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Textiles — Dyestuffs —

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

Textiles — Colorants —

Partie 2: Méthode générale de détermination des colorants extractibles, notamment les colorants allergènes et cancérigènes (méthode utilisant un mélange pyridine/eau)

Reference number
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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. www.iso.org/directives

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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

ISO 16373-2 was prepared by the European Committee for Standardization (CEN) Technical Committee CEN/TC 248, *Textiles and textile products*, in collaboration with ISO Technical Committee TC 38, *Textiles*, in accordance with the Agreement on technical cooperation between ISO and CEN (Vienna Agreement).

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 (method using pyridine-water)*

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

Introduction

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

ISO 16373-1[1)] includes the definition of the dyestuff classes and 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 this part of ISO 16373, the principle of the test method is based on the 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 ISO 16373-3, the principle of the test method is based on extraction using triethylamine/methanol solution. This solution has been found efficient to extract some dyestuffs in some cases.

Additional information related to the recovery rate (to characterize the extraction efficiency) obtained from the application of ISO 16373-3 and this part of ISO 16373 will be summarized in ISO 16373-1.

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

The percentage of recovery using the method of this part of ISO 16373 is shown in Annex F for the dyestuff classes (to be defined in ISO 16373-1) acid, basic, direct, disperse, solvent dyestuffs and "mordant dyestuffs" on different textile fibres.

1) To be published.

Textiles — Dyestuffs —

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

1 Scope

This part of ISO 16373 specifies the analyses used to detect extractable dyestuffs in textile products, with the extraction performed for all kind of fibres and types of dyestuffs using pyridine/water (1:1). It lists (see Annexes A and B) the allergenic and carcinogenic dyestuffs which can be analysed using this method; the lists of dyestuffs are expandable.

2 Normative references

The following documents, in whole or in part, are normatively referenced in this document and are indispensable for its application. For dated references, only the edition cited applies. For undated references, the latest edition of the referenced document (including any amendments) applies

ISO 3696, *Water for analytical laboratory use — Specification and test methods*

3 Terms and definitions

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

3.1
allergenic dyestuff
dyestuff which may cause an allergic skin reaction

3.2
carcinogenic dyestuff
dyestuff which is classified as carcinogenic substance

Note 1 to entry: Harmonized classification according to the *Globally harmonized system of classification and labelling of chemicals (GHS)*,[2] incorporated in EU Regulation 1272/2008 (CLP)[3].

4 Principle

A coloured test specimen is selected from the textile article and extracted with pyridine/water at 100 °C. The extract is analysed by liquid chromatography/diode array detection (LC/DAD) and/or by liquid chromatography/mass spectrometry (LC/MS).

5 Safety precautions

WARNING —The substances listed in Tables A.1, B.1 and B.2 are classified as substances known to be or suspected to be human allergens or carcinogens.

Ensure that any handling and disposal of these substances is 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 a single use dust respirator while handling powder dyestuffs.

Attention is drawn to any national and local safety regulations.

6 Reagents

Unless otherwise specified, analytical grade chemicals shall be used.

6.1 Pyridine.

6.2 Acetonitrile, chromatographic grade.

6.3 Ammonium acetate.

6.4 Tetrabutylammonium dihydrogen phosphate.

6.5 Deionized water, grade 3 in accordance with ISO 3696.

6.6 Pyridine/water (1:1), prepared by mixing 500 ml pyridine (6.1) and 500 ml water (6.5).

Keep the solution in a brown glass bottle.

6.7 Individual stock solutions, prepared in pyridine-water 1:1, of all reference substances listed in Annexes A and B

It is recommended that reference substances (including those listed in Annexes A and B) of the highest purity grade available on the market be used. The given purity has to be considered for the calculation (see Clause 9).

7 Apparatus

7.1 Apparatus and auxiliaries for sample preparation

7.1.1 Standard laboratory equipment.

7.1.2 Analytical balance, resolution at 0,01 g.

7.1.3 Glass vials (20 ml to 40 ml), with tight closure.

7.1.4 Heating source that generates heat at a temperature of (100 ± 2)°C (thermal block or laboratory sand-bath, controllable).

7.1.5 Autosampler glass vials, with tight closure.

7.1.6 Thermo-sensing device, e.g. thermocouple, to measure at 100 °C with a resolution of 0,1 °C.

7.2 Chromatographic equipment (selected from the following)

7.2.1 Equipment for LC/DAD

— High performance liquid chromatograph (HPLC),

— DAD detector,

— separating column,

— guard column.

