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**INTERNATIONAL STANDARD**



**5531**

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INTERNATIONAL ORGANIZATION FOR STANDARDIZATION • МЕЖДУНАРОДНАЯ ОРГАНИЗАЦИЯ ПО СТАНДАРТИЗАЦИИ • ORGANISATION INTERNATIONALE DE NORMALISATION

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## **Wheat flour — Determination of wet gluten**

*Farines de blé — Détermination du gluten humide*

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## FOREWORD

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International Standard ISO 5531 was developed by Technical Committee ISO/TC 34, *Agricultural food products*, and was circulated to the member bodies in October 1976.

It has been approved by the member bodies of the following countries :

Australia	France	Portugal
Austria	Germany	Romania
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Bulgaria	India	Thailand
Canada	Israel	Turkey
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The member body of the following country expressed disapproval of the document on technical grounds :

Belgium

This International Standard is based on Standard No. 106 of the International Association for Cereal Chemistry (ICC).

# Wheat flour – Determination of wet gluten

## 0 INTRODUCTION

The alternative techniques specified in this International Standard for isolation of the wet gluten, i.e. washing out by hand and mechanical washing out, do not give equivalent results. Consequently the test report shall always indicate the technique used. In the case of mechanical washing out, the type of machine used shall also be indicated.

## 1 SCOPE AND FIELD OF APPLICATION

1.1 This International Standard specifies a method for the determination of wet gluten in wheat flour.

1.2 This method is applicable to wheat flours (commercial and experimental flours) but not to the coarse whole meal of wheats.

## 2 REFERENCE

ISO 2170, *Cereals and pulses – Sampling of milled products*.

## 3 DEFINITION

**wet gluten in wheat flour**: A plastic-elastic substance, consisting of gliadin and glutenin, obtained by the method specified in this International Standard.

## 4 PRINCIPLE

Preparation of dough from a sample of flour and a buffered solution of sodium chloride. Isolation of the wet gluten by washing this dough with a buffered solution of sodium chloride, followed by removal of excess washing solution and weighing of the residue.

## 5 REAGENTS

The reagents shall be of recognized analytical quality. The water used shall be distilled water or water of at least equivalent purity.

5.1 **Sodium chloride**, 20 g/l solution, buffered to pH 6,2.

Dissolve 200 g of sodium chloride in water; add 7,54 g of potassium dihydrogen phosphate ( $\text{KH}_2\text{PO}_4$ ) and 2,46 g of disodium hydrogen phosphate dihydrate ( $\text{Na}_2\text{HPO}_4 \cdot 2\text{H}_2\text{O}$ ).

Dilute to 10 litres with water.

Prepare a fresh solution daily.

5.2 **Iodine**, approximately 0,001 N solution.

## 6 APPARATUS

Ordinary laboratory equipment and in particular :

6.1 **Porcelain mortar**, glazed inside, or **enamelled metal vessel** of diameter 10 to 15 cm.

6.2 **Burette**, 10 ml, graduated in 0,1 ml, complying with the requirements of ISO/R 385.

6.3 **Spatula**, of horn, plastic or stainless steel, 18 to 20 cm in length.

6.4 **Glass plate**, about 40 cm × 40 cm, with slightly roughened surface.

6.5 **Gloves** of thin rubber and having a smooth surface.

6.6 **Wooden frame**, about 30 cm × 40 cm, covered with No. 56 grit gauze (mesh 315 μm) (for washing out by hand).

6.7 **Gluten washing machine** (for mechanical washing out).

6.8 **Container** with adjustable outflow, for the sodium chloride solution (5.1) used for washing out (8.3).

6.9 **Gluten press**.

6.10 **Stop-clock**.

6.11 **Balance**, accurate to 0,01 g.

## 7 SAMPLING

See ISO 2170.

## 8 PROCEDURE

### 8.1 Test portion

Weigh, to the nearest 0,01 g, 10,00 g of the test sample and transfer it quantitatively to the mortar or metal vessel (6.1).

