Essential Oils and Aroma Chemicals from **Eucalyptus Globulus** Labill.

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Eucalyptus globulus Labill., which is used in Spain for cellulose and paper production, is grown under an integrated reforestation program. In addition to its importance in the pulp and paper industry, the leaves of *E. globulus* are used to produce an important essential oil from which 1,8-cineole is isolated. Essential oils can also be obtained from the flowers and fruits of this same tree. Commercial amounts of a resinoid absolute are also obtained from the leaves.

History

The species, *Eucalyptus globulus*, was discovered in Tasmania in 1792 by Labillardiere.[1] The origin of the name globulus is not clear, but the leaves do contain globular oil glands, which can be seen by permeating light.[2]

In 1803 a *Eucalyptus* species was cultivated in the monastery of Camaldules in Naples, whereafter this tree received the name *Eucalyptus camaldulensis* and also *Eucalyptus rostrata.*[3]

The *Eucalyptus globulus* species was already present in Portugal in 1829, probably introduced by missionaries who brought seeds over from Tasmania. Ramel imported the *Eucalyptus globulus* in Spain in 1854.[4]

In the second half of the nineteenth century—not in the 18th century as mentioned by Guenther and Gildemeister and Hoffmann—*Eucalyptus* species were introduced in many countries around the world with tropical and subtropical climates. The *Eucalyptus* species, mainly *Eucalyptus globulus* and *Eucalyptus camaldulensis,* were originally brought to Spain and Portugal for the purpose of drying out swampy grounds.[5] In the beginning, the people thought that the balsam-scented leaves were useful to keep off malaria-carrying mosquitos,[6] therefore the *Eucalyptus globulus* received the name *fever tree.* Later on it was discovered that the mosquitos diminished because of the disappearance

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of the swamplands.

Formerly the drying-out properties of the trees were attributed to the evaporation capacity of the leaves. Griffon, however, proved that the transpiration of *Eucalyptus* leaves was rather small in comparison with other trees, like birch or willow.[7] The drying-out capacity of the *Eucalyptus* trees is elevated because the tree grows quickly and in a short time (5 to 10 years) gets a lot leaves.[8]

Botany

The *Eucalyptus* species, which originates from Australasia (mainly Australia and Tasmania), belongs to the family Myrtaceae, which comprises about 80 genera and more than 3,000 species. The Myrtaceae are either shrubs or trees with leaves possessing essential oil glands. They are all evergreen with leathery (coriaceous) leaves. The most economically important genera of the Myrtaceae are *Eucalyptus* (ca. 600 species), *Eugenia* (ca. 700 species), *Melaleuca* (over 100 species), *Myrtus* (ca. 100 species) and *Psidium* (ca. 100 species).

The knowledge of the *Eucalyptus* species has been developed during the last two centuries. In 1788, the genus was named *Eucalyptus* by L'Heritier, the word being derived from the Greek *eu* = well, and *kalypto* = I cover, in allusion to the lid of the flower-bud, which covers the stamens until they are fully developed.[9]

Morphologically, the trees possess two kinds of opposite, coriaceous leaves. The juvenile leaves, which are broad, egg or lancet-shaped, are covered with a white wax, which on the green leaves gives them a bluish appearance. The mature leaves, which are mostly petiolate, are usually longer than the juvenile leaves and more sickle-lanceolate in shape. They are generally greyish-green in colour. The opercular flower-buds are formed by the growing together of the sepals and petals. The operculum covering the many stamen falls off when the flower is fully developed. The flowers, which can be white, yellow, pink or red, are borne in the axil between the leaf and the twig. They are generally 3-4 cm across, possess a great many stamen and bloom in the autumn or winter. It is characteristic for the *Eucalyptus* to shed its bark revealing a smooth greyish-white trunk.

The botanical study of the *Eucalyptus* genus is truly a subject for specialists (Eucalyptologists) and classification by breeds comprises a veritable code, with six-letter indicators that correspond to subgenus, section, major series, sub-series, species and sub-species.[3] The *Eucalyptus* genus, of which almost all grow in Australia, are the tallest trees (up to 100 M) in the world. For example, a *Eucalyptus regnans* tree, which was felled in Australia in the late 19th century, measured 132 M in height, and was possibly the world's tallest tree.

