
**Horizontal methods for molecular
biomarker analysis — Methods
of analysis for the detection of
genetically modified organisms and
derived products —**

**Part 2:
Construct-specific real-time PCR
method for detection of event FP967
in linseed and linseed products**

*Méthodes horizontales d'analyse moléculaire de biomarqueurs —
Méthodes d'analyse pour la détection des organismes génétiquement
modifiés et des produits dérivés —*

*Partie 2: Méthode PCR en temps réel spécifique de la construction
pour la détection d'un événement FP967 dans les graines de lin et les
produits à base de graines de lin*



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Contents

Page

Foreword	iv
1 Scope	1
2 Normative references	1
3 Terms and definitions	1
4 Principle	1
5 Reagents and materials	2
5.1 PCR reagents	2
6 Apparatus	3
6.1 General	3
6.2 PCR device	3
7 Sampling	3
8 Procedure	3
8.1 Test sample preparation	3
8.2 Preparation of the DNA extracts	3
8.3 DNA extraction	3
8.4 PCR setup	3
8.5 Temperature time programme	4
9 Accept/reject criteria	4
9.1 General	4
9.2 Identification	5
10 Validation status and performance criteria	5
10.1 Robustness of the method	5
10.2 Intralaboratory trial	5
10.3 Collaborative trial	5
10.4 Sensitivity	7
10.5 Specificity	7
11 Test report	8
Bibliography	9

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.

International Standards are drafted in accordance with the rules given in the ISO/IEC Directives, Part 2.

The main task of technical committees is to prepare International Standards. Draft International Standards adopted by the technical committees are circulated to the member bodies for voting. Publication as an International Standard requires approval by at least 75 % of the member bodies casting a vote.

In other circumstances, particularly when there is an urgent market requirement for such documents, a technical committee may decide to publish other types of document:

- an ISO Publicly Available Specification (ISO/PAS) represents an agreement between technical experts in an ISO working group and is accepted for publication if it is approved by more than 50 % of the members of the parent committee casting a vote;
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An ISO/PAS or ISO/TS is reviewed after three years in order to decide whether it will be confirmed for a further three years, revised to become an International Standard, or withdrawn. If the ISO/PAS or ISO/TS is confirmed, it is reviewed again after a further three years, at which time it must either be transformed into an International Standard or be withdrawn.

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.

ISO/TS 21569-2 was prepared by Technical Committee ISO/TC 34, *Food products*, Subcommittee SC 16, *Horizontal methods for molecular biomarker analysis*.

ISO/TS 21569 consists of the following parts, under the general title *Horizontal methods for molecular biomarker analysis — Methods of analysis for the detection of genetically modified organisms and derived products*:

- *Part 2: Construct-specific real-time PCR method for detection of event FP967 in linseed and linseed products*

Horizontal methods for molecular biomarker analysis — Methods of analysis for the detection of genetically modified organisms and derived products —

Part 2:

Construct-specific real-time PCR method for detection of event FP967 in linseed and linseed products

1 Scope

This method describes a procedure for the detection of a DNA sequence present in a genetically modified linseed (*Linum usitatissimum*) line (event FP967, also named as “CDC Triffid”). For this purpose, extracted DNA is used in a real-time PCR and the genetic modification (GM) is specifically detected by amplification of a 105 bp DNA sequence representing the transition between the nopaline synthase gene terminator (*Thos*) from *Agrobacterium tumefaciens* and the dihydrofolate reductase gene (*dfrA1*) from a Class 1 integron of *Escherichia coli*.

The method described is applicable for the analysis of DNA extracted from foodstuffs. It may also be suitable for the analysis of DNA extracted from other products such as feedstuffs and seeds. The application of this method requires the extraction of an adequate amount of amplifiable DNA from the relevant matrix for the purpose of analysis.

2 Normative references

ISO 21569, *Foodstuffs — Methods of analysis for the detection of genetically modified organisms and derived products — Qualitative nucleic acid based methods*

ISO 21571:2005, *Foodstuffs — Methods of analysis for the detection of genetically modified organisms and derived products — Nucleic acid extraction*

ISO 24276, *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 terms and definitions given in ISO 24276 apply.

4 Principle

DNA is extracted from the test sample applying a suitable method. The DNA analysis consists of two parts:

- Verification of the amount, quality and amplifiability of the extracted DNA, e.g. by means of a target taxon specific real-time PCR with primers amplifying a 68 bp long fragment from the linseed-specific (*Linum usitatissimum*) stearoyl-acyl carrier protein desaturase 2 gene (SAD) (Reference [1]).
- Detection of the *Thos-dfr* construct in a real-time PCR (Reference [1]).