

Indian Standard

METHODS FOR MEASUREMENT OF AIR POLLUTION

PART XII POLYNUCLEAR AROMATIC HYDROCARBONS IN AIR PARTICULATE MATTER

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PART XII POLYNUCLEAR AROMATIC HYDROCARBONS IN AIR PARTICULATE MATTER

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0. F O R E W O R D

0.1 This Indian Standard (Part XII) was adopted by the Indian Standards Institution on 26 September 1974, after the draft finalized by the Air Pollution Sectional Committee had been approved by the Chemical Division Council.

0.2 Polynuclear hydrocarbons are commonly regarded with extreme suspicion as possible carcinogenus. In air, these may exist adsorbed on carrier particles or in free state.

0.3 This method is based on ASTM D 2682-72 'Method of test for polynuclear aromatic hydrocarbons in air particulate matter', published by American Society for Testing and Materials.

0.4 In reporting the result of a test or analysis made in accordance with this standard, if the final value, observed or calculated, is to be rounded off, it shall be done in accordance with IS:2-1960*.

1. SCOPE

1.1 This standard (Part XII) prescribes a method which covers the determination of polynuclear aromatic hydrocarbons in air particulate matter.

2. PRINCIPLE OF THE METHOD

2.1 The polynuclear aromatic hydrocarbon content of air particulate matter is determined by a lengthy process involving air sampling, Soxhlet extraction, column chromatography, and ultraviolet-visible spectrophotometry.

2.2 Air particulate samples are collected on glass-fibre filters using a high-volume air sampler. The samples are then extracted with benzene in a

*Rules for rounding off numerical values (*revised*).

Soxhlet extractor to obtain a benzene-soluble fraction of air particulate matter which is normally referred to as the 'organic fraction'. This organic fraction is then further separated into approximately 30 to 40 subfractions by means of column chromatography on alumina using increasing amounts of diethyl ether in pentane as the eluent. Each of the subfractions is dried, dissolved in pentane, and spectrophotometrically analyzed for polynuclear aromatic hydrocarbon content. The polynuclear aromatic hydrocarbons that can be effectively separated and analyzed by this technique, if they are present in a particular air sample, are anthracene, phenanthrene, fluoranthene, pyrene, benz (a) anthracene, chrysene, benzo(a)pyrene, benzo(e)pyrene, perylene, benzo (g, h, i) perylene, anthanthrene, and coronene.

2.3 This method will easily determine microgram quantities of the various hydrocarbons per 1 000 m³ of air. The exact sensitivity depends on the volume of air sampled and the type of pollution. If hydrocarbons are present in microgram quantities, they can be determined. The range of the method can be easily extended by simple dilution of the fraction.

3. INTERFERENCES

3.1 Sometimes the impurities cannot be removed by simple recrystallization, and other techniques such as chromatography or destruction of the impurity by reaction may be necessary. These problems in purification will have to be individually coped with as they arise.

3.2 Any material eluted from the column along with a particular hydrocarbon that exhibits a spectral band at the wavelength at which the hydrocarbon is measured will probably mask the peak to be measured and of course be an interference. So far this type of interference has been found to be minimum.

4. APPARATUS

4.1 Chromatographic Columns—All-glass columns with an internal diameter of 13 mm and an overall height of approximately 380 mm are satisfactory. They shall be equipped at the top with a glass joint to accommodate a solvent reservoir and on the bottom with a drip tip.

4.2 Fraction Collector—A fraction collector capable of collecting 15 to 25-ml fractions in test tubes. A time flow collector is recommended rather than a drop counter collector because most drop counters employ a photoelectric cell in counting. The light intensity of the photoelectric cell may induce changes in the polynuclear hydrocarbons that are light-sensitive.

4.3 Vacuum Oven—A vacuum oven or other device capable of evaporating the pentane-ether subfractions in the dark at room temperature can be used.