Blakely mentioned that 638 species and varieties of *Eucalyptus* exist.[10] Penfold and Willis described the *Eucalyptus* genus in detail.[11] In 1973, Chippendale mentioned that some 500 species of *Eucalyptus* produced terpenoid hydrocarbons.[12] The biology of *Eucalyptus* and the distribution of *Eucalyptus globulus* in the world was published by Prior in 1976.[13]

The *Eucalyptus* trees are of such importance, that every five years a World Eucalyptus Conference is organized. When the first conference was held in Rome in 1956 there were about 700,000 hectares of replanted Eucalyptus forests in the world. In 1974, according to F.A.O. estimates, this figure stood at 3.7 million hectares. Because there are new plantings every year, one may assume the area of reforestation of Eucalyptus trees at this moment on more than five million hectares (or 50.000Km^2), an area which could completely cover countries like The Netherlands or Switzerland. This may seem an enormous figure, however, at this moment every year the same area of woods disappear.

Eucalyptus tree reforestation is opening up new possibilities for industrialization and use of land in tropical and subtropical countries. Moreover this implies major consequences for the world's timber supply, with relaxed pressure on natural forests, and with regard to the location of processing plants, which until now are almost monopolized by some highly developed countries, like the U.S.A., Canada, and Scandinavia, which have large conifer forests.[3]

It was recently reported that *Eucalyptus globulus*, which was introduced in the Mediterranean region in 1860, was the most widely planted gum tree.[6]

Essential Oil Production

Usually, the trees are felled for the collection of the leaves because gathering of leaves from live trees is costly. The leaves are steam distilled only when the owner decides to fell the trees and sell the wood.[5] Sometimes leaves are collected from old live trees that contain abundant foliage. On the stumps of felled trees often grow young shoots, so-called suckers, bearing juvenile leaves (which, for example, are steam distilled in California). It is surprising that these shoots have not been harvested or collected for steam distillation of an essential oil in Spain and Portugal where they are abundantly available too.

Three essential oils derived from *Eucalyptus globulus* are known in literature, namely steam distilled oils from the leaves, the flowers and the fruits. In practice, however, the most important commercial essential oil is obtained from the leaves.

Formerly the oil was obtained in poor yield (up to 0.5%) by direct heating of the branches with the leaves in water, isolating the distillate and separating the oil. Later on, typical field stills were used such as were described in detail by Guenther and Gildemeiser and Hoffmann.[5,14] In these stills, the twigs with the leaves are on a kind of grill not in direct contact with the water which is heated at the bottom of the still where the steam is produced and transported through the material. After condensation and separation, a wet oil is obtained in 0.7% to 1.2% yield. Some of these stills are in production today.[15] In a modern distillery, the plant material is treated in stainless steel equipment with high pressure steam (5-10 bar), which is generated outside the still. The yield of oil by this latter method is 1.0% to 1.5%.

The essential oil of *Eucalyptus globulus* leaves can also be obtained in 1-1.5% yield by the modern hydrodiffusion method (DBCH) by treating the branches and leaves with low pressure steam.

Because the crude oil is wet, coloured (greenish-yellow) and contains lower aliphatic aldehydes with off-odours, it is necessary to purify it. Normally the oil is dried over anhydrous Na_2SO_4

and distilled over a few percent (1-2%) caustic soda (NaOH), which causes polymerisation of the lower aldehydes and subsequently distilled off the very (low) light ends.

Apart from the method of isolation, the yield of the essential oil of *Eucalyptus globulus* is dependent on the type of plant material, juvenile or adult leaves, and its conditions, time of harvesting, and dryness (see Table I). Yields of steam distilled oil of leaves have been reported from 0.5% to over 8%. Juvenile leaves should give a higher yield (up to 3%) than adult leaves (up to 1.5%).

Table I. Yield of Essential Oil of *Eucalyptus globulus* Leaves

Authors	% Fresh Leaves	% Dried Leaves	Origin
Baker and Smith (16)	0.92	-	Australasia
von Muller (17)	0.71	-	Australasia
Schimmel (18)	-	1.6-3.0	Mediterranean
Puran Singh (19)	1.16	2.28	India
(1 yr juvenile)	0.83	1.61	India
(10 yr adult)	0.80	1.48	India
Pellini and Morani (20)	0.635	1.252	Sicily
Tompkins (21)			
(1 yr)	0.09	-	California
(5 yr)	0.21	-	California
(10 yr)	0.84	-	California
	Leaves harvested in		
Invidiato (22)	April 0.7%	December 0.76%	Sicily
Timofeew (23)	March 1.65%	June 1.0%	Russia
Esdorn (24)			Mediterranean
after 1 day in water	5.97		
after 5 days in water	8.22		
Nishimura and Calvin (25)			California
from fruits	1.00 (low cineole content)		
from leaves and branches	1.20 (medium cineole content)		
from shoots	1.20 (high cineole content)		

Physicochemical Properties

The physicochemical properties of *Eucalyptus globulus* leaf oil such as specific gravity, refractive index, optical rotation and solubility in ethanol are more or less related to the cineole content and the amount of monoterpenes, as depicted in Table II.

