

Picric acid

Analyte:	Picric acid	Method No:	S228
Matrix:	Air	Range:	0.036 - 0.189 mg/cu m
OSHA Standard:	0.1 mg/cu m	Precision (\overline{CV}_T):	0.082
Procedure:	Filter collection, extraction with 70% aqueous methanol, HPLC	Validation Date:	11/25/77

1. Principle of the Method

- 1.1 A known volume of air is drawn through a mixed cellulose ester membrane filter to trap the picric acid aerosol present. This method is not applicable for sampling environments where significant picric acid vapor may be present.
- 1.2 The filter is transferred to a jar and extracted with 70% aqueous methanol.
- 1.3 An aliquot of the sample is injected into a high performance liquid chromatograph (HPLC) equipped with a variable wavelength UV detector set at 360 nm.
- 1.4 The area of the resulting sample peak is used as a measure of analyte concentration by comparison with corresponding areas obtained from the injection of standards.

2. Range and Sensitivity

- 2.1 This method was validated over the range of 0.036 - 0.189 mg/cu m at an atmospheric temperature and pressure of 22°C and 772 mm Hg, using a 180-liter sample.
- 2.2 The method may be extended to higher values by further dilution of the sample solution. The detection limit of the analytical method is estimated to be at least 10 ng per ml.

3. Interferences

- 3.1 When two or more compounds are known or suspected to be present in the air, such information, including their suspected identities, should be transmitted with the sample.
- 3.2 It must be emphasized that any other compound which has the same retention time as the analyte at the operating conditions

described in this method is an interference. Retention time data based on a single set of conditions cannot be considered as proof of chemical identity.

4. Precision and Accuracy

4.1 The Coefficient of Variation (\overline{CV}_T) for the total analytical and sampling method in the range of 0.036 - 0.189 mg/cu m was 0.082. This value corresponds to a 0.008 mg/cu m standard deviation at the OSHA standard level. Statistical information can be found in Reference 11.1. Details of the test procedure are found in Reference 11.2.

4.2 A collection efficiency of at least 99% was determined for the collection medium; thus, no significant bias was introduced in the sample collection step. There was also no bias in the analytical method--the average recovery from the filters was 99.3%. In addition, the samples were found to be stable when stored for seven days. Thus, \overline{CV}_T is a satisfactory measure of both accuracy and precision of the sampling and analytical method.

5. Advantages and Disadvantages of the Method

5.1 The sampling device is small, portable and involves no liquids. Interferences are minimal, and most of those which do occur can be eliminated by altering chromatographic conditions. The filters are analyzed by means of a quick, instrumental method.

5.2 This sampling method is applicable for particulate picric acid only; in operations where significant vapor may also be present, this method will not apply.

6. Apparatus

6.1 Sampling Equipment. The sampling unit for the collection of personal air samples for the determination of organic aerosol has the following components:

6.1.1 Filter. The filter unit consists of a mixed cellulose ester membrane filter, 0.8 micrometer pore size and 37-mm diameter, supported by a cellulose backup pad, and a 37-mm, three-piece filter holder held together by tape or a shrinkable band.

6.1.2 Personal Sampling Pump. A calibrated personal sampling pump whose flow can be determined to an accuracy of $\pm 5\%$ at the recommended flow rate is needed. The pump must be calibrated with a representative filter holder and filter in the line.

- 6.1.3 Thermometer.
- 6.1.4 Barometer.
- 6.1.5 Stopwatch.
- 6.2 High pressure liquid chromatograph equipped with a detector capable of UV detection at 360 nm.
- 6.3 Column (30-cm x 3.9-mm I.D. stainless steel) packed with μ Bondapak C₁₈ or equivalent.
- 6.4 Syringe. Twenty- μ l, for HPLC injection.
- 6.5 An electronic integrator or some other suitable method for measuring peak areas.
- 6.6 Microliter syringes. One hundred-microliter and other convenient sizes for making standard solutions.
- 6.7 Ointment jars. Use squat form with Teflon film gaskets and screw cap.
- 6.8 Volumetric flasks. Twenty-five milliliter and other convenient sizes for making standard solutions.

