
**Essential oil of *Melaleuca*, terpinen-4-
ol type (Tea Tree oil)**

*Huile essentielle de Melaleuca, type terpinén-4-ol (huile essentielle de
Tea Tree)*





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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.

The procedures used to develop this document and those intended for its further maintenance are described in the ISO/IEC Directives, Part 1. In particular the different approval criteria needed for the different types of ISO documents should be noted. This document was drafted in accordance with the editorial rules of the ISO/IEC Directives, Part 2 (see www.iso.org/directives).

Attention is drawn to the possibility that some of the elements of this document may be the subject of patent rights. ISO shall not be held responsible for identifying any or all such patent rights. Details of any patent rights identified during the development of the document will be in the Introduction and/or on the ISO list of patent declarations received (see www.iso.org/patents).

Any trade name used in this document is information given for the convenience of users and does not constitute an endorsement.

For an explanation on the voluntary nature of standards, the meaning of ISO specific terms and expressions related to conformity assessment, as well as information about ISO's adherence to the World Trade Organization (WTO) principles in the Technical Barriers to Trade (TBT) see the following URL: www.iso.org/iso/foreword.html.

This document was prepared by Technical Committee ISO/TC 54, *Essential oils*.

This third edition cancels and replaces the second edition (ISO 4730:2004), which has been technically revised.

Essential oil of *Melaleuca*, terpinen-4-ol type (Tea Tree oil)

1 Scope

This document specifies certain characteristics of the essential oil of *Melaleuca*, terpinen-4-ol type (Tea Tree oil), in order to facilitate assessment of its quality.

2 Normative references

The following documents are referred to in the text in such a way that some or all of their content constitutes requirements of this document. For dated references, only the edition cited applies. For undated references, the latest edition of the referenced document (including any amendments) applies.

ISO/TS 210, *Essential oils — General rules for packaging, conditioning and storage*

ISO/TS 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 11024 (all parts), *Essential oils — General guidance on chromatographic profiles*

3 Terms and definitions

For the purposes of this document, the following terms and definitions apply.

ISO and IEC maintain terminological databases for use in standardization at the following addresses:

— IEC Electropedia: available at <http://www.electropedia.org/>

— ISO Online browsing platform: available at <http://www.iso.org/obp>

3.1

essential oil of *Melaleuca*, terpinen-4-ol type

Tea Tree oil

essential oil obtained by steam distillation of the leaves and terminal branchlets of *Melaleuca alternifolia* (Maiden et Betche) Cheel or of *M. linariifolia* Sm.

Note 1 to entry: For information on the CAS number, see ISO/TR 21092.

4 Requirements

4.1 Essential oil of *Melaleuca*, terpinen-4-ol type (Tea Tree oil) shall meet the requirements as given in [Table 1](#).

Table 1 — Requirements for the essential oil of *Melaleuca*, terpinen-4-ol type (Tea Tree oil)

Characteristics	Requirements	ISO test method
Appearance	Clear, mobile liquid	—
Colour	Colourless to pale yellow	—
Odour	Characteristic	—
Relative density at 20 °C, d_{20}^{20}	0,885 to 0,906	ISO 279
Refractive index at 20 °C	1,475 to 1,482	ISO 280
Optical rotation	Between +7° and +12°	ISO 592
Miscibility in ethanol 85 % (volume fraction) at 20 °C	It shall not be necessary to use more than 2 volumes of ethanol, 85 % (volume fraction), to obtain a clear solution with 1 volume of essential oil	ISO 875

4.2 Chromatographic profile

Carry out the analysis of the essential oil by gas chromatography. Determine the chromatographic profile in accordance with the ISO 11024 series. Identify in the chromatogram obtained, the representative and characteristic components shown in [Table 2](#). The proportions of these components, indicated by the integrator, shall be as shown in [Table 2](#). This constitutes the chromatographic profile of the essential oil.