From the data shown in Table II it is evident that the specific gravity will be higher and the solubility in ethanol better for an oil with a high cineole content, whereas the refractive index and the specific rotation are just the opposite.

Table II. A Comparison of the Physicochemical Properties of Crude Oil of *Eucalyptus globulus* with Those of 1,8-cineole

Physicochemical properties	Crude Oil of E. globulus	1,8-cineole
spec. gravity d_{20}^{20}	0.916	0.928-0.930
refract. index n_D^{20}	1.4645	1.454-1.461
spec. rotation $alpha_D^{20}$	+4.2°	0°
solubility L_{70}°	1 in 5 and more	1 in 1.5 and more
1.8-cineole content	65-70%	Over 98%
monoterpene content	20-25%	Less than 2%

Little is know about the acid-, alcohol-, carbonyl- and ester numbers of the *Eucalyptus globulus* oil. Authors Baker and Smith found an acid number of 1.1 and ester number of 1.0 in an oil of Australasian origin.[16] Invidiato found acid numbers of 1.2 to 1.5, alcohol numbers of 8% to 14%, and ester numbers of 8.8 to 18.3 in a Sicilian oil.[22]

A summary of the published physicochemical properties of *E. globulus* leaf oil can be seen in Table III.

A few publications appeared about the essential oil from the flowers and fruits from *Eucalyptus globulus.* For example, Pande et al. reported

Table III. Physicochemical Properties of Essential Oils of Leaves of *Eucalyptus globulus*

Origin	Specific Gravity	Refractive Index	Optical Rotation	1,8-Cineole Content	Solubility in Ethanol 70%	Authors
Australasia	0.913	1.4663	+ 8.4°	-	1 in 1.5	Baker & Smith (16)
Australasia	0.9116-0.9166	1.4980	- 6°6'	-	-	Briggs & Short (27)
Australasia	0.910 -0.930	1.460-1.470	+0°18' to 15°	70-75%	1 in 1.5-3	Penfold (11)
Spain	0.905 -0.922	-	+1°48' to +9°	71.3-84.5%	1 in 1.5-10	Guenther (5)
Spain	0.910 -0.930	1.457-1.469	+8° (max +15°)	-	1 in 2-3	Schimmel (18)
Spain	0.915 -0.925	1.4663-1.4669	+1° to +3°5'	-	1 in 2.3	Fesneau (28)
Spain	0.9175-0.930	-	+2° to +7°12'	66.5-73.3%	1 in 2.2-11	Bordas (29)
Portugal	0.920 -0.927	1.461-1.466	+0.5° to +4°	min. 70%	1 in 4	Laurent & Rato (30)
Portugal	0.910 -0.925	1.459-1.467	+0° to +5°	min. 70%	1 in 5	Carmo (15)
Sicily	0.9301/0.9284	1.415/1.423	+5°28 / +9°39'	-	-	Invidiato (22)
Corsica	0.9185	1.4633	+1.0°	66%	1 in 4.5	Zola & Le Vanda (31)
Brasil	0.9070-0.9105	1.4612-1.4653	+0.46° to +0.68°	68.2-69.9%	completely	Raoul (32)
Ecuador	0.9261	1.45932	+1°41'	ca 86%	1 in 1.9	Parry & Ferguson (33)
India	0.9065-0.9155	1.4630-1.4660	+5°28' to 9°39'	60%	-	Puran Singh (19)
India	0.9005-0.9075	1.4548-1.4656	-5° to +10°	min. 60%	1 in 1 (80°)	Puran Singh (19)
Spain/Portugal Standard	0.906 -0.925	1.459 - 1.467	0 to +10	min. 70%	min 1 in 5	ISO no. R 770-1968 British Pharm. 1981

that the essential oil from the fruits of *Eucalyptus globulus* had the following characteristics: acid number 0.4, alcohol number 31.8, carbonyl number 0.1 and ester number (after acetylation) 31.6.[26] Thus the essential oil from the fruits can differ quite a lot from that of the leaves, as was also reported by Nishimura and Calvin.[25]

Chemical Composition

In 1870 Cloez for the first time investigated the steam distilled oil of the leaves of *Eucalyptus globulus* and isolated a substance, with a boiling point of 175°C, which he called Eucalyptol,[34] but which was probably accompanied by terpenes. Jans in 1884 discovered the true identity of Eucalyptol—$C_{10}H_{18}O$—which was the main constituent of the essential oil of leaves of *Eucalyptus globulus*, and gave it the name cineole.[35]

In 1902 Baker and Smith did research on the Eucalypts especially with respect to their essential oils.[16] Maiden in 1909 gave a critical review of the genus *Eucalyptus*.[36] In 1930 Penfold and Morrison made their notes on the essential oils from some cultivated *Eucalyptus* and in 1952 they gave a guide to the extraction of Eucalyptus oils in the field.[11]

A survey of the literature reveals that the oil of *E. globulus* has been the subject of considerable study. A summary of the work can be seen below.

