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Plastics — Determination of average molecular weight and molecular weight distribution of polymers using size-exclusion chromatography —

Part 4: High-temperature method

Plastiques — Détermination de la masse moléculaire moyenne et de la distribution des masses moléculaires de polymères par chromatographie d'exclusion stérique —

Partie 4: Mesurage aux températures élevées

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ISO copyright office
CP 401 • Ch. de Blandonnet 8
CH-1214 Vernier, Geneva
Phone: +41 22 749 01 11
Fax: +41 22 749 09 47
Email: copyright@iso.org
Website: www.iso.org

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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.

The procedures used to develop this document and those intended for its further maintenance are described in the ISO/IEC Directives, Part 1. In particular, the different approval criteria needed for the different types of ISO documents should be noted. This document was drafted in accordance with the editorial rules of the ISO/IEC Directives, Part 2 (see www.iso.org/directives).

Attention is drawn to the possibility that some of the elements of this document may be the subject of patent rights. ISO shall not be held responsible for identifying any or all such patent rights. Details of any patent rights identified during the development of the document will be in the Introduction and/or on the ISO list of patent declarations received (see www.iso.org/patents).

Any trade name used in this document is information given for the convenience of users and does not constitute an endorsement.

For an explanation of the voluntary nature of standards, the meaning of ISO specific terms and expressions related to conformity assessment, as well as information about ISO's adherence to the World Trade Organization (WTO) principles in the Technical Barriers to Trade (TBT) see www.iso.org/iso/foreword.html.

This document was prepared by Technical Committee ISO/TC 61, *Plastics*, Subcommittee SC 5, *Physical-chemical properties*.

This third edition cancels and replaces the second edition (ISO 16014-4:2012), which has been technically revised. The main changes compared to the previous edition are as follows:

— publication dates of references have been removed;

— molecular mass has been changed to molecular weight according to IUPAC rule;

— the operating temperature of the columns has been changed into 60 °C to 220 °C.

A list of all parts in the ISO 16014 series can be found on the ISO website.

Any feedback or questions on this document should be directed to the user's national standards body. A complete listing of these bodies can be found at www.iso.org/members.html.

Plastics — Determination of average molecular weight and molecular weight distribution of polymers using size-exclusion chromatography —

Part 4: High-temperature method

1 Scope

This document specifies a method for determining the average molecular weight and the molecular weight distribution of polymers by size-exclusion chromatography (SEC) using an organic eluent at temperatures between 60 °C and 220 °C (see Annex A). The average molecular weight and the molecular weight distribution are calculated from a calibration curve prepared using polymer standards. Therefore, this test method is classified as a relative method (see ISO 16014-1).

2 Normative references

The following documents are referred to in the text in such a way that some or all of their content constitutes requirements of this document. For dated references, only the edition cited applies. For undated references, the latest edition of the referenced document (including any amendments) applies.

ISO 472, *Plastics — Vocabulary*

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

ISO 5725-2, *Accuracy (trueness and precision) of measurement methods and results — Part 2: Basic method for the determination of repeatability and reproducibility of a standard measurement method*

ISO 16014-1, *Plastics — Determination of average molecular weight and molecular weight distribution of polymers using size-exclusion chromatography — Part 1: General principles*

ISO 16014-2, *Plastics — Determination of average molecular mass and molecular mass distribution of polymers using size-exclusion chromatography — Part 2: Universal calibration method*

3 Terms and definitions

For the purposes of this document, the terms and definitions given in ISO 472 and in ISO 16014-1 apply.

ISO and IEC maintain terminological databases for use in standardization at the following addresses:

— ISO Online browsing platform: available at https://www.iso.org/obp

— IEC Electropedia: available at http://www.electropedia.org/

4 Principle

According to ISO 16014-1.

5 Reagents

5.1 Eluent.

For a general discussion of eluents, see ISO 16014-1.

For examples of eluents used for SEC measurements at temperatures > 60 °C, see Annex B.

5.2 Reagent for column evaluation, according to ISO 16014-1.

There are several low molecular weight compounds that can be used, for example diphenylmethane when 1,2-dichlorobenzene is used as eluent or ethylbenzene when 1,2,4-trichlorobenzene is used as eluent.

