INTERNATIONAL STANDARD

ISO 13885-1

First edition
1998-12-15

Binders for paints and varnishes — Gel permeation chromatography (GPC) —

Part 1:
Tetrahydrofuran (THF) as eluent

Liants pour peintures et vernis — Chromatographie par perméation de gel (GPC) —

Partie 1: Utilisation de tétrahydrofurane (THF) comme éluant

Contents

1 Scope 1

2 Normative references 1

3 Definition 2

4 Principle 2

5 Apparatus 2

6 Eluent 5

7 Calibration of the apparatus 6

8 Sampling 8

9 Preparation for the test 8

10 Conditions of analysis 9

11 Data acquisition and evaluation 9

12 Precision 12

13 Test report 13

Annex A (informative) **Further information** 17

Annex B (informative) **Bibliography** 23

Annex C (informative) **Example of a data sheet for a polymer standard** 24

International Organization for Standardization
Case postale 56 • CH-1211 Genève 20 • Switzerland
Internet iso@iso.ch

Printed in Switzerland

Foreword

ISO (the International Organization for Standardization) is a worldwide federation of national standards bodies (ISO member bodies). The work of preparing International Standards is normally carried out through ISO technical committees. Each member body interested in a subject for which a technical committee has been established has the right to be represented on that committee. International organizations, governmental and non-governmental, in liaison with ISO, also take part in the work. ISO collaborates closely with the International Electrotechnical Commission (IEC) on all matters of electrotechnical standardization.

Draft International Standards adopted by the technical committees are circulated to the member bodies for voting. Publication as an International Standard requires approval by at least 75 % of the member bodies casting a vote.

International Standard ISO 13885-1 was prepared by Technical Committee ISO/TC 35, *Paints and varnishes,* Subcommittee SC 10, *Test methods for binders for paints and varnishes*.

ISO 13885 will consist of the following parts, under the general title *Binders for paints and varnishes — Gel permeation chromatography*:

— *Part 1: Tetrahydrofuran (TMF) as eluent*

— *Part 2:* N,N-*dimethylacetamide (DMAC) as eluent*

— *Part 3: Water as eluent*

At the time of publication of this part of ISO 13885, parts 2 and 3 were still at the planning stage.

Annexes A to C of this part of ISO 13885 are for information only.

Binders for paints and varnishes — Gel permeation chromatography (GPC) —

Part 1:
Tetrahydrofuran (THF) as eluent

WARNING — This part of ISO 13885 may involve hazardous materials, operations and equipment. It does not purport to address all of the safety problems associated with its use. It is the responsibility of the user of this part of ISO 13885 to establish appropriate safety and health practices and determine the applicability of regulatory limitations prior to use. A specific hazard statement appears in clause 6.

1 Scope

This part of ISO 13885 is one of a series of standards dealing with the sampling and testing of paints, varnishes and related products.

It describes conditions for the determination of the molecular-mass distribution, number-average molecular mass M_n and mass-average molecular mass M_w of polymers that are soluble in THF (tetrahydrofuran) by gel permeation chromatography (GPC)[1)].

It is possible that, in spite of the good repeatability obtained with this method, it cannot be used with certain polymer types because of specific interactions, such as adsorption within the sample/eluent/column system.

The method is not an absolute one and requires calibration with commercially available unbranched-polystyrene standards that have been characterized by absolute methods. The results for samples of polymers other than polystyrene are therefore only comparable within groups of samples of the same type.

The conditions specified in this part of ISO 13885 are not suitable for the GPC analysis of polymer samples with M_w values greater than 10^6 (see annex A).

No correction methods, e.g. for the elimination of peak broadening, are included in this part of ISO 13885. If absolute molecular-mass values are required, an absolute method, e.g. membrane osmometry for M_n or light scattering for M_w, must be used.