7. Reagents

- 7.1 Picric acid, reagent grade.
- 7.2 Distilled water.
- 7.3 Methanol in distilled water, 70%. Prepare by diluting 700 ml of methanol to 1000 ml with distilled water. This solution is used for sample extraction and also as the mobile phase for the HPLC analysis, but should be degassed prior to such use.
- 7.4 Picric acid stock solution, 1.8 mg/ml. Dissolve 0.18g of picric acid in 100 ml of 70% methanol.

8. Procedure

- 8.1 Cleaning of Equipment. All glassware used for the laboratory analysis should be detergent washed and thoroughly rinsed with tap water and distilled water.
- 8.2 Calibration of Personal Pumps. Each personal pump must be calibrated with a representative filter holder in the line. This will minimize errors associated with uncertainties in the sample volume collected.

8.3 Collection and Shipping of Samples

- 8.3.1 Assemble the filter in the three-piece filter holder and close firmly to insure that the center ring seals the edge of the filter. The cellulose membrane filter is held in place by a cellulose backup pad and the filter holder is held together by plastic tape or a shrinkable cellulose band. If the middle piece of the filter holder does not fit snugly into the bottom piece of the filter holder, sample leakage will occur around the filter. A piece of flexible tubing is used to connect the filter holder to the pump.
- 8.3.2 Remove the filter holder plugs and attach to the personal sampling pump tubing. Clip the filter holder to the worker's lapel.
- 8.3.3 Air being sampled should not be passed through any hose or tubing before entering the filter holder.
- 8.3.4 A sample size of 180 liters is recommended. Sample at a flow rate of 1.5 liters per minute. The flow rate should be known with an accuracy of at least $\pm 5\%$.
- 8.3.5 Turn the pump on and begin collection. Set the flow rate as accurately as possible using the manufacturer's directions. Since it is possible for filters to become plugged by heavy particulate loading or by the presence of oil mists or other liquids in the air, the pump rotameter should be checked frequently and readjusted as needed. If the rotameter cannot be readjusted, terminate sampling.
- 8.3.6 Terminate sampling at the predetermined time and note sample flow rate, collection time and ambient temperature and pressure. If pressure reading is not available, record the elevation.
- 8.3.7 After sampling, holders should be firmly sealed with filter holder plugs in both the inlet and outlet.
- 8.3.8 Carefully record sample identity and all relevant sample data.
- 8.3.9 With each batch of samples, submit one filter which is subjected to exactly the same handling as the samples except that no air is drawn through it. Label this as a blank. Submit one blank for every ten samples.
- 8.3.10 The filter holders should be shipped in a suitable container designed to prevent damage in transit.

8.3.11 A bulk sample of the suspected material should be submitted to the laboratory in a glass container lined with a Teflon cap. Label of the bulk sample should match air samples for identification purposes.

8.4 Analysis of Samples

8.4.1 Preparation of Samples

1. Open the filter holder. Carefully remove the cellulose membrane filter from the holder with the aid of appropriate tweezers and transfer filter to the 2-oz. ointment jar.
2. Add 5 ml of 70% methanol to the jar and properly cap unit. Gently swirl the jar to ensure that the filter is thoroughly wetted.

8.4.2 Analysis by high pressure liquid chromatography. The mobile phase is 70% aqueous methanol. The typical operating conditions for the liquid chromatograph are:

1. 1.0 ml/min solvent flow rate
2. Ambient column temperature
3. 2250 psi system pressure
4. 360 nm UV detection wavelength
5. Capacity ratio: 2.2

8.4.3 Injection. The first step in the analysis is the injection of the sample into the liquid chromatograph. A 20 μ l-sample aliquot is recommended for this analysis. The sample may be injected either by using an appropriate syringe or by filling a fixed volume sample loop provided that reproducibility requirements are satisfied. Duplicate injections of each sample and standard should be made. No more than a 3% difference in area is to be expected.

8.4.4 Measurement of Area. The area of the sample peak is measured by an electronic integrator or some other suitable form of area measurement, and results are read from a standard curve prepared as discussed in Section 9.

8.5 Determination of Analytical Method Recovery

8.5.1 Need for Determination. To eliminate any bias in the analytical method, it is necessary to determine the

recovery of the compound. The sample recovery should be determined in duplicate and should cover the concentration range of interest. If the recovery is less than 95%, the appropriate correction factor should be used to calculate the "true" value.

