BS ISO 15163:2012

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Milk and milk products — Calf rennet and adult bovine rennet — Determination by chromatography of chymosin and bovine pepsin contents

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National foreword

This British Standard is the UK implementation of ISO 15163:2012.

The UK participation in its preparation was entrusted to Technical Committee AW/5, Chemical analysis of milk and milk products.

A list of organizations represented on this committee can be obtained on request to its secretary.

This publication does not purport to include all the necessary provisions of a contract. Users are responsible for its correct application.

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Milk and milk products — Calf rennet and adult bovine rennet — Determination by chromatography of chymosin and bovine pepsin contents

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Contents

Page

Foreword iv
Foreword v
Introduction vi
1 Scope 1
2 Normative references 1
3 Principle 1
4 Reagents 2
5 Apparatus 3
6 Sampling 4
7 Procedure 4
7.1 Check 4
7.2 Preparation of a fresh column with Fractogel 4
7.3 Regeneration and equilibration of the Fractogel resin in the column 4
7.4 Storage of the column with Fractogel 5
7.5 Preparation of test sample 5
7.6 Analysis of the desalted rennet 6
7.7 Determination of the clotting times 7
8 Calculation and expression of results 8
8.1 Calculation of the activity of chymosin and pepsin, expressed as a percentage 8
8.2 Calculation of active chymosin and active bovine pepsin, in milligrams per litre 9
8.3 Expression of results 10
9 Precision 10
9.1 Interlaboratory test 10
9.2 Repeatability 10
9.3 Reproducibility 11
10 Test report 11
Annex A (informative) **Qualitative determination by double immunodiffusion of milk-coagulating enzymes in commercial coagulants** 12
Annex B (informative) **Interlaboratory test** 17
Bibliography 19

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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.

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.

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 15163 | IDF 110 was prepared by Technical Committee ISO/TC 34, *Food products*, Subcommittee SC 5, *Milk and milk products and* the International Dairy Federation (IDF). It is being published jointly by IDF and ISO.

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Foreword

IDF (the International Dairy Federation) is a non-profit organization representing the dairy sector worldwide. IDF membership comprises National Committees in every member country as well as regional dairy associations having signed a formal agreement on cooperation with IDF. All members of IDF have the right to be represented on the IDF Standing Committees carrying out the technical work. IDF collaborates with ISO in the development of standard methods of analysis and sampling for milk and milk products.

The main task of Standing Committees is to prepare International Standards. Draft International Standards adopted by the Standing Committees are circulated to the National Committees for endorsement prior to publication as an International Standard. Publication as an International Standard requires approval by at least 50 % of IDF National Committees casting a vote.

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.

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All work was carried out by an ISO-IDF Project Group on *Chymosin and bovine pepsin determination,* of the Standing Committee on *Analytical methods for processing aids and indicators,* under the aegis of its project leader, Prof. A. Andrén (SE).

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Introduction

Calf rennet and adult bovine rennet preparations contain both chymosin and bovine pepsin in various amounts as main clotting enzymes. The proportion of chymosin decreases relative to pepsin in the abomasum (the fourth “true” stomach) with age and at weaning of the calf.

The ratio of abomasa from young cattle to that of old cattle in the raw material for rennet production thus highly influences the composition of chymosin and pepsin in the final rennet. The higher the abomasa from young milk-fed calves, the higher the proportion of chymosin and vice versa[5][6].

Both chymosin and pepsin have special characteristics relevant to milk-clotting activity and suitability for cheese making. The milk-clotting activity of pepsin is, for example, much more pH-dependent than chymosin and pepsin also has a more general proteolytic activity than chymosin.

Therefore, it is very important to analyse the content of chymosin and pepsin in addition to the strength (total milk-clotting activity) of the rennet[6][7].

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INTERNATIONAL STANDARD

ISO 15163:2012(E)
IDF 110:2012(E)

Milk and milk products — Calf rennet and adult bovine rennet — Determination by chromatography of chymosin and bovine pepsin contents

1 Scope

This International Standard specifies a reference method for the determination of the amounts of chymosin and bovine pepsin present in a test sample of calf rennet and adult bovine rennet. In addition, it can be used for mixtures of calf/bovine rennet with fermentation-produced bovine chymosin (FPC).

2 Normative references

The following referenced document is indispensable for the application 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 11815 | IDF 157:2007, *Milk — Determination of total milk-clotting activity of bovine rennets*

3 Principle

As a first step, the rennet sample is desalted and the enzymes chymosin and bovine pepsin separated on an anion exchange column[8][9]. In a second step, the milk-clotting activity of each of the separated two enzymes is determined by ISO 11815 | IDF 157 (reconstituted milk with pH 6,5). The enzymatic composition of the rennet sample is expressed in percentage chymosin activity and percentage pepsin activity of the sum of the activities in International Milk-Clotting Units (IMCU) of both components, or the results are expressed in milligrams per litre of active chymosin and milligrams per litre of active pepsin.

The total milk-clotting activity of the first batch of calf rennet reference standard powder and the first batch of adult bovine rennet reference standard powder has once and for all been set at 1 000 IMCU/g. Future preparations of reference standards shall be set relative to the previous reference standards (see ISO 11815 | IDF 157).

This International Standard specifies both a manual set-up of the anion exchange chromatography and an alternative automated set-up.

This is a reference method and changes may therefore only be made if confirmed to give the same result, and repeatability and reproducibility at least as high as the original standard method. Any change to what is stated in this International Standard method shall also be mentioned in the test report (see Clause 10).

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