2 Normative references

The following standards contain provisions which, through reference in this text, constitute provisions of this part of ISO 13885. At the time of publication, the editions indicated were valid. All standards are subject to revision, and parties to agreements based on this part of ISO 13885 are encouraged to investigate the possibility of applying the most recent editions of the standards indicated below. Members of IEC and ISO maintain registers of currently valid International Standards.

ISO 1513:1992, *Paints and varnishes — Examination and preparation of samples for testing.*

ISO 5725-1:1994, *Accuracy (trueness and precision) of measurement methods and results — Part 1: General principles and definitions.*

---

1) Also known as size exclusion chromatography (SEC).

ISO 15528:—[2], *Paints and varnishes — Sampling*.

ASTM D 3536-91, *Test method for molecular weight averages and molecular weight distribution by liquid exclusion chromatography (gel permeation chromatography — GPC).*

ASTM D 5296-92, *Test method for molecular weight averages and molecular weight distribution of polystyrene by high performance size-exclusion chromatography*.

3 Definition

For the purposes of this part of ISO 13885, the following definition applies.

3.1
gel permeation chromatography
a chromatographic method in which the completely dissolved molecules of a polymer sample are fractionated on a porous column material, separation taking place according to the size of the molecule (or more precisely the size of the polymer coil which forms in this elution solvent)

NOTE 1 Small molecules diffuse into the pores of the column material more frequently and are therefore retarded more than large molecules. Thus large molecules are eluted earlier, small molecules later. Under the test conditions given, the retention volume is solely a function of the size of the molecule.

NOTE 2 This is a special form of liquid chromatography.

4 Principle

The polymer content of a sample is determined, the sample is then diluted with eluent to give a concentration of less than 5 g/l and an aliquot of the diluted sample is injected into the GPC system. The concentration of the molecules eluted from the column is measured in order of decreasing coil size with a concentration-sensitive detector, typically a differential refractometer. The molecular-mass distribution, the quantities M_n and M_w and the heterogeneity or polydispersity M_w/M_n are calculated from the resultant chromatogram with the aid of a calibration curve that has been determined for the particular GPC system.

5 Apparatus

The apparatus shall consist of the components shown in figure 1, which are described below.

It is essential that all components which come into contact with the eluent or the sample solution, are resistant to them and do not exhibit adsorption or memory effects in any form. The individual components of the GPC apparatus, which in this case uses THF as eluent, shall be linked with stainless-steel capillary tubes.

5.1 Eluent supply

The eluent reservoir shall provide the eluent with adequate protection against external influences such as the atmosphere and light, if necessary by means of a blanket of inert gas over the surface of the liquid. The eluent reservoir shall have sufficient capacity for the apparatus to be brought to the equilibrium between elution solvent and the surface of the column material and for several analyses to be conducted.

The eluent shall be degassed, either before it is introduced into the reservoir or by use of a device fitted between the reservoir and the pump, to prevent malfunctions of the pump or the formation of bubbles in the detector. The method of degassing used, e.g. bubble trap, online purging with helium, or vacuum degassing, is open to choice but shall be stated in the test report.

2) To be published. (Revision of ISO 842:1984 and ISO 1512:1991)

5.2 Pump

The pump ensures that the eluent flow through the column is as smooth and pulse-free as possible. The flow rate shall be 1 ml/min. To fulfil these requirements, the pump shall operate at optimum efficiency at this flow rate.

Either the pump shall be designed to ensure that the level of detector noise specified in 5.6 is maintained or a pulse dampener shall be fitted immediately downstream of the pump.

The parameter which characterizes a polymer molecule of a particular size is the volume eluted between injection of the sample solution and elution of the polymer. The reproducibility of measurement of this volume shall be better than 0,3 %. If the retention volume is not measured by a flow meter whose design provides adequate accuracy, but only indirectly from the elution time, the constancy of the pumping rate and the reproducibility of the pumps used are more critical: the constancy and reproducibility of about 1 % that can currently be achieved over long operating times is inadequate for the molecular-mass measurement reproducibility required. When the chromatograms are evaluated on the basis of time, it is therefore necessary to check that the flow conditions during calibration and analysis are the same, e.g. by using internal standards in the calibration and sample solutions, and, if the flow conditions are not the same, making appropriate corrections. The internal standards used shall be stated in the test report.