- 8.5.2 Procedure for Determining Recovery. A known amount of the analyte, preferably equivalent to the sample concentration expected, is added to a representative cellulose membrane filter and air-dried. The analyte is then extracted from the filter with 5 ml of 70% methanol and analyzed as described in Section 8.4.

For the validation studies conducted to determine the precision and accuracy of this method, an amount of the analyte equivalent to that present in a 180-liter sample at the selected level was used to determine the analytical method recovery. A stock solution containing 0.975 milligrams of picric acid per milliliter of 70% aqueous methanol was prepared. Ten, 20 and 40-microliter aliquots of the solution were added to the cellulose membrane filters and air-dried to produce samples equivalent to 180-liter collections at 0.5, 1 and 2X the OSHA standard level. The analytical samples were allowed to stand overnight. A parallel blank filter was also prepared except that no sample was added to it. All filters were then extracted and analyzed as described in Section 8.4.

The sample recovery equals the average weight in μg recovered from the filter divided by the weight in μg added to the filter, or

$$\text{Recovery} = \frac{\text{Average Weight } (\mu\text{g}) \text{ recovered} - \text{Blank } (\mu\text{g})}{\text{Weight } (\mu\text{g}) \text{ added}}$$

The recovery value is used in Section 10.3 if the recovery is less than 95%.

9. Calibration and Standards

- 9.1 From the stock standard solution, prepare at least 6 working standards to cover the concentration range of 9-36 $\mu\text{g}/5 \text{ ml}$. Transfer 25 to 100 μl -aliquots of the stock standard into 25-ml volumetric flasks and dilute to volume with 70% methanol.
- 9.2 This series of standards is analyzed under the same HPLC conditions and during the same time period as the unknown samples. It is convenient to express concentration of standards in $\mu\text{g}/5 \text{ ml}$ 70% methanol, because samples are extracted in this amount of 70% methanol. Curves are established by plotting concentrations in micrograms per 5.0 ml versus peak area.

NOTE: To minimize effect of variations in LC conditions and detector response due to sample cell conditions, frequent standardization should be practiced.

10. Calculations

10.1 Read the concentration, in $\mu\text{g}/5\text{ ml}$, corresponding to the peak area from the standard curve. No volume corrections for sample aliquots analyzed are needed, because the standard curve is based on μg per 5.0 ml and the volume of sample injected is identical to the volume of the standards injected.

10.2 Corrections for the blank must be made for each sample.

$$\mu\text{g} = \mu\text{g sample} - \mu\text{g blank}$$

where:

$$\mu\text{g sample} = \mu\text{g found in sample filter}$$

$$\mu\text{g blank} = \mu\text{g found in blank filter}$$

10.3 Divide the total weight by the recovery (Section 8.5.2) to obtain the corrected $\mu\text{g}/\text{sample}$.

$$\text{Corrected } \mu\text{g}/\text{sample} = \frac{\text{Total Weight}}{\text{Recovery}}$$

10.4 For personal sampling pumps with rotameters only, the following correction should be made.

$$\text{Corrected Volume} = f \times t \left(\sqrt{\frac{P_1}{P_2} \times \frac{T_2}{T_1}} \right)$$

where:

f = sampling flow rate

t = sampling time

P_1 = pressure during calibration of sampling pump (mm Hg)

P_2 = pressure of air sampled (mm Hg)

T_1 = temperature during calibration of sampling pump ($^{\circ}\text{K}$)

T_2 = temperature of air sampled ($^{\circ}\text{K}$)

10.5 The concentration of the analyte in the air sampled can be expressed in mg per cu m (μg per liter = mg per cu m).

$$\text{mg}/\text{cu m} = \frac{\text{Corrected } \mu\text{g (Section 10.3)}}{\text{Volume of Air Sampled in Liters}}$$

11. References

- 11.1 Documentation of NIOSH Validation Tests, National Institute for Occupational Safety and Health, Cincinnati, Ohio (DHEW-NIOSH-Publication No. 77-185), 1977. Available from Superintendent of Documents, Washington, D. C., Order No. 017-033-00231-2.
- 11.2 S228 Backup Data Report for Picric acid, prepared under NIOSH Contract No. 210-76-0123, November 25, 1977.