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Screening of genetically modified organisms (GMOs) in cotton and textiles

Criblage pour la détection des organismes génétiquement modifiés (OGM) dans le coton et les textiles

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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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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 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.

International Workshop Agreement IWA 32 was approved at a workshop hosted by the Netherlands Standardization Institute (NEN), in association with the Organic Cotton Accelerator, held in New Delhi, India, in January 2019.

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

0.1 General

This purpose of this document is to provide guidance to laboratories worldwide to assess, in a standardized way, whether cotton, cotton fibre and/or cotton-derived materials are produced from, or contain materials from, genetically modified (GM) cotton plants. This document is intended for non-GM cotton and textiles production lines, but it can be applied to any production line that wants to check the presence of GM cotton.

0.2 Protocol

The GM screening protocol described in this document is based on Polymerase Chain Reaction (PCR)-based methods, as these methods are the minimal set of DNA-based methods to cover all known GM-cotton events. The protocol is written for and tested to work on all four of the major commercial cotton species: *Gossypium hirsutum*, *G. barbadense*, *G. arboreum G. herbaceum*.

Cotton (*Gossypium* spp.) has been cultivated for lint for over 8 000 years. There are over 50 species in the *Gossypium* genus (Wendel et al., 2009). The *Gossypium* genome is complex, containing 2,25 to 2,43 gigabase (Arumuganathan and Earle, 1991). While GM-cotton cultivation covers a large part of global cotton production today, there are countries where the cultivation of GM cotton is not allowed by law as well as voluntary private and/or public standards that do not allow the intentional use of genetically modified organisms (GMOs) in the cotton and textile production process. This creates a need for an adequate and harmonized protocol on the screening of cotton and cotton-derived materials for the potential presence of GM-cotton related sequences.

This document describes a procedure to screen seed, leaf and (processed) fibre samples in the cotton production chain for the potential presence of GM-related DNA elements. The protocol describes three major steps:

a) an effective way to isolate DNA from cotton materials;

b) a method to confirm that the isolated DNA consists of amplifiable cotton DNA, i.e. suitable for PCR, preferably a low copy nuclear target;

c) A screening method consisting of a minimum set of detection methods covering all the currently known GM cotton events, to be performed on the cotton DNA isolate.

If the results of the screening methods described in this protocol are 'not detected', the likelihood that the cotton sample is (at least partly) derived from GM cotton is minimal, based on the ability of the screening methods to detect elements and constructs of the GM cotton events. GM cotton levels below the detection limit of the method or unknown GM cotton events that do not contain any of the elements or the construct tested cannot be determined by this detection method. When one or more screening methods indicate that GM elements are present, the sample should be considered as derived from GM cotton.

Further investigation for the identification of GM-cotton events present in the sample is not part of this document as such, but some guidance is provided in Annex A as to how further identification of the related cotton events can be achieved.

0.3 Structure

The structure of this document is illustrated in Figure 1. Clause 4 describes the principle of the screenings protocol. Clause 5 describes sample preparation for different types of material. Clause 6 describes the DNA isolation method that allows for successful DNA isolation from the respective cotton-related products. Clause 7 describes the DNA quality control for the different cotton species. Clause 8 describes the screening of GM-related DNA sequences in a cotton sample. Clause 9 describes recommendations on the test report (outcome). Annex A gives an overview of known GMO cotton events. Annex B gives an overview of detection methods applied by RIKILT[1)]. Annex C provides

1) https://www.wur.nl/en/Research-Results/Research-Institutes/rikilt.htm

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more information on the inhouse validation as carried out by RIKILT. Annex D provides a list of the contributos to the International Workshop.

