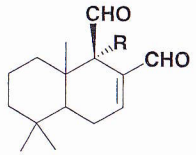
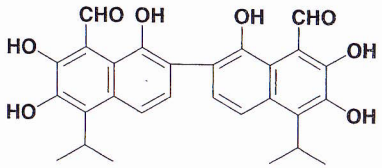
due to the (–) isomer. In China, it has been administered to male volunteers. After two months of treatment, oligospermia is observed, as well as a high frequency of spermatozoid abnormalities. Gossypol acts by destroying the seminiferous tubules, and it is efficacious, but it has non-trivial side effects: hypokalemia, gastrointestinal distress, altered libido, and prolonged sterility. The toxic symptoms are linked mainly to the (+)-isomer, and might be diminished by decreasing the daily dose. Studies in the rat tend to demonstrate the benefits of combining with gossypol the total, anti-inflammatory, and immunosuppressive glycosides of the bark-free root of *Tripterygium wilfordii* Hook., Celastraceae.



R = H : *Polygodial*
R = OH : *Warburganal*



Gossypol

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Monoterpenes and Sesquiterpenes

Essential Oils

1. DEFINITIONS

According to the 8th edition of the French Pharmacopoeia (1965), essential oils (= essences = volatile oils = oils) are: "products, generally of rather complex composition, comprising the volatile principles contained in the plants, and more or less modified during the preparation process. To extract these volatile principles, there are various procedures. Of these, only two may be used to prepare official oils: steam distillation of oil-containing plants or of selected plant parts, and expression." Next, the Pharmacopoeia specifies that the latter procedure is recommended for obtaining the *Citrus* fruit oils. Beginning with the 9th edition (1972), the French Pharmacopoeia has only used the term essential oil (in French, *huile essentielle*).

More recently (October, 1987), the French standard or *Norme Française* (= NF) T 75-006 gave the following definition of an essential oil: "Product obtained from a plant starting material, either by steam distillation, or by mechanical procedures from the epicarp of *Citrus* fruits, or by simple distillation. The essential oil is subsequently separated from the aqueous phase by physical methods."

This definition *by procedure* is restrictive: it excludes the products obtained by solvent extraction, as well as those obtained by any other process (pressurized gas, enfleurage, and others) *. Yet these other processes supply a large part of the following industrial markets: pharmaceuticals, hygiene products, cosmetics, perfumes, as well as many areas of food technology. Therefore, we find it useful to first define the terms most commonly used in this field.

* Note, however, that the products obtained from fruit juices during their concentration, or during their brief high-temperature treatment (= "flash pasteurization") are entitled to the appellation of essential oil. To be exact, they are referred to as: *fruit juice essential oils* or *juice oils*.

- ***concrete***: extract of characteristic odor, obtained from a fresh starting material of vegetable origin, by extraction with a non-aqueous solvent. In French, this is also referred to as a concrete oil, or *essence * concrète*.

- ***pomade***: perfumed fat obtained from flowers either by "cold enfleurage" (i.e., diffusion of the odoriferous constituents of the flowers into the fat), or by "hot enfleurage" (i.e., the digestion or immersion of the flowers in the melted fat).

- ***resinoid***: extract of characteristic odor, obtained from a dried starting material of natural origin, by extraction with a non-aqueous solvent. Here, starting material of natural origin means: "of vegetal, animal, or microbiological origin, including products derived from these starting materials by enzymatic routes" (AFNOR).

- ***absolute***: product of characteristic odor, obtained from a concrete, a pomade, or a resinoid, by ethanol extraction at ambient temperature. The resulting ethanol solution is generally cooled and filtered in order to eliminate waxes; the ethanol is then removed by distillation.

Essential oils may undergo a subsequent treatment designed to partially or completely eliminate a constituent or group of constituents: the result is a so-called "terpeneless", "sesquiterpeneless", "rectified", or "Xless" essential oil. We shall indicate below (p. 579) the definition of plant starting materials which combine volatile compounds and other constituents (e.g., oleoresins).

Although a fair number of essential oils are used for their medicinal virtues, many are used—because of their organoleptic characteristics—in food preparation. These are often spices or herbs.

- ***spices***: natural plant products or their mixtures, free of foreign matter, that are used to impart flavor and aroma, and to season food; the term applies both to the entire product and to the powder (French standard V 00-001, 1990). In practice and in France, the term "spice" includes traditional spices and herbs **.

* Comment: in current practice, the term oil (or essential oil) also designates odoriferous products that do not occur in the plant, but result from the enzymatic degradation of a substrate after the tissues are altered. This is especially the case for mustards and their isothiocyanates, released upon hydrolysis of the glucosinolates, or for garlic and the volatile sulfur-containing products arising from the decomposition of alliin. In the case of fruits, the term aroma is preferred.

