

INTERNATIONAL
STANDARD

ISO
3214

Second edition
2000-07-15

Oil of *Litsea cubeba* (*Litsea cubeba* Pers.)

Huile essentielle de Litsea cubeba (Litsea cubeba Pers.)

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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 other than those identified above. ISO shall not be held responsible for identifying any or all such patent rights.

International Standard ISO 3214 was prepared by Technical Committee ISO/TC 54, *Essential oils*.

This second edition cancels and replaces the first edition (ISO 3214:1974), which has been technically revised.

Annexes A and B of this International Standard are for information only.

Oil of *Litsea cubeba* (*Litsea cubeba* Pers.)

1 Scope

This International Standard specifies certain characteristics of the oil of *Litsea cubeba* (*Litsea cubeba* Pers.), in order to facilitate assessment of its quality.

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/TR 210, *Essential oils — General rules for packaging, conditioning and storage.*

ISO/TR 211, *Essential oils — General rules for labelling and marking of containers.*

ISO 212, *Essential oils — Sampling.*

ISO 279, *Essential oils — Determination of relative density at 20 °C — Reference method.*

ISO 280, *Essential oils — Determination of refractive index.*

ISO 592, *Essential oils — Determination of optical rotation.*

ISO 875, *Essential oils — Evaluation of miscibility in ethanol.*

ISO 1279, *Essential oils — Determination of carbonyl value — Potentiometric methods using hydroxylammonium chloride.*

ISO 11024-1, *Essential oils — General guidance on chromatographic profiles — Part 1: Preparation of chromatographic profiles for presentation in standards.*

ISO 11024-2, *Essential oils — General guidance on chromatographic profiles — Part 2: Utilization of chromatographic profiles of samples of essential oils.*

NF T 75-414, *Oils of Litsea cubeba and lemongrass — Determination of citral (neral + geranial) content — Gas chromatographic method on packed and capillary columns.*

3 Term and definition

For the purposes of this International Standard the following term and definition applies.

3.1

oil of *Litsea cubeba*

essential oil obtained by steam distillation of the fresh fruits of *Litsea cubeba* Pers., from the Lauraceae family

NOTE CAS number of oil of *Litsea cubeba*: 90063-59-5.

4 Requirements

4.1 Appearance

Clear, mobile liquid.

4.2 Colour

Pale yellow to dark yellow.

4.3 Odour

Characteristic, fresh, reminiscent of citral.

4.4 Relative density at 20 °C, d_{20}^{20}

Minimum: 0,880 0

Maximum: 0,892 0

4.5 Refractive index at 20 °C

Minimum: 1,480 0

Maximum: 1,490 0

4.6 Optical rotation at 20 °C

Between +3° and +12°.

4.7 Miscibility in 70 % ethanol (volume fraction) at 20 °C

It shall not be necessary to use more than 3 volumes of 70 % ethanol (volume fraction) to obtain a clear solution with 1 volume of essential oil.

4.8 Carbonyl index

Minimum: 272

This corresponds to a content of carbonyl constituents, expressed as citral, of

minimum: 74 %

4.9 Citral content (neral + geranial) by gas chromatography using the internal standard method

Minimum: 70 %

4.10 Chromatographic profile

Analysis of the essential oil shall be carried out by gas chromatography. In the chromatogram obtained, the representative and characteristic components shown in Table 1 shall be identified. The proportions of these components, indicated by the integrator, shall be as shown in Table 1. This constitutes the chromatographic profile of the essential oil.

Table 1 — Chromatographic profile

Component	Minimum %	Maximum %
α-Pinene	—	1,5
Limonene	9	15
2-Methyl-2-heptene-6-one	1,8	3
Citronellal	—	1,5
Linalol	1,5	3
Neral	25	33
Geranial	38	45
Citronellol	0,5	1,5
Nerol	0,2	1,2
Geraniol	0,5	1,5

NOTE The chromatographic profile is normative, contrary to typical chromatograms given for information in annex A.