Monoterpenes
alpha-pinene[15,31,37-42]
camphene[15,18,40]
beta-pinene[15,25,31,40,41a]
limonene[15,40,41a]
p-cymene[40,41]
alpha-terpinene[25]
phellandrene[40,41]
beta terpinene[25]
gamma-terpinene[25]
myrcene[32]
alpha-terpineol[40,41a,54]
isopulegol[41b]
trans-carveol[41b]
terpinen-4-ol[25,41b]
p-cymenol (cumenic alcohol)[41b,54]
globulol[15,18a,25,48,55]
epiglobulol[48]
eudesmol[38]
ledol[48]
vividiflorol[48]

Sesquiterpenes
aromadendrene[25,27,40-48]
allo-aromadendrene[25,47,48]
gamma/delta-cadinenes[15,25,49]
caryophyllene[50]
eremophyllene[25]
S-guaiazulene[40]
alpha-gurjunene[25]

Terpenoid Ethers
1,8-cineole[11a,15,25,32,34,35,38,40-42,51,52]

Alcohols
ethanol[53]
isoamyl alcohol[15,18a,39]
geraniol[41a]
pinocarveol[15,39,40,54]
linalool[41]
myrtenal[40,54]
isopinocampheol[41]

Aldehydes
isovaleraldehyde[15,39,40]
valeraldehyde[40]
capronaldehyde[40]
butyraldehyde[40,54]
myrtenal[54]
cuminaldehyde[11b]
phellandral[11b]

Ketones
l-pinocarvone[40,54]
d-verbenone[40,54]
pipertone[25]
l-carvone[54]
3-isopropyl-1-acetyl-cyclopentene-5[54]
cryptone[41,56]

Esters
bornyl acetate[40]
alpha-terpinyl acetate[40,41b]

A crude oil of *Eucalyptus globulus* leaves was examined using modern analytical techniques. The oil contains up to 90 constituents, from which 58 were identified, comprising 99% of the oil. A summary of the results obtained from this study can be seen below.

Aliphatic Hydrocarbons (0.76%)
undecane 0.11
dodecane 0.07
tridecane 0.07
tetradecane 0.08
pentadecane 0.14
hexadecane 0.10
heptadecane 0.06
octadecane 0.05
nonadecane 0.05
eicosane 0.03

Aliphatic Aldehydes (0.15%)
3-methylbutanal 0.15

Monoterpenes (21.15%)
α-pinene 14.73
camphene 0.14
β-pinene 0.35
myrcene 0.09
α-fenchene 0.04
pinadiene 0.07
α-phellandrene 0.31
p-cymene 1.87
limonene 3.01
γ-terpinene 0.15
terpinolene + p-isopropenyltoluene 0.35
unidentified 0.04

Monoterpenoid Ethers (66.12%)
1,8-cineole 66.1
anhydrolinalooloxide 0.02

Monoterpene Alcohols (4.00%)
fenchol 0.10
trans-pinocarveol 2.64
verbenol 0.04
α/δ-terpineol 0.44
borneol 0.08
terpinen-4-ol 0.13
p-cymene-8-ol 0.04
1-(7)-8-p-menthadienol 0.26
myrtenol 0.04
carveol 0.08
a menthadienol 0.15

Monoterpenoid Carbonyls (1.40%)
α-campholenic aldehyde 0.06
isopinocamphone 0.01
pinocarvone 1.02
myrtenal 0.07
verbenone 0.04
1-acetyl-3-isopropyl-cyclopent-5(1)-ene 0.12
carvone 0.08

Esters (1.10%)
3-methylbutyl-3-methylbutyr. (isoamyl isovalerate) 0.02
1,8-epoxy-p-menthyl acetate 0.08
α-terpinyl acetate 0.88
β-phenylethyl 3-methyl butyrate 0.12

Sesquiterpenes (3.10%)
α-gurjunene 0.04
β-caryophyllene 0.12
β-gurjunene 0.11
aromadendrene 2.21
humulene 0.05
allo-aromadendrene 0.45
eremophyllene 0.10
δ-guaiene 0.02

