

## SCHEDULE

### PART II

#### METHOD OF TESTING SOLID MINERAL HYDROCARBON FOR THE PRESENCE OF POLYCYCLIC HYDROCARBON

##### Principle of method

1. The method is based on the maximum extinction within four wavelength ranges of an extract prepared by various physical and chemical processes on an iso-octane solution of a sample of solid mineral hydrocarbon.

##### General Instructions

2. Because of the sensitivity of the test, the possibility of errors arising from contamination is great. All glassware including stoppers and stopcocks shall be scrupulously cleaned to remove all organic matter such as oil, grease, detergent residues, etc. and shall be examined under ultra-violet light to detect any residual fluorescent contamination. All glassware shall be rinsed with purified iso-octane immediately before use. No grease shall be used on stopcocks or joints. Great care shall be taken to avoid contamination of the samples in handling and to ensure absence of any extraneous material arising from inadequate packaging. Because some of the polynuclear hydrocarbons sought in this test are very susceptible to photo-oxidation, the entire procedure shall be carried out under subdued light.

##### Apparatus

3.—(1) The following apparatus shall be used—

- (a) separatory funnels of 250 millilitre, 500 millilitre, 1 litre and 2 litre capacity, equipped with tetrafluoroethylene polymer stopcocks;
- (b) a reservoir of 500 millilitre capacity, equipped with a 24/29 British Standard 572:1960 taper male fitting at the bottom and a suitable balljoint at the top for connecting to the nitrogen supply; the male fitting shall be equipped with glass hooks;
- (c) a chromatographic tube, 180 millimetres in length, inside diameter to be 15.7 millimetres  $\pm 0.1$  millimetre, equipped with a coarse, fritted-glass disc, a tetrafluoroethylene polymer stopcock, and a female 24/29 British Standard 572:1960 tapered fitting at the opposite end; the female fitting shall be equipped with glass hooks;
- (d) a tetrafluoroethylene polymer disc, 5 centimetres diameter and approximately 5 millimetres thick with a hole bored in the centre closely to fit the stem of the chromatographic tube;
- (e) a heating jacket, conical, for a 500 millilitre separatory funnel, with variable transformer heat control;
- (f) a suction flask being either a 250 or 500 millilitre filter flask;
- (g) a condenser with 24/29 British Standard 572:1960 joints, fitted with a drying tube of optional length;
- (h) a vacuum distillation assembly, all glass (for purification of dimethyl sulphoxide) consisting of a 2 litre distillation flask with heating mantle; a Vigreux vacuum-jacketed condenser (or equivalent) 45 centimetres in length and distilling head with separable cold

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finger condenser; tetrafluoroethylene polymer sleeves on the glass joints shall be used to prevent freezing;

- (i) spectrophotometric cells of fused quartz, optical path length in the range of 4.000 centimetres  $\pm$ 0.005 centimetres;
- (j) a spectrophotometer with a spectral range of 250–400 millimicrons;
- (k) a cylinder of nitrogen (water-pumped or of equivalent purity) which shall be equipped with regulator and valve to control the flow at 5 pounds per square inch gauge.

(2) The following additional apparatus may be used, namely an evaporation flask of 250 millilitre or 500 millilitre capacity, equipped with British Standard 572:1960 taper stopper having inlet and outlet tubes to permit passage of nitrogen across the surface of contained liquid to be evaporated.

## Reagents

4.—(1) The following reagents shall be used. They shall be prepared in accordance with any instructions contained in the following sub-paragraphs and shall comply with the appropriate specifications and tests described therein.

### (a) Organic Solvents

- (i) Iso-octane (2,2,4-trimethylpentane) shall pass the test described in subparagraph (v) of this sub-paragraph. For this test, 180 millilitres of iso-octane shall be used. It shall be purified, if necessary, by passage through a column of activated silica gel about 90 centimetres in length and 5–8 centimetres in diameter.
- (ii) Benzene shall be of reagent grade and shall pass the test described in subparagraph (v) of this sub-paragraph. For this test, 150 millilitres of benzene shall be used. It shall be purified, if necessary, by distillation or otherwise.
- (iii) Acetone shall be of reagent grade and shall pass the test described in subparagraph (v) of this sub-paragraph. For this test, 200 millilitres of acetone shall be used. It shall be purified, if necessary, by distillation.
- (iv) Methyl alcohol shall be of reagent grade and shall pass the test described in subparagraph (v) of this sub-paragraph. For this test, 10.0 millilitres of methyl alcohol shall be used. It shall be purified, if necessary, by distillation.
- (v) Test for Organic Solvents. The purity test for organic solvents shall be carried out as follows—

