INTERNATIONAL STANDARD

ISO 16373-3

First edition
2014-06-01

Textiles — Dyestuffs —

Part 3: Method for determination of certain carcinogenic dyestuffs (method using triethylamine/methanol)

Textiles – Colorants —

Partie 3: Méthode de détermination de certains colorants cancérigènes (méthode à la triéthylamine et au méthanol)

Reference number
ISO 16373-3:2014(E)

ISO copyright office
Case postale 56 • CH-1211 Geneva 20
Tel. + 41 22 749 01 11
Fax + 41 22 749 09 47
E-mail copyright@iso.org
Web www.iso.org

Published in Switzerland

Contents

Page

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. 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. 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 on the meaning of ISO specific terms and expressions related to conformity assessment, as well as information about ISO's adherence to the WTO principles in the Technical Barriers to Trade (TBT) see the following URL: Foreword - Supplementary information

The committee responsible for this document is ISO/TC 38.

ISO 16373 consists of the following parts, under the general title *Textiles — Dyestuffs*:

— *Part 1: General principles of testing coloured textiles for dyestuff identification*

— *Part 2: General method for the determination of extractable dyestuffs including allergenic and carcinogenic dyestuffs (method using pyridine-water)*

— *Part 3: Method for determination of certain carcinogenic dyestuffs (method using triethylamine/methanol)*

Introduction

Due to concerns of consumers over safety and hygiene, many countries have introduced regulations regarding carcinogenic dyestuffs in textile articles. To support international and national regulations the development of a test method is very important and this part of ISO 16373 does just that.

The ISO 16373 series deal with dyestuffs used in textile for qualification and quantification.

— ISO 16373-1[1)] includes the definition of the dyestuff, and classes the description of some procedures to identify qualitatively the dyestuff class used in textile material. The other parts of ISO 16373 are related to the quantification of some dyestuffs.

— In ISO 16373-2, the principle of the test method is based on extraction using pyridine-water solution, which has been found to be the most efficient solution to extract a large range of dyestuffs, including allergenic and carcinogenic dyestuffs.

— In this part of ISO 16373, the principle of the test method is based on extraction using triethylamine-methanol solution. This solution has been found to be efficient at extracting some dyestuffs in some cases.

Additional information related to the recovery rate (to characterize the extraction efficiency) obtained from the application of ISO 16373-2 and this part of ISO 16373 is summarized in ISO 16373-1:—, Annex B.

It is important to note that there are other test methods related to azo dyes, for which a reduction of the extracted azo dyes leads to the release of some aromatic amines to be detected and determined using chromatography.[6][7]

1) To be published.

Textiles — Dyestuffs —

Part 3: Method for determination of certain carcinogenic dyestuffs (method using triethylamine/methanol)

1 Scope

This part of ISO 16373 specifies a method for the detection and quantitative determination of the presence of carcinogenic dyestuffs as listed in Table 1 in dyed, printed or coated textile products by chromatographic analysis of their extracts.

2 Terms and definitions

For the purposes of this document, the following terms and definitions apply.

2.1
textile
woven fabric, knitted fabric, etc., formed by the interlocking of fibres and yarns having a certain cohesion and which is generally intended for clothing or furniture applications

Note 1 to entry: Textiles often include certain types of non-woven fabrics.

2.2
carcinogenic dyestuff
substance yielding a dye that is a substance known to be or suspected of being a human carcinogen

3 Principle

The dyestuff of a coloured test specimen is extracted by means of a solvent in an ultrasonic bath under specified conditions. The extract is analysed using either a high-performance liquid chromatography photodiode array detector (HPLC-DAD) or a high-performance liquid chromatography mass spectrometer (HPLC-MSD).

The carcinogenic dyestuffs are listed in Table 1.

Table 1 — List of carcinogenic dyestuffs

C.I. Generic name	CAS number	C.I. Constitution number
C.I. Basic Red 9	569–61–9	42500
C.I. Disperse Orange 11	82–28–0	60700
C.I. Disperse Yellow 3	2832–40–8	11855
C.I. Acid Red 114	6459–94–5	23635
C.I. Acid Red 26	3761–53–3	16150
C.I. Direct Black 38	1937–37–7	30235
C.I. Direct Red 28	573–58–0	22120
C.I. Disperse Blue 1	2475–45–8	64500

Table 1 *(continued)*

C.I. Generic name	CAS number	C.I. Constitution number
C.I. Basic Violet 14	632–99–5	42510
C.I. Direct Blue 6	2602–46–2	22610
C.I. Direct Brown 95	16071–86–6	30145

4 Safety precautions

4.1 General

Warning — The dyestuffs targeted in this part of ISO 16373 are classified as substances known to be or suspected of being human carcinogens.