5.3 Molecular weight standards, according to ISO 16014-1.

Some examples of commercially available molecular weight standards are given in Annex B.

5.4 Reagent for flow rate marker (internal standard), according to ISO 16014-1.

An example of a compound suitable for use as a flow rate marker is 2,6-di-*tert*-butyl-4-methylphenol when 1,2-dichlorobenzene or 1,2,4-trichlorobenzene is used as eluent. When an IR detector is used as concentration detector, a low molecular weight aliphatic, like heptane, can be used.

5.5 Additives.

Phenol-type antioxidants such as 2,6-di-*tert*-butyl-4-methylphenol should preferably be added to avoid degradation of the polymer sample. For further examples of antioxidants, see Annex B.

6 Apparatus

6.1 General

A schematic diagram of an SEC system is shown in ISO 16014-1.

Either commercially available or assembled SEC systems may be used, provided they meet the component requirements specified and have the capability to maintain a constant column temperature between 60 °C and 220 °C. However, components connected upstream of the injector, such as the eluent reservoir, the pumping system and tubing, does not need to be kept at the same temperature as the columns.

6.2 Eluent reservoir, according to ISO 16014-1.

As mentioned in 6.1, it is not necessary to keep the eluent reservoir at the same temperature as the columns.

6.3 Pumping system, according to ISO 16014-1.

In order to maintain the flow rate accurate to within ±0,3 %, the pumping system shall be kept at a constant temperature. As mentioned in 6.1, it is not, however, necessary to keep the pumping system at the same temperature as the columns.

6.4 Injector, according to ISO 16014-1.

In order to ensure that the polymer solution remains clear, without any precipitation, the injector temperature-control equipment shall be capable of keeping the injector at the same temperature, to within ±1 K, as the columns. As manual injection is impossible at such temperatures, an automatic injection system shall be used.

6.5 Columns. according to ISO 16014-1.

Organic or inorganic packing materials may be used, and there are no limitations on particle size or shape except that, when analysing high molecular weight and/or shear-sensitive polymers, the particle size should be large enough for no rupture of the polymer chain to occur during elution of the polymers.

The set of columns used shall have a theoretical plate number > 12 000/m, and the resolution factor R shall be > 1,5 close to the polymer peak. The asymmetry factor shall be within the range 1,00 ± 0,15. The set of columns used should preferably cover the whole range of molecular weights being determined, and the calibration curve shall be as linear as possible (the correlation factor shall be very close to 1). Determination of the theoretical plate number, the resolution factor and the asymmetry factor of the columns shall be carried out as described in ISO 16014-1.

The column temperature-control equipment shall be capable of keeping the columns within ±0,5 K of the operating temperature, which shall be between 60 °C and 220 °C.

6.6 Detector, according to ISO 16014-1.

The detector temperature-control equipment shall be capable of keeping the detector within ±0,5 K of the temperature set, in order to meet the requirements for flow rate and baseline stability (sensitivity). It is recommended that the columns and detector be kept at the same temperature.

6.7 Tubing, according to ISO 16014-1.

The temperature of the tubing shall be kept constant and high enough to ensure that the column performance requirements are met, but it is not necessary to keep the tubing at the same temperature as the column.

6.8 Temperature-control unit.

Refer to 6.4 for the injector temperature-control equipment. Refer to 6.5 and 6.6 for the columns and detector.

One of the important factors in SEC is that all components need to be kept at a constant temperature and, with the method described in this document, some of them need to be kept at a high temperature.

6.9 Recorder and plotter, according to ISO 16014-1.

6.10 Data-processing system, according to ISO 16014-1.

6.11 Other components.

In addition to the components described above, a column guard filter, a pressure monitor, a pulse damper or related components can be used, if necessary.

7 Procedure

7.1 Preparation of solutions of molecular weight standards

The molecular weight standards used to prepare the calibration curve should preferably be selected so as to cover the range of molecular weights of the polymer being analysed and so that there are at least two standards in each molecular weight decade. Solutions may be prepared which contain more than one narrow molecular weight distribution standard, but only if the standards are perfectly separated from each other on the chromatogram. Solutions of standards of molecular weight > 1 000 000 shall be prepared separately.