To the specified quantity of solvent in a 250 millilitre Erlenmeyer flask, 1 millilitre of purified n-hexadecane shall be added, and the mixture shall be evaporated on the steam bath under a stream of nitrogen. A loose aluminium foil jacket may be placed around the flask to speed evaporation. Evaporation shall be discontinued when not more than 1 millilitre of residue remains.

In the case of the residue from benzene, a 10 millilitre portion of purified iso-octane shall be added, and the mixture shall be re-evaporated. A further 10 millilitre portion of purified iso-octane shall be added, and the mixture re-evaporated to ensure complete removal of benzene.

If the evaporation time is reduced by using the optional evaporation flask, the solvent and n-hexadecane shall be placed in the flask on the steam bath, the tube assembly shall be inserted, and a stream of nitrogen shall be fed through the inlet tube while the outlet tube is connected to a solvent trap and vacuum line in such a way as to prevent any flow-back of condensate into the flask.

The 1 millilitre of n-hexadecane residue shall be dissolved in iso-octane and made to 25 millilitres volume. The extinction in the 4 centimetre path length cells compared

to iso-octane as reference shall be determined. The extinction of the solution of the solvent residue (except for methyl alcohol) shall not exceed 0.01 per centimetre path length between 280 and 400 millimicrons. For methyl alcohol this extinction value shall be 0.00.

(b) Eluting mixtures:

- (i) 10 per cent. benzene in iso-octane. The mixture shall be prepared by placing by means of a pipette 50 millilitres of benzene in a 500 millilitre glass-stoppered volumetric flask, adjusting to volume with iso-octane, and mixing.
- (ii) 20 per cent. benzene in iso-octane. The mixture shall be prepared by transferring by means of a pipette 50 millilitres of benzene into a 250 millilitre glass-stoppered volumetric flask, adjusting to volume with iso-octane, and mixing.
- (iii) Acetone-benzene-water mixture. The mixture shall be prepared by adding 20 millilitres of water to 380 millilitres of acetone and 200 millilitres of benzene, and mixing.

(c) n-Hexadecane shall be 99 per cent. olefine-free and shall pass the following test—

1.0 millilitre of n-hexadecane shall be diluted to 25 millilitres with iso-octane. The extinction in a 4 centimetre path length cell compared to iso-octane as reference point between 280 and 400 millimicrons shall be determined. The extinction per centimetre path length shall not exceed 0.00 in this range. The n-hexadecane shall be purified, if necessary, by percolation through activated silica gel or by distillation.

(d) Dimethyl sulphoxide shall be of pure grade, clear water-white, and shall have a melting point of not less than 18°C. It shall pass the following test—

120 millilitres of dimethyl sulphoxide shall be diluted with 240 millilitres of distilled water in a 500 millilitre separatory funnel, mixed and allowed to cool for 5–10 minutes. 40 millilitres of iso-octane shall be added to the solution which shall be extracted by shaking the funnel vigorously for 2 minutes. The lower aqueous layer shall be drawn off into a second 500 millilitre separatory funnel and the extraction repeated with 40 millilitres of iso-octane. The aqueous layer shall be drawn off and discarded. Each of the 40 millilitre extractives shall be washed three times with 50 millilitre portions of distilled water. Each wash shall be shaken for 1 minute. The aqueous layers shall be discarded. The first extractive shall be filtered through anhydrous sodium sulphate pre-washed with iso-octane into a 250 millilitre Erlenmeyer flask, or optionally into the evaporating flask. The filter shall be prepared as described in sub-paragraph (j) of this paragraph. The first separatory funnel shall be washed with the second 40 millilitre iso-octane extractive, and passed through the sodium sulphate into the flask. The second and first separatory funnels shall be washed with a 10 millilitre portion of iso-octane, and the solvent passed through the sodium sulphate into the flask. 1 millilitre of n-hexadecane shall be added and the iso-octane evaporated on the steam bath under nitrogen. Evaporation shall be discontinued when not more than 1 millilitre of residue remains. A 10 millilitre portion of iso-octane shall be added to the residue and the mixture shall be re-evaporated to 1 millilitre of n-hexadecane. 10 millilitres of iso-octane shall be added to the residue and the mixture evaporated to 1 millilitre of n-hexadecane to ensure complete removal of all volatile materials. The 1 millilitre of n-hexadecane shall be dissolved in iso-octane and made up to 25 millilitre volume. The extinction in 4 centimetre path length cells compared to iso-octane as reference shall be determined. The extinction of the solution shall not exceed 0.02 per centimetre path length between 280 and 400 millimicrons. (Note: Difficulty in meeting this extinction specification may be due to organic impurities in the distilled water. Repetition of the test omitting the dimethyl sulphoxide will disclose their presence. If necessary to meet the specification, the water shall be purified by re-distillation over

