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# Milk and milk products — Determination of nitrogen content

Part 4: Determination of protein and non-protein nitrogen content and true protein content calculation (Reference method)



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AS ISO 8968.4:2021

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**Part 4: Determination of protein and non-protein  
nitrogen content and true protein content  
calculation (Reference method)**

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## Preface

This Standard was prepared by the Standards Australia Committee FT-024, Food Products.

The objective of this document is to specify a method for the direct and indirect determination of the protein nitrogen content of liquid, whole or skimmed milk.

This document is identical with, and has been reproduced from, ISO 8968-4:2016, *Milk and milk products — Determination of nitrogen content — Part 4: Determination of protein and non-protein nitrogen content and true protein content calculation (Reference method)* which was jointly published by ISO and the International Dairy Federation as IDF 20-4:2016.

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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](http://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](http://www.iso.org/patents)).

Any trade name used in this document is information given for the convenience of users and does not constitute an endorsement.

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](#)

The committee responsible for this document is ISO/TC 34, *Food products*, Subcommittee SC 5, *Milk and milk products* and the International Dairy Federation (IDF). It is being published jointly by ISO and IDF.

This second edition of ISO 8968-4|IDF 20-4 cancels and replaces ISO 8968-4|IDF 20-4:2001 and ISO 8968-5|IDF 20-5:2001, which have been technically revised.

ISO 8968|IDF 20 consists of the following parts, under the general title *Milk and milk products — Determination of nitrogen content*:

- *Part 1: Kjeldahl principle and crude protein calculation*
- *Part 3: Block-digestion method (Semi-micro rapid routine method)<sup>1)</sup>*
- *Part 4: Determination of protein and non-protein nitrogen content and true protein content calculation (Reference method)*

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1) It is intended that upon revision, the main element of the title of ISO 8968-3|IDF 20-3 (i.e. "Milk") will be aligned with the main element of the titles of ISO 8968-1|IDF 20-1 and ISO 8968-4|IDF 20-4.

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.

Attention is drawn to the possibility that some of the elements of this document may be the subject of patent rights. IDF 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](http://www.iso.org/patents)).

Any trade name used in this document is information given for the convenience of users and does not constitute and endorsement

ISO 8968-4|IDF 20-4 was prepared by the IDF Standing Committee on *Analytical methods for composition* and ISO Technical Committee ISO/TC 34, *Food products*, Subcommittee SC 5, *Milk and milk products*.

The work was carried out by the IDF/ISO Project Group (C13) of the Standing Committee on *Analytical methods for composition* under the aegis of its project leaders, D. Barbano (CH) and P. Trossat (FR).

This ISO|IDF International Standard cancels and replaces ISO 8968-4|IDF 20-4:2001 and ISO 8968-5|IDF 20-5:2001, which have been technically revised.

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# Australian Standard®

## Milk and milk products — Determination of nitrogen content

### Part 4: Determination of protein and non-protein nitrogen content and true protein content calculation (Reference method)

**WARNING** — The use of this part of ISO 8968|IDF 20 can involve the use of hazardous materials, operations and equipment. This part of ISO 8968|IDF 20 does not purport to address all the safety risks associated with its use. It is the responsibility of the user of this part of ISO 8968|IDF 20 to establish appropriate safety and healthy practices and determine the applicability of local regulatory limitations prior to use.

#### 1 Scope

This part of ISO 8968|IDF 20 specifies a method for the direct and indirect determination of the protein nitrogen content of liquid, whole or skimmed milk.

#### 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 8968-1|IDF 20-1, *Milk and milk products — Determination of nitrogen content — Part 1: Kjeldahl principle and crude protein calculation*

#### 3 Terms and definitions

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

##### 3.1

##### **non-protein nitrogen content**

##### **NPN**

mass fraction of substances determined by the specified procedure

Note 1 to entry: The non-protein nitrogen content is expressed as a percentage by mass.

##### 3.2

##### **protein nitrogen content**

mass fraction of substances determined by the specified procedure, directly or, alternatively, indirectly

Note 1 to entry: The protein nitrogen content is expressed as a percentage by mass.

#### 4 Principle

##### 4.1 Indirect protein nitrogen

Precipitation of protein from a test portion by addition of trichloroacetic acid solution such that the final concentration of trichloroacetic acid in the mixture is approximately 12 %. Removal of the precipitated milk protein by filtration, with the remaining filtrate containing the non-protein nitrogen components. Determination of the nitrogen content of the filtrate by the procedure described in ISO 8968-1|IDF 20-1.

Where the total nitrogen content of the milk sample has previously been determined, the true protein nitrogen content may be calculated as the difference between the total nitrogen content and the non-protein nitrogen content.