If molecular weight standards having the same chemical structure as the polymer being analysed are not available, the calibration curve may be prepared using standards consisting of a different type of polymer, and a universal calibration curve prepared for this different type of polymer (according to ISO 16014-2).

If gentle shaking and/or stirring or heating is required to accelerate dissolution, the duration shall be as short as possible to avoid any rupture of the polymer chains.

Filtration of the solutions is recommended to protect the column from clogging. In such cases, membrane filters or sintered-metal filters with a pore size between 0,2 µm and 1 µm shall be used. If solid material is observed on the filter, indicating incomplete dissolution, repeat the dissolution process. If a membrane filter is used, the membrane and backing shall be inert to the solvent being used. By filtration, sample polymer may cause shear degradation. In such a case, use a filter having larger the pore size and prevent shear degradation.

In general, use solutions within 48 h of preparation. However, longer storage times are allowed if the solution is kept in a cool, dark place to prevent polymer degradation and solvent evaporation.

Recommended concentrations for solutions of molecular weight standards are as follows:

$M_p < 5 \times 10^4$ 0,4 mg/cm^3

$5 \times 10^4 \leq M_p < 10^6$ 0,2 mg/cm^3

$10^6 \leq M_p$ 0,1 mg/cm^3

If a viscometric detector is used, higher molecular weight standard concentrations are required in the lower molecular weight region. Sample elution times should preferably be measured at lower concentrations, however.

7.2 Preparation of sample solutions

Prepare sample solutions by weighing accurately 10 mg to 250 mg of sample into a 10 cm^3 to 50 cm^3 flask. Add eluent and, if necessary, an internal standard and dissolve, in the same way as for the molecular weight standard solutions, within 30 min. In general, samples with molecular weights > 10^5 have a slow rate of dissolution, however, and it might be necessary to continue beyond 30 min to ensure complete dissolution. Filtration of solutions is recommended to avoid clogging of the column. When filtering, be careful about the shear deterioration of the polymer by the filtration.

If solid material is observed on the filter, indicating incomplete dissolution, repeat the dissolution process. Following preparation, transfer sample solutions to vials and store at room temperature.

The sample shall be heated sufficiently to dissolve it completely. Excessive or long heating shall be avoided, however, as this might lead to thermal or oxidative degradation. The optimum dissolution temperature and time should preferably be determined experimentally. For example, except for high molecular weight or high-density samples, polyethylene can be dissolved in 1,2-dichlorobenzene by heating at 140 °C for 1,5 h. Polyethylene with a molecular weight > 10^6 is more difficult to dissolve, requiring higher temperatures and longer times.

Before injection, sample solutions may be kept at a temperature between room temperature and the dissolution temperature. The temperature of the injector shall be sufficiently high, and the dwell time of the solution in the injector sufficiently long, to ensure that the sample remains dissolved and no precipitate forms, but the temperature shall not be so high, or the dwell time so long, that degradation of the sample occurs.

Sample solution concentrations shall not exceed the following limits:

$M_w < 1 \times 10^5$	5,0 mg/cm^3
$1 \times 10^5 \leq M_w < 10^6$	2,0 mg/cm^3
$10^6 \leq M_w$	0,5 mg/cm^3

7.3 Preparation of solutions for column performance evaluation

Prepare a 10 mg/cm^3 solution of a suitable low molecular weight compound to determine the theoretical plate number, asymmetry factor and resolution factor of the set of columns. When SEC signal became the over-scale, reduce quantity of injection volume or lower a polymer concentration.

7.4 Setting up the apparatus

Place the amount of eluent required for the SEC measurements in the reservoir and degas. Flush all the SEC components, except for the columns, with fresh eluent. Connect the set of columns into the system. Inspect all connections for leakage under the test conditions.

Keep the system at the test conditions (e.g. flow rate, detection sensitivity and temperature) until a flat baseline is obtained, with no drift or noise.