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alkaline permanganate, passage through an ion-exchange resin, or by any other suitable method.)

The dimethyl sulphoxide shall be purified, if necessary, by the following procedure:

To 1,500 millilitres of dimethyl sulphoxide in a 2 litre glass-stoppered flask, shall be added 6.0 millilitres of phosphoric acid and 50 grams of alkaline decolourising carbon. The flask shall be stoppered, and using a magnetic stirrer (tetrafluoroethylene polymer coated bar) the solvent shall be stirred for 15 minutes. The dimethyl sulphoxide shall be filtered through four thicknesses of fluted paper 18.5 centimetres in diameter. If the initial filtrate contains carbon fines, the filtrate shall be refiltered through the same filter until a clear filtrate is obtained. The sulphoxide shall be protected from air and moisture during this operation by covering the solvent in the funnel and collection flask with a layer of iso-octane. The filtrate shall be transferred to a 2 litre separatory funnel and the dimethyl sulphoxide drawn off into the 2 litre distillation flask of the vacuum distillation assembly and distilled at approximately 3 millimetres mercury pressure or less. The first 200 millilitre fraction of the distillate shall be discarded and the distillate collection flask replaced with a clean one. The distillation shall be continued until 1 litre of the sulphoxide has been collected. Because the reagent is very hygroscopic and will react with some metal containers in the presence of air, it shall be stored at the completion of distillation in glass-stoppered bottles.

- (e) Phosphoric acid shall be of 85 per cent. strength, reagent grade.
- (f) Sodium borohydride shall be of 98 per cent. purity.
- (g) Heavy magnesium oxide shall be of reagent grade and shall be prepared as follows—  
100 grams of the magnesium oxide shall be placed in a large beaker, 700 millilitres of distilled water shall be added to make a thin slurry, and the mixture shall be heated, with intermittent stirring, on a steam bath for 30 minutes. The mixture shall be stirred well initially to ensure that all the magnesium oxide is completely wetted. The mixture shall be filtered with suction, using a Buchner funnel and a filter paper of suitable diameter. Suction shall be continued until water no longer drips from the funnel. The magnesium oxide shall be transferred to a glass trough lined with aluminium foil which shall be free from rolling oil. The magnesium oxide shall be broken up with a clean spatula and spread out on the aluminium foil in a layer 1–2 centimetres thick. It shall be dried for 24 hours at 160°C. ±1°C. The magnesium oxide shall be pulverised with mortar and pestle. The pulverised magnesium oxide shall be sieved through a 60 mesh British Standard 410:1962 sieve.
- (h) Diatomaceous earth.
- (i) Magnesium oxide-diatomaceous earth mixture shall be a mixture of the reagents described in sub-paragraphs (g) and (h) of this paragraph and shall be prepared as follows—  
The magnesium oxide and the diatomaceous earth in two to one proportions, respectively, by weight shall be placed in a glass-stoppered flask large enough for adequate mixing, which shall be shaken vigorously for 10 minutes. The mixture shall be transferred to a glass trough lined with aluminium foil which shall be free from rolling oil, and spread out in a layer 1–2 centimetres thick. The mixture shall be reheated at 160°C. ±1°C. for 2 hours, and stored in a tightly closed flask.
- (j) Sodium sulphate shall be anhydrous of reagent grade, in granular form and shall be prepared as follows—  
For each bottle of sodium sulphate used, the necessary sodium sulphate pre-wash shall be established as follows to provide the filters required in the method. 35 grams of anhydrous sodium sulphate shall be placed in a 30 millilitre coarse, fritted-glass funnel or in a 65 millimetre filter funnel with glass wool plug and washed with successive 15 millilitre portion of the indicated solvent until a 15 millilitre portion of the wash shows 0.00