4.11 Flashpoint

Information on the flashpoint is given in annex B.

5 Sampling

See ISO 212.

Minimum volume of test sample: 30 ml.

NOTE This volume allows each of the tests specified in this International Standard to be carried out at least once.

6 Test methods

6.1 Relative density at 20 °C, d_{20}^{20}

See ISO 279.

6.2 Refractive index at 20 °C

See ISO 280.

6.3 Optical rotation at 20 °C

See ISO 592.

6.4 Miscibility in 70 % ethanol (volume fraction) at 20 °C

See ISO 875.

6.5 Carbonyl index

See ISO 1279.

Test sample: 1,2 g to 2 g.
 Standing time: 15 min without heating.
 Relative molecular mass of citral: 152,2.

6.6 Citral content by gas chromatography

See NF T 75-414.

6.7 Chromatographic profile

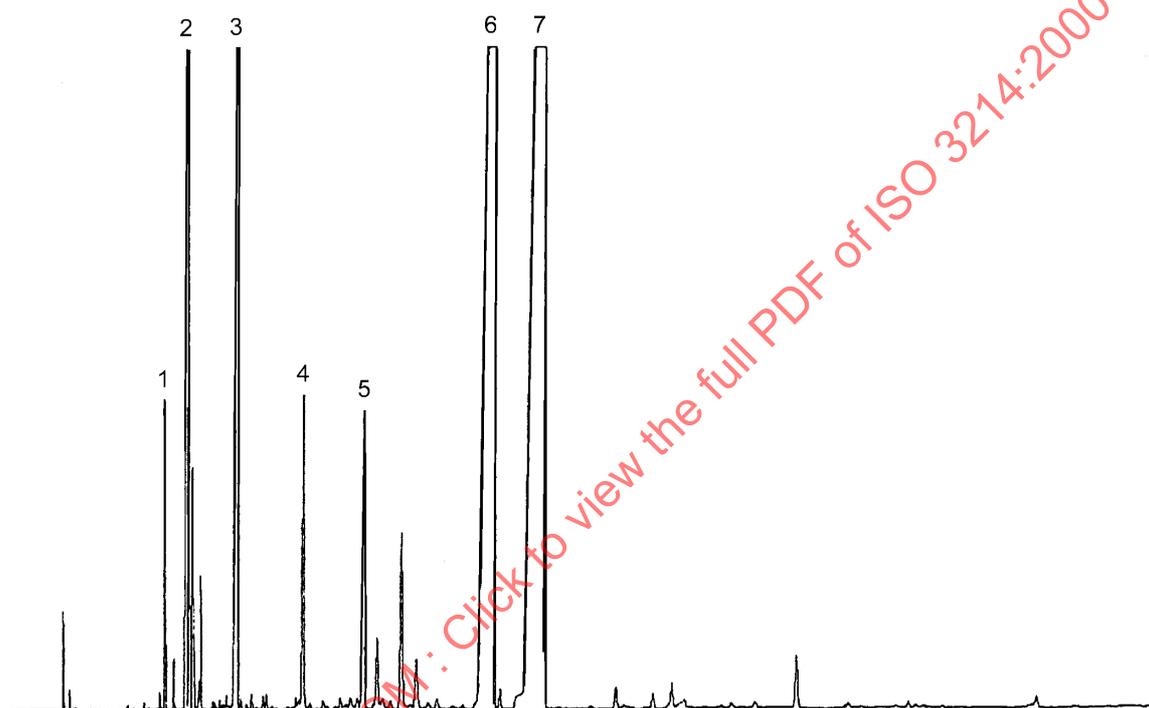
See ISO 11024-1 and ISO 11024-2.

7 Packaging, labelling, marking and storage

See ISO/TR 210 and ISO/TR 211.

Annex A (informative)

Typical chromatograms of the analysis by gas chromatography of the essential oil of *Litsea cubeba* (*Litsea cubeba* Pers.)