Sesquiterpene Alcohols (1.00%)
epiglobulol 0.15
globulol 0.47
β-eudesmol 0.08
unidentified 0.30

Oil of Eucalyptus Globulus

Oleum Eucalipti Globulus
Essence de Eucalyptus Globulus
Oel von Eucalyptus Globulus
Aceite Esencial de Eucaliptus Globulus
Olio essenziale de Eucalipto Globulus
CAS reg. no. 8000-48-4
COE no. 185
FEMA no. 2466
ISO/R 770-1968

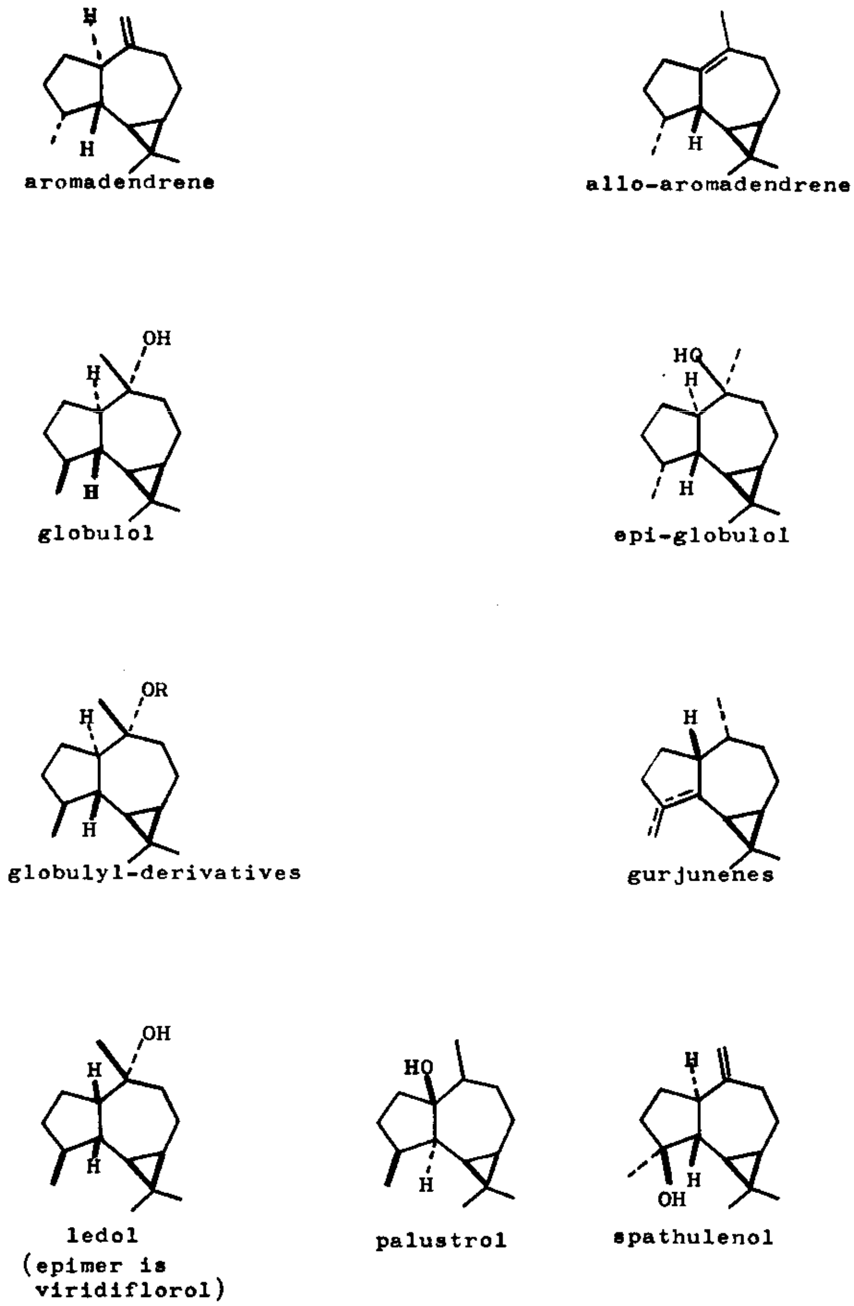

Figure 1. Sesquiterpinoids and Derivatives from *Eucalyptus Globulus* Oil

According to the published data and our own investigation the essential oil obtained from *Eucalyptus globulus* leaves may contain up to one hundred constituents, which can be divided into the groups seen in Table IV.

