage.

10.4 Add the weights found in the sampler stages to determine the total weight (W_t) collected.

10.5 The concentration of 4-nitrobiphenyl in air (C, µg/m^3) is given by

$$C = \frac{W_t}{V} \times 10^3$$

where V is the volume of air sampled in liters and 10^3 is the factor to convert from liters to m^3.

11. Reference

Raul Morales, J. F. Stampfer, Jr., R. E. Hermes, E. E. Campbell, and H. J. Ettinger, "Development of Sampling and Analytical Methods for Carcinogens, October 1, 1976-December 31, 1977," Progress Report LA-7375-PR, National Technical Information Service, Springfield, Virginia, 1978.

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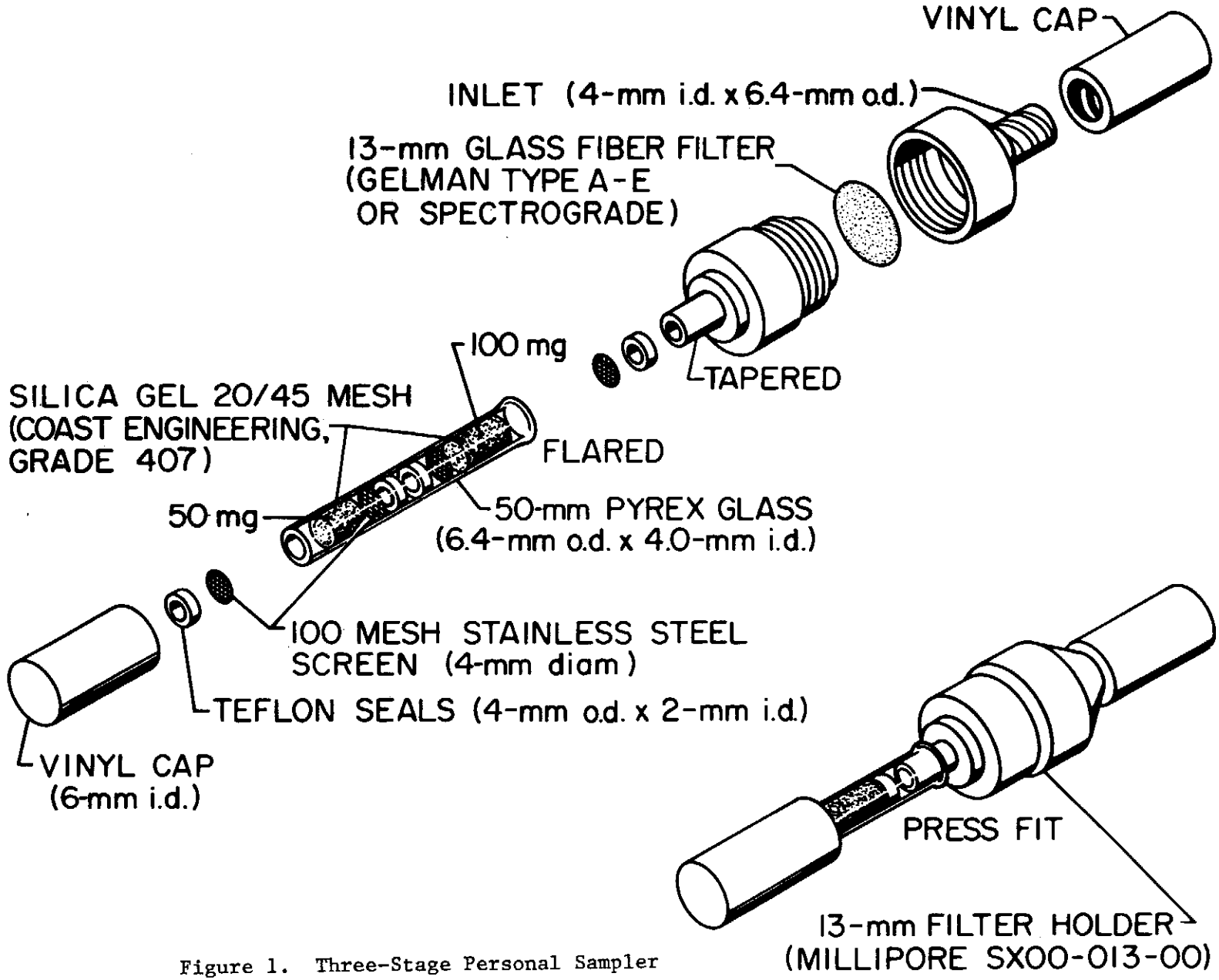

Figure 1. Three-Stage Personal Sampler