Peak identification

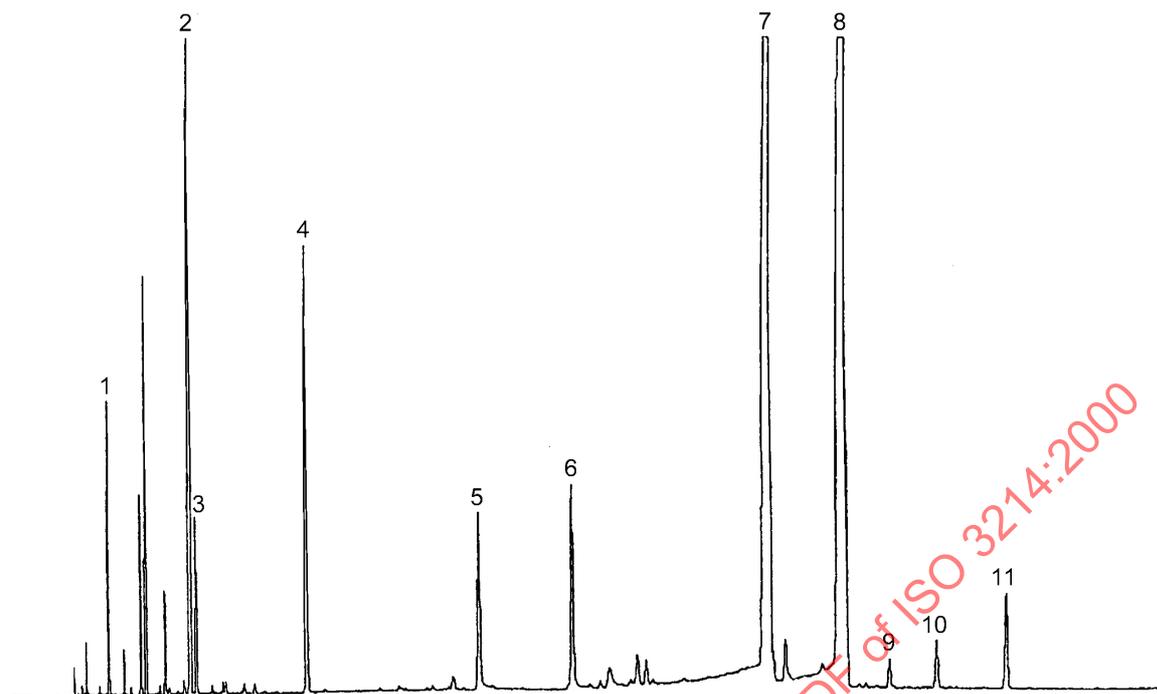
- | | |
|---|-----------------------------|
| 1 | α -Pinene |
| 2 | 2-Methyl-2-heptene-6-one |
| 3 | Limonene + 1,8-cineole |
| 4 | Linalol |
| 5 | Citronellal |
| 6 | Neral + nerol + citronellol |
| 7 | Geraniol + geranial |

Operating conditions

Column: capillary, silica, 30 m long and of 0,25 mm internal diameter
 Thickness of film: not provided
 Stationary phase: dimethyl polysiloxane (SE 30)
 Oven temperature: temperature programming from 70 °C to 250 °C
 at a rate of 2 °C/min
 Injector temperature: 250 °C
 Detector temperature: 250 °C
 Detector: flame ionization type
 Carrier gas: nitrogen
 Volume injected: 0,3 μ l
 Carrier gas flow rate: 1 ml/min
 Split ratio: 1/100

NOTE With this kind of column, neral, nerol and citronellol are not always well separated. In this case, it is recommended to carry out the analysis using a column of different polarity.