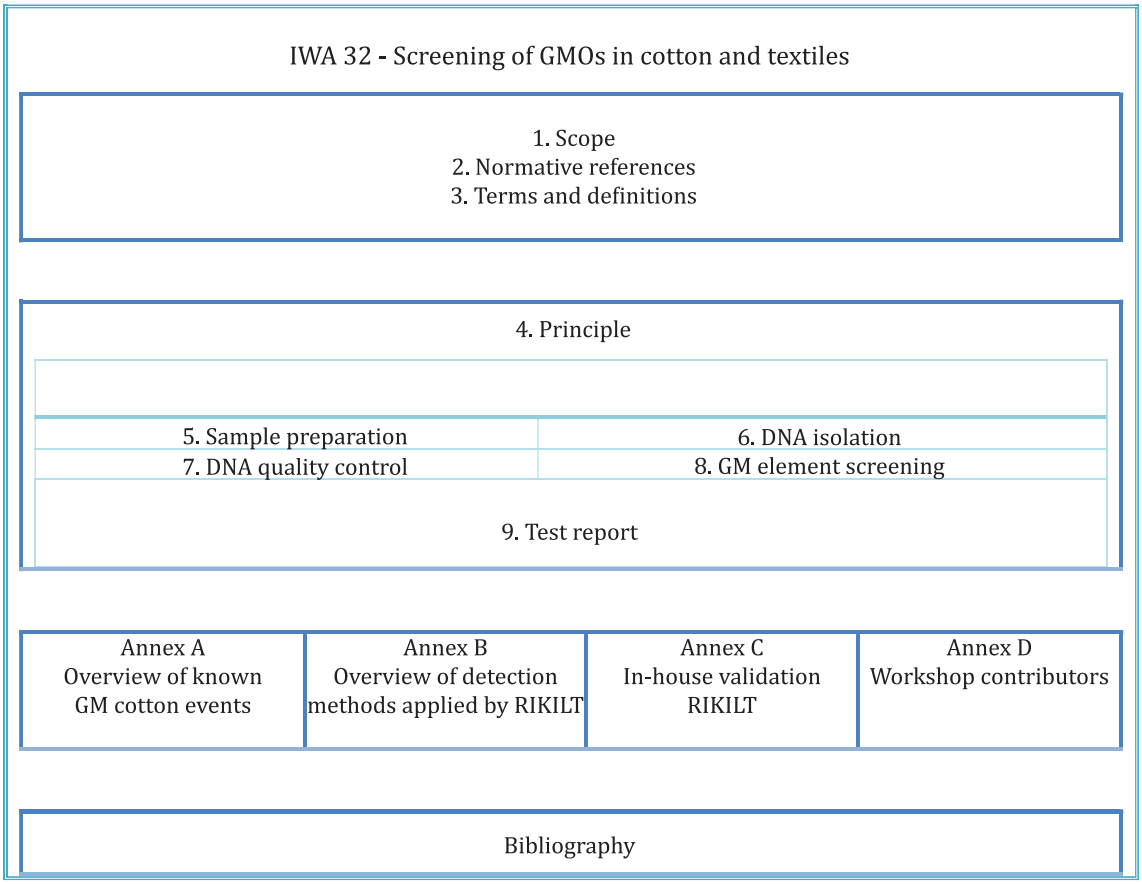

Figure 1 — Structure of this document

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Screening of genetically modified organisms (GMOs) in cotton and textiles

WARNING — The method described in this document implies the use of reagents that pose a hazard to health. This document does not claim to address all associated safety problems. It is the responsibility of the user of this document to take appropriate measures for health and safety protection.

1 Scope

This document provides requirements and recommendations to laboratories that perform genetically modified organism (GMO) analyses in cottonseed, leaf, cotton fibre and cotton fibre-derived materials.

The following are within the scope of this document:

a) identifying the materials to be assessed, based on the probability of obtaining good quality, fit for purpose DNA from the materials in subsequent steps in the cotton cloth production process;

b) specifying a method for efficient DNA isolation from cotton and cotton-derived materials described under point a);

c) specifying the cotton-specific method(s) to be used as control for amplifiable DNA;

d) specifying the screening procedure that provides optimal chances to detect GMOs as a result of the performance of the lowest number of genetically modified (GM) element screening assays.

NOTE 1 The protocol allows for the screening of all currently known GM cotton events and is set up in a way that optimizes the probability of also detecting unknown GM cotton events that possibly contain similar DNA sequences. Further information is given in CEN/TS 16707.

Sampling is outside of the scope of this document.

NOTE 2 A recommended sampling method is given in ISO 6497. General guidance for the sampling of bulk materials or for cotton-based products is available in standards such as ASTM D1441-12 and CEN/TS 15568.

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 21570:2005, *Foodstuffs — Methods of analysis for the detection of genetically modified organisms and derived products — Quantitative nucleic acid based methods*

ISO 21571, *Foodstuffs — Methods of analysis for the detection of genetically modified organisms and derived products — Nucleic acid extraction*

ISO 24276:2006, *Foodstuffs — Methods of analysis for the detection of genetically modified organisms and derived products — General requirements and definitions*

3 Terms and definitions

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

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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
cottonseed
seed from cotton plants

3.2
cotton leaf
leaves from the cotton plant

3.3
seed cotton
raw cotton that contains both the seed and the fibre before it has been ginned

3.4
cotton lint
raw fibre that has gone through the ginning process

3.5
greige yarn
unprocessed long continuous length of interlocked cotton lint that results from the cleaning and subsequent spinning of the cotton lint

3.6
greige fabric
unprocessed textiles formed by weaving, knitting or crocheting the yarn and non-wovens

3.7
processed yarn
yarn that has undergone processing, to develop its full textile potential

3.8
processed fabric
fabric that has undergone processing, to develop its full textile potential

4 Principle

This document describes a method for the screening of GMO in cotton and textiles. The screening is based on realtime PCR methods which depends on obtaining good quality amplifiable DNA. Good quality DNA samples (those fit for purpose) are defined as those where the amplification of an endogenous cotton gene (positive control) is observed. The amplification and detection of endogenous cotton is achieved through isolation methods that result in good quality DNA, applied to cotton and textiles, while the targeted amplification of six genetic elements can allow for the detection of GM-cotton in these samples.

NOTE Experimental results have shown that good quality DNA can be isolated from the production stages of cottonseed up to greige yarn and greige fabric, while it showed not to be possible to isolate amplifiable DNA in processed yarn and processed fabric. Processed yarn and processed fabric are therefore excluded from this protocol. See Clause C.3 for the assessment of isolation of good quality DNA at different cotton production stages by RIKILT.

5 Sample preparation

Homogenize the sample using suitable methods and avoiding excessive heating.

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