** The ordinary dictionaries of the American language differentiate a spice from a condiment by specifying their origin. According to the Webster's New World Dictionary of the American Language, Second College Edition, a spice is a "vegetable substance [...] used to season food", whereas a condiment is "a seasoning or relish for food". The same dictionary seems to indicate that the concepts of herb and aromatic are broader: an herb is (other than in the botanical sense) any plant used as medicine, seasoning, or flavoring (this allows for uses other than as seasoning); an aromatic is a plant, chemical, etc. which has an aroma, smells sweet or spicy, or is fragrant or pungent. The definition of the term "herb" as it is used in food technology is also broad: "vegetable or part of a vegetable which naturally contains flavorful and fragrant principles"; furthermore "included in herbs are spices and aromatic condiment plants...". The gourmet reader will probably prefer the definition by Pierre Delaveau: spices are "for impact", and herbs are "for seduction". (Delaveau, P. [1987]. Les épices - Histoire, description et usage des différents épices, aromates et condiments. Albin Michel, Paris).

LAURUS NOBILIS L.

- ***aroma:*** the concept of aroma is at the same time different from, and broader than, that of essential oil, since it applies to any fragrant principle emanating from natural substances or which is generated by a physical, chemical, or enzymatic process (examples include roasted coffee, grilled meat, fish, cheese).

In a pharmaceutical context, flavors are products or substances designed to be added to medicines to mask or improve the taste or smell, excluding substances that only have a sweet, acidic, or salty taste. French legislators consider the following to be flavors: 1. natural flavors (e.g., (–)-menthol from extraction); 2. identical flavors (e.g., synthetic vanillin); 3. artificial flavors (e.g., ethylvanillin); 4. flavoring preparations; 5. process flavors; 6. the smoke flavor. The first four categories are defined by the French Pharmacopoeia (10th Ed.), which matches the current European legislation (Directive 88/388/CEE –> decree 91-366 of 17 April 1991 [amended by decree 92-814 of 17 August 1992]), which in turn specifies that flavors for medicinal use must be, at a minimum, of food quality.

2. DISTRIBUTION, LOCALIZATION, AND FUNCTION

Distribution. Essential oils occur virtually only in higher plants: according to Lawrence, there are 17,500 aromatic species. The genera capable of elaborating the compounds that constitute essential oils are distributed in a limited number of families, for example Myrtaceae, Lauraceae, Rutaceae, Lamiaceae, Asteraceae, Apiaceae, Cupressaceae, Poaceae, Zingiberaceae, and Piperaceae.

Essential oils accumulate in all of the types of vegetable organs: flowers of course (bergamot tree, tuberose), but also leaves (citronella, eucalyptus, laurel), and, although this is less common, barks (cinnamon), woods (rosewood, sandalwood), roots (vetiver), rhizomes (turmeric, ginger), fruits (allspice, anise, star anise), and seeds (nutmeg).

All of the organs of a given species may contain an essential oil, but the composition of this oil may vary with the localization. Thus, in the case of the bitter orange tree (*C. aurantium* L., ssp. *aurantium*, Rutaceae), the zest, that is the fresh pericarp of the fruit, is used to produce bitter orange oil or "*essence de Bigarade*", the flower is used to produce "neroli, neroli oil, or orange flower essential oil", and steam distillation of the leaves, twigs, and small fruits produces "petitgrain oil". These three essential oils have different compositions.

Quantitatively, the levels of essential oil are rather low, and quite often they are lower than 10 mL/kg. High levels, such as those of the flower bud of the clove (150 mL/kg and more of the dried drug), are exceptional.

Localization. The synthesis and accumulation of essential oils are generally associated with the presence of specialized histological structures, often located on or near the surface of the plant: oil cells of the Lauraceae or Zingiberaceae, glandular trichomes of the Lamiaceae, secretory cavities of the Myrtaceae or Rutaceae, and secretory canals of the Apiaceae or Asteraceae.

Function. In most cases, the biological function of the terpenoids of essential oils remains obscure. It is conceivable, however, that they have an ecological role. This hypothesis is supported by experiments which proved the role of some terpenoids in plant interactions (allelopathic agents, particularly germination inhibitors), as well as in plant-animal interactions: protection against predators (insects, fungi), and attraction of pollinating species. For some authors, the terpenoids might support some sort of "communication", especially since their structural variety allows for the transfer of selective "biological messages".

3. PHYSICAL PROPERTIES

Essential oils are liquids at ambient temperature, but they are also volatile, which is what differentiates them from "fixed oils". They are only very rarely colored. Their density is generally lower than that of water (the essential oils of sassafras, clove, or cinnamon are the exceptions). They have a high refractive index, and most of them rotate the plane of polarized light. They are soluble in common organic solvents and liposoluble.