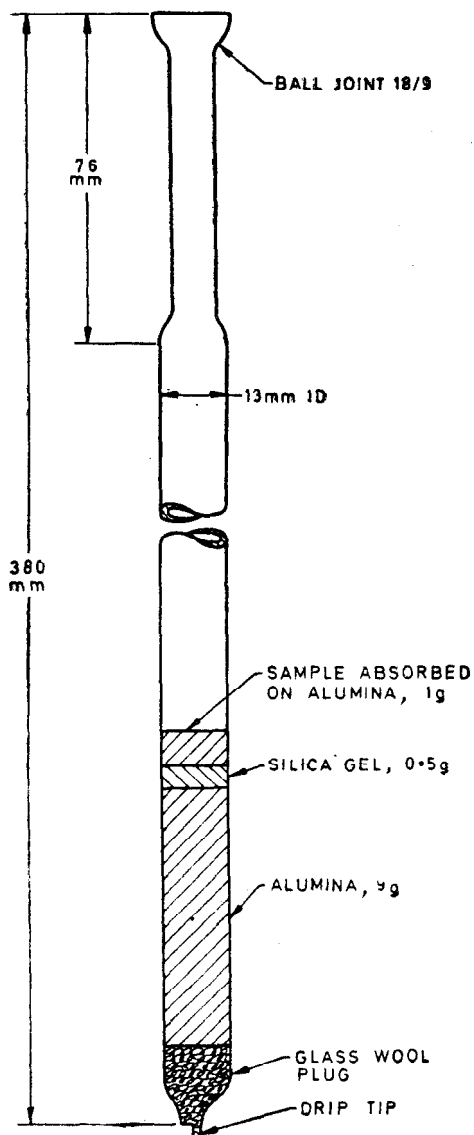


FIG. 1 CHROMATOGRAPHIC COLUMN USED FOR SEPARATION

4.4 Spectrophotometer — A ratio-recording spectrophotometer capable of measuring absorbance between 220 and 450 nm is suitable for analysis. A manual spectrophotometer is highly impractical because of the many absorbance maxima and minima measured for each subfraction.

4.5 Spectrophotometric Cells for Analysis — Cells having a 10-mm light path and minimum transmittance of 80 percent at 200 nm are used for spectrophotometric analysis. For convenience, the cells are also used as miniature volumetric flasks and shall be calibrated to contain a volume of 3 ml. The 10-mm cylindrical cells contain 3 ml when filled just to the bottom of the glass joint.

5. REAGENTS

5.1 Alumina — acid-washed, suitable for chromatography. If a great number of samples are to be analyzed, a large batch of alumina can be prepared since the water content of the prepared alumina will remain relatively stable for at least 6 months if stored in a tightly stoppered glass container. Place the quantity of alumina desired in a beaker, cover with diethyl ether and stir for 5 minutes. Allow the alumina to settle, decant the supernatant ether, and filter by vacuum using a Büchner funnel. Repeat the process four times on the settled alumina and after the final washing transfer the entire quantity of alumina to the funnel using diethyl ether as a wash. Leave the alumina drying on the funnel until there is no noticeable odour of ether in the alumina. Transfer the alumina to an evaporating dish and further dry in an oven at 130°C for 30 hours. The percentage of water (approximately 11.5 to 12 percent) remaining in the alumina is determined by heating an accurately weighed sample (1 to 2 g) in a crucible at red heat (800°C) for 10 minutes, cooling in a desiccator over phosphorus pentoxide or anhydrous, and reweighing. Adjust the water content of the large batch of alumina to 13.7 percent by adding the required amount of water directly to the alumina. Mix the adjusted alumina well by shaking and allow to stand for 12 hours in a stoppered container to attain equilibrium. Repeat the determination of water in the alumina as described and readjust the water again, if necessary.

5.2 Benzene — The benzene shall be redistilled before use in an all-glass apparatus. The fraction boiling at 80.1°C shall be used for extraction.

5.3 Chloroform

5.4 Ether — The diethyl ether shall be anhydrous. Sodium-lead has proved effective in drying ether.

5.5 Pentane — The pentane shall be redistilled before use in all-glass apparatus. The fraction boiling at 35 to 36°C shall be used for analysis.

