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Fruit and vegetable products — Determination of ethanol content

Produits dérivés des fruits et légumes — Détermination de la teneur en éthanol

Reference number
ISO 2448:1998(E)

Foreword

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International Standard ISO 2448 was prepared by Technical Commitee ISO/TC 34, *Agricultural food products*, Subcommittee SC 3, *Fruit and vegetable products.*

This second edition cancels and replaces the first edition (ISO 2448:1973), which has been technically revised.

Annex A forms an integral part of this International Standard.

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Fruit and vegetable products — Determination of ethanol content

1 Scope

This International Standard specifies a method for the chemical determination of ethanol in fruit and vegetable products.

The method is not applicable to products containing more than 5 % (*m/m*) of ethanol.

In the case of products containing essential oils, it is necessary to remove the latter (see annex A).

2 Definition

For the purposes of this International Standard, the following definition applies.

2.1
ethanol content
mass of the substances determined by the method specified in this International Standard, expressed as a percentage by mass for solid products or as grams per 100 ml for liquid products

3 Principle

Separation of the ethanol by distillation, followed by oxidation by potassium dichromate in a sulfuric acid medium. Determination of the excess dichromate by ammonium iron(II) sulfate in the presence of iron(II)-1,10-phenanthroline as indicator.

4 Reagents

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

4.1 **Sulfuric acid** (H_2SO_4), ρ_{20} = 1,84 g/ml.

4.2 **Dilute sulfuric acid,** ρ_{20} = 1,49 g/ml, containing 500 ml of sulfuric acid (4.1) per litre.

4.3 **Calcium hydroxide** [$Ca(OH)_2$], suspension obtained by shaking 110 g to 112 g of calcium oxide in 1 litre of water.

4.4 **Potassium dichromate solution,** containing 42,572 g of $K_2Cr_2O_7$ per litre. 1 ml of this solution is equivalent to 0,01 g of ethanol.

4.5 **Potassium permanganate *solution*,** containing 1,372 g of $KMnO_4$ per litre. 10 ml of this solution is equivalent to 1 ml of ammonium iron(II) sulfate solution (4.6).

4.6 **Ammonium iron(II) sulfate solution** [$(NH_4)_2Fe(SO_4)$].

Dissolve 170,2 g of ammonium iron(II) sulfate hexahydrate in water. Add 20 ml of sulfuric acid (4.1) and dilute to 1 litre with water.

Stabilize by the addition of aluminium chips. 2 ml of this solution is equivalent to 1 ml of potassium dichromate solution (4.4).

4.7 Iron(II)-1,10-phenanthroline solution.

Dissolve 0,695 g of iron(II) sulfate heptahydrate ($FeSO_4 \cdot 7H_2O$) in 100 ml of water. Add 1,485 g of 1,10-phenanthroline monohydrate ($C_{12}H_8N_2 \cdot H_2O$) and heat to aid solution.

The solution keeps well.

5 Apparatus

Usual laboratory apparatus and, in particular, the following.

5.1 Distillation apparatus, comprising a 500 ml flask surmounted by a fractionation column and a condenser ending in a slightly tapered extension piece long enough to reach to the bottom of a 100 ml volumetric flask.

Any other steam distillation apparatus may be used, provided that it satisfies the following criterion.

No loss of ethanol greater than 0,02 % shall occur in the course of a distillation; i.e. 200 ml of a 10 % (*V/V*) ethanol/water mixture distilled five times in succession shall contain at least 9,9 % of ethanol after the last distillation.

5.2 Heating apparatus, which does not cause even slight decomposition of the extractable materials contained in the flask.

5.3 Volumetric flasks, of capacity 100 ml.

5.4 Pipettes, of capacities 5 ml, 10 ml and 20 ml.

5.5 Burettes, of capacity 50 ml, fitted with a stopcock.

5.6 Wide-necked flasks, of capacity 250 ml, fitted with ground glass stoppers, clean, dry, free from grease and airtight (absolute tightness can be ensured with the aid of a polytetrafluorethylene sleeve).

5.7 Blender.

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

6 Sampling

It is important the laboratory receive a sample which is truly representative and has not been damaged or changed during transport or storage.

Sampling is not part of the method specified in this International Standard. As there is no specific International Standard dealing with fruit and vegetable products, it is recommended that the parties concerned come to an agreement on the subject.

7 Preparation of test sample

7.1 Solid or thick products (purée, marmalade or jam, fruits, vegetables)

Blend mechanically the whole of the sample supplied. Take care that the temperature of the product does not rise. Take a sufficient quantity of the product to enable two parallel determinations to be carried out.

7.2 Liquid products (juices, pulps and syrups)

Thoroughly mix the sample. Take a sufficient quantity of the product to enable two parallel determinations to be carried out.

8 Procedure

NOTE If it is required to check whether the repeatability requirement (clause 10) is met, carry out two determinations in accordance with 8.2.1 to 8.2.3.

8.1 Test portion

Weigh, to the nearest 0,01 g, a mass of the prepared sample, or take a volume of it such that the quantity of ethanol collected in 100 ml of distillate is less than 1 g.

8.2 Determination

8.2.1 Distillation

Dilute the test portion with about 50 ml of water and transfer it quantitatively to the flask of the distillation apparatus (5.1). Rinse the vessel used to take the test portion with not more than 120 ml of water, collecting the rinsing water in the flask.

Make the product slightly alkaline (pH 8 ± 0,2) with the calcium hydroxide suspension (4.3), shaken before use.

Add glass beads or pieces of porcelain to control the rate of boiling. Pour 10 ml of water into a 100 ml volumetric flask (5.3) and insert the end of the tapered extension of the distillation apparatus so that it is immersed in the liquid.