7.5 Operating parameters

7.5.1 Flow rate

A flow rate of approximately 1 cm^3/min is recommended for a series of two or three high-performance columns of approximately 30 cm in length and 8 mm in diameter. For high molecular weight and/or shear-sensitive polymers, the flow rate should preferably be reduced so that no chain rupture will occur during elution of the polymer.

7.5.2 Injection masses and injection volumes

The mass of polymer sample and volume of sample solution injected depend on the column dimensions and the detector sensitivity. The optimum sample injection mass has been found experimentally to be approximately 0,01 mg/cm^3 of empty column (without packing). The maximum mass injected shall be < 0,1 mg/cm^3 of empty column. The maximum injection volume shall be < 0,01 cm^3/cm^3 of empty column.

The injection volumes of the solutions of molecular weight standards shall be the same as for the sample solution.

The recommended injection volume of the solution of low molecular weight compound is the same as that of the sample solution.

7.5.3 Temperature of, and dwell time in, injector

The temperature of the injection port should be the same as that of the columns, and it shall be demonstrated that the dwell time of sample solutions in the injector (including the autosampler, if used) does not cause degradation of the sample.

7.5.4 Column temperature

The column temperature should be selected based mainly on the solubility of the sample, the viscosity and boiling point of the eluent, and the ambient temperature.

7.5.5 Detector sensitivity

The signal intensity depends on the amount of sample injected and on the specific refractive index increment d_n/d_c for an RI detector and the absorbance per unit mass concentration for a UV detector or IR detector. The detector sensitivity should be set to obtain a strong peak signal for the sample to ensure accurate data handling.

The linear relationship between solute concentration and peak height shall be maintained by keeping the sensitivity at the same setting.

7.6 Number of determinations

Carry out at least two sample runs to demonstrate the repeatability of the positions and shapes of the peaks in the chromatogram.

8 Data acquisition and processing

According to ISO 16014-1.

9 Expression of results

According to ISO 16014-1.

10 Precision

10.1 General

The precision of this test method was determined in an interlaboratory testing carried out in 1999 in accordance with ISO 5725-1 and ISO 5725-2.

10.2 Experimental conditions

The test samples, which included three types of polyethylene and one type of polypropylene, and the calibration standards of narrow molecular weight distribution were distributed to the participating laboratories by the organizer. The details of the interlaboratory tests are as follows:

Polymer samples	Sample A	Polyethylene (high molecular weight, broad MMD sample)
	Sample E	Polyethylene (narrow MMD sample, NIST SRM-1475)
	Sample F	Polyethylene (low molecular weight, broad MMD sample)
	Sample G	Polypropylene (broad MMD sample)
Calibration	14 polystyrene standards	
Column packing material	Polystyrene gel	
Eluents	1,2-Dichlorobenzene and 1,2,4-trichlorobenzene	
Column temperature	135 °C or 140 °C	
Number of laboratories	11	

NOTE The higher column temperature of 140 °C was used for the PE samples containing components of molecular weight > 1×10^6.

10.3 Results of interlaboratory tests

The results, expressed as repeatability and reproducibility, are summarized in Table 1. The raw data are shown in Annex C.

Table 1 — Results of interlaboratory tests for high-temperature SEC

Polymer	Average values of M_n and M_w[a]		Repeatability, s_r[a] %		Reproducibility, s_R[a] %	
	Tosoh[b]	PL[c]	Tosoh[b]	PL[c]	Tosoh[b]	PL[c]
Sample A (polyethylene)	M_n = 130 000 (5/11)[d]	M_n = 145 000 (6/10)	1,72	2,19	7,21	14,22
	M_w = 526 000 (9/11)	M_w = 574 000 (9/10)	2,18	3,08	11,35	12,95
Sample E (polyethylene)	M_n = 39 200 (11/11)	M_n = 39 100 (10/10)	3,50	4,68	11,26	11,99
	M_w = 120 000 (10/11)	M_w = 128 000 (10/10)	1,40	1,52	9,75	13,23
Sample F (polyethylene)	M_n = 57 400 (10/11)	M_n = 55 100 (10/10)	3,26	7,15	14,55	14,56
	M_w = 218 000 (11/11)	M_w = 239 000 (10/10)	1,67	2,14	7,86	11,21
Sample G (polypropylene)	M_n = 68 100 (10/11)	M_n = 69 100 (10/10)	5,86	5,73	21,33	17,49
	M_w = 323 000 (9/11)	M_w = 363 000 (10/10)	1,29	2,34	4,59	11,24

NOTE Sample A shows a very low standard deviation for M_n because of the existence of six outliers. If the results from all 11 of the laboratories are used, this gives a value of s_R of 35,61 % for M_n. Sample A was found to be unsuitable because it contains components of extremely high molecular weight beyond the effective range of the calibration curve.

