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IULTCS/IUC 20-1

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**Leather — Chemical tests for the
determination of certain azo colorants
in dyed leathers —**

**Part 1:
Determination of certain aromatic
amines derived from azo colorants**

*Cuir — Essais chimiques pour le dosage de certains colorants
azoïques dans les cuirs teints —*

*Partie 1: Dosage de certaines amines aromatiques dérivées des
colorants azoïques*



Reference numbers
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Contents

Page

Foreword	iv
1 Scope	1
2 Normative references	1
3 General	1
4 Principle	2
5 Safety precautions	3
6 Apparatus	3
7 Reagents	4
8 Sampling and preparation of samples	5
9 Procedure	5
9.1 Degreasing	5
9.2 Reductive cleavage	5
9.3 Liquid-liquid extraction	5
9.4 Check of the analytical system	6
10 Chromatographic analyses	6
11 Calibration	6
12 Evaluation	6
12.1 Calculation of amine in the sample	6
12.2 Reliability of the method	7
13 Test report	7
Annex A (informative) Chromatographic analyses	8
Annex B (informative) Reliability of the method	12
Annex C (informative) Assessment guide — Interpretation of analytical results	13
Bibliography	15

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).

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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 17234-1 was prepared by the Chemical Test Commission of the International Union of Leather Technologists and Chemists Societies (IUC Commission, IULTCS) in collaboration with the European Committee for Standardization (CEN) Technical Committee CEN/TC 289, *Leather*, the secretariat of which is held by UNI, in accordance with the Agreement on technical cooperation between ISO and CEN (Vienna Agreement). This method is technically similar to the method in IUC 20, which was declared an official method at the IULTCS Delegates meeting on 31st May 2003 in Cancun, Mexico.

IULTCS, originally formed in 1897, is a world-wide organization of professional leather societies to further the advancement of leather science and technology. IULTCS has three Commissions, which are responsible for establishing international methods for the sampling and testing of leather. ISO recognizes IULTCS as an international standardizing body for the preparation of test methods for leather.

This second edition of ISO 17234-1 cancels and replaces the first edition (ISO 17234-1:2010), which has been technically revised to include two new aromatic amines from the Chinese Standard GB 20400 in [Table 1](#). The layout for this part of ISO 17234 has been re-arranged and updated to be the same as ISO 17234-2.

ISO 17234 consists of the following parts, under the general title *Leather — Chemical tests for the determination of certain azo colorants in dyed leathers*:

- *Part 1: Determination of certain aromatic amines derived from azo colorants*
- *Part 2: Determination of 4-aminoazobenzene*

Leather — Chemical tests for the determination of certain azo colorants in dyed leathers —

Part 1:

Determination of certain aromatic amines derived from azo colorants

1 Scope

This part of ISO 17234 specifies a method for determining the use of certain azo colorants which can release certain aromatic amines.

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 2418, *Leather — Chemical, physical and mechanical and fastness tests — Sampling location*

ISO 3696:1987, *Water for analytical laboratory use — Specification and test methods*

ISO 4044, *Leather — Chemical tests — Preparation of chemical test samples*

ISO 17234-2, *Leather — Chemical tests for the determination of certain azo colorants in dyed leathers — Part 2: Determination of 4-aminoazobenzene*

3 General

Certain azo colorants can release, by reductive cleavage of azo group(s), one or more of the following aromatic amines, which are listed in Appendix 8 of EU Regulation 1907/2006, Annex XVII of the European Parliament, and/or the Chinese Standard GB 20400-2006.