Table 2 — Chromatographic profile

Components	Min. %	Max. %
α -Pinene	1,0	4,0
Sabinene	traces ^a	3,5
α -Terpinene	6,0	12,0
Limonene	0,5	1,5
<i>p</i> -Cymene	0,5	8,0
1,8-Cineole	traces ^a	10,0
γ -Terpinene	14,0	28,0
Terpinolene	1,5	5,0
Terpinen-4-ol	35,0	48,0
α -Terpineol	2,0	5,0
Aromadendrene	0,2	3,0
Ledene (syn. viridiflorene)	0,1	3,0
δ -Cadinene	0,2	3,0
Globulol	traces ^a	1,0
Viridiflorol	traces ^a	1,0
^a traces: <0,01 %. NOTE The chromatographic profile is normative, contrary to typical chromatograms given for information in Annex A .		

5 Additional information

5.1 Flashpoint

Information on the flashpoint is given in [Annex B](#).

5.2 Enantiomeric distribution

Information concerning enantiomeric distribution is given in [Annex C](#).

6 Sampling

Sampling shall be performed in accordance with ISO 212. Minimum volume of test sample is 50 ml.

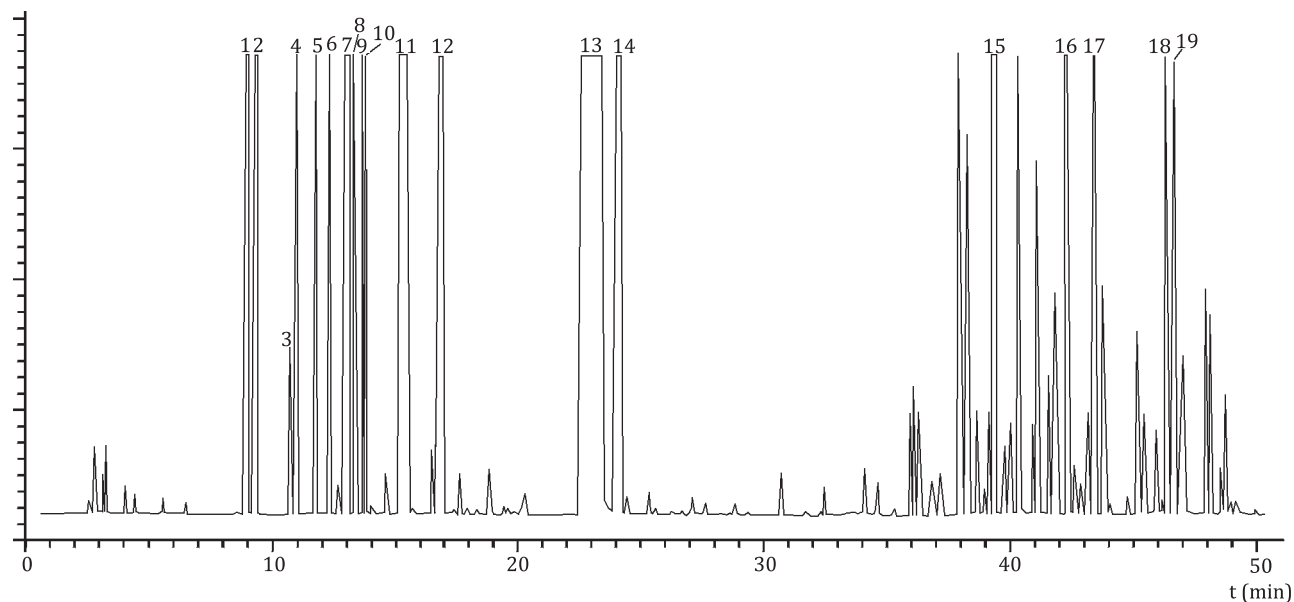
NOTE This volume allows each of the tests specified in this document to be carried out at least once.

7 Packaging, labelling, marking and storage

These items shall be in accordance with ISO/TS 210 and ISO/TS 211.