5.3 Injection system

The injection system serves to introduce a predetermined, precise amount of the sample solution into the eluent stream in a rapid and smooth fashion.

When filling the sample loop with sample solution and subsequently introducing the sample solution into the eluent stream, the volume of liquid used shall be great enough to ensure that, even if laminar-flow effects occur, the sample loop is completely filled with the sample solution and subsequently completely flushed out.

Memory effects from the previous sample solution in the injection system shall be avoided by suitable design or by adequate flushing.

5.4 Columns

The apparatus shall have one or more columns connected in series and packed with spherical porous material, the diameter of the pores corresponding to the size of the polymer molecules being analysed.

The packing material typically consists of a styrene/divinylbenzene copolymer (S/DVB), produced by a special polymerization process, which swells only slightly in the solvent and therefore does not deform under the pressure developed at the flow rate of 1 ml/min.

In addition to these macroporous spherical S/DVB particles, packing materials based on other organic monomers or on silicon dioxide (silica) are also used. The criterion for their use is that no adsorptive interaction shall occur between their surface and the polymer molecules in the sample. Furthermore, the sample being analysed shall not be changed, either chemically or structurally, within the chromatographic system.

Certain polymers can interact with the surface of the packing material, e.g. by adsorption, and other effects can sometimes interfere with the GPC separation mechanism. Details of such effects and notes on possible remedies are discussed in annex A. If it is intended to compare analyses by different laboratories of such polymers, the laboratories shall agree on details of the test conditions that are not covered by this part of ISO 13885.

It is practically impossible to obtain two columns with the same pore radius distribution and quality of packing. To meet the objective of this part of ISO 13885 of obtaining results that agree as well as possible in different laboratories using different GPC apparatus with the same sample, it is necessary to adhere to the minimum requirements specified below with regard to peak broadening (expressed in terms of a number of theoretical plates) and separation performance. The values actually obtained shall be stated in the test report.

a) Number of theoretical plates

The number of plates shall be determined, for the apparatus used, from the peak width at half height (see figure 2). Inject 20 µl of a solution of ethylbenzene (concentration 1 g/l) on to the column and evaluate the chromatogram obtained under the same conditions as are used for analysing polymers, according to equation (1):

$$\text{Theoretical plate number } N = 5{,}54 \times \left(\frac{V_e}{W_{1/2}}\right)^2 \times \frac{100}{L} \quad \ldots (1)$$

where

V_e is the retention volume or time to the peak maximum;

$W_{1/2}$ is the peak width at half height (see figure 2) — use the same units for V_e and W;

L is the length, in cm, of the column/column system.

Determine the peak width at half height either electronically from at least 30 data points per peak or manually on a chromatogram where the peak is at least 2 cm wide at half height and at least 15 cm high at the peak maximum.

Express the result as the number of theoretical plates per metre of total column length. To meet the requirements of this part of ISO 13885, a column system shall have at least 20 000 plates/m.

NOTE Please consult annex A with regard to tailing and fronting (asymmetry) of the peak used to calculate the plate count.

b) Separation performance

To ensure adequate resolution, the $\log_{10}M$ versus retention volume V_e calibration curve for the column system used shall not exceed a specified gradient. This parameter shall be measured using a pair of polystyrene standards which elute in the area of the peak maximum for the polymer sample under investigation or shall be obtained from the calibration curve and evaluated as

$$\text{Separation performance} = \frac{V_{e,Mx} - V_{e,(10\times Mx)}}{\text{Column cross-sectional area}} > 6{,}0 \quad \ldots (2)$$

where

$V_{e,Mx}$ is the retention volume for polystyrene of molecular mass M_x, in cm^3;

$V_{e,(10 \times Mx)}$ is the retention volume for 10 times that molecular mass, in cm^3;

the column cross-sectional area is in cm^2.