### 8.2 Preparation of dough

8.2.1 Add, drop by drop 5,5 ml of the sodium chloride solution (5.1) from the burette (6.2) while continuously stirring the flour with the spatula (6.3).

8.2.2 After adding the sodium chloride solution, compress the mixture with the spatula and form a dough ball, taking care to avoid loss of flour. Dough residues adhering to the wall of the vessel or to the spatula shall be collected with the dough ball.

8.2.3 To homogenize, roll out the ball to a length of 7 to 8 cm with the flat of the hand on the roughened glass plate (6.4), then fold it.

During this operation the hands shall be covered with rubber gloves (6.5) in order to protect the dough from warmth and perspiration from the hands.

8.2.4 Repeat this operation (8.2.3) five times.

### 8.3 Washing out

Washing out may be carried out either using mechanical washing followed by hand washing (8.3.2) or, if a gluten washing machine is not available, entirely by hand washing (8.3.1).

#### 8.3.1 Washing out by hand

8.3.1.1 The operations described in 8.3.1.2 and 8.3.1.3 shall be carried out over the wooden frame covered with gauze (6.6) to avoid the possible loss of dough.

8.3.1.2 Take the dough ball (8.2) in the hand and allow the sodium chloride solution (5.1) to drip onto it from the container (6.8) at a rate such that 750 ml flow in 8 min. During this time, successively roll out the dough ball, flatten it, stretch it to make two pieces, then mould them together into one piece; repeat these operations seven times.

8.3.1.3 The washing out time depends on the gluten content, but in general is about 8 min.

#### 8.3.2 Mechanical washing out

8.3.2.1 Place the dough ball (8.2) in the gluten washing machine (6.7) and moisten it with a few drops of the

sodium chloride solution (5.1) from the container (6.8). Wash the dough in the machine, in accordance with the manufacturer's instructions, for 10 min with the sodium chloride solution. Use about 400 ml of the solution for this operation.

8.3.2.2 The mechanical washing out shall be followed by hand washing, which generally does not last longer than 2 min.

### 8.4 Verification of completeness of washing

The washing is considered to be complete when more sodium chloride solution (5.1) pressed out from the gluten ball obtained as in 8.3.1 or 8.3.2 contains only traces of starch. Use the iodine solution (5.2) for detection of starch.

### 8.5 Removal of excess washing solution

8.5.1 Eliminate most of the washing solution adhering to the gluten ball by holding it between the fingers of one hand and compressing it briefly three times.

8.5.2 Form the gluten ball into a laminate shape and place it in the gluten press (6.9).

Close the gluten press and re-open it after 5 s; transfer the gluten leaf, without deforming it, to another dry spot in the press and press it again. Repeat this operation 15 times; dry the glass plates of the gluten press after each operation.

### 8.6 Determination of the mass of the wet gluten

Weigh the pressed gluten to the nearest 0,01 g.

### 8.7 Number of determinations

Carry out two determinations on the same test sample.

## 9 EXPRESSION OF RESULTS

### 9.1 Method of calculation and formula

The wet gluten, expressed as a percentage by mass of the original product, is equal to

$$\frac{m \times 100}{10} = 10 m$$

where  $m$  is the mass of wet gluten (8.6).

NOTE – Generally the result of the determination is not referred to the dry matter content.

Take as the result the mean of the two determinations provided that the requirement for repeatability (see 9.2) is satisfied. If it is not, carry out a third determination on the same test sample and take as the result the mean of the three determinations if the difference between the lowest and highest values obtained does not exceed 1 % wet gluten. If the difference exceeds 1 %, carry out a fourth determination on the same test sample and take as the result the mean of all four values obtained.

**9.2 Repeatability**

The difference between the results of two determinations carried out simultaneously or in rapid succession by the same analyst using the same apparatus shall not exceed 0,5 % wet gluten.

technique : mechanical or manual washing out; type of apparatus, etc.) and the result obtained. It shall also mention all operating details not specified in this International Standard, or regarded as optional, as well as any circumstances which may have influenced the result.

**10 TEST REPORT**

The test report shall show the method used (including the

The report shall include all details necessary for the complete identification of the sample.

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