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# INTERNATIONAL STANDARD **ISO** 2294



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## **Meat and meat products — Determination of total phosphorus content (Reference method)**

*Viandes et produits à base de viande — Détermination de la teneur en phosphore (Méthode de référence)*

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## FOREWORD

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Draft International Standards adopted by the Technical Committees are circulated to the Member Bodies for approval before their acceptance as International Standards by the ISO Council.

International Standard ISO 2294 was drawn up by Technical Committee ISO/TC 34, *Agricultural food products*, and circulated to the Member Bodies in April 1971.

It has been approved by the Member Bodies of the following countries :

Austria	France	Poland
Belgium	Germany	Portugal
Brazil	Hungary	South Africa, Rep. of
Bulgaria	India	Spain
Chile	Ireland	Thailand
Czechoslovakia	Israel	Turkey
Denmark	Netherlands	United Kingdom
Egypt, Arab Rep. of	New Zealand	

This International Standard has also been approved by the Association of Official Analytical Chemists (AOAC).

No Member Body expressed disapproval of the document.

# Meat and meat products – Determination of total phosphorus content (Reference method)

## 1 SCOPE AND FIELD OF APPLICATION

This International Standard specifies a reference method for the determination of the total phosphorus content of meat and meat products.

## 2 REFERENCES

ISO/R 936, *Meat and meat products – Determination of ash*.

ISO 3100, *Meat and meat products – Sampling*.<sup>1)</sup>

## 3 DEFINITION

**total phosphorus content of meat and meat products** : The phosphorus content determined by the procedure described, and expressed as a percentage by mass of phosphorus pentoxide.

## 4 PRINCIPLE

Mineralization of a test portion with sulphuric and nitric acids. Precipitation of the phosphorus as quinoline phosphomolybdate. Drying and weighing of the precipitate.

An alternative method of mineralization is described in clause 10.

## 5 REAGENTS

All reagents shall be of recognized analytical reagent quality. Distilled water or water of equivalent purity shall be used in the test.

**5.1 Sulphuric acid**,  $\rho_{20}$  1,84 g/ml.

**5.2 Nitric acid**,  $\rho_{20}$  1,40 g/ml.

**5.3 Precipitating reagent**

**5.3.1** Dissolve 70 g of sodium molybdate dihydrate ( $\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$ ) in 150 ml of water.

**5.3.2** Dissolve 60 g of citric acid monohydrate [ $\text{CH}_2(\text{CO}_2\text{H})\text{COH}(\text{CO}_2\text{H})\text{CH}_2(\text{CO}_2\text{H}) \cdot \text{H}_2\text{O}$ ] in 150 ml of water and add 85 ml of nitric acid (5.2).

**5.3.3** Gradually add solution 5.3.1 to solution 5.3.2, while stirring.

**5.3.4** To 100 ml of water add successively 35 ml of nitric acid (5.2) and 5 ml of distilled quinoline.

Gradually add this solution to the mixture 5.3.3, while stirring. Leave for 24 h at room temperature.

Filter, add 280 ml of acetone and dilute to 1 000 ml with water.

Store the reagent in a well-stoppered plastics bottle in the dark.

## 6 APPARATUS

Usual laboratory equipment not otherwise specified, and

**6.1 Mechanical meat mincer**, laboratory size, fitted with a plate with holes of diameter not exceeding 4 mm.

**6.2 Analytical balance**.

**6.3 Kjeldahl flask**, 250 ml capacity, or a **long-necked round-bottom flask**.

**6.4 Heating device**, on which the flask (6.3) can be heated in an inclined position in such a way that the source of heat only touches the part of the wall of the flask which is below the level of the liquid. For heating by gas, a suitable device is a plate of asbestos provided with a circular hole, such that only the lower part of the flask is exposed to the flame.

**6.5 Suction device**, to remove the acid fumes evolved during the digestion.

**6.6 Fritted glass filter**, pore diameter 5 to 15  $\mu\text{m}$  (P. 16).

1) At present at the stage of draft.

**6.7 Electrically heated drying oven**, with temperature control, which can be adjusted to  $260 \pm 20$  °C.

**6.8 Conical suction flask**.

**6.9 Desiccator**, provided with an effective desiccant.

**6.10 Pasteur pipette**.

## 7 SAMPLE

Proceed from a representative sample of at least 200 g. See ISO 3100.

Store the sample in such a way that deterioration and change in composition are prevented.