[a] Outliers were eliminated by Grubbs' and Cochran's methods[1].

[b] Polystyrene calibration standards supplied by Tosoh Co. (Japan).

[c] Polystyrene calibration standards supplied by Polymer Laboratories Ltd. (United Kingdom).

[d] Numbers in brackets indicate (total minus outliers)/(total number of laboratories).

11 Test report

According to ISO 16014-1.

In addition to the information required by ISO 16014-1, on the apparatus and measurement parameters, the test report shall contain the following:

a) the temperature of the injection port and the dwell time of the sample solution in it;

b) the temperature at which the sample was dissolved and the length of time necessary.

Annex A
(informative)

Further information on applicability of method

The method described in this document is suitable for measurements at temperatures between 60 °C and 220 °C and assumes the sample is a linear homopolymer. However, because it is a relative method, it is also applicable to nonlinear homopolymers, such as branched, star-shaped, comb-like, stereo-regular and stereo-irregular polymers, and to other types of polymer, such as random, block, graft and heterophasic copolymers. The method is applicable to molecular weights ranging from that of the monomer to 3 000 000 from that of the monomer to exclusion limit of the used columns, but is not applicable to samples that contain > 30 % of components having a molecular weight < 1 000.

The method cannot be used with water as eluent, i.e. for water-soluble polymers, or at column temperatures > 220 °C (e.g. for polyphenylenesulfide) or with polymers that exhibit appreciable secondary effects such as adsorption of the polymer molecules on the column packing material or repulsion between the polymer molecules and the packing material.

Annex B
(informative)

Further information on reagents

B.1 Examples of suitable eluents

The following are examples of eluents for SEC measurements at temperatures > 60 °C:

a) 1,2-dichlorobenzene;

b) 1,2,4-trichlorobenzene;

c) 1-chloronaphthalene;

d) toluene;

e) *N*,*N*-dimethylformamide.

B.2 Narrow molecular weight distribution standards

According to ISO 16014-1.

B.3 Additives

The following are examples of additives which may be added to the sample as an antioxidant for SEC measurements at temperatures > 60 °C:

— 2,6-Di-*tert*-butyl-4-methylphenol

— 4-Hydroxymethyl-2,6-di-*tert*-butylphenol

— 1,1,3-Tri(*tert*-butylhydroxymethylphenyl)butane

— 4,4′-Thio-*bis*-(6-*tert*-butyl-*m*-cresol)

Inorganic salts like lithium bromide are also used as additives in high-temperature SEC when *N*,*N*-dimethylformamide is used as eluent.

Annex C
(informative)

Interlaboratory test

C.1 Raw data from round robin test

Tables C.1 to C.4 present the raw data (molecular weight averages and variances) obtained in the interlaboratory test. Three sample runs (see 7.6) were carried out for each determination (i.e. $n = 3$).

Table C.1 — Molecular weight averages and variances obtained for polyethylene (sample A)