Table 1 — Aromatic amines listed in Appendix 8 of EU regulation 1907/2006, Annex XVII, and/or GB 20400-2006

No.	CAS number	Index number	EC number	Substances
1	92-67-1	612-072-00-6	202-177-1	biphenyl-4-ylamine 4-aminobiphenyl xenylamine
2	92-87-5	612-042-00-2	202-199-1	benzidine
3	95-69-2		202-441-6	4-chloro-o-toluidine
4	91-59-8	612-022-00-3	202-080-4	2-naphthylamine
5 ^a	97-56-3	611-006-00-3	202-591-2	o-aminoazotoluene 4-amino-2',3-dimethyla- zobenzene 4-o-tolylazo-o-toluidine
6 ^a	99-55-8		202-765-8	5-nitro-o-toluidine
7	106-47-8	612-137-00-9	203-401-0	4-chloroaniline

Table 1 (continued)

No.	CAS number	Index number	EC number	Substances
8	615-05-4		210-406-1	4-methoxy-m-phenylenediamine
9	101-77-9	612-051-00-1	202-974-4	4,4'-methylenedianiline 4,4'-diaminodiphenylmethane
10	91-94-1	612-068-00-4	202-109-0	3,3'-dichlorobenzidine 3,3'-dichlorobiphenyl-4,4'-ylenediamine
11	119-90-4	612-036-00-X	204-355-4	3,3'-dimethoxybenzidine o-dianisidine
12	119-93-7	612-041-00-7	204-358-0	3,3'-dimethylbenzidine 4,4'-bi-o-toluidine
13	838-88-0	612-085-00-7	212-658-8	4,4'-methylenedi-o-toluidine
14	120-71-8		204-419-1	6-methoxy-m-toluidine p-cresidine
15	101-14-4	612-078-00-9	202-918-9	4,4'-methylene-bis-(2-chloro-aniline) 2,2'-dichloro-4,4'-methylene-dianiline
16	101-80-4		202-977-0	4,4'-oxydianiline
17	139-65-1		205-370-9	4,4'-thiodianiline
18	95-53-4	612-091-00-X	202-429-0	o-toluidine 2-aminotoluene
19	95-80-7	612-099-00-3	202-453-1	4-methyl-m-phenylenediamine
20	137-17-7		205-282-0	2,4,5-trimethylaniline
21	90-04-0	612-035-00-4	201-963-1	o-anisidine 2-methoxyaniline
22 ^b	60-09-3	611-008-00-4	200-453-6	4-aminoazobenzene
23 ^c	95-68-1	612-027-00-0	202-440-0	2,4-xylylidine 2,4-dimethylbenzene-1-amine
24 ^c	87-62-7	612-161-00-X	201-758-7	2,6-xylylidine 2,6-dimethylbenzene-1-amine

^a The CAS-numbers 97-56-3 (No. 5) and 99-55-8 (No. 6) are further reduced to CAS-numbers 95-53-4 (No. 18) and 95-80-7 (No. 19).

^b Azo colorants that are able to form 4-aminoazobenzene, generate under the condition of this method aniline and/or 1,4-phenylenediamine. The presence of these colorants shall be tested using ISO 17234-2.

^c Additional aromatic amines in the Chinese Standard GB 20400-2006

4 Principle

After degreasing, the leather sample is treated with sodium dithionite in an aqueous buffer solution (pH 6) at 70 °C in a closed vessel. The amines released in the process of reductive cleavage are transferred to a *t*-butyl methyl ether phase by means of liquid-liquid extraction using Kieselgur columns. The *t*-butyl methyl ether extract is then concentrated under mild conditions in a rotary vacuum evaporator and the residue is dissolved in a suitable solvent, depending on the method used to determine the amines.

Determination of the amines is performed by means of high-performance liquid chromatography (HPLC) using a diode array detector (DAD) or mass selective detector (HPLC/MS), capillary gas chromatography with a mass-selective detector (GC-MS) or by capillary electrophoresis with a diode array detector (CE/DAD), or qualitatively with thin layer chromatography (TLC, HPTLC).

The amines shall be identified by means of at least two different chromatographic separation methods in order to avoid any possible misinterpretations caused by interfering substances (such as position isomers of the amines to be identified) and hence any incorrect statements. Amine quantification shall be performed by HPLC/DAD or GC/MS.

5 Safety precautions

5.1 WARNING — The aromatic amines listed in [Clause 3](#) are classified as substances known to be or suspected to be human carcinogens.