They can be steam distilled, and are sparingly soluble in water; they are water-soluble enough, however, to impart a distinct fragrance to water. Such water is referred to in French as "*eau distillée florale*", literally "floral distilled water", an aromatic water. According to the French Pharmacopoeia, 10th edition, "*eau aromatisée florale*" or "floral aromatized water" is a similar preparation, obtained by dissolving flavors in purified water.

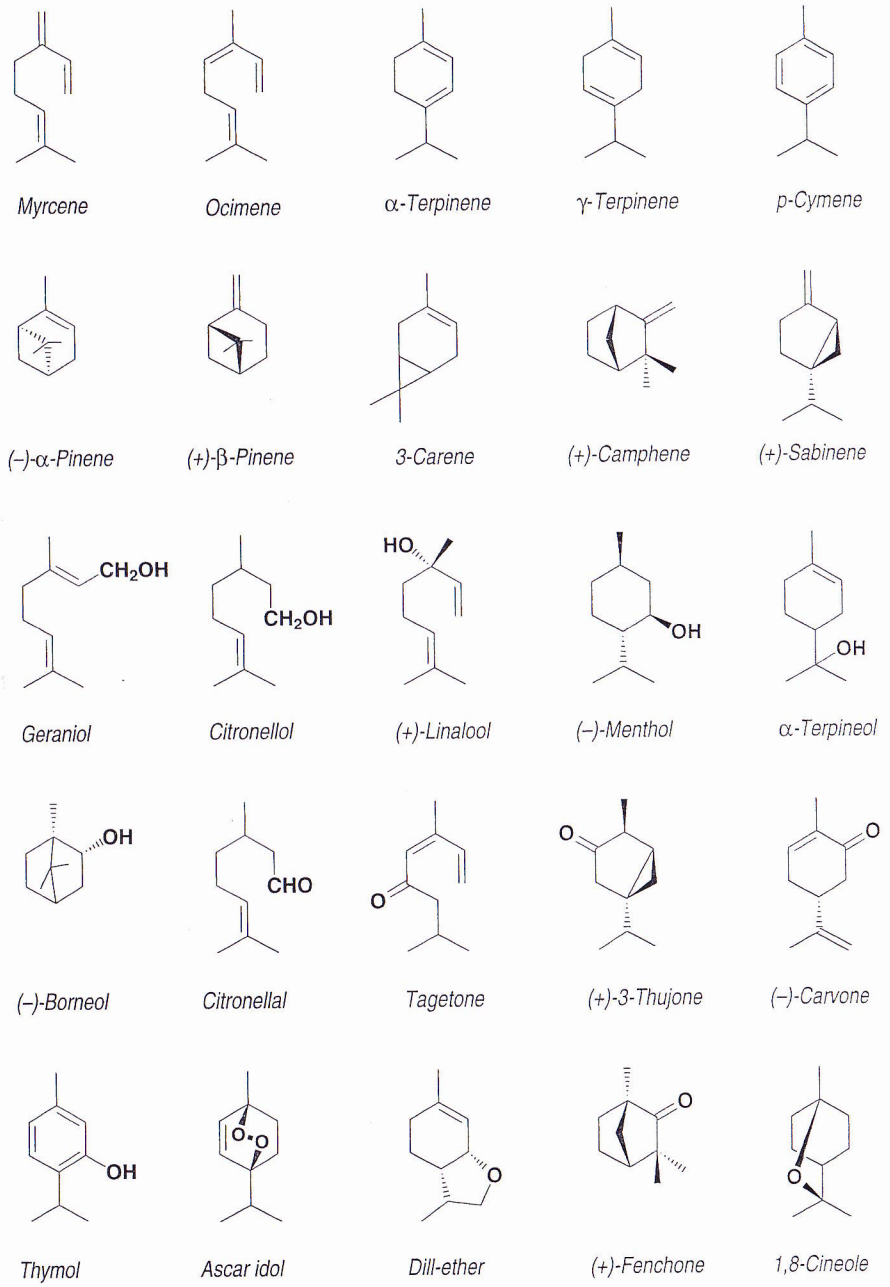
4. CHEMICAL COMPOSITION

Essential oils are complex and highly variable mixtures of constituents which belong, virtually exclusively, to two groups characterized by distinct biogenetic origins: the group of terpenoids, and the group, far less common, of aromatic compounds derived from phenylpropane. Some essential oils contain degradation products of non-volatile constituents.

A. Terpenoids

Essential oils contain only the most volatile terpenes, in other words those whose molecular weight is not too high: mono- and sesquiterpenes. We have seen above (p. 475 *sqq*) how the high reactivity of the intermediate cationic species explains the structural variety: several thousand compounds have been described in these two series.