Laboratory	M_n (× 10^{-4})		M_w (× 10^{-4})	
	Average	Variance	Average	Variance
Sample A (polyethylene) **Calibration: polystyrene standards (Tosoh)**				
A	2,29	(0,015 7)	57,4	0,26
B	2,06	(0,007 2)	48,4	0,21
E	1,29	0,000 1	49,9	0,21
F	1,38	(0,061 0)	45,5	(9,62)
G	1,19	0,000 7	54,6	2,73
H	1,42	0,000 8	58,4	1,95
I	1,40	0,000 8	46,8	2,21
J	(1,75)	0,000 4	43,7	0,08
K	(3,17)	0,008 7	(114,9)	0,64
L	1,35	0,000 1	61,9	3,18
M	1,30	(0,030 4)	51,9	0,97
Sample A (polyethylene) **Calibration: polystyrene standards (Polymer laboratories)**				
A	1,57	0,001 9	52,2	3,87
B	(2,21)	0,001 7	58,9	2,50
F	1,40	(0,038 1)	47,5	10,89
G	1,21	0,000 7	62,0	3,50
H	1,55	0,000 2	71,9	1,78
I	1,43	0,000 7	54,0	3,06
J	1,74	0,000 4	50,3	0,12
K	(3,32)	(0,009 9)	(135,1)	0,88
L	1,24	0,002 1	61,8	2,22
M	1,30	(0,008 1)	58,1	0,20
NOTE Values in brackets are outliers.				

Table C.2 — Molecular weight averages and variances obtained for polyethylene (sample E)

Laboratory	M_n (× 10^{-4})		M_w (× 10^{-4})	
	Average	Variance	Average	Variance
Sample E (polyethylene) **Calibration: polystyrene standards (Tosoh)**				
A	4,64	0,008	13,0	0,08
B	4,13	0,016	13,4	0,01
E	3,80	0,004	12,6	0,01
F	3,28	0,035	10,8	0,00
G	4,07	0,028	12,3	0,06
H	3,28	0,081	10,6	0,07
I	3,90	0,001	11,8	0,00
J	3,63	0,003	10,0	0,01
K	4,58	0,002	13,6	(0,46)
L	3,82	0,010	12,6	0,01
M	3,98	0,020	12,6	0,02
Sample E (polyethylene) **Calibration: polystyrene standards (Polymer laboratories)**				
A	3,19	0,003	10,2	0,01
B	4,38	0,033	15,1	0,03
F	3,40	0,038	11,9	0,00
G	4,14	0,029	13,3	0,08
H	3,42	0,074	11,5	0,09
I	3,98	0,002	12,8	0,01
J	3,67	0,003	10,7	0,02
K	4,63	0,082	14,9	0,04
L	4,17	0,020	14,2	0,09
M	4,08	0,052	13,0	0,00
NOTE Values in brackets are outliers.				

Table C.3 — Molecular weight averages and variances obtained for polyethylene (sample F)

Laboratory	M_n (× 10^{-4})		M_w (× 10^{-4})	
	Average	Variance	Average	Variance
Sample F (polyethylene) **Calibration: polystyrene standards (Tosoh)**				
A	7,32	0,009	23,80	0,244
B	5,31	0,109	21,90	0,003
E	5,19	0,003	21,10	0,004
F	5,68	0,022	19,30	0,223
G	5,90	0,005	22,30	0,513
H	3,87	(0,445)	20,30	0,111
I	5,33	0,095	20,10	0,003
J	6,14	0,013	21,60	0,001
K	6,82	0,061	25,40	0,061
L	4,57	0,000	22,10	0,166
M	5,11	0,033	21,70	0,135
Sample F (polyethylene) **Calibration: polystyrene standards (Polymer laboratories)**				
A	5,17	0,033	19,0	0,19
B	5,77	0,312	25,0	0,27
F	5,90	0,024	21,6	0,28
G	6,05	0,005	24,7	0,67
H	4,17	0,347	23,0	0,16
I	5,45	0,105	22,4	0,00
J	6,25	0,015	23,9	0,00
K	6,62	0,546	29,2	0,39
L	4,24	0,163	25,2	0,10
M	5,52	0,006	24,7	0,53
NOTE Values in brackets are outliers.				

Table C.4 — Molecular weight averages and variances obtained for polypropylene (sample G)

Laboratory	M_n (× 10^{-4})		M_w (× 10^{-4})	
	Average	Variance	Average	Variance
Sample G (polyethylene) **Calibration: polystyrene standards (Tosoh)**				
A	9,69	0,062	34,70	0,10
B	6,89	0,086	31,80	0,62
E	7,27	0,001	32,20	0,02
F	6,35	0,194	32,60	(1,40)
G	6,60	0,032	32,90	0,49
H	4,00	0,139	31,50	0,06
NOTE Values in brackets are outliers.				