The main constituents of *E. globulus* oil will certainly have been formed biosynthetically. The biogenesis of 1,8-cineole in different *Eucalyptus* species starting from marked mevalonate, via monoterpenoids has been proved by Birch.[15,46] Lawrence postulated biosynthetic pathways for the formation of bicyclic compounds from α- and β-pinene, especially for terpenoid alcohols and carbonyls.[57]

However, one can question whether certain mono- and sesquiterpene derivatives (see figures 1 and 2) have not been formed in the plants by normal chemical reactions under the influence of

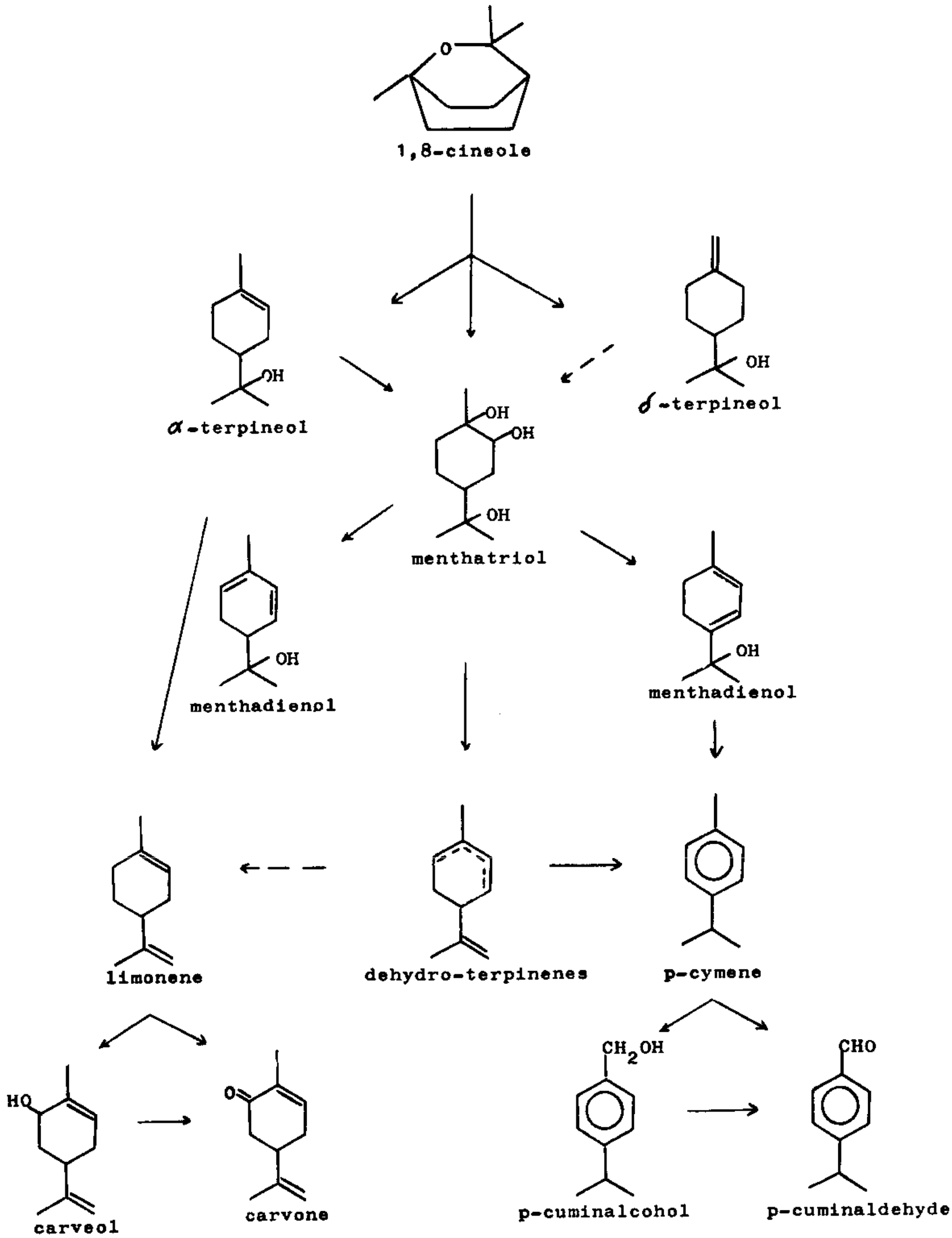

Figure 2. Possible Degradation of 1,8-Cineole

light and air.