Select M_x such that the peak maximum for the polymer sample under investigation lies approximately halfway between these two retention volumes.

NOTE See annex A regarding the minimum resolution required by ASTM D 5296-92, clause 12, equation (3).

5.5 Column temperature control

Carry out the test at room temperature or at a temperature of up to 40 °C. The temperature of the column shall not change by more than 1 °C during the analysis (see annex A). Conduct the calibration and sample analyses at the same temperature. When analyses are to be carried out by different laboratories for comparison, the column temperature shall be agreed upon.

5.6 Detector

Use a differential refractometer detector. The cell volume shall not exceed 0,010 ml.

NOTE Concerning the restriction to a single detector type, see annex A.

If samples consisting of copolymers or polymer blends are to be analysed, ensure that all the components give a similar response factor (ratio of detector signal to concentration of analyte in the eluate or, in the case of the differential refractometer, specific refractive index increment ν (usually expressed as dn/dc), i.e. mathematically:

$$0{,}2 \leqslant \frac{k_i}{k_j} \leqslant 5 \qquad \text{. . (3)}$$

where

k_i and k_j are the response factors for components i and j, respectively;

dn/dc is the change in the refrative index n related to the change in the concentration c.

If the ratio of the response factors does not fall within this range in the analysis of a set of samples, a different detector or combination of detectors may be used. If it is intended to compare the results obtained by different laboratories for such a set of samples, the type of detector shall be agreed upon. If a different detector is used, the reasons for using it shall be stated in the test report. See annex A.

The detector response obtained using the sample loadings specified in this part of ISO 13885 should, at the lowest setting for electronic damping, exhibit a noise level of less than 1 % of the maximum height of the polymer peak. As the noise level is influenced by variations in pressure, temperature and flow rate, particularly in the differential refractometer, suitable measures shall be taken to maintain a constant temperature and to damp out pulses.

5.7 Flowrate meter

As described in 5.2, the most important parameter in the elution of a certain size of molecule is the retention volume. The type of flowrate meter used to measure this parameter shall be stated in the test report. If the retention volume is determined indirectly, e.g. from the elution time, the assumptions made and the measurements carried out shall also be explained in the test report.

5.8 Data acquisition

In the simplest setup, the signals from the detector are recorded by a chart recorder, though usually they will be recorded by means of an electronic data system together with information on the retention volume (see clause 11 for details).

6 Eluent

The eluent shall consist of tetrahydrofuran (THF) with the following specification:

Assay > 99,5 %;

Water < 0,05 %;

Peroxides < 0,005 %.

It may be stabilized with max. 250 ppm of 2,6-di-*tert*-butyl-4-methylphenol to prevent the formation of peroxides.

The peroxide level in the tetrahydrofuran shall be checked before use, e.g. with test strips, and stated in the test report.

WARNING — THF is highly flammable. The user of this part of ISO 13885 should refer to appropriate safe handling procedures.

In exceptional cases, which shall be explained in the test report, it may be necessary to incorporate additives in the THF eluent up to a maximum of 10 g/l, to avoid problems in the analysis of certain samples (see annex A for details).

Discard the eluent after using it to condition the column and for the actual analyses, and do not return it to the eluent reservoir.

7 Calibration of the apparatus

Calibrate the GPC apparatus with a series of unbranched-polystyrene standards of narrow molecular-mass distribution (see annex A) and whose molecular masses have been determined by independent, absolute methods. The result is a calibration curve for the evaluation of GPC analyses of polystyrene samples. If this calibration curve is used to analyse samples of other compositions, containing molecules with other structures, the results shall be expressed as the "polystyrene-equivalent molecular mass" [1].