## 8 PROCEDURE

### 8.1 Preparation of test sample

Make the sample homogeneous by passing it at least twice through the meat mincer (6.1) and mixing. Keep the homogeneous sample in a completely filled, air-tight, closed container and store it in such a way that deterioration and change in composition are prevented. Analyse the sample as soon as possible, but in any case within 24 h, according to the method given in 8.2 to 8.4, or clause 10.

If the sample is not immediately analyzed after passage through the mincer, liquid separation may occur on standing. Therefore, homogenize the test sample thoroughly with a fork immediately before taking the test portion.

### 8.2 Test portion

Weigh, to the nearest 0,001 g, about 3 g of the test sample into the flask (6.3). See also the note to 8.4.

### 8.3 Mineralization

Add 20 ml of nitric acid (5.2) and some glass beads or boiling chips.

Place the flask in an inclined position (at an angle of about 40° from the vertical) on the heating device (6.4). Heat for 5 min, cool, and then add 5 ml of sulphuric acid (5.1).

Heat the flask gently until the foaming has ceased. Then heat somewhat more strongly. As soon as the mixture starts to carbonize, add more nitric acid by means of a Pasteur pipette (6.10) and continue the heating. Repeat the operation until the evolution of brown fumes has ceased.

Finally, when the liquid has become colourless, heat until white fumes appear.

Cool, add 15 ml of water and boil gently for 10 min, minimizing the evaporation of water (for example by means of a pear-shaped glass bulb inserted in the neck of the flask).

Transfer the liquid quantitatively into a 250 ml beaker or conical flask, rinsing the flask (6.3) with water. Add 10 ml of nitric acid. The total liquid volume should then be about 50 ml.

### 8.4 Determination

Add 50 ml of precipitating reagent (5.3) to the liquid in the beaker or conical flask.

Cover with a watch glass and boil for 1 min on a hotplate placed in the suction device (6.5).

Allow to cool to room temperature; during cooling, swirl the contents three or four times.

Filter under suction through the fritted glass filter (6.6), which has been previously heated for 30 min at a temperature of  $260 \pm 20$  °C, cooled in the desiccator (6.9) and weighed to the nearest 1 mg.

Wash the precipitate on the filter five times with 25 ml portions of water, using this water, at the same time, to wash any remaining precipitate from the conical flask onto the filter.

Dry in the oven (6.7) at a temperature of  $260 \pm 20$  °C for 1 h.

Cool in the desiccator (6.9) and weigh to the nearest 1 mg.

Carry out two determinations on the same test sample.

NOTE — If the mass of the dried precipitate is more than 750 mg, repeat the analysis with a smaller test portion.

### 8.5 Blank test

Carry out a blank test in parallel with the analysis itself, using the same procedure and the same quantities of all the reagents, but omitting the test portion.

## 9 EXPRESSION OF RESULTS

### 9.1 Method of calculation and formula

Calculate the phosphorus content, expressed as percentage phosphorus pentoxide by mass, by means of the formula

$$0,032\ 07 \times m_1 \times \frac{100}{m_0} = 3,207 \frac{m_1}{m_0}$$

where

$m_0$  is the mass, in grams, of the test portion;

$m_1$  is the mass, in grams, of the quinoline phosphomolybdate precipitate.

Take as the result the arithmetic mean of the two determinations, provided that the conditions of repeatability are satisfied (see 9.2).

Report the result to the nearest 0,01 g of phosphorus pentoxide per 100 g of sample.

**9.2 Repeatability**

The difference between the results of two determinations carried out simultaneously or in rapid succession by the same analyst shall not be greater than 0,02 g of phosphorus pentoxide per 100 g of sample.

**10 NOTE ON PROCEDURE**

If desired, the mineralization can be carried out by incineration using the method described in ISO/R 936. Then proceed as follows :

Take up the ash in 15 ml of concentrated nitric acid (5.2), using a stirring rod to aid dissolution. Transfer the liquid to a 250 ml conical flask. Wash the ashing dish and the stirring

rod several times with water, and add the washings to the contents of the conical flask. Dilute to about 75 ml.

Heat for 30 min on a boiling water bath. Allow to cool, and proceed according to 8.4.

**11 TEST REPORT**

The test report shall show the method used and the result obtained. It shall also mention any operating conditions not specified in this International Standard, or regarded as optional, as well as any circumstances that may have influenced the result.

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



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