extinction per centimetre path length between 280 and 400 millimicrons when tested as prescribed in paragraph 4(1)(a)(v) of this Part of this Schedule. (*Note:* It will be found that three portions of wash solvent are normally sufficient.)

- (k) Pre-equilibrated sulphoxide-phosphoric acid mixture and pre-equilibrated iso-octane shall be prepared as follows—

300 millilitres of dimethyl sulphoxide shall be placed in a 1 litre separatory funnel and 75 millilitres of phosphoric acid added. The contents of the funnel shall be mixed and allowed to stand for 10 minutes. (As the reaction between the sulphoxide and the acid is exothermic, the pressure shall be released after mixing, and the funnel thereafter kept stoppered.) 150 millilitres of iso-octane shall be added and the mixture shaken to pre-equilibrate the solvents. The individual layers shall be drawn off and stored in glass-stoppered flasks.

(2) Before proceeding with analysis of a sample, the extinction in a 4 centimetre path length cell between 250 and 400 millimicrons for the reagent blank shall be determined by carrying out the procedure, without a solid mineral hydrocarbon sample, at room temperature, recording the spectra after the extraction stage and after the complete procedure as described. The extinction per centimetre path length following the extraction stage shall not exceed 0.040 in the wavelength range between 280 and 400 millimicrons; the extinction per centimetre path length following the complete procedure shall not exceed 0.070 in the wavelength range between 280 and 299 millimicrons inclusive, nor 0.045 in the wavelength range between 300 and 400 millimicrons. If either spectrum shows the characteristic benzene peaks in the 250–260 millimicrons region, the benzene shall be removed by the procedure described in paragraph 4(1)(a)(v) of this Part of this Schedule and the extinction recorded again.

### **Method of Conducting Test**

5. The test shall be carried out as follows—

A representative 1 kilogram sample of solid mineral hydrocarbon or, if this amount is not available, the entire sample shall be placed in a beaker of a capacity three times the volume of the sample and heated with occasional stirring on a steam bath until the sample is completely melted and homogeneous. Four 25 gram  $\pm 0.2$  gram portions of the melted sample shall be weighed in separate 100 millilitre beakers. Three of the portions shall be reserved for later replicate analysis as necessary. One weighed portion shall be poured immediately after remelting on the steam bath into a 500 millilitre separatory funnel containing 100 millilitres of the pre-equilibrated sulphoxide-phosphoric acid mixture which has been heated in the heating jacket at a temperature just high enough to keep the sample melted. (*Note:* In pre-heating the sulphoxide-acid mixture, the stopper of the separatory funnel shall be removed at intervals to release the pressure.)

The transfer of the sample to the funnel in the jacket with portions of the pre-equilibrated iso-octane shall be promptly completed, warming the beaker, if necessary, and using a total volume of just 50 millilitres of the solvent. If the sample comes out of solution during these operations, the stoppered funnel shall be left in the jacket until the sample redissolves. The stopper shall be removed from the funnel at intervals to release pressure. When the sample is in solution, the funnel shall be removed from the jacket and shaken vigorously for 2 minutes. Three 250 millilitre separatory funnels shall be set up, each containing 30 millilitres of pre-equilibrated iso-octane. After separation of the liquid phases, the contents shall be allowed to cool until the main portion of the sample in iso-octane solution begins to show a precipitate. The funnel shall be gently swirled when precipitation first occurs on the inside surface of the funnel to accelerate the process. The lower layer shall be carefully drawn off, filtered through a thin layer of glass wool fitted loosely in a filter funnel into the first 250 millilitre separatory funnel, and washed in tandem with the 30 millilitre portions of iso-octane contained in the 250 millilitre separatory funnels. The shaking time for each wash shall be 1 minute. The extraction operation shall be repeated with two additional portions of the sulphoxide-

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acid mixture. The funnel shall be replaced in the jacket after each extraction to keep the sample in solution and each extractive shall be washed in tandem through the same three portions of iso-octane.