7.1 Specification for the calibration standard

The molecular-mass distribution of the standard shall be narrower than the limits given below as a function of the peak-maximum molecular mass M_p:

$M_p < 2\,000$ g/mol	$M_w/M_n \leqslant 1{,}20$
$2\,000$ g/mol $\leqslant M_p < 10^6$ g/mol	$M_w/M_n \leqslant 1{,}05$
10^6 g/mol $\leqslant M_p$	$M_w/M_n \leqslant 1{,}20$

The peak-asymmetry factor A/B for each chromatogram, calculated from the peak half-widths A and B at half height before and after the perpendicular through the peak maximum shall lie in the range

$$\frac{A}{B} = 1{,}00 \pm 0{,}15 \quad \ldots (4)$$

The half-widths A and B shall be determined either from electronically acquired data on peaks defined by at least 60 data points or manually on a peak with a width of at least 2 cm at half height and a height of at least 15 cm.

The following minimum requirements shall be fulfilled in the characterization of each individual polystyrene standard used for calibration:

a) At least one average molecular-mass value M_n, M_w or M_z (see equations in 11.2) shall be determined by an absolute method. The M_p-values are used for calibration, but there is no absolute method of determining M_p. Therefore the procedure for obtaining the M_p-values (e.g. calculation by M_n and M_w or iterative GPC calibration, starting with the M_w-values associated with the peak maximum, and re-evaluation of M_w) must be specified in the data sheet of the standard.

b) At least one method shall be used to determine the molecular-mass distribution.

c) All the parameters involved in these methods and used in the calculations shall be stated in the test report.

d) The results and data for each batch analysed shall be presented in a form that can be re-evaluated by the user.

NOTE An example of a data sheet of this type is given in annex C.

Should the calibration standards give a shoulder on either side of the peak, pre-peaks or a tailing peak, the area represented by these anomalies shall be less than 2,0 % of the peak area, otherwise the calibration standard shall be rejected.

Hexylbenzene (M = 162) shall be used as the standard with the lowest molecular mass on the calibration curve.

If the calibration standards in the low-molecular range are separated so well that the peaks of the individual oligomers can be recognized, their actual molecular mass, including the terminal groups, shall be used in the calculations.

7.2 Preparation of the calibration solutions for injection

Shake the calibration standards in the eluent at room temperature as described in 9.1, and store at room temperature.

Filter the solutions manually through a 0,2 mm to 0,5 mm membrane filter. If the filter shows signs of blocking, the solution is unsuitable for calibration purposes.

The solutions shall be used within 48 h.

Several calibration standards may be injected and analysed at the same time, as long as all the peaks are separated down to the baseline.

The concentration of the individual calibration standards in the injection solution, as a function of the peak-maximum molecular mass, shall be

$M_p < 50\,000$ g/mol	1,0 g/l
$50\,000$ g/mol $\leqslant M_p < 10^6$ g/mol	0,5 g/l
10^6 g/mol $\leqslant M_p$	0,1 g/l

The quantities injected on to the column shall be matched to the capacity of the column by adjusting the injection volume, and not the concentration. The injection volumes determined in accordance with the requirements of clause 10 shall be used both in calibration runs and in sample analyses.

7.3 Conditions for calibration runs

The conditions for a calibration run shall, with the exception of the concentration of the injection solutions, be identical to those for the sample analyses.

7.4 Measurement of retention volume/time

The retention volume or retention time shall be measured from the start of injection to the point on the baseline at which the peak reaches its maximum height. In determining this point, a baseline drift of 5 % of the peak height, measured from injection to after the impurity peaks, is acceptable. If the drift is greater or the baseline is unsteady in the area of the peak, the analysis shall be repeated.

The repeatability of the analysis time shall be better than 0,3 %. When the retention time is measured rather than the retention volume, it shall be checked against an internal standard of known retention time and, if necessary, a correction made.