Figure A.1 — Typical chromatogram taken on an apolar column

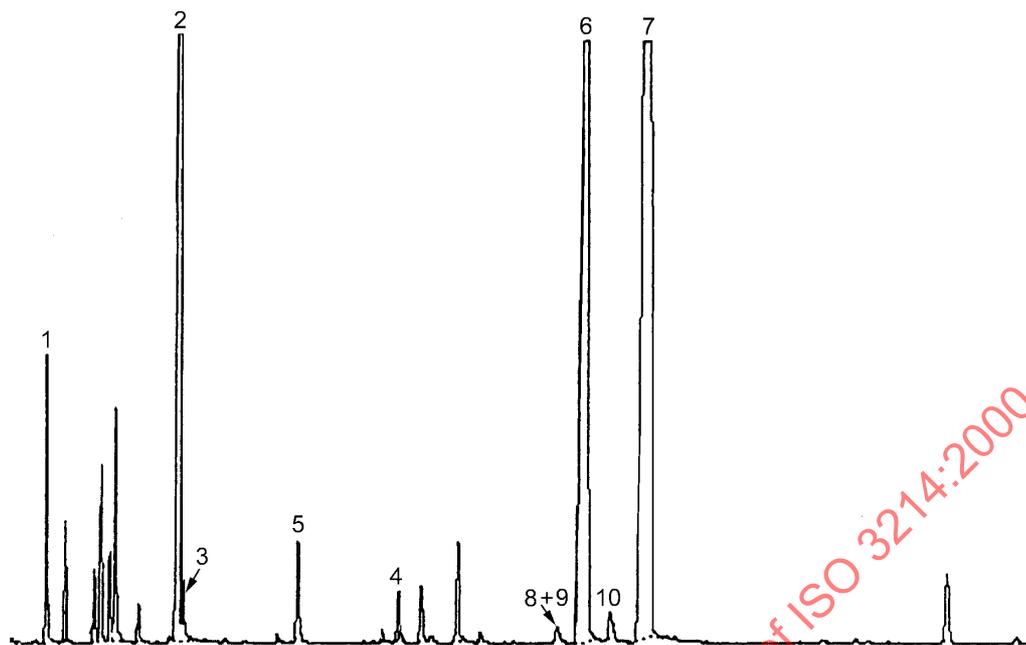
**Peak identification**

- 1 α -Pinene
- 2 Limonene
- 3 1,8-Cineole
- 4 2-Methyl-2-heptene-6-one
- 5 Citronellal
- 6 Linalol
- 7 Neral
- 8 Geranial
- 9 Citronellol
- 10 Nerol
- 11 Geraniol

Operating conditions

Column: capillary, silica, 30 m long and of 0,25 mm internal diameter
 Thickness of film: not provided
 Stationary phase: polyethylene glycol (Supelcowax 10) cross-linked
 Oven temperature: temperature programming from 70 °C to 250 °C
 at a rate of 2 °C/min
 Injector temperature: 250 °C
 Detector temperature: 250 °C
 Detector: flame ionization type
 Carrier gas: nitrogen
 Volume injected: 0,3 μ l
 Carrier gas flow rate: 1 ml/min
 Split ratio: 1/100

Figure A.2 — Typical chromatogram taken on a polar column

**Peak identification**

- 1 α -Pinene
- 2 Limonene
- 3 1,8-Cineole
- 4 Citronellal
- 5 Linalol
- 6 Neral
- 7 Geranial
- 8 Citronellol
- 9 Nerol
- 10 Geraniol

Operating conditions

Column: capillary, silica, 30 m long and of 0,25 mm internal diameter
 Thickness of film: 0,25 μ m
 Stationary phase: DB5TM
 Oven temperature: temperature programming from 60 °C to 200 °C
 at a rate of 3 °C/min
 Injector temperature: 210 °C
 Detector temperature: 210 °C
 Detector: flame ionization type
 Carrier gas: nitrogen at 140 kPa
 Volume injected: 0,1 μ l
 Carrier gas flow rate: 0,38 m/s on the *n*-hexane peak
 Split ratio: 1/80

Figure A.3 — Typical chromatogram taken on an apolar column