INTERNATIONAL STANDARD

ISO 22195-2

Second edition
2023-02

Textiles — Determination of index ingredient from coloured textile —

Part 2: Turmeric

Textiles — Détermination d'indicateurs d'ingrédients de textiles colorés —

Partie 2: Curcuma

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Foreword

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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).

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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 38, *Textiles*.

This second edition cancels and replaces the first edition (ISO 22195-2:2020), which has been technically revised.

The main changes are as follows:

— changed extraction solvent from co-solvent to single organic solvent;

— added GC-MS analysis method for the detection and qualification of index material.

A list of all parts in the ISO 22195 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.

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Introduction

There is no doubt that dyeing plays the most important role in expressing the colour of clothes. Until the invention of synthetic dyes capable of expressing diverse colours, materials obtained from nature to dye fabric have been used. Typically, colourants were obtained from plants or various materials were extracted from minerals or insects. When dyeing fabrics using materials derived from these natural substances, it becomes necessary to identify which substances the colourant was derived from. In other words, there has been a demand to confirm whether a fabric is dyed using a natural substance.

A test method is developed to identify which types of natural substances have been used.

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Textiles — Determination of index ingredient from coloured textile —

Part 2: Turmeric

1 Scope

This document specifies a test method which determines the index ingredient of chemicals in coloured fabric with turmeric.

2 Normative references

The following document is 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 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.

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 https://www.electropedia.org/

3.1
turmeric
Curcuma longa Linne
type of rhizomatous herbaceous perennial plant of the ginger family, Zingiberaceae

Note 1 to entry: Plants are gathered annually for their rhizomes and propagated from some of those rhizomes in the following season. When not used fresh, the rhizomes are boiled and then dried in ovens, after which they are ground into a deep-orange- yellow powder.

3.2
coloured
expressing of colours to textiles by dyeing or printing

3.3
natural colourant
colourant obtained from plants, wood, rocks, soil, insects or any other thing existing on earth without any chemical reaction adopted before colouring of textiles

4 Principle

Natural colourants usually contain several chemical constituents. Depending on the type of natural colourant, each contains a distinctive chemical. This characteristic chemical remains in the fabric dyed

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with natural colourant. Therefore, analysis of natural coloured fabrics by chromatography can detect characteristic chemicals depending on the kind of natural colourant.

NOTE On the other hand, if the index component curcumin is detected through this test method, it cannot be said that it is necessarily stained with turmeric alone. However, based on this principle, applying this test method to unknown coloured fabrics or textiles is useful to provide a minimum amount of information that can be used to confirm whether the fabric is coloured using turmeric.

5 Reagent

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

5.1 Water, glass double distilled water or grade 2 water complying with ISO 3696.

5.2 Methanol.

5.3 Acetonitrile.

5.4 Formic acid, volume fraction of 30 %.

5.5 Curcumin, reference standard with percentage purity indication, e.g. 95 % or more.

The Safety Data Sheet for Chemicals (FISPQ) of each reagent should be observed.

6 Apparatus

6.1 Analytical balance, resolution at 0,001 g.

6.2 Ultrasonic water bath, to be set up at (30 ± 2) °C.

6.3 Borosilicate glass container with cap, 50 ml.

6.4 Membrane filter, with 0,2 µm pore size.

6.5 Liquid chromatograph (LC) with mass spectroscopy (MS).

6.6 Gas chromatograph (GC) with mass spectroscopy (MS).

7 Procedure

7.1 Standard preparation

Stock solution of curcumin is prepared in methanol containing 1 000 mg/l.

7.2 Preparation of test specimen

Cut the test specimen into pieces of approximately 5 mm × 5 mm and approximately 1 g. Weigh it to the nearest 0,01 g, and then place it into the glass container (6.3).

Add 10 ml of methanol each into the other glass container and it poured to cut test specimen containing glass container. Place the glass container containing the test specimen into an ultrasonic bath at (30 ± 2) °C for (20 ± 1) min. Afterwards, let the extract cool down to room temperature. Dilute if necessary.

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Filter about 1 ml of the diluted solution into a HPLC or GC vial using disposable syringe equipped with a membrane filter (6.4).

7.3 Analysis

The detection and qualification of curcumin is conducted using LC-MS (6.5) with ESI mass spectrometer or GC-MS (6.6).

The recommended chromatographic conditions are given in Annex A.

7.4 Determination and calculation

7.4.1 Determination of curcumin

Comparison between analyses of standard and test specimen through 7.3 may show the result of existence of curcumin in test specimen.

Detection of curcumin may vary due to conditions of coloured test specimen. In this case, the amount of test specimen and extraction solution can be modified, and concentration of extracted solution can be adopted. The modified test specimen preparation conditions should be described in test result.