The successive extractives (totalling 300 millilitres) shall be collected in a 2 litre separatory funnel, containing 480 millilitres of distilled water, mixed, and allowed to cool for a few minutes after the last extractive has been added. 80 millilitres of iso-octane shall be added to the solution and extracted by shaking the funnel vigorously for 2 minutes. The lower aqueous layer shall be drawn off into a second separatory funnel (preferably 2 litre) and the extraction repeated with 80 millilitres of iso-octane. The aqueous layer shall be drawn off and discarded. Each of the 80 millilitre extractives shall be washed three times with 100 millilitre portions of distilled water. The shaking time for each wash shall be 1 minute. The aqueous layers shall be discarded. The first extractive shall be filtered through anhydrous sodium sulphate pre-washed with iso-octane prepared as directed in paragraph 4(1)(j) of this Part of this Schedule into a 250 millilitre Erlenmeyer flask (or optionally into the evaporation flask). The first separatory funnel shall be washed with the second 80 millilitre iso-octane extractive and passed through the sodium sulphate. The second and first separatory funnels shall be washed successively with a 20 millilitre portion of iso-octane, and the solvent passed through the sodium sulphate into the flask. 1 millilitre of n-hexadecane shall be added to the contents of the flask and the iso-octane evaporated on the steam bath under nitrogen. Evaporation shall be discontinued when not more than 1 millilitre of residue remains. A 10 millilitre portion of iso-octane shall be added to the residue and the mixture re-evaporated to 1 millilitre of n-hexadecane. This operation shall be repeated once.

The residue shall be quantitatively transferred with iso-octane to a 25 millilitre volumetric flask, made to volume and mixed. The extinction of the solution in the 4 centimetre path length cells compared to iso-octane as reference between 280 and 400 millimicrons shall be determined, taking care to lose none of the solution in filling the sample cell. The extinction values shall be corrected for any extinction derived from reagents as determined by carrying out the procedure without a solid mineral hydrocarbon sample. If the corrected extinction does not exceed the limits prescribed in paragraph 6 of this Part of this Schedule, the sample shall be deemed to satisfy the requirements of the test. If the corrected extinction per centimetre path length exceeds the limits prescribed, the test shall be continued as follows:

The iso-octane solution shall be quantitatively transferred to a 125 millilitre flask, equipped with 24/29 British Standard 572:1960 joint, and the iso-octane evaporated on the steam bath under a stream of nitrogen to a volume of 1 millilitre of n-hexadecane. 10 millilitres of methyl alcohol and approximately 0.3 gram of sodium borohydride shall be added to the contents of the flask. (In order to minimize exposure of the borohydride to the atmosphere, a measuring dipper may be used.) A water-cooled condenser equipped with a 24/29 British Standard 572:1960 joint and with a drying tube shall be fitted to the flask immediately. The flask shall be shaken until the borohydride is dissolved, and allowed to stand for 30 minutes at room temperature, with intermittent swirling. At the end of this period, the flask shall be disconnected and the methyl alcohol evaporated on the steam bath under nitrogen until the sodium borohydride begins to come out of the solution. 10 millilitres of iso-octane shall then be added and the mixture evaporated to a volume of about 2–3 millilitres. A further 10 millilitres of iso-octane shall be added and the mixture concentrated to a volume of approximately 5 millilitres. The flask shall be swirled repeatedly to ensure adequate washing of the sodium borohydride residues.