4.2 Handling

It is the user's responsibility to ensure any handling and disposal of these substances are in strict accordance with the appropriate national health and safety regulations.

It is the user's responsibility to use safe and proper techniques in handling materials in this test method. Consult manufacturers for specific details, such as material safety data sheets and other recommendations.

Good laboratory practice should be followed. Wear safety glasses in all laboratory areas and single-use dust respirator while handling the dyestuff powder.

5 Apparatus

5.1 Ultrasonic bath, capable of heating to and maintaining at (50 ± 5) °C and output power of 40 W, oscillating frequency, 42 kHz, or equivalent.

5.2 Coil condenser, for chemical testing use.

5.3 Vacuum rotary evaporator, capable of operating at water evaporation capacity of a maximum of 25 ml/min, or equivalent.

5.4 Round bottom flask, of 200 ml in capacity.

5.5 Pipettes, of 1 ml and 10 ml in capacity.

5.6 Volumetric flask, of 10 ml, 100 ml and 1 l in capacity.

5.7 High-performance liquid chromatography (HPLC) system and **diode array detector (DAD) or mass spectroscope (MSD)**.

5.8 Test tube, of 100 ml in capacity, with a silicone plug.

NOTE For details of the high-performance liquid chromatography equipment, see Annex A.

5.9 Analytical balance, of 0,001 g in resolution.

6 Reagents

Unless otherwise specified, use only reagents of recognized analytical grade.

6.1 Acetonitrile.

6.2 Methanol.

6.3 Hexane.

6.4 0,25 % tri-ethylamine methanol solution, 2,5 ml triethylamine is dissolved in methanol and made up to 1 l.

6.5 10 mmol/l ammonium acetate aqueous solution, 0,77 g ammonium acetate is dissolved in water and made up to 1 l.

6.6 Carcinogenic dyestuffs. Use only carcinogenic dyestuffs of reagent grade of the highest purity available on the market, or dyestuffs of which quantities of the dye are manufactured in controlled environments within Europe under the control of the EU creating standard dyestuffs.

6.7 Standard solution of carcinogenic dyestuffs.

An amount of each carcinogenic dyestuff is weighed accurately in the range of 1 mg to 10 mg and transferred quantitatively to a 10 ml volumetric flask, and then made up to volume with methanol (6.2) to prepare a standard solution in the range of 100 ug/ml to 1 000 ug/ml.

The standard solution may be diluted properly and four solutions with known concentrations may be made to draw the calibration curve. As an example, the range of concentration of standard solutions for the calibration curve can be recommended to be from 1 µg/ml to 100 µg/ml.

7 Test specimen sampling and preparation

7.1 General

The test specimen shall be selected based on the following criteria:

— parts of the textile article;

— nature of the fibre component (fibre composition);

— colours.

Prepare a test specimen of maximum 1,0 g (≤1,0 g) by cutting the laboratory sample up into small pieces no larger than 1 cm^2. Determine the mass of the test specimen to the nearest 0,01 g and record it as m_E (see 8.2).

8 Procedure

8.1 Extraction

8.1.1 Cleansing

If required, remove oil, grease or other fatty matter from the surface of the test specimen by soaking it in 100 ml hexane (6.3) for 5 min in an ultrasonic bath (5.1) at ambient temperature.

Remove and drain the test specimen.

8.1.2 Extraction of dyestuff

Place 1,0 g of the test specimen in a 100 ml test tube. Add 100 ml of the 0,25 % tri-ethylamine methanol solution (6.4) and seal the test tube using a silicone plug. Heat the tube in an ultrasonic bath until a temperature of 50 °C ± 2 °C is reached and maintained this temperature for 3 h.

8.1.3 Concentration of extract and preparation of analysis solution

Transfer the extract obtained according to 8.1.2 to a 200 ml round bottom flask (5.4) and place it in a vacuum rotary evaporator (5.3) in the water bath at 40 °C ± 2 °C until all the liquid has evaporated.

Dissolve the residue in 1 ml of methanol. Filter the solution through a 0,45 µm PTFE filter. If the resultant measurement is higher than the calibrated range of the chromatograph, dilute the solution further with methanol.

8.2 Detection, identification and quantification of carcinogenic dyestuffs

Detection of carcinogenic dyestuffs is performed by the chromatographic analysis using the apparatus described in 5.7. When the carcinogenic dyestuffs are identified by comparing with peaks of reference carcinogenic dyestuffs, quantification is performed using a calibration curve, which is drawn by using a minimum of four points obtained from an HPLC analysis of the standard solution (6.7) and the correlation coefficient of the linear curve should be 0,99 in the range of concentration of 1 µg/ml to 100 µg/ml. Quantification is executed by the method of HPLC/DAD. When a large amount of foreign substances are detected, HPLC/MSD is recommended for identification and quantification.