Distil the diluted test portion (previously made alkaline) in such a way that the distillate, when it reaches the volumetric flask, is at a relatively low temperature (15 °C to 20 °C).

Collect about 80 ml to 85 ml of distillate. Stop the distillation and rinse the condenser and extension with a few millilitres of water.

Shake the volumetric flask to mix the contents. If necessary, immerse the flask in cold water (15 °C to 20 °C) for a few minutes.

Dilute the contents of the volumetric flask to the mark with water and shake.

8.2.2 Oxidation

See note

Pour 20 ml (V_1) of potassium dichromate solution (4.4), accurately measured (see note), and 20 ml of dilute sulfuric acid (4.2) into a 250 ml flask with a ground glass stopper (5.6) and shake.

Add 10 ml (V_0) of distillate (8.2.1), accurately measured. Stopper the flask, moistening the stopper with a drop of sulfuric acid (4.1). Shake the flask and wait for at least 30 min, shaking the flask from time to time.

The resultant mixture should in no case assume the green coloration of the chromium cation as this would indicate that the ethanol content of the test portion was too high. If this occurs, recommence the oxidation taking a smaller amount of distillate (for example 5 ml). If necessary, recommence both distillation and oxidation taking a smaller test portion. Take account of any such changes in the calculations.

If the test portion contains too little ethanol, a smaller amount of potassium dichromate solution (4.4) may be used, i.e. 10 ml or 5 ml of this solution, diluted with 10 ml or 15 ml of distilled water, respectively. Take account of any such change in the calculations.

8.2.3 Titration

Titrate the excess of dichromate, using the ammonium iron(II) sulfate solution (4.6). The excess of dichromate should be at least equal to 20 % of the quantity used for the blank test. Shake the flask after each addition.

When the colour changes to greenish blue, add 4 drops of iron(II)-1,10-phenanthroline solution (4.7). (Another appropriate coloured indicator may be used, at the discretion of the operator, see note.) Continue the addition of the ammonium iron(II) sulfate solution (4.6) until the colour of the medium changes from greenish blue to brown. If the endpoint is passed, return to it precisely by adding potassium permanganate solution (4.5).

Deduct, from the volume of the ammonium iron(II) sulfate solution used, one-tenth of the volume of potassium permanganate solution added. Let V_2 be the volume remaining after this deduction, which represents the exact volume of ammonium iron(II) sulfate solution equivalent to the excess potassium dichromate.

NOTE — The titration of the excess of dichromate with the solution of ammonium iron(II) sulfate (4.6) can also be carried out in the presence of a mixture of

- 1 ml of orthophosphoric acid (85 %), $\rho_{20} = 1{,}71$ g/ml; and
- 1 ml of a solution of barium diphenylaminesulfonate containing 0,5 g per 100 ml.

Carry out two determinations on the same prepared sample.

8.2.4 Blank test

Carry out a blank test under the same conditions as for the titration, replacing the volume, V_0, of distillate by the same volume of distilled water. Let V_3 be the volume of ammonium iron(II) sulfate solution (4.6) used, following the procedure of 8.2.2 and 8.2.3.

9 Expression of results

9.1 Solid products

The ethanol content, expressed as a percentage by mass, is equal to

$$0{,}01\ V_1 \times \frac{V_3 - V_2}{V_3} \times \frac{100}{V_0} \times \frac{100}{m}$$

where

m is the mass, in grams, of the test portion;

V_0 is the volume, in millilitres, of distillate taken for the titration;

V_1 is the volume, in millilitres, of potassium dichromate solution used for the oxidation;

V_2 is the volume, in millilitres, of ammonium iron(II) sulfate solution used for the back titration of the dichromate;

V_3 is the volume, in millilitres, of ammonium iron(II) sulfate solution used in the blank test.

9.2 Liquid products

The ethanol content, expressed in grams per 100 ml of product, is equal to

$$0{,}01\ V_1 \times \frac{V_3 - V_2}{V_3} \times \frac{100}{V_0} \times \frac{100}{V_4}$$

where

V_0, V_1, V_2 and V_3 have the same meaning as in 9.1;

V_4 is the volume, in millilitres, of the test portion.

10 Repeatability

The absolute difference between two independent single test results, obtained using the same method on identical test material in the same laboratory by the same operator using the same equipment within a short interval of time, will in not more than 5 % of cases be greater than 1 % of the arithmetic mean of the two results.

11 Test report

The test report shall specify:

- all information necessary for the complete identification of the sample;
- the sampling method used, if known;
- the test method used, together with reference to this International Standard;
- all operating details not specified in this International Standard, or regarded as optional, together with details of any incidents which may have influenced the test result(s);
- the test result(s) obtained;
- if the repeatability has been checked, the final quoted result obtained.

Annex A
(normative)

Modified procedure for products containing essential oils

In the presence of essential oils, the distillate is turbid, with drops of essential oil floating on the surface. In such cases the procedure shall be modified as follows.

Collect the distillate in a 100 ml volumetric flask and allow it to stand for 2 h. Dilute to the mark with water, the interface between the two phases (essential oil and water) being at the level of the mark. Allow to stand for a further 1 h to 2 h. Discard the small quantity of essential oil collected on the surface, either by suction with a fine pipette or by filtration through paper in a covered funnel.

Transfer the still-turbid filtrate to a 150 ml flask and add 10 g of polystyrene granules (granule size 1 mm to 2 mm). Shake the stoppered flask for 15 min and then filter the mixture through gauze in a covered funnel. The liquid should then have become clear and have lost its odour almost completely.

Proceed with the determination on this liquid.

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