The tetrafluoroethylene polymer disc shall be fitted on the upper part of the stem of the chromatographic tube. The tube with the disc shall be placed on the suction flask and the vacuum applied (approximately 135 millimetres mercury pressure). 14 grams of the 2:1 magnesium oxide-diatomaceous earth mixture shall be weighed and the adsorbent mixture poured into the chromatographic tube in approximately 3 centimetre layers. After the addition of each layer, the top of the adsorbent shall be levelled off with a flat glass rod or metal plunger by pressing down firmly until the adsorbent is well packed. The topmost few millimetres of each adsorbent layer shall be loosened with the end of a metal rod before the addition of the next layer. Packing in this

manner shall be continued until all the 14 grams of the adsorbent have been added to the tube. The top of the adsorbent shall be levelled off by pressing down firmly with a flat glass rod or metal plunger to make the depth of the adsorbent bed approximately 12.5 centimetres. The vacuum shall then be turned off and the suction flask removed. The 500 millilitre reservoir shall be fitted onto the top of the chromatographic column and the column pre-wetted by passing 100 millilitres of iso-octane through the column. The nitrogen pressure shall be adjusted so that the rate of descent of the iso-octane coming off the column is between 2–3 millilitres per minute. Pressure shall be discontinued just before the last of the iso-octane reaches the level of the adsorbent. (*Caution:* The liquid level must not, at any time, recede below the adsorbent level.) The reservoir shall be removed and the 5 millilitre iso-octane concentrate solution decanted onto the column. With slight pressure again the liquid level shall be allowed to recede to barely above the adsorbent level. The flask and the residue shall be thoroughly washed with two 5 millilitre portions of iso-octane which shall be rapidly transferred to the column in a similar manner. Just before the final 5 millilitres wash reaches the top of the adsorbent, 100 millilitres of iso-octane shall be added to the reservoir, and the percolation continued at the 2–3 millilitres per minute rate. Just before the last of the iso-octane reaches the adsorbent level, 100 millilitres of 10 per cent. benzene in iso-octane shall be added to the reservoir, and the percolation at the aforementioned rate continued. Just before the solvent mixture reaches the adsorbent level, 25 millilitres of 20 per cent. benzene in iso-octane shall be added to the reservoir and the percolation continued at 2–3 millilitres per minute until all this solvent mixture has been removed from the column. All the elution solvents collected up to this point shall be discarded. 300 millilitres of the acetone-benzene-water mixture shall be added to the reservoir, and the mixture percolated through the column to elute the polynuclear compounds. The eluate shall be collected in a clean 1 litre separatory funnel. The column shall be allowed to drain until most of the solvent mixture has been removed. The eluate shall be washed three times with 300 millilitre portions of distilled water, shaking well for each wash. (Small amounts of sodium chloride may be added to facilitate separation.) The aqueous layer shall be discarded after each wash. After the final separation, the residual benzene shall be filtered through anhydrous sodium sulphate pre-washed with benzene (prepared as directed in paragraph 4(1)(j) of this Part of this Schedule) into a 250 millilitre Erlenmeyer flask (or into the evaporation flask). The separatory funnel shall be washed with two additional 20 millilitre portions of benzene which shall also be filtered through the sodium sulphate. 1 millilitre of n-hexadecane shall be added and the benzene completely removed by evaporation under nitrogen, using the special procedure to eliminate benzene described in paragraph 4(1)(a)(v) of this Part of this Schedule. The residue shall be quantitatively transferred with iso-octane to a 25 millilitre volumetric flask and adjusted to volume. The extinction of the solution in the 4 centimetre path length cells compared to iso-octane as reference between 250 and 400 millimicrons shall be determined. Correction shall be made for any extinction derived from the reagents, as determined by carrying out the procedure without a solid mineral hydrocarbon sample. If either spectrum shows the characteristic benzene peaks in the 250–260 millimicrons region, the solution shall be evaporated to remove benzene by the procedure described in paragraph 4(1)(a)(v) of this Part of this Schedule. The residue shall be dissolved and transferred quantitatively, and adjusted to volume in iso-octane in a 25 millilitre volumetric flask. The extinction shall again be recorded. If the corrected extinction does not exceed the limits prescribed in paragraph 6 of this Part of this Schedule, the sample shall be deemed to satisfy the requirements of the test.

### **Result of test**

6. The tested solid mineral hydrocarbon shall be deemed to have satisfied the test if the following result is obtained, namely if the light extinction of the extract per centimetre path length does not exceed the following limits:

Wavelength range (millimicrons)

280–289, 290–299, 300–359, 360–400.

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Extinction limit

0.15, 0.12, 0.08, 0.02.