5.6 Solvent — All of the hydrocarbons can be recrystallized from either the nonpolar solvents such as heptane or ethylcyclohexane; or from the polar solvents such as methanol and methanol containing increasing amounts of methyl cellosolve; or water depending upon the melting point and molecular mass of the hydrocarbon. If the impurity is polar in nature, a nonpolar solvent shall be selected and vice versa.

5.7 Silica Gel (90 to 150 microns) — The silica gel shall be thoroughly washed with diethyl ether and dried in the same manner as described for the alumina (*see* 5.1) and heated at 130°C for 1 hour. Silica gel shall be stored in tightly stoppered glass bottles.

6. SAMPLING

6.1 Air particulate matter is usually collected on flash-fired glass-fibre filter paper using a high-volume air sampler although other methods of collecting particulate matter may also be employed. The amount of particulate matter collected by a high-volume sampler depends upon many variables (for example, the particulate loading in the air, sampler location, volume and rate of air sampled, etc.). On the average, when a high-volume sampler is located in an urban area, the sampler will collect approximately 250 to 350 mg of particulate matter while sampling 2 000 to 2 400 m³ of air during a 24 hour period. Of the 250 to 350 mg of particulate matter approximately 10 percent will be soluble in benzene. Hence, the benzene-soluble portion of air particulate matter (hereinafter referred to as the organic fraction) for an average high-volume sample will amount to approximately 25 mg. A 50 to 150-mg quantity of the organic fraction is needed for the analysis of polynuclear aromatic hydrocarbons in air particulate matter. A typical sample shall weigh 100 mg. Therefore, to obtain this needed amount of organic fraction it will probably become necessary to pool together organic fractions of several individual high-volume air samples.

6.2 To obtain the organic fraction, the collected particulate matter is extracted with benzene using a Soxhlet extractor for approximately 5 to 6 hours. The benzene extract is filtered through a sintered glass funnel to remove any glass fibre particles and dried in an oven at 60°C.

7. CALIBRATION

7.1 In order to calibrate the analytical procedure, it is essential to become familiar with the quantitative ultraviolet-visible spectra of the polynuclear hydrocarbons being analyzed. Although the spectra and absorptivity of these polynuclear hydrocarbons in pentane that have been published can be used for comparison purposes, a new spectrum of each pure hydrocarbon in pentane shall be determined in order to allow for any small operative or instrumental differences that may occur.

7.2 Next to be determined for calibration purposes are the column retentions of the hydrocarbons (that is, the amount of each hydrocarbon eluted from the column as compared to the amount placed on the column) and the relative positions of elution of the hydrocarbons (that is, the percentage of ether in pentane necessary to elute the hydrocarbon) for each batch of treated alumina. For this reason a large batch of alumina shall be prepared.

7.3 Prepare solutions containing 100 μg of hydrocarbon per millilitre of chloroform or dichloromethane. Prepare individual solutions for each hydrocarbon to be tested. Disperse 1 ml (100 μg) of the pure hydrocarbon in 1.0 g of pretreated alumina and carry out the described analytical procedure using the individual hydrocarbons instead of an organic fraction of air particulate matter. The spectrophotometric analysis of the fractions obtained will show (a) the amount of ether in the pentane necessary to elute the hydrocarbon from the column, and (b) the amount of hydrocarbon retained on the column as calculated by the base line technique.

7.4 Prepare a synthetic mixture containing 100 μg of all the hydrocarbons previously tested per millilitre of chloroform. Add 1 ml (containing 100 μg of each hydrocarbon) of the mixture to 1.0 g of alumina and again carry out the analytical procedure. Examination of the ultraviolet-visible curves of the fractions obtained will demonstrate the effectiveness of the chromatographic separation.

7.5 Once an analyst becomes familiar with the procedure, it is better to use an actual organic fraction of air particulate matter as the standard. This is accomplished by pooling individual organic fractions into one large fraction and performing repeated analysis. In this manner, the standard is an actual air sample and will contain most of the interfering substances normally found in ambient atmospheres. This material can be used at a later time if recalibration becomes necessary since it will remain stable for at least a year when stored in the dark in a refrigerator.