Any handling and disposal of this substance shall be in strict accordance with the appropriate national health and safety regulations.

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

5.3 Good laboratory practice should be followed. Wear safety glasses in all laboratory areas, and a dust respirator and single-use gloves while handling powder colorants and aromatic amines.

5.4 Users should comply with any national and local safety regulations.

6 Apparatus

Usual laboratory equipment and, in particular, the following:

6.1 Suitable reaction vessel, of temperature-resistant glass with a gas-tight closure.

6.2 Hot cabinet with sand bath (sea sand, 0,1 mm to 0,3 mm) or **water bath** with thermostat.

6.3 Thermometer, 0,5 °C accuracy at 70 °C.

6.4 Volumetric flasks, different volumes.

6.5 Polypropylene or glass column ¹⁾ with 25 mm to 30 mm inner diameter and 140 mm to 150 mm length, glass filter at the outlet, filled with porous granulated Kieselgur.

6.6 Polypropylene or polyethylene syringe, 2 ml.

6.7 Vacuum rotary evaporator with vacuum control and water bath.

6.8 Pipettes, 10 ml, 5 ml, 2 ml, 1 ml.

6.9 Ultrasonic bath with thermostat.

1) The EXtrelut® NT20 prefilled column supplied by Merck is an example of a suitable product available commercially. This information is given for the convenience of users of this part of ISO 17234 and does not constitute an endorsement by ISO of this product. Equivalent products can be used if they can be shown to lead to the same results.

6.10 Round-bottomed flask, of 100 ml with standard ground joint NS 29/32.

6.11 Chromatographic equipment selected from the following:

6.11.1 High performance liquid chromatography (HPLC) with gradient controller and DAD or MS detector.

6.11.2 Capillary gas chromatography (GC) with mass selective detector (MS).

6.11.3 Capillary electrophoresis (CE) with DAD.

6.11.4 Thin layer chromatography (TLC) or high performance thin layer chromatography (HPTLC).

NOTE A description of the chromatographic equipment is given in [Annex A](#).

7 Reagents

Unless otherwise specified, analytical grade chemicals shall be used.

7.1 Methanol.

7.2 *t*-butyl methyl ether.

7.3 Sodium dithionite, minimum 87 % purity (mass fraction).

7.4 Aqueous sodium dithionite solution, $\rho = 200 \text{ mg/ml}^2$), freshly prepared, to be used immediately after resting for 1 h in a closed vessel.

7.5 *n*-hexane.

7.6 Amines, listed in [Table 1](#) (highest available purity standard).

7.7 Stock solution of the amines ([7.6](#)): 400 mg/l in ethyl acetate for TLC.

7.8 Stock solution of the amines ([7.6](#)): 200 mg/l in methanol for GC, HPLC, CE.

7.9 Citrate buffer solution³⁾, 0,06 mol/l, pH = 6, preheated to $(70 \pm 5) ^\circ\text{C}$.

7.10 Standard solution for amine process control: 30 μg amine per millilitre solvent, freshly prepared from stock solutions [7.7](#) or [7.8](#) depending on the analytical method.

7.11 20 % methanolic NaOH solution, 20 g NaOH dissolved in 100 ml methanol.

7.12 Water, Grade 3 according to ISO 3696:1987.

2) ρ = mass concentration.

3) The solution No. 1.09437.1 000 supplied by Merck is an example of a suitable product available commercially. This information is given for the convenience of users of this part of ISO 17234 and does not constitute an endorsement by ISO of this product. Equivalent products can be used if they can be shown to lead to the same results.

8 Sampling and preparation of samples

Sample in accordance with ISO 2418 and either grind the leather in accordance with ISO 4044 or cut into small pieces (approx. 25 mm²). If sampling in accordance with ISO 2418 is not possible (e.g. in the case of leathers from finished products like shoes, garments, etc.), details about sampling shall be given in the test report. Any traces of adhesives shall be removed mechanically.