Annex A
(informative)

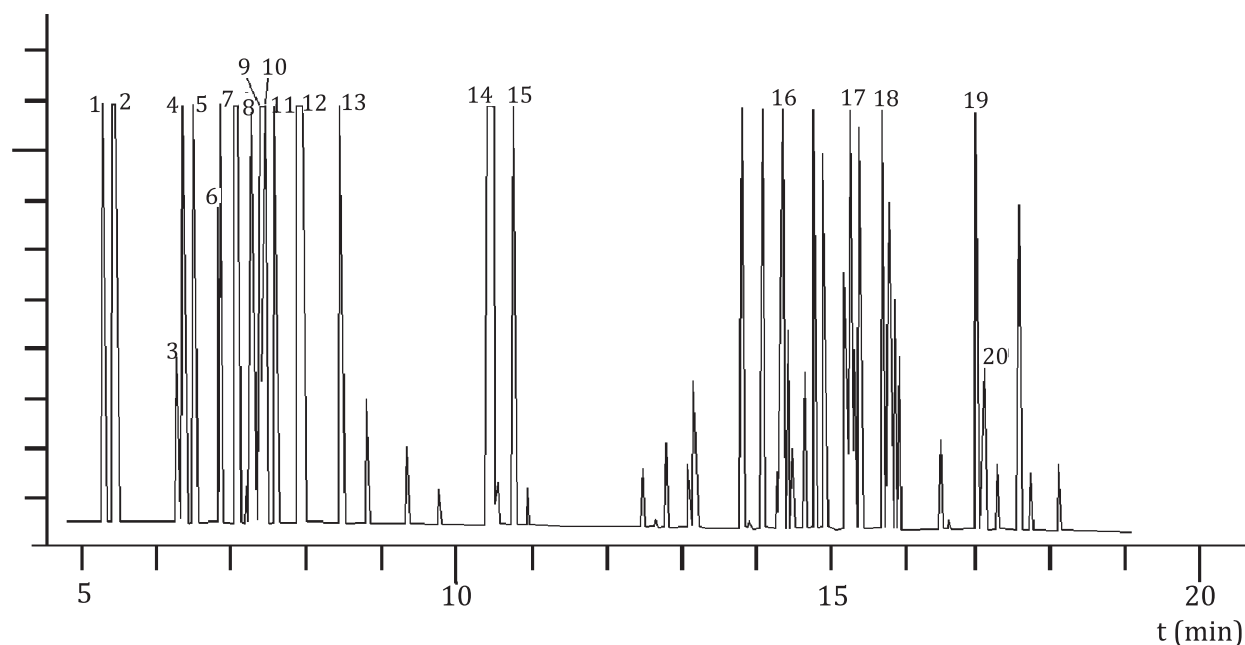
**Typical chromatograms of the analysis by gas chromatography of
the essential oil of *Melaleuca*, terpinen-4-ol type (Tea Tree oil)**



Peak identification	Operating conditions
1 α -Thujene	Column: fused silica capillary; length 50 m; internal diameter 0,20 mm
2 α -Pinene	Stationary phase: poly(dimethyl siloxane) (OV-101 [®] a)
3 Sabinene	Film thickness: 0,25 μ m
4 β -Pinene	Oven temperature: temperature programming from 70 °C to 220 °C at a rate of 2 °C/min
5 Myrcene	Injector temperature: 230 °C
6 α -Phellandrene	Detector temperature: 250 °C
7 α -Terpinene	Detector: flame ionization type
8 <i>p</i> -Cymene	Carrier gas: hydrogen
9 1,8-Cineole + β -phellandrene	Volume injected: 0,2 μ l
10 Limonene	Carrier gas flow rate: 1,0 ml/min
11 γ -Terpinene	Split ratio: 1/100
12 Terpinolene	
13 Terpinen-4-ol	
14 α -Terpineol	
15 Aromadendrene	
16 Ledene (viridiflorene)	
17 δ -Cadinene	
18 Globulol	
19 Viridiflorol	

a OV-101[®] is an example of a suitable product available commercially. This information is given for the convenience of users of this document and does not constitute an endorsement by ISO of this product.