7.6 Determine the amount of hydrocarbon retained on the column during actual analysis by adding known amounts to an actual organic fraction. Add 100 μg of each hydrocarbon to be determined to a previously analyzed organic fraction. Make repeated analyses of the enriched mixture. Then subtract the average amount recovered from the average amount present in the original fraction. The difference is the amount retained on the column.

8. PROCEDURE

8.1 Dissolve the entire sample collected under 6, using about 5 ml of chloroform for 50 to 150 mg of sample. Add 1 g of the pretreated

alumina to the solution. Evaporate the chloroform so that the organic fraction is homogeneously dispersed on the alumina. Transfer this dry mixture to the top of a chromatographic column containing an ether-washed and dried glass wool plug at the bottom, a middle layer of 9.0 g of pretreated alumina, and an upper layer of 0.5 g of silica gel. Clamp the column into position on the fraction collector and clamp the solvent reservoir to the ball joint at the top of the column. The column shall be protected from light during fractionation. Add each succeeding volume of pentane-ether after the previous volume has drained to a level about 1 cm about the top of the alumina so as not to disturb the top of the column.

8.2 Elute the column through the reservoir with successive 100-ml volumes of pentane containing 0, 3, 6, 9, 12, 15, 18 and 25 percent diethyl ether. Finally, elute 100 ml of pure diethyl ether through the column to ensure complete recovery of the sample.

8.3 Collect about 40 to 45 chromatographic fractions of 15 to 20 ml each in test tubes using the fraction collector. Some solvent may be lost through evaporation. Evaporate the fractions in a vacuum oven at room temperature in the dark.

8.4 Dissolve the residue in each test tube in about 0.5 to 1.0 ml of pentane. Transfer the solute into a precalibrated 3-ml spectral cell of 1.0-cm light path. A disposable capillary pipette is quite adequate for the transfer. Repeat this procedure three or four times to ensure quantitative transfer of the residue to the cell. Adjust the volume in the cell to 3 ml with pentane. Determine the ultraviolet-visible absorption spectra of each fraction from 240 to 400 nm against a pentane blank. Extend absorption spectra determined to 450 nm after emergence of chrysene. Comparison of these spectra with predetermined standards enables the determination of the polynuclear hydrocarbon content of air particulate matter.

8.5 Prepare a new chromatographic column for each analysis.

NOTE 1 — The organic fraction to be analysed is dispersed in the alumina prior to fractionation because it is only slightly soluble in the primary eluting solvent.

NOTE 2 — Care shall be taken in plugging the column with glass wool. If not, small particles of alumina will channel through the wool and into the subfractions. It is better to plug the column with two or three small pieces of glass wool rather than one large one to eliminate any channelling.

NOTE 3 — Upon the addition of the first 100-ml portion of 100 percent pentane, the interaction of the solvent and alumina produces a small amount of heat which sometimes causes the pentane to vaporize slightly. If this happens, some pentane vapor pockets may form in the sample area of the column which essentially results in column splitting. This can be easily controlled by cooling the column until the column becomes saturated with pentane. The cooling of the column can be simply achieved by rubbing the column with an ice cube wrapped in a towel or by a piece of towelling which is cooled by an evaporating solvent such as acetone.

NOTE 4— The hydrocarbons are eluted from the column in the following order: aliphatics, olefins, benzene derivatives, naphthalene derivatives, dibenzofuran fraction, anthracene fraction, pyrene fraction, benzofluorene fraction, chrysene fraction, benzopyrene fraction, benzoperylene fraction, and coronene fraction. The smaller hydrocarbons (aliphatics, olefins, benzene, naphthalene, and dibenzofuran) are not analyzed by this procedure because they come off the column together in the initial phases of chromatography. Anthracene and phenanthrene are volatile under the conditions of air sampling and therefore are not found in normal ambient air samples. However, they are found in air polluted by coal tar pitch fumes.

