
**Milk and milk powder —
Determination of aflatoxin M₁ content
— Clean-up by immunoaffinity
chromatography and determination
by high-performance liquid
chromatography**

*Lait et lait en poudre — Détermination de la teneur en aflatoxine
M₁ — Purification par chromatographie d'immunoaffinité et
détermination par chromatographie en phase liquide à haute
performance*





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Forewords

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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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 34, *Food products*, Subcommittee SC 5, *Milk and milk products*, and the International Dairy Federation (IDF), in collaboration with the European Committee for Standardization (CEN) Technical Committee CEN/TC 302, *Milk and milk products - Methods of sampling and analysis*, in accordance with the Agreement on technical cooperation between ISO and CEN (Vienna Agreement). It is being published jointly by ISO and IDF.

This third edition cancels and replaces the second edition (ISO 14501 | IDF 171:2007), which has been technically revised. The main changes compared with the previous edition are as follows:

- the lack of detailed explanation in some clauses was leading to variability in the way the method was executed from one laboratory to another. Practical information from skilled end users familiar with routine analysis using this protocol was taken into account in this revision to clarify those ambiguous clauses.

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.

IDF (the International Dairy Federation) is a non-profit private sector organization representing the interests of various stakeholders in dairying at the global level. IDF members are organized in National Committees, which are national associations composed of representatives of dairy-related national interest groups including dairy farmers, dairy processing industry, dairy suppliers, academics and governments/food control authorities.

ISO and IDF collaborate closely on all matters of standardization relating to methods of analysis and sampling for milk and milk products. Since 2001, ISO and IDF jointly publish their International Standards using the logos and reference numbers of both organizations.

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Milk and milk powder — Determination of aflatoxin M₁ content — Clean-up by immunoaffinity chromatography and determination by high-performance liquid chromatography

CAUTION 1 — The method described in this document requires the use of solutions of aflatoxin M₁. Aflatoxins are carcinogenic to humans. Attention is drawn to the statement made by the International Agency for Research on Cancer (WHO)^{[1][2]}.

CAUTION 2 — Adequately protect the laboratory in which the analyses are performed from daylight and keep aflatoxin M₁ standard solutions protected from light, e.g. by using aluminium foil.

CAUTION 3 — The use of non-acid-washed glassware (e.g. tubes, vials, flasks, beakers, syringes) for aqueous aflatoxin solutions can cause loss of aflatoxin M₁. Moreover, brand new laboratory glassware, before coming into contact with aqueous solutions of aflatoxin M₁, should be soaked in dilute acid (e.g. sulfuric acid, $c = 2 \text{ mol/l}$) for several hours, then rinsed well with distilled water to remove all traces of acid (check to ensure pH is in the range 6 to 8).

CAUTION 4 — Use decontamination procedures for laboratory wastes such as solid compounds, solutions in organic solvents, aqueous solutions and spills, and for glassware coming into contact with carcinogenic materials. Suitable decontamination procedures have been developed and validated by the International Agency for Research on Cancer (WHO)^{[1][2]}.

1 Scope

This document specifies a method for the determination of aflatoxin M₁ content in milk and milk powder. The lowest level of validation is 0,08 µg/kg for whole milk powder, i.e. 0,008 µg/l for reconstituted liquid milk. The limit of detection (LOD) is 0,05 µg/kg for milk powder and 0,005 µg/kg for liquid milk. The limit of quantification (LOQ) is 0,1 µg/kg for milk powder and 0,01 µg/kg for liquid milk.

The method is also applicable to low-fat milk, skimmed milk, low-fat milk powder and skimmed milk powder.

2 Normative references

There are no normative references in this document.

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:

- IEC Electropedia: available at <http://www.electropedia.org/>
- ISO Online browsing platform: available at <https://www.iso.org/obp>

3.1

aflatoxin M₁ content

concentration or mass fraction of substances determined by the procedure specified in this document

Note 1 to entry: Concentration is expressed in µg/l and mass fraction is expressed in µg/kg.