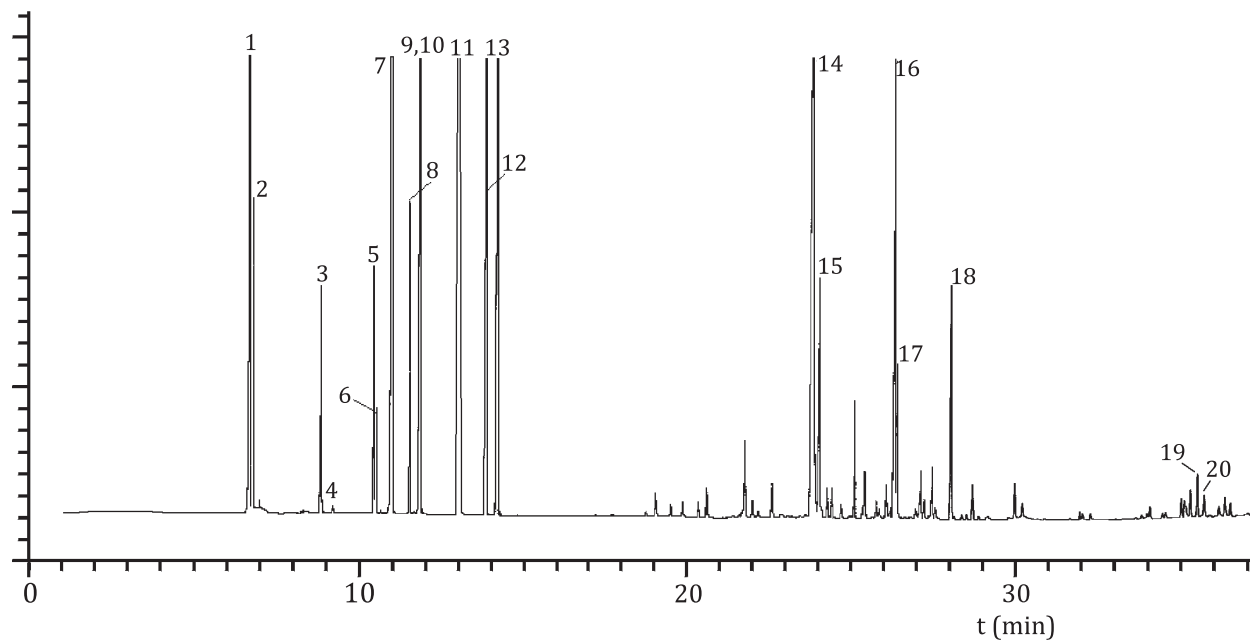
Figure A.1 — Typical chromatogram taken on an apolar column



Peak identification	Operating conditions
1 α -Thujene	Column: FSOT; length 60 m; internal diameter 0,25 mm
2 α -Pinene	Stationary phase: (35 %)-diphenyl-(65 %)-methylsiloxane copolymer
3 Sabinene	(AT-35® a)
4 β -Pinene	Film thickness: 0,25 μ m
5 Myrcene	Oven temperature: isothermal at 50 °C for 1 min, then temperature
6 α -Phellandrene	programming from 50 °C to 250 °C at a rate of 10 °C/min and isothermal
7 α -Terpinene	at 250 °C for 9 min
8 Limonene	Injector temperature: 200 °C
9 β -Phellandrene	Detector temperature: 300 °C
10 <i>p</i> -Cymene	Detector: flame ionization type
11 1,8-Cineole	Carrier gas: hydrogen
12 γ -Terpinene	Volume injected: 1 μ l (1 % in ethanol)
13 Terpinolene	Carrier gas flow rate: 1,0 ml/min
14 Terpinen-4-ol	Split ratio: 1/50
15 α -Terpineol	
16 Aromadendrene	
17 Ledene (viridiflorene)	
18 δ -Cadinene	
19 Globulol	
20 Viridiflorol	

^a AT-35® is an example of a suitable product available commercially. This information is given for the convenience of users of this document and does not constitute an endorsement by ISO of this product.