In the case of leather patchwork fabrics with varicoloured patterns, the various colours have to be taken into account separately as far as possible. For commodities consisting of various leather qualities, specimens of the various qualities shall be analysed separately.

For the analytical procedure, accurately weigh a representative sample of more than 1,0 g of this ground leather or cut pieces in the reaction vessel (6.1).

9 Procedure

9.1 Degreasing

Treat more than 1,0 g of the leather cut into pieces or a ground leather sample in a closed 50 ml vessel (6.1) with 20 ml *n*-hexane (7.5) in an ultrasonic bath (6.9) at $(40 \pm 2)^\circ\text{C}$ for 20 min.

Decant the *n*-hexane layer from the leather sample. Any loss of leather particles during decanting shall be avoided. Directly after decanting, treat the sample again in the same way as before with 20 ml *n*-hexane. Evaporate the residual *n*-hexane at least overnight in the open vessel.

9.2 Reductive cleavage

Add a quantity of 17 ml buffer solution (7.9) preheated to $(70 \pm 5)^\circ\text{C}$ to the sample. Tightly seal the reaction vessel (6.1) and, after shaking, keep it in a ventilated oven, in a sand bath, or in a heatable bath (6.2) for (25 ± 5) min at $(70 \pm 2)^\circ\text{C}$. The reaction temperature of 70°C shall be reached inside the reaction vessel. This shall be checked with an additional vessel with thermometer inside.

Add 1,5 ml aqueous sodium dithionite solution (7.4) with a syringe (6.6) and keep the vessel at 70°C for 10 min. Afterwards, add another 1,5 ml sodium dithionite solution and heat the vessel for another 10 min. Then cool it with water to room temperature (20°C to 25°C) within 2 min.

9.3 Liquid-liquid extraction

Using a glass pestle squeeze the reaction solution out of the fibres, decant on the Kieselgur column (6.5), and allow to be absorbed by the column for 15 min.

Add 5 ml of *t*-butyl methyl ether (7.2) and 1 ml of 20 % methanolic NaOH (7.11) to the leather fibre residue in the vessel. Close the vessel, shake vigorously and transfer the solution to the Kieselgur column (6.5).

Wash the reaction vessel and fibre residues with 1×15 ml and 1×20 ml *t*-butyl methyl ether and transfer to the Kieselgur column to begin eluting the amines. Afterwards, directly flush 40 ml *t*-butyl methyl ether on the column. Collect the eluate in a 100 ml round-bottomed flask with standard ground joint (6.10).

Concentrate the *t*-butyl methyl ether extract to approximately 1 ml (not to dryness) in a rotary vacuum evaporator (6.7) in a slight vacuum at not more than 50°C . Then, evaporate the remainder of the ether to dryness using a slight flow of inert gas.

With methanol, immediately transfer the residue to a 2 ml volumetric flask (6.4) and made up to volume with methanol (or ethyl acetate for TLC analytical method). This solution is ready for the instrumental analysis, which shall be performed within 24 h.

9.4 Check of the analytical system

To check the analysis procedure, add 1,0 ml of the standard solution (7.10) to a reaction vessel (6.1) containing 16 ml of the preheated buffer solution (7.9). Then, carry out the procedure described in 9.2 and 9.3. Amine recovery rates shall comply with the following minimum requirements:

- amines No. 1 to 4, 7, 9 to 17, and 20 to 21: recovery rate 70 %;
- amine No. 8: recovery rate 20 %;
- amines No. 18, 19, 23, and 24: recovery rate 50 %;
- amines No. 5, 6, and 22, see footnotes to Table 1.

10 Chromatographic analyses

The detection of the aromatic amines can be performed using the chromatographic techniques listed in 6.11. Other validated methods can be used. The quantification of the aromatic amines is performed by means of HPLC/DAD or GC-MS; where gas chromatography is used; appropriate internal standards as described in ISO 17234-2 shall be employed.

If any amine is detected by one chromatographic method, then confirmation shall be made using one or more alternative methods. The result is positive only if both methods give a positive result.