The concentration of carcinogenic dyestuff in the specimen is calculated as a mass fraction of the specimen, w (µg/g), as given by Formula (1):

$$w = \frac{\rho_s \times V}{m_E} \quad (1)$$

where

ρ_s is the interpolated concentration of carcinogenic dyestuff, in micrograms per millilitre (µg/ml);

V is the final solution volume made up to according to 8.1.2, in millilitres (ml);

m_E is the mass of the test specimen, in millilitres (ml);

9 Test report

The test report shall include the following:

a) reference to this part of ISO 16373, i.e. ISO 16373-3;

b) kind, origin and designation of the specimen (partial specimen, if applicable);

c) detection method and quantification method;

d) results reported as level and detection limit for each of the carcinogenic dyestuffs (µg/g);

e) any deviation from the procedure.

Annex A
(informative)

Chromatographic analysis

A.1 Chromatographic analysis — General

As the instrumental equipment of laboratories might vary, no generally applicable instructions can be provided for chromatograph analysis. Therefore, the following parameters have been successfully tested and used. See Figures A.1 to A.14 and Table A.1.

A.2 High-performance liquid chromatography/diode array detector (HPLC/DAD)

See Table A.1.

Table A.1 — Condition of HPLC/DAD

Eluent 1:	10 mmol/l ammonium acetate
Eluent 2:	Acetonitrile
Column	Inertsil ODS-3, 150 mm × 3,0 mm, 5 µm
Flow rate:	0,8ml/min
Gradient Time programme	Time (min) Eluent 2 concentrations Initial 5 % 30 60 % 40 60 % 40,1 5 % 50 5 %
Column temperature:	45 °C
Injection volume:	5,0 µl
Determination:	DAD
Quantification:	540 nm (for Basic Red 9) 480 nm (for Disperse Orange 11) 350 nm (for Disperse Yellow 3) 510 nm (for Acid Red 114) 510 nm (for Acid Red 26) 600 nm (for Direct Black 38) 500 nm (for Direct Red 28)
Remark Columns of equivalent quality may be used.	

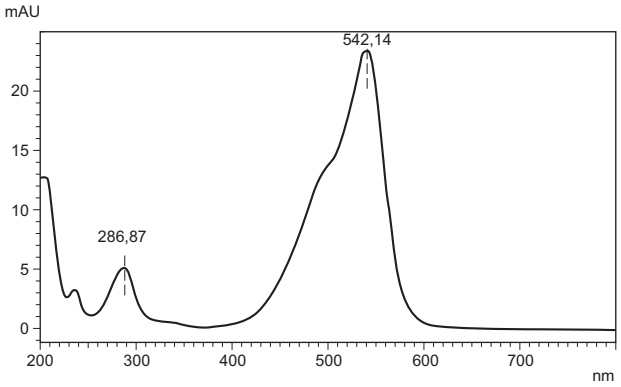

Figure A.1 — UV spectrum of Basic Red 9

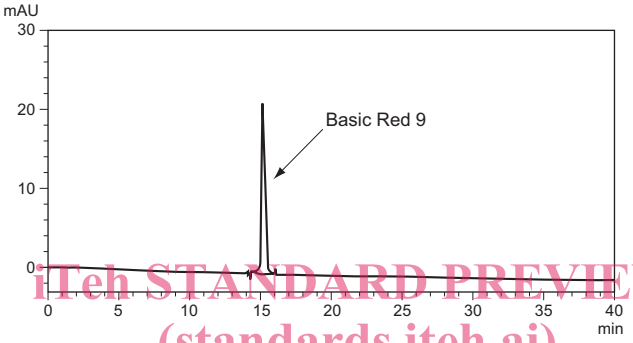

Figure A.2 — HPLC/DAD Chromatogram at 540 nm detection

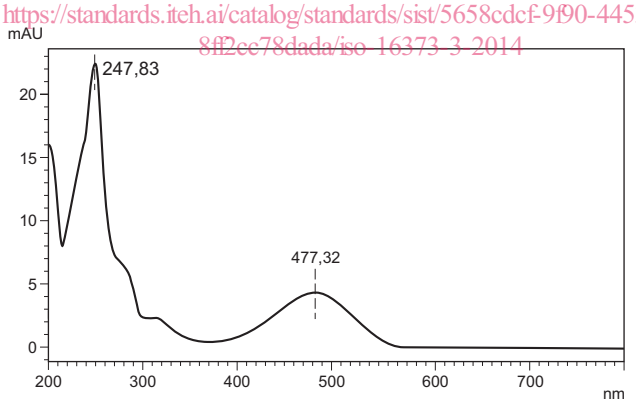

Figure A.3 — UV spectrum of Disperse Orange 11