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Textiles — Qualitative and quantitative proteomic analysis of some animal hair fibres —

Part 3: Peptide detection using LC-MS without protein reduction

Textiles — Analyse protéomique qualitative et quantitative de certaines fibres animales —

Partie 3: Détection des peptides par LC-MS sans réduction protéique

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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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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 (see www.iso.org/directives).

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

A list of all parts in the ISO 20418 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.

Introduction

Cashmere is a long slender fibre obtained from cashmere goats and is expensive because of its high quality and rarity. Mislabelling or adulteration of cashmere products blended with other cheaper animal fibres such as sheep wool and yak have been repeatedly reported worldwide.

Current official methods to identify specific animal fibres are based on microscopic observations. However, the microscopy-based identification is becoming increasingly difficult due to a wider use of chemical or physical treatments in the manufacturing process. Given these issues, several other methods have also been studied either to distinguish fibre structures by the use of near-infrared spectroscopy or terahertz spectroscopy, or to distinguish DNA sequences by the use of polymerase chain reaction. Nevertheless, each method has shown some complications when applied. Therefore, it is required to develop novel identification methods.

Animal fibres consist mainly of proteins called keratins and some associated proteins. Therefore, the most promising methods to identify fibres are based on the analysis of proteins contained in textiles. Commonly, proteins are analysed by being subjected to digestion by trypsin, resulting in smaller molecules, i.e. peptides, which will be later characterized through mass spectrometry. Accordingly, identification methods using either matrix-assisted laser desorption/ionization time-of-flight mass spectrometer or liquid chromatography/mass spectrometer (LC-MS) have been studied. When comparing these options, the latter type of instrument is less expensive and more readily available in testing laboratories as a versatile analytical instrument than the former. Moreover, LC-MS has a high quantitative capability, and is therefore preferable to calculate the blending ratio of animal fibres.

Keratins are highly insoluble due to the disulphide bonds they tend to form, both at an intramolecular as well as at an intermolecular level. Thus, keratins are generally extracted in the presence of reducing agents. However, this reducing step is considered as time-consuming and arduous. In this document, an alternative method in which cysteine-free peptides are selected for identification markers is used, thereby eliminating the need of the reducing step and enabling rapid preparation of LC-MS samples.

Both ISO 20418-1 and this document describe procedures using LC-MS, but they differ regarding the method utilized to extract the peptides. In ISO 20418-1, proteins are first extracted from fibres with a thiourea/urea/dithiothreitol (DTT) solution, and then digested by trypsin to obtain peptides. In the process described here, peptides are directly extracted by trypsin digestion of mechanically powdered fibres. The method has been shown to be useful even for highly processed samples and is applicable to various types of animal hairs such as goat (cashmere or mohair), wool and yak.

Textiles — Qualitative and quantitative proteomic analysis of some animal hair fibres —

Part 3:
Peptide detection using LC-MS without protein reduction

1 Scope

This document specifies a qualitative and quantitative procedure to determine the composition of animal hair fibre blends (made of wool, cashmere, yak, alpaca, camel or angora) by LC-MS without protein reduction.

NOTE 1 The composition of non-animal hair fibres can be measured by ISO 1833 (all parts). Both results are combined to determine the total fibre composition.

The method is based on a preliminary identification, by light microscopy, of all fibres in the blend on the basis of their morphology, according to ISO/TR 11827[4]. It is not applicable if fibres of the same animal species (such as blends of cashmere and mohair) are present.

NOTE 2 In this case, the quantitative analysis is performed using microscopical analysis [for example, ISO 17751 (all parts)].

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 1833-1, *Textiles — Quantitative chemical analysis — Part 1: General principles of testing*

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

ISO 17751 (all parts), *Textiles — Quantitative analysis of cashmere, wool, other specialty animal fibers and their blends*

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 http://www.electropedia.org/

3.1
animal hair fibre
type of keratin fibre for textile use, such as wool, cashmere, yak, alpaca, camel or angora

3.2
Bovidae
biological family of cloven-hoofed, ruminant mammals including cashmere goat, sheep and yak