Figure A.2 — Typical chromatogram taken on an intermediate polarity column



Peak identification	Operating conditions
1 α -Pinene	Column: fused silica capillary; length 50 m; internal diameter 0,33 mm
2 α -Thujene	Stationary phase: poly(ethylene glycol) 20 000 (BP-20® a)
3 β -Pinene	Film thickness: 0,50 μ m
4 Sabinene	Oven temperature: isothermal at 50 °C for 1 min, then temperature
5 Myrcene	programming from 50 °C to 220 °C at a rate of 5 °C/min and isothermal at
6 α -Phellandrene	220 °C for 5 min
7 α -Terpinene	Injector temperature: 240 °C
8 Limonene	Detector temperature: 240 °C
9 β -Phellandrene	Detector: flame ionization type
10 1,8-Cineole	Carrier gas: helium
11 γ -Terpinene	Volume injected: 1,5 μ l (3 % in hexane)
12 p-Cymene	Carrier gas flow rate: 1,0 ml/min
13 Terpinolene	Split ratio: 1/100
14 Terpinen-4-ol	
15 Aromadendrene	
16 α -Terpineol	
17 Ledene (viridiflorene)	
18 δ -Cadinene	
19 Globulol	
20 Viridiflorol	

^a BP-20® is an example of a suitable product available commercially. This information is given for the convenience of users of this document and does not constitute an endorsement by ISO of this product.

Figure A.3 — Typical chromatogram taken on a polar column

Annex B **(informative)**

Flashpoint

B.1 General information

For safety reasons, transport companies, insurance companies and people in charge of safety services require information on the flash points of essential oils, which in most cases are flammable products.

A comparative study on the relevant methods of analysis (see ISO/TR 11018) concluded that it was difficult to recommend a single apparatus for standardization purposes, given that:

- there is a wide variation in the chemical composition of essential oils;
- the volume of the sample needed in certain requirements would be too costly for high-priced essential oils.
- as there are several different types of equipment which can be used for the determination, users cannot be expected to use one specified type only.

Consequently, it was decided to give a mean value for the flashpoint annexed to each document, for information, in order to meet the requirements of the interested parties.

The equipment with which this value was obtained shall be specified. For further information, see ISO/TR 11018.

B.2 Flashpoint of the essential oil of *Melaleuca*, terpinen-4-ol (Tea Tree oil)

The mean value is +59 °C.

NOTE Obtained with “closed cup” equipment.

Annex C (informative)

Enantiomeric distribution — General information

Some essential oil components can exist in two enantiomeric forms designated as (R) or (S), D or L or (+) or (-) isomers. Many enantiomers have distinctly different properties and hence their presence in the right form is critical. Also, pure natural essential oils contain enantiomers in characteristic ratios. This ratio is upset by the addition of adulterants including synthetic major components of different enantiomeric ratios.

Consequently, the measurement of enantiomeric excess or enantiomeric ratio as per ISO 22972^[4] in an informative annex of appropriate isolates in International Standards, provides an extra measure of essential oil authenticity.

The enantiomeric distribution for terpinen-4-ol is (R)(+) 67 % - 71 % and (S)(-) 29 % - 33 %.

Bibliography

- [1] ISO 3218, *Essential oils — Principles of nomenclature*
- [2] ISO/TR 11018, *Essential oils — General guidance on the determination of flashpoint*
- [3] ISO/TR 21092, *Essential oils — Characterization*
- [4] ISO 22972, *Essential oils — Analysis by gas chromatography on chiral capillary columns — General method*