Monoterpene alcohols may be formed by hydration or oxidation of monoterpenes. Moreover hydrolysis, followed by dehydration, of 1,8-cineole can possibly be a route to some of these alcohols, for instance menthadienols (see figure 3). The presence of the dominant monoterpenoid alcohol, trans-pinocarveol, may be due to the bio-oxidation of β-pinene.[57] However this product could also be formed by epoxidation of α-

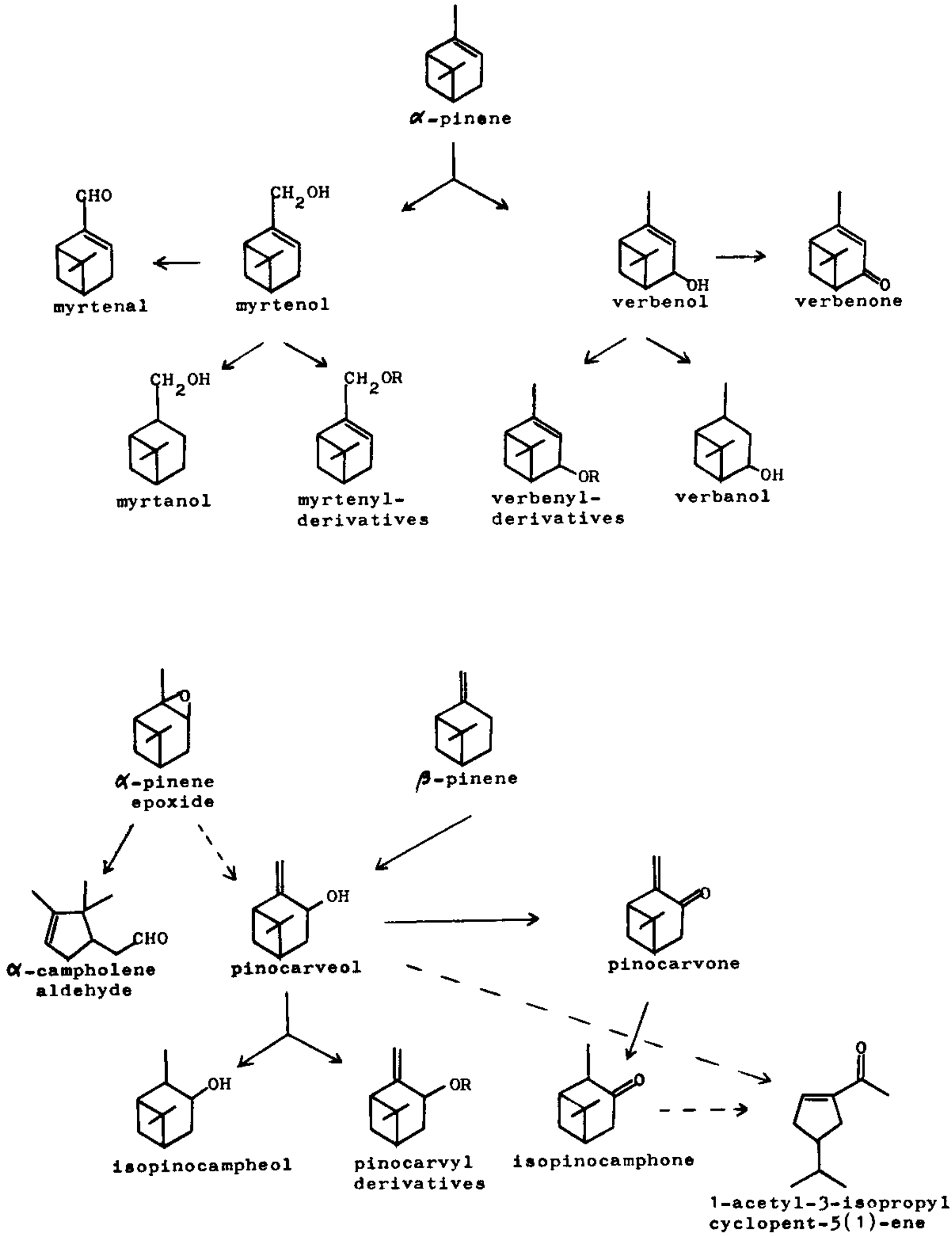

Figure 3

pinene, followed by dehydration.

From a chemical point of view the formation of monoterpenoid carbonyls is interesting. The most important precursors for these compounds are α- and β-pinene. Here again the question can be put forward whether these compounds, aldehydes and ketones, are formed biochemically in the plants, without outside influences, or under the influences of external conditions, like light, air and temperature, by normal chemical reactions. The carbonyl compounds, for instance, may be formed by free-radical allyl-oxidation of the monoterpenes. A summary of the biosynthetic ideas of this author can be seen in figure 3.

