
**Essential oils — Determination of content
of phenols**

Huiles essentielles — Détermination de la teneur en phénols

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Printed in Switzerland

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 3.

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 International Standard may be the subject of patent rights. ISO shall not be held responsible for identifying any or all such patent rights.

International Standard ISO 1272 was prepared by Technical Committee ISO/TC 54, *Essential oils*.

This second edition cancels and replaces the first edition (ISO 1272:1973), which has been technically revised.

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Essential oils — Determination of content of phenols

1 Scope

This International Standard specifies a method for the determination of the percentage, by volume, of phenols in essential oils.

NOTE See 8.2.2 which establishes some limits to the application of this method.

2 Normative references

The following normative documents contain provisions which, through reference in this text, constitute provisions of this International Standard. For dated references, subsequent amendments to, or revisions of, any of these publications do not apply. However, parties to agreements based on this International Standard are encouraged to investigate the possibility of applying the most recent editions of the normative documents indicated below. For undated references, the latest edition of the normative document referred to applies. Members of ISO and IEC maintain registers of currently valid International Standards.

ISO 356:1996, *Essential oils — Preparation of test sample*.

ISO 648, *Laboratory glassware — One-mark pipettes*.

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ISO 1773, *Laboratory glassware — Narrow-necked boiling flasks*.

3 Principle

The phenolic compounds contained in a known volume of an essential oil are transformed into their alkaline phenol esters, which are soluble in the aqueous phase. The volume of the unabsorbed portion of the essential oil is measured.

4 Reagents

Unless indicated otherwise, use only reagents of recognized analytical grade, and distilled or demineralized water or water of equivalent purity.

4.1 **Tartaric acid**, pulverized.

4.2 **Potassium hydroxide**, free from silica and alumina, 1 mol/l aqueous solution.

4.3 **Xylene**, free from impurities soluble in the potassium hydroxide solution (4.2).

5 Apparatus

Usual laboratory apparatus and, in particular, the following.

5.1 **One-mark pipettes**, of 2 ml and 10 ml capacities, conforming to class A of ISO 648.

5.2 **Conical flasks**, of 100 ml capacity, conforming to ISO 1773.

5.3 **Cassia flask**, of 150 ml capacity, with a neck graduated in 0,1 ml, the graduated portion being of 10 ml capacity and at least 150 mm in length.

The zero graduation mark shall be a little above the base of the cylindrical portion of the neck. The angle made by the conical wall with the vertical shall be about 30° (see Figure 1).

5.4 **Glass rod**, of very small diameter.