7.5 Plotting the calibration curve

The calibration curve shall be plotted with $\log_{10} M_p$ as the ordinate and the retention volume V_e or corrected retention time t_R as abscissa. At least two calibration points shall be measured per decade of molecular mass and there shall be at least five calibration points altogether. In the low molecular mass range, the calibration curve shall be extrapolated from the hexylbenzene peak to the impurity peaks. In the high molecular mass range, the peak of the first calibration standard eluted shall lie before the high molecular mass limit of the sample, and the volume for the exclusion column shall be determined.

The results of the calibration runs can be fed into a computer or recorded in the form of a table or in the form of one or more regression curves. They shall be available at all times in the form of hard copy for direct checking. Since the evaluation of the chromatograms involves their conversion into differential distribution curves in which the reciprocal of the first derivative of the calibration curve is required (see 11.3), the following requirements shall be met:

a) If the calibration curve is expressed as an equation of the form $\log_{10} M = f(V_e$ or $t_R)$, it shall be possible to differentiate the equation.

b) In all other cases:

— the calibration curve shall be described by at least 20 equidistantly spaced coordinate pairs per decade of M, and the values in one of the sets of coordinates $\log_{10} M$, V_e or t_R shall be equidistant;

— the first derivative shall be calculated by regression analyses over a maximum of five consecutive coordinate pairs.

To check how well the calibration curve thus produced fits the measurements, the percentage deviation for each calibration point, given by

$$\frac{M_{\text{p,calibration value}} - M_{\text{p,calculated}}}{M_{\text{p,calibration value}}} \times 100 \qquad \ldots (5)$$

shall be plotted against V_e or t_R. From this graph, it should be posible to assess whether the positive or negative deviations are random along the V_e or t_R axis. Calibration-curve fits which exhibit trends in the deviation plot over particular elution ranges are unsuitable. If such distributions of residuals cannot be improved upon with the regression models (see annex A) available in a laboratory, the results must be expected to contain greater errors and this shall be stated in the test report.

The test for the distribution of residuals is not appropriate to calibration curves obtained by methods in which the measured points and those on the calibration curve automatically coincide, as is the case with a connected series of straight lines and with uncompensated spline algorithms. With these methods, other means must be used to ensure that the calculated calibration curves contain no physically impossible areas, e.g. regions with a positive slope.

8 Sampling

Take a representative sample of the product to be tested, as described in ISO 15528.

Examine and prepare samples of paints and varnishes for testing, as described in ISO 1513.

9 Preparation for the test

9.1 Preparation of the injection solution

Weigh an aliquot of the polymer sample and dissolve in THF from the eluent reservoir of the chromatograph in which it is to be analysed. Store the solution at room temperature. The concentration of the injection solution is not an independent quantity. It depends on the total volume of the column used, and the injection volume. See clause 10 for details.

Shake the solution at room temperature to ensure complete dissolution and homogenization; in the case of samples with a mean molecular mass of less than 700 000 g/mol, a magnetic stirrer may be used. The use of ultrasonic energy is not permitted because of the risk of degradation. The use of heat should preferably also be avoided. Exceptions, e.g. for PVC, shall be justified in the test report.

As a rule, polymer samples shall be weighed free of solvent. If the sample contains solvent and if it is sensitive, the original solution can be used at its original concentration, or it shall be concentrated carefully under vacuum at room temperature before weighing. The polymer content of the original solution shall be determined separately; the method used shall be stated in the test report. If such samples give overlapping solvent and polymer peaks, the evaluation shall be restricted to the unaffected polymer area and the limit of the evaluation stated in the test report in terms of molecular mass. When several samples are analysed and compared, the evaluation limit selected shall be identical in each case.

Remove insoluble foreign matter, e.g. pigments, extender materials and high-impact components, from the injection solution by suitable methods, e.g. ultracentrifugation, filtration or membrane filtration. Even if the solution appears clear to the eye, filtration through membrane filters with a pore size between 2 µm and 0,2 µm is always