11 Calibration

Use the standard solution (7.10) containing 30 µg amine/ml for calibration. This procedure shall be carried out with each batch of samples.

12 Evaluation

12.1 Calculation of amine in the sample

Calculate the amine concentration based on the peak areas of the individual amine components with reference to the 30 µg/ml calibration group of amines (7.10). Calculate the content of the amine as a mass fraction, w , in milligrams of the individual component per kilogram (mg/kg) of leather material according to Formula (1):

$$w = \rho_c \times \frac{A_s \times V}{A_c \times m_E} \quad (1)$$

where

ρ_c is the concentration of the amine in the calibration solution, in micrograms per millilitre (µg/ml);

A_s is the peak area of the amine in the sample solution, in area units;

A_c is the peak area of the amine in the calibration solution, in area units;

V is the volume of the specimen according to 9.3 (final sample volume), in millilitres (ml); here: 2 ml;

m_E is the mass of the leather sample, in grams (g).

12.2 Reliability of the method

For the reliability of the method, see [Annex B](#).

13 Test report

The test report shall refer to this official method and state at least the following particulars:

- a) a reference to this part of ISO 17234, i.e. ISO 17234-1;
- b) type, origin, and designation of the specimen (partial specimen, if applicable);
- c) date of receipt and date of analysis;
- d) sampling procedure;
- e) any deviations from the analytical procedure, particularly any additional steps performed;
- f) declaration of the performed separation procedure and methods used for detection and determination (a second method shall be used for confirmation of a positive result);
- g) the analytical results for the amines in milligrams per kilogram (see [Clause 12](#)), shall be individually listed and reported according to the identification threshold values as follows:

In the case of levels per amine component ≤ 30 mg/kg:

According to the analysis as carried out, azo colorants which release the listed aromatic amines were not detected.

In the case of levels per amine component > 30 mg/kg:

The analysis result suggests that the leather submitted has been manufactured or treated using azo colorants which release one or more of the listed amines.

In the case of levels of 4-aminodiphenyl and/or 2-naphthylamine > 30 mg/kg:

Use of this analytical method has detected 4-aminodiphenyl and/or 2-naphthylamine. According to the current state of knowledge it cannot be unequivocally confirmed without additional information that azo colorants which release amines were used.

NOTE Care should be taken in the interpretation of less than 30 mg/kg of amines as these can be due to false-positive results. For the interpretation of results, see [Annex C](#).

Annex A (informative)

Chromatographic analyses

A.1 Preliminary remark

As the instrumental equipment ([6.11](#)) of the laboratories can vary, no generally applicable instructions can be provided for chromatographic analyses. The following parameters have been successfully tested and used.

A.2 High-performance liquid chromatography (HPLC)

A.2.1 High-performance liquid chromatography/diode array detector (HPLC/DAD)

Eluent 1:	methanol;
Eluent 2:	0,575 g of ammonium dihydrogen phosphate + 0,7 g of disodium hydrogen phosphate in 1 000 ml of water, pH = 6,9;
Stationary phase:	LiChrospher 60 RP-select B ^a (5 µm), (250 × 4,6) mm;
Flow rate:	(0,7 to 1,0) ml/min;
Gradient:	start: 15 % eluent 1, linear increase to 80 % eluent 1 within 45 min;
Column temperature:	40 °C;
Injection volume:	10,0 µl;
Detection:	DAD, spectrograph;
Quantification:	at 240 nm, 280 nm, and 305 nm.

^a LiChrospher 60 RP-select B is an example of a suitable product available commercially. This information is given for the convenience of users of this part of ISO 17234 and does not constitute an endorsement by ISO of this product. Equivalent products can be used if they can be shown to lead to the same results.