NOTE 5— Three factors affect the chromatography of the organic fraction. These are (a) the retardation volume, that is, the volume of eluent passing through the column per gram of adsorbent before the substance in question leaves the column, (b) the volume spread of the eluted substance, that is, the volume of eluent in which the substance is found, and (c) the separation, that is, the volume of eluent separating one eluted substance from another. These factors can be affected by three variables namely percentage of water in the alumina, percentage of ether in the pentane, and possible miscellaneous factors such as the composition and mass of the organic fraction. An increase in either the amount of water in the alumina or the percentage of ether in the eluent decreases the retardation volume. Conversely, a decrease in either the amount of water in the alumina or the percentage of ether in the eluent increases the retardation volume. The retardation volume, the volume spread, and the volume separation of aliphatic hydrocarbons, and mono-, di-, tri-, and tetracyclic aromatic hydrocarbons are optimal in alumina containing 12 to 13 percent water while alumina containing 14 to 15 percent water works best for penta-, hexa-, and heptacyclic aromatic hydrocarbons, when eluted with 100-ml volumes of pentane containing 0, 3, 6, 9, 12, 15, 18, and 25 percent ether successively. Therefore, alumina containing 13.7 percent water is used for the fractionation.

Although the relative location of the hydrocarbons in the eluted fractions is almost always the same, unknown variables (probably due to the nature of the sample or discrete differences in alumina) sometimes cause the hydrocarbons to be eluted sooner or later than expected. For this reason, the retardation volume, the volume spread, and the volume separation should be determined whenever, (a) the procedure is being initially performed, (b) a new sample source is to be evaluated, and (c) a new batch of alumina is to be prepared for analysis.

9. CALCULATION

9.1 Since the contents of the tubes to be analyzed for polynuclear hydrocarbons are dissolved in 3 ml of pentane, it is convenient for final calculations to determine the micrograms of each individual hydrocarbon necessary to give an absorbance of 1.00 when dissolved in 3 ml of pentane and measured at a specified wavelength by the baseline technique. The baseline technique involves the drawing of a base line between two designated wavelengths beneath the specified wavelength maximum to be measured for each hydrocarbon. A perpendicular line is dropped from the apex of the curve to the base line drawn beneath the curve. The absorbance covered by the perpendicular is directly proportional to the micrograms of the hydrocarbons present. Thus, the micrograms of hydrocarbon necessary to give an absorbance covered by the perpendicular of 1.00 can be easily calculated.

9.1.1 Once the spectral curves are obtained and the presence of the hydrocarbons in the various tubes is established use the base-line

technique of calculation. Draw the base line between wavelengths and draw the perpendicular from the specified measuring wavelength to the base line. Do this for each hydrocarbon in every tube it appears. Find the total absorbance exhibited by any one hydrocarbon by adding together the absorbance found in the various tubes due to the hydrocarbon. Multiply this total absorbance by the calibration factor and the column factor to give the total micrograms of hydrocarbon present.

$$\text{Hydrocarbons, } \mu\text{g} = A \times F_1 \times F_2 \quad \dots \quad (1)$$

where

A = Total absorbance exhibited by the hydrocarbon on fractionation;

F_1 = Calibration factor, micrograms of hydrocarbon in 3 ml of pentane necessary to give an absorbance of 1.00; and

F_2 = Column factor, factor to correct amount eluted from the column to the amount placed on the column.

9.2 Perform the following calculations on air particulate samples:

$$X = (A/B) \times 1000 \quad \dots \quad (2)$$

$$Y = XC/D \quad \dots \quad (3)$$

$$Z = (XC/E) \times 1000 \quad \dots \quad (4)$$

where

X = Micrograms of hydrocarbon per gram of benzene-soluble (organic fraction),

A = Micrograms of hydrocarbon found (Equation 1),

B = Milligrams of organic fraction analyzed,

Y = Micrograms of hydrocarbon per gram of air particulate matter,

C = Total grams of organic fraction,

D = Grams of particulate matter,

Z = Micrograms of hydrocarbon per 1 000 m³ of air, and

E = Cubic metres of total air samples to obtain organic fraction.

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