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### **Wheat and wheat flour – Gluten content – Part 2: Determination of wet gluten by mechanical means (ISO 21415-2:2006)**

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EUROPEAN STANDARD

**EN ISO 21415-2**

NORME EUROPÉENNE

EUROPÄISCHE NORM

January 2008

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ICS 67.060

English Version

## Wheat and wheat flour - Gluten content - Part 2: Determination of wet gluten by mechanical means (ISO 21415-2:2006)

Blé et farines de blé - Teneur en gluten - Partie 2:  
Détermination du gluten humide par des moyens  
mécaniques (ISO 21415-2:2006)

Weizen und Weizenmehl - Glutengehalt - Teil 2:  
Bestimmung des Feuchtglutens durch mechanische  
Verfahren (ISO 21415-2:2006)

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**Management Centre: rue de Stassart, 36 B-1050 Brussels**

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## **Foreword**

The text of ISO 21415-2:2006 has been prepared by Technical Committee ISO/TC 34 "Agricultural food products" of the International Organization for Standardization (ISO) and has been taken over as EN ISO 21415-2:2008 by Technical Committee CEN/TC 338 "Cereal and cereal products", the secretariat of which is held by AFNOR.

This European Standard shall be given the status of a national standard, either by publication of an identical text or by endorsement, at the latest by July 2008, and conflicting national standards shall be withdrawn at the latest by July 2008.

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### **Endorsement notice**

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## **Introduction**

The alternative techniques specified in this part of ISO 21415 and in ISO 21415-1 for the isolation of wet gluten (i.e. washing out by hand and mechanical washing out) do not usually give equivalent results. The reason is that, for complete development of gluten structure it is necessary to rest the dough. Therefore, the result obtained by hand washing is usually higher than that obtained by mechanical washing, mainly in the case of wheat which has high gluten content. Consequently, the test report should always indicate the technique used.



# Wheat and wheat flour — Gluten content —

## Part 2: Determination of wet gluten by mechanical means

### 1 Scope

This part of ISO 21415 specifies a method for the determination of the wet gluten content of wheat flour (*Triticum aestivum* L. and *Triticum durum* Desf.) by mechanical means. This method is directly applicable to flour. It is also applicable to semolina and wheat after grinding, if their particle size distribution meets the specification given in Table B 1.

### 2 Normative references

The following referenced documents are 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 712, *Cereals and cereal products — Determination of moisture content — Routine reference method*

### 3 Terms and definitions

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

#### 3.1

##### **wet gluten**

visco-elastic substance, composed principally of two protein fractions (gliadin and glutenin) in hydrated form, obtained as specified in this part of ISO 21415 or ISO 21415-1

#### 3.2

##### **ground wheat**

product of small-scale milling of whole wheat which meets the particle size distribution shown in Table B.1.

#### 3.3

##### **semolina**

coarsely milled wheat endosperm

#### 3.4

##### **flour**

finely milled wheat endosperm with a particle size of less than 250 µm

### 4 Principle

Dough is prepared from a sample of flour or reground semolina, or ground wheat and a solution of sodium chloride, in the chamber of the equipment. The wet gluten is separated by washing the dough with sodium chloride solution, followed by removal of excess washing solution by centrifugation. The residue is weighed.

## 5 Reagents

Use only reagents of recognized analytical grade, unless otherwise specified, and distilled or demineralized water or water of equivalent purity.

### 5.1 Sodium chloride solution, 20 g/l.

Dissolve 200 g of sodium chloride (NaCl) in water then dilute it to 10 l. The temperature of the solutions when used should be  $22\text{ °C} \pm 2\text{ °C}$ .

It is advisable to prepare this solution freshly every day.

### 5.2 Potassium iodide/iodine solution (Lugol's solution).

Dissolve 2,54 g of potassium iodide (KI) in water. Add 1,27 g of iodine (I<sub>2</sub>) to this solution and, after complete dissolution of the integral parts, dilute to 100 ml with water.

## 6 Apparatus

Usual laboratory apparatus and, in particular, the following.

**6.1 Automatic apparatus for gluten separation**<sup>1)</sup>, single or double, consisting of washing chamber(s), kneader(s) (see Figures A.1 and A.2) and a dispensing device under electronic control for gluten separation.

