
**Ginger and its oleoresins — Determination
of the main pungent components (gingerols
and shogaols) — Method using high-
performance liquid chromatography**

*Gingembre et oléorésines de gingembre — Dosage des principaux
composés piquants (gingérols et shogaols) — Méthode par
chromatographie en phase liquide à haute performance*



Foreword

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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.

International Standard ISO 13685 was prepared by Technical Committee ISO/TC 34, *Agricultural food products*, Subcommittee SC 7, *Spices and condiments*.

Annexes A to E of this International Standard are for information only.

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Printed in Switzerland

Introduction

This method, initially based on Wood's publications [Overseas Development Natural Resources Institutes (ODNRI)], enables one to know without any ambiguity the identity of the peaks obtained by HPLC, for the quality control of ginger extracts. It is particularly useful in this case as the pungent compounds of interest are very sensitive to heat.

The method has been validated by two international interlaboratory tests carried out by seven laboratories.

After examination of the results, it is estimated that the method may be considered as the best that can be achieved for the moment but it could be improved, in particular for the precision data.

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Ginger and its oleoresins — Determination of the main pungent components (gingerols and shogaols) — Method using high-performance liquid chromatography

1 Scope

This International Standard describes a method for the determination of gingerols [6]-G, [8]-G and [10]-G and the corresponding shogaols [6]-S, [8]-S and [10]-S in dried ginger or in oleoresins of ginger, by high-performance liquid chromatography (HPLC) in the reverse phase.

NOTE - Chemical structures of gingerols and shogaols are shown in annex A.

2 Normative references

The following standards contain provisions which, through reference in this text, constitute provisions of this International Standard. At the time of the publication, the editions indicated were valid. All standards are subject to revision, and parties to agreements based on the International Standard are encouraged to investigate the possibility of applying the most recent editions of the standards indicated below. Members of IEC and ISO maintain registers of currently valid International Standards.

ISO 939:1980, *Spices and condiments - Determination of moisture content - Entrainment method.*

ISO 2825:1981, *Spices and condiments - Preparation of a ground sample for analysis.*

3 Principle

From ground and dried ginger, extraction of the pungent components by methanol at atmospheric temperature and concentration at reduced pressure of part of the extract. The oleoresins are dissolved in the methanol.

The resulting solutions are analysed directly (without any cleaning) by reverse-phase HPLC on a column filled with octadecylsilyl silica, with a mixture of acetonitrile and aqueous acetic acid as the mobile phase and by UV detection at 280 nm.

Quantification is carried out using an external standard, nonanoic acid vanillylamide, which has a retention time comparable to that of gingerol-[6] (and a similar UV absorption spectrum).

4 Reagents and products

Use only reagents of recognized analytical grade, unless otherwise specified, and water of HPLC grade.

4.1 External standard: Nonanoic acid vanillylamide (NVA), with melting point between 42 °C and 44 °C and giving primarily a unique peak (at least 98 % of the total area of the peak between 3 min and 8 min) when analysed by HPLC under the conditions described in this International Standard.

WARNING: Nonanoic acid vanillylamide is a product which can cause irritation.

NOTE — NVA is also known as nonyl vanillylamide or pelargonic acid vanillylamide.

4.2 Solvents

WARNING: Acetonitrile is a toxic product which must be used with caution. Studies are underway to replace it by another product.

4.2.1 Methanol, HPLC grade, for preparation of the NVA standard.

4.2.2 Methanol, analytical grade, for dilution and extraction of the test samples.

4.2.3 Acetonitrile, HPLC grade.

4.2.4 Glacial acetic acid.

5 Apparatus

Usual laboratory equipment and, in particular, the following.

5.1 Chromatograph

Apparatus equipped for HPLC analysis:

- an injection system capable of delivering portions of 20 µl of the sample solution with considerable precision;
- an ultraviolet (UV) detector measuring absorbance at a wavelength of 280 nm;
- a recorder/integrator capable of precisely measuring the area of the peaks with appropriate baseline corrections, with a sensitivity of 0,25 to 0,5 a.u.f.s.

5.2 Stainless-steel column, of 250 mm length and 4,6 mm diameter (or of comparable dimensions) filled with type C₁₈ stationary phase with particles of 5 µm.¹⁾

¹⁾ ODS, distributed by Phase Separations Ltd., UK, is an example of a suitable product available commercially. This information is given for the convenience of users of International Standard and does not constitute an endorsement by ISO of this product.