A.2.2 High-performance liquid chromatography/mass selective detector (HPLC/DAD/MS)

Eluent 1:	acetonitrile;
Eluent 2:	ammonium acetate in 1 000 ml of water, 5 mmol, pH = 3,0;
Stationary phase:	Zorbax Eclipse XDB C18 ^b (3,5 µm); (50 × 2,1) mm;
Flow rate:	300 µl/min;
Gradient:	see Table A.1 ;
Column temperature	40 °C;
Injection volume:	2,0 µl;
Detection:	quadrupole - and/or ion-trap mass detector, scanning mode and/or MS daughter ion MS detection; DAD: for wavelengths, see A.2.1 ;
Spray gas:	nitrogen (bottled/generator);
Ionization:	API electrospray positive, fragmentor 120 V.

^b Zorbax Eclipse XDB C18 is an example of a suitable product available commercially. This information is given for the convenience of users of this document and does not constitute an endorsement by ISO of this product.

Table A.1 — Gradient programme

Time min	Eluent 1 %	Eluent 2 %
0	10	90
1,5	20	80
7,5	90	10

A.3 Capillary gas chromatography (GC-MS)

Capillary column:	medium polarity, e.g. SE 54 or DB 5, length: 50 m, inside diameter: 0,32 mm, film thickness: 0,5 µm;
Injector system:	split/splitless;
Injector temperature:	250 °C;
Carrier gas:	helium;
Temperature programme:	70 °C (2 min), 70 °C to 280 °C (at 10 °C/min), 280 °C (5 min);
Injection volume:	1,0 µl, splitless 2 min;
Detection:	MSD, scan 45 – 300 amu.

A.4 Capillary electrophoresis (CE/DAD)

250 µl of the sample solution (9.3) is mixed with 50 µl HCl ($c = 0,01$ mol/l) and passed through a membrane filter (0,2 µm). This solution is analysed by means of capillary zone electrophoresis.

Capillary 1:	56 cm, uncoated, inside diameter 50 µm, with extended light path;
Capillary 2:	56 cm, coated with polyvinyl alcohol (PVA), inside diameter 50 µm, with extended light path;
Buffer solution:	phosphate buffer solution ($c = 50$ mmol/l), pH = 2,5;
Column temperature:	25 °C;
Voltage:	30 kV;
Injection time:	4 s;
Flushing time:	5 s;
Detection:	DAD spectrograph at 214 nm, 240 nm, 280 nm, 305 nm.

A.5 Thin-layer chromatography (TLC); HPTLC or TLC only for semiquantitative confirmation