7.2.2 Equipment for LC/MS

— High performance liquid chromatograph (HPLC),

— electrospray ion source,

— MS detector,

— separating column,

— guard column.

8 Procedure

8.1 Preparation of test specimen

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 1,0 g max. 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).

8.2 Extraction

Add 7,5 ml of pyridine/water (1:1) (6.6) to the test specimen and close the vial tightly. Heat the vial in the heating source until the temperature of the solvent reaches (100 ± 2) °C, and hold this temperature for (35 ± 5) min.

Check the time taken for the temperature of the solvent to reach the required temperature using a glass vial (see 8.1) containing 7,5 ml of pyridine/water (1:1)(6.6) with a thermo-sensing element (7.1.6), which is plunged in the solvent and sealed with a septum.

Allow the vial to cool down to 40 °C or below before opening it.

Transfer approximately 1 ml of (test) liquid from the vial to a smaller vial for further analysis.

NOTE This step could be done with a syringe through the closed septum to minimize contact with pyridine.

8.3 Detection and quantification of dyestuff

Dyestuff detection can be performed using the above-specified chromatographic techniques (7.2). If other analytical techniques are used it shall be reported.

Dyestuff quantification is performed by means of HPLC/DAD/MS.

NOTE Some dyes can be quantified with HPLC/DAD.

8.4 Calibration

For the calibration, the mixed standards of the reference substances listed in Table A.1 (and respectively in Tables B.1 and B.2) are prepared in pyridine/water (1:1). The stock solutions are used to prepare mixed standards with concentrations levels of 1 mg/l, 5 mg/l, 10 mg/l and 20 mg/l related to dyestuff content.

9 Calculation and expression of the results

Amounts of dyestuff are usually calculated by means of a software program. The calculation can be carried out manually as described in Annex C.

Amounts of dyestuff are expressed in mg dyestuff per kg of textile (mg/kg).

If the detected amount of a dye is over 100 mg/kg, it is certain that this dye is used – such as one of those listed in Table A.1, B.1 or B.2.

10 Test report

The test report shall refer to this method and shall include at least the following:

a) reference to this part of ISO 16373, i.e. ISO 16373-2;

b) all information necessary for the identification of the test specimen;

c) date of sample receipt and date of analysis;

d) sampling procedure;

e) detection method and quantification method;

f) results reported as level and detection limit per dyestuff, in mg/kg;

g) any deviation from the given procedure.

Annex A
(normative)

List of carcinogenic dyestuffs

See Table A.1.

Table A.1 — Reference carcinogenic dyestuffs

Number[a]	Carcinogenic dyestuff[b]	C. I. Number[c]	CAS Number	Molecular formula
1	Disperse Blue 1	64500	2475–45–8	$C_{14}H_{12}N_4O_2$
2	Solvent Yellow 1 4-aminoazobenzene	11000	60–09–4	$C_{12}H_{11}N_3$
3	Solvent Yellow 2	11020	60–11–7	$C_{14}H_{15}N_3$
4	Solvent Yellow 3 o-aminoazotoluene	11160	97–56–3	$C_{14}H_{15}N_3$
5	Basic Red 9	42500	569–61–9	$C_{19}H_{17}N_3HCl$
6	Basic Violet 14	42500	632–99–5	$C_{20}H_{19}N_3HCl$
7	Disperse Yellow 3	11855	2832–40–8	$C_{15}H_{15}O_2N_3$
8	Acid Red 26	16150	3761–53–3	$C_{18}H_{14}N_2Na_2O_7S_2$
9	Direct Black 38	30235	1937–37–1	$C_{34}H_{25}N_9Na_2O_7S_2$
10	Direct Blue 6	22610	2602–46–2	$C_{32}H_{24}N_6O_{14}S_4Na_4$
11	Direct Red 28	22120	573–58–0	$C_{32}H_{22}N_6Na_2O_6S_2$
12	Disperse Orange 11	60700	82–28–0	$C_{15}H_{11}NO_2$
13	Acid Red 114	23635	6459-9-5	$C_{37}H_{28}N_4Na_2O_{10}S_3$

a Numbering used in Tables D.1, D.4, D.5.

b Classified according to (GHS)[2] (and to CLP).[3]

c Colour index number.[4]