Olfactive Properties and Application

The crude oil of *Eucalyptus globulus* has an olfactive topnote principally of isovaleraldehyde which manifests itself by an irritating action and causes coughing.

The olfactive quality of the rectified oil of *Eucalyptus globulus* is agreeable, refreshing, typical eucalpytus, 1,8-cineole-like.[5,14] Often a characteristic camphoraceous connotation is added to this odour description.[4,58] However, if one compared the olfactive quality of a rectified oil of *Eucalyptus globulus* (70-75% 1,8-cineole) with that of pure 1,8-cineole isolated from the oil, remarkable differences can be observed. The oil has a terpene-like, harsh topnote, immediately followed by the characteristic fresh, somewhat minty, camphoraceous cineole connotation. These olfactive qualities often don't last for more than one hour on a blotter at room temperature. The next odour connotation is somewhat hay- and cumic-like, reminiscent of rosemary due to the presence of monoterpenoid carbonyls, these notes last for 2-8 hours. Finally the dry-out odour quality, after 5-20 hours, is characteristic soft-woody, powdery, for which the sesquiterpenoids are responsible. The olfactive properties are summarized in Table V.

Table IV

	Approximate Number	Approximate Percentages
aliphatic compounds	20	1
monoterpenes	15	20–25
monoterpenoid alcohols	15	3–5
monoterpenoid carbonyls	15	1–2
monoterpenoid ethers	5	65–70
monoterpenoid esters	5	1–2
sesquiterpenes	10	3–5
sesquiterpenoid alcohols	10	1–2
benzenoid compounds	5	0.5

Table V. Olfactive Properties

1st topnote	: terpene-like, harsh, conifer	0–1/4 hr
2nd topnote	: fresh, characteristic 1,8-cineole, minty, camphoraceous	1/4–1 hr
middle note	: hay- and cumic-like, reminiscent of rosemary	2–8 hr
dry-out note	: woody, dusty, powdery	5–20 hr

Table VI

	Odour	Taste	Cooling Effect
general threshold	0.0002% (2 ppm)	0.001% (10 ppm)	0.02%
specific threshold	0.002% (20 ppm)	0.023% (230 ppm)	-

The performance of *Eucalyptus globulus* oil in various media is described as follows: on detergent: moderate to good; liquid alkaline (pH = 13): moderate to good; liquid acid (pH = 0.5): poor to moderate; liquid acid (pH = 2): good; cold-wave end product: good; extrait perfumes and cosmetic compounds: excellent.

Today the medical use of *Eucalyptus globulus* oil is mainly for the preparation of menthol and eucalyptus inhalation products.[58,59] Some time ago, Hall described the properties of eucalyptus oils especially in relation to their bacterial power.[60]

1,8-Cineole

The olfactive properties of 1,8-cineole have been described as fresh, camphoraceous, minty and medicinal. 1,8-Cineole is used in perfume compounds for room sprays, lotions and in all kinds of cosmetic preparations. Several publications have appeared about the odour threshold value of 1,8-cineole and values have been mentioned from 0.0002 to 7 mg/M^3 air.[61] Skramlik carried out sensory analysis of 1,8-cineole with three observers and found the values shown in Table VI.[62]

Amoore and Venstrom found the detection threshold value of 1,8-cineole to be 0.012 mg/kg water (12 ppb).[63] Later on Amoore published an average olfactory detection threshold for normal observers in 2/5 tests of 0.020 mg/1 water.[64] Pelosi discovered the specific anosmia to 1,8-cineole,[65] which may partly explain the great variance in the published threshold values.

1,8-Cineole is also used widely in pharmaceutical preparations applied internally and locally. Internally, cineole serves as a stimulating expectorant, e.g., in cases of bronchitis. Locally, cineole serves as a mild anaesthetic and antiseptic in the treatment of inflammatory conditions.

Resinoid Absolute of Eucalyptus globulus

The olfactive properties of a resinoid absolute from the leaves of *Eucalyptus globulus* are quite different from those of the steam distilled oil. The resinoid absolute has a natural green topodour with connotations of "bourgeon de cassis" (blackcurrant) and of oil of fenugreek and lovage, with other longlasting balsamic, woody, dusty and powdery odour characteristics. The odour-note of "bourgeon de cassis" (blackcurrant) is also clearly recognized at the fruit buds of *Eucalyptus globulus* and may be due to hydrogen sulfide adducts to monoterpenoid carbonyls, for example, thiopinocamphone.

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