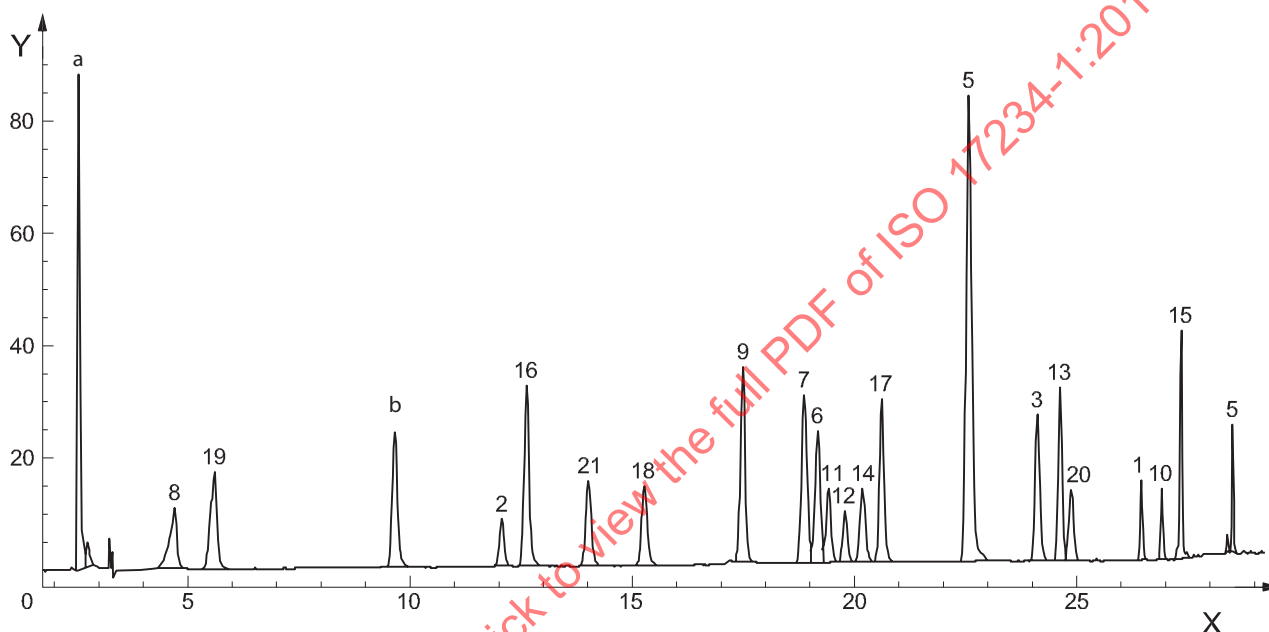
A.5.1	Plates (HPTLC):	silica gel 60 with fluorescence indicator F254, (20 × 10) cm;
	Applied volume:	5 µl, applied as a line with automatic applicator;
	Mobile solvent 1:	chloroform/acetic acid (90 + 10) parts per volume.
A.5.2	Plates (TLC):	silica gel 60, (20 × 10) cm, saturated chamber;
	Applied volume:	10,0 µl, applied as a dot with an automatic applicator;
	Mobile solvent 2:	chloroform/ethyl acetate/acetic acid (60 + 30 + 10) parts by volume;
	Mobile solvent 3:	chloroform/methanol (95 + 5) parts per volume;
	Mobile solvents 2 and 3:	successively without drying of the plates.
A.5.3	Detection:	1. ultraviolet (UV) lamp; 2. after successive treatment with reagents 2 and 3, reaction time approximately 5 min;
	Reagent 1:	0,1 % NaNO ₂ in KOH ($c = 1$ mol/l);
	Reagent 2:	0,2 % α-naphthol in KOH ($c = 1$ mol/l);
	Reagent 3:	0,5 – 1,0 % of ammonium sulphamate in methanol.

NOTE Derivatization procedure:

After developing TLC plate, it is to be dried in air or by a hand-held hot air drier (e.g. hair dryer) for a minute or two, then the plate is immersed in reagent 1 for 30 s to 1 min followed by immersing it in reagent 3 for 30 s to 1 min. The plate is dried like earlier and then immersed in reagent 2 for 1 min. The plate is dried by a hot air drier. Instead of immersion, spraying the reagents using an atomizer is also possible.

A.6 Example of HPLC chromatogram

An example of a HPLC chromatogram is shown in [Figure A.1](#).



Key

- X time in min
- Y absorbance in mAU at 240 nm
- a 1,4-phenylenediamine
- b aniline

NOTE For aromatic amines 1 to 21, see [Table 1](#) (aromatic amines 23 and 24 are not shown).

Figure A.1 — HLPC/DAD-chromatogram⁴⁾

⁴⁾ From EN 14362-1:2012, Annex A.

Annex B (informative)

Reliability of the method

The data indicated in [Table B.1](#) were obtained in an interlaboratory collaborative trial on different kinds of leathers. The data were obtained by using HPLC with DAD. The samples were ground according to ISO 4044. For liquid-liquid extraction Merck columns, type EXtrelut NT201) Part No. 11737, were used.

Table B.1 — Interlaboratory trial — Precision data

Leather sample	Detected amines	Mean mg/kg	Repeatability	Reproducibility
			mg/kg <i>r</i>	mg/kg <i>R</i>
A	Benzidine	13,5	5,4	8,4
	3,3'-Dimethoxybenzidine	15,4	4,4	6,4
	3,3'-Dimethylbenzidine	20,5	7,1	9,5
B	Benzidine	12,9	3,8	8,9
	2-Toluidine	37,5	15,4	38,5
C	3,3'-Dimethylbenzidine	25,6	8,0	17,0
	2-Toluidine	50,1	20,2	42,1
D	Benzidine	16,5	3,0	7,1