Dimensions in millimetres
Graduations in millilitres

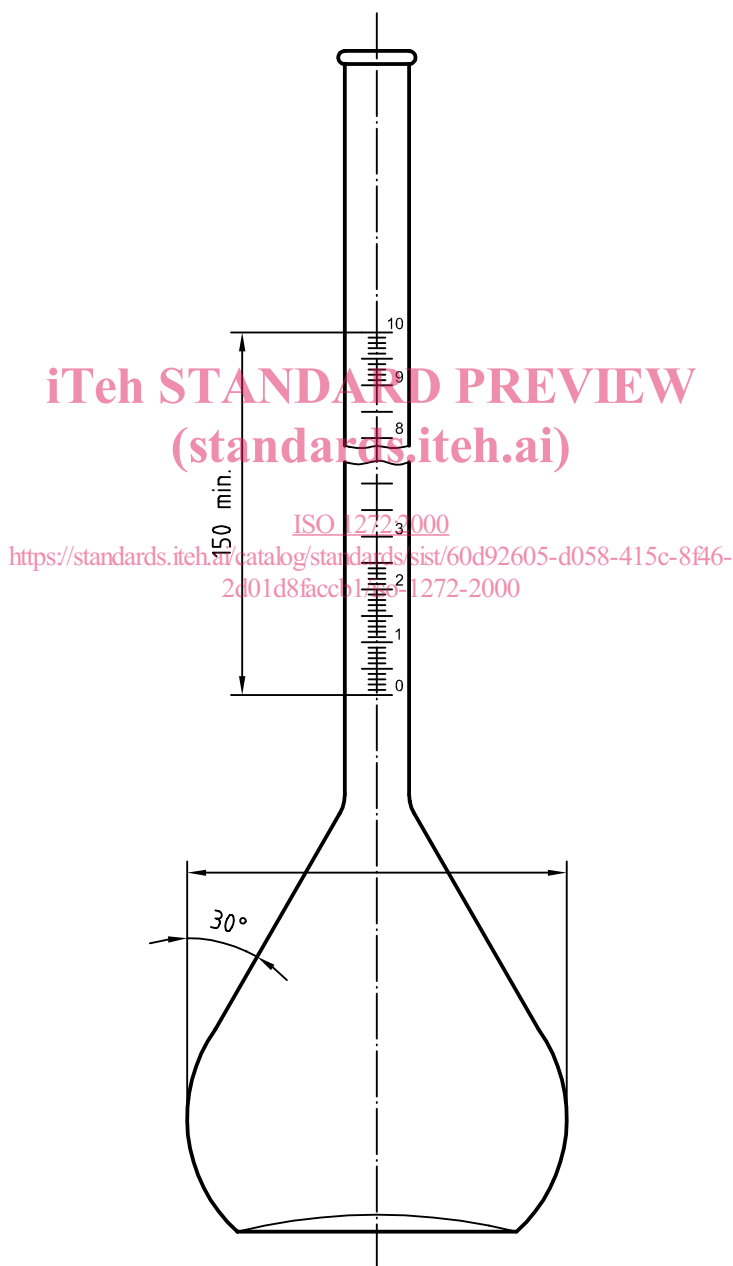


Figure 1 — Cassia flask with a graduated neck

6 Sampling

Sampling is not part of the method specified in this International Standard. A recommended sampling method is given in ISO 212.

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

7 Preparation of test sample

Prepare the test sample according to ISO 356.

In order to avoid interphase emulsions at the moment of determination, pretreat the essential oil with tartaric acid as follows:

- take a volume of essential oil greater than 10 ml, then add 0,02 g of tartaric acid (4.1) per millilitre of oil;
- shake vigorously and without filtering, then dry with magnesium sulfate according to 5.3 of ISO 356:1996.

8 Procedure

NOTE If verification of the repeatability is desired, carry out two separate determinations.

8.1 Test sample

Take with the pipette (5.1) 10 ml of the prepared sample (see clause 7) and transfer it to the Cassia flask (5.3) after having introduced approximately 75 ml of the potassium hydroxide solution (4.2).

Shake the mixture six times at 5-min intervals. Operate at a constant temperature of 20 °C except in particular cases (see 8.2.2).

NOTE The shaking time will be specified in each monograph.

8.2 Determination

8.2.1 Add to the mixture a supplementary quantity of potassium hydroxide (4.2) so that the portion of the oil not transformed into alkaline phenates soluble in the aqueous phase is raised into the graduated neck of the Cassia flask.

Facilitate the separation of the oil drops attached to the walls by rotating the flask between the hands and gently tapping.

After allowing the flask to stand for a few hours, read off the volume of unabsorbed oil if all of it is gathered into the neck.

If any emulsion is observed, add 2 ml of xylene (4.3) measured with the pipette (5.1). Agitate the emulsified layer by means of the glass rod (5.4) and allow to stand. If the emulsion disappears, read the volume of unabsorbed oil.

If the emulsion persists, repeat the test with the addition of 2 ml of xylene to the test portion before the initial shaking.

8.2.2 In some cases heating is required for the determination. In the case of essential oils containing phenol esters, there is sometimes a partial or total saponification of such esters (for example in the case of essential oils of clove-containing eugenol acetate, the ester is completely saponified during the measurement).

Indications related to the modifications to be introduced in the procedure described in this International Standard, such as shaking time, intervals between shakes, and temperature, will be stated in the standard specific for the corresponding essential oil in the particular cases indicated above.

9 Expression of results

The phenol content, w , of the sample is given, as percentage by volume, by the formula:

$$w = 10 (10 - V)$$

where V is the volume, in millilitres, of the unabsorbed portion of the oil read in 8.2.1.

If 2 ml of xylene were added, subtract 2 ml from the volume V .

Express the result to the nearest whole number.

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 %.

11 Test report

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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, with reference to this International Standard;
- all operating details not specified in this International Standard, or regarded as optional, together with details or 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.

Bibliography

- [1] ISO 212, *Essential oils — Sampling*.

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