7.4.2 Calibration curve

Calibration curves with standards of curcumin at 1 mg/l, 10 mg/l, 20 mg/l, 50 ml/l, and 100 mg/l are prepared with at least 5 calibration points.

NOTE Concentration ranges for the calibration standards are subject to change upon the need of each laboratory and equipment used.

For quantification, the calibration curve shall have a correlation coefficient greater than 0,995(R^2 greater than 0,990).

7.4.3 Calculation of curcumin

Calculated the concentration of each alizarin in the test specimen, in mg/kg from Formula (1):

$$C_S = \frac{C_1}{W} \times f \times V \quad (1)$$

where

C_S is the concentration of each curcumin in the test specimen, in mg/kg;

C_1 is the concentration of each curcumin in the test specimen solution, in mg/l;

W is the mass of the test specimen, in g;

f is the dilution factor;

V is the final extraction volume, in ml.

8 Test report

The test report shall include the following information:

a) a reference to this document, i.e. ISO 22195-2:2023;

b) identification of the test specimen;

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c) the concentration of curcumin, in mg/kg

d) conditions of chromatographic analysis;

e) any deviation from the specified procedure in this document;

f) date of the test.

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Annex A
(informative)

Example of test result

A.1 Analysis of turmeric colourant

A.1.1 General

The powdered turmeric colourant was dissolved with water and methanol tested according to 7.3. The result chromatogram is shown Figure A.1.

A.1.2 Chromatographic conditions for the LC-MS

As the instrumental equipment of the laboratories may vary, no general applicable parameters can be provided for chromatographic analyses.

— Mobile Phase: (phase A) water (phase B) acetonitrile, both in 0,1 % formic acid

— Column: Kinetics EVO C18 (100 mm × 2,1 mm) 1,7 µm

— Column Oven: 35 °C

— Flow rate: 0,3 ml/min

— Injection: 2 µl

— Ionization: ESI+

— Data acquisition: scan 100 mass number to charge number (m/z) to 1 000 m/z

— Nebulizer gas flow: 1,5 l/min

— Interface temperature: 250 °C

— Interface voltage: 3 500 V

— Ratio of the eluent: 10 mg/l

See Table A.1.

Table A.1 — Condition of the mobile phase

Time (min)	Phase A, water (%)	Phase B, acetonitrile (%)
0	70	30
10	50	50
20	50	50
30	70	30

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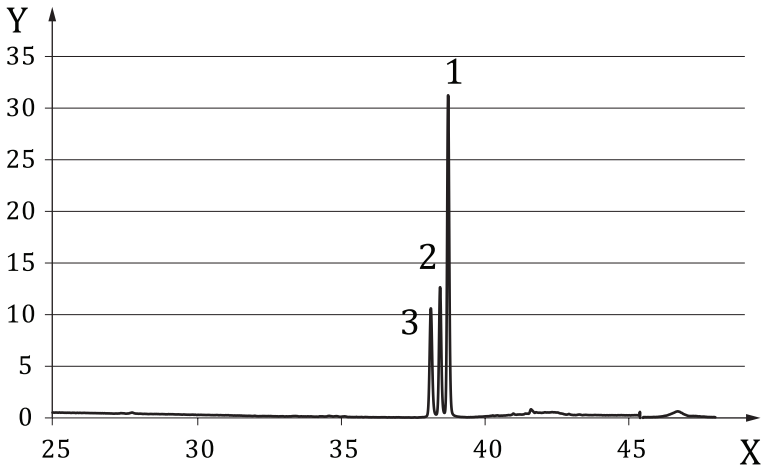

Key

X time, expressed in minutes
Y intensity
1 curcumin
2 dimethoxycurcumin
3 bis- dimethoxycurcumin

Figure A.1 — Chromatogram of turmeric colourant by LC-MS

A.1.3 Chromatographic conditions for the HPLC-DAD

The HPLC-DAD analysis is adopted to find out the specified wavelength in 430 nm by diode array detector (DAD). Its chromatographic conditions are as follows:

— Detection wavelength: 430 nm;

— Column: Phenomenex C18(2)(particle size: 5 µm, length: 150 mm, inner diameter: 4,6 mm);

— Mobile Phase: (phase A) water (phase B) acetonitrile, both in 0,1 % formic acid;

— Flow rate: 0,3 ml/min.

See Table A.2.

Table A.2 — Condition of the mobile phase

Time (min)	Phase A, water (%)	Phase B, acetonitrile (%)
0	90	10
5	90	10
35	50	50
40	5	95
43	5	95
43,1	90	10
48	90	10

A.1.4 Chromatographic conditions for the GC-MS

As the instrumental equipment of the laboratories may vary, no general applicable parameters can be provided for chromatographic analyses.

— Column: DB-EUPAH(length: 20 m, internal diameter: 0,18 nm, film thickness: 0,14 µm)

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