**6.1.1 Washing chamber(s)**, equipped with interchangeable chromed sieve holder(s) and with 88 µm aperture polyester sieves or 80 µm aperture metal sieves, and 840 µm aperture polyamide sieves or 800 µm aperture metal sieves.

**6.1.2 Kneader hook**, at a distance from the chromed sieve holder of  $0,7\text{ mm} \pm 0,05\text{ mm}$ . This value should be checked using the stamped metal plates supplied.

**6.1.3 Plastic container**, of capacity 10 l, containing a reservoir of sodium chloride solution (5.1) connected to the equipment by plastic tubing.

**6.1.4 Dispensing device**, consisting of a peristaltic pump, to deliver the sodium chloride solution (5.1) for gluten washing at a constant flow rate of 50 ml/min to 56 ml/min.

For a detailed description of the equipment and for detailed operating instructions, users of this part of ISO 21415 should consult the manufacturer's manual for the equipment employed.

**6.2 Adjustable dispenser**, for the sodium chloride solution, capable of dispensing 3 ml to 10 ml with an accuracy of  $\pm 0,1\text{ ml}$ .

**6.3 Centrifuge**, capable of maintaining a rotational frequency of  $6\ 000 \pm 5$  per minute and of producing a radial acceleration of 2 000 g, equipped with perforated plates having apertures 500 µm in diameter.

**6.4 Balance**, capable of weighing to the nearest 0,01 g.

**6.5 Spatula**, made of stainless steel.

**6.6 Beakers**, of capacity 500 ml (to collect washings).

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1) Glutomatic equipment (types 2100 and 2200) produced by Perten Instruments AB (Sweden) is the most widely used mechanical means for this purpose. This information is given for the convenience of users of this part of ISO 21415 and does not constitute an endorsement by ISO of this equipment. Other equipment may also be used if it gives similar results to Glutomatic equipment or to the method specified in ISO 21415-1.

### 6.7 Metal tweezers.

**6.8 Small-scale mill**, capable of producing ground product with a particle size that meets the requirements given in Table B.1.

## 7 Sampling

A representative sample should have been sent to the laboratory. It should not have been damaged or changed during transport or storage.

Sampling is not a part of the method specified in this part of ISO 21415. A recommended sampling method is given in ISO 6644 or ISO 13690.

## 8 Preparation of test sample

Homogenize the samples and determine their moisture content in accordance with ISO 712. Prior to the gluten content measurement, grind the wheat grain and semolina using a small-scale mill (6.8) as specified in Annex B. To avoid significant changes in the moisture content of the samples, take special care during grinding and storage.

## 9 Procedure

### 9.1 General

The operations of preparation and washing of the dough are carried out as a continuous process by the automatic apparatus (6.1). Work in accordance with the manufacturer's operating instructions for the apparatus in use.

### 9.2 Test portion

Weigh 10 g of the test sample to the nearest 0,01 g and transfer it quantitatively to the washing chamber of the apparatus (6.1.1). Make sure that the washing chamber is equipped with the appropriate sieve, which has been previously cleaned and moistened.

Fine (88 µm aperture) polyester or (80 µm aperture) metal sieve(s) is/are used for testing flour and reground semolina samples. When testing ground wheat, chromed sieve holder(s) with an indented ring around the base is/are also required with 840 µm aperture polyamide or 800 µm metal sieve(s). The aperture size of the sieve(s) used should be indicated in the test report.

Shake the chamber gently to spread the flour evenly.

### 9.3 Dough preparation

As a suggested starting point, add 4,8 ml of the sodium chloride solution (5.1) to the test sample using the adjustable dispenser (6.2). Direct the stream of salt solution against the side wall of the chamber so that it does not go through the sieve. Shake the washing chamber gently so that the salt solution is spread evenly over the flour.

It may be necessary to adjust the amount of salt solution used to accommodate high and low gluten content or very weak gluten samples. Where difficulties in mixing cohesive dough are experienced (the chamber is flooded during the washing period), the amount of added salt solution should be reduced (minimum of 4,2 ml). Where a very strong, tough gluten is formed during mixing, the amount of solution may be increased to 5,2 ml.