Monoterpenes. The hydrocarbons are almost always present. They may be acyclic (e.g., myrcene, ocimene), monocyclic (e.g., α and γterpinene, *p*-cymene), or bicyclic (e.g., pinenes, Δ-carene, camphene, sabinene). Sometimes, they constitute over 90% of the essential oil, for example in the *Citrus* oils and in the [illegible]

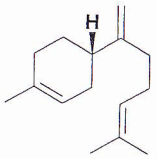


Examples of structures of acyclic and cyclic monoterpenes found in essential oils

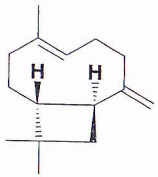
The reactivity of the intermediate cations (see above, p. 475) explains the occurrence of multiple functionalized molecules:

- alcohols: acyclic (e.g., geraniol, linalool, citronellol), monocyclic (e.g., menthol, αterpineol, terpin-1-en-4-ol), or bicyclic (e.g., borneol, fenchol);
- aldehydes: most often acyclic (e.g., geranial, neral, citronellal);
- ketones: acyclic (e.g., tagetone), monocyclic (e.g., menthone, isomenthone, carvone, pulegone), or bicyclic (e.g., camphor, fenchone, thujones);
- esters: acyclic (e.g., linalyl acetate or propionate, citronellyl acetate), monocyclic (e.g., menthyl or αterpinyl acetate), or bicyclic (e.g., isobornyl acetate);
- ethers: 1,8-cineole (also known as eucalyptol), dill ether; cyclic, tetrahydrofuran, di- or tetrahydropyran ethers, some of which play a major role in fruit flavors (linalol oxide, rose oxides);
- peroxides: ascaridol;
- phenols: thymol, carvacrol.

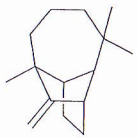
When the molecule is optically active—which is almost always the case—the proportions of the two enantiomers vary considerably depending on the plant species. One of the two enantiomers can be the major one by far or even the only one (i.e., >99%); examples include (–)-Δ-carene of the volatile fraction of black pepper oleoresin, (+)-Δ-carene of turpentine, (+)-(*S*)-linalool, the major enantiomer of coriander, nearly pure (–)-(*R*)-linalool of sweet basil and lavender, nearly racemic linalool of passion fruit, nearly pure (+)-(*S*)-terpin-1-en-4-ol of lavender, and (–)-(*R*)-terpin-1-en-4-ol, predominant in *Eucalyptus globulus*.

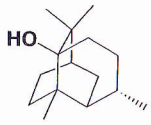


β-*Bisabolene*

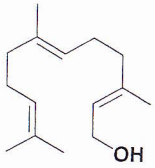


β-*Caryophyllene*

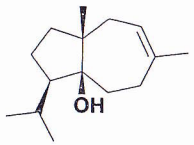

Longifolene



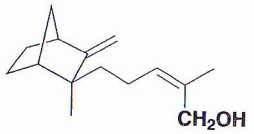
Patchoulol



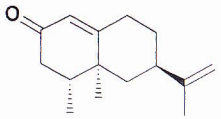
trans, trans-Farnesol



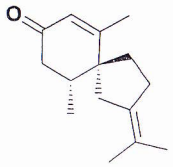
Carotol



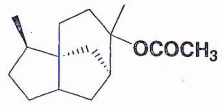
β-*Santalol*



(+) Nootkatone



β-vetivone

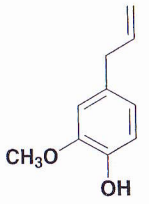


Cedryl acetate

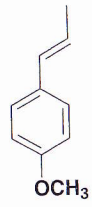
Sesquiterpenes. Structural variations in this series are of the same nature as in the monoterpenes, with hydrocarbons, alcohols, and ketones being the most common. It is noteworthy that the elongation of the chain (FPP) increases the number of possibilities for cyclization, hence the very large variety of known structures (over a hundred different skeletons have been described). Examples of sesquiterpenes characteristic of essential oils will appear below: mono- or polycyclic hydrocarbons (βbisabolene, βcaryophyllene, longifolene), alcohols (farnesol, carotol, βsantalol, patchouli alcohol), ketones (nootkatone, *cis*-longipinane-2,7-dione, βvetivone), aldehydes (sinensals), and esters (cedryl acetate).

B. Aromatic Compounds

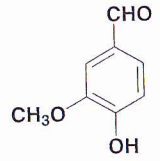
Phenylpropanoids (C_6-C_3) are far less common than terpenoids. Very often, they are allyl- and propenylphenols, and sometimes, they are aldehydes characteristic of certain Apiaceae oils (anise, fennel, parsley: anethole, anisaldehyde, apiole, methylchavicol [=estragole]), but also of those of clove, nutmeg, tarragon, sweet basil, calamus, cinnamons, and more (eugenol, safrole, asarones, cinnamaldehyde). Also found in essential oils are C_6-C_1 compounds such as vanillin (rather common) or methyl anthranilate. Lactones derived from cinnamic acids (i.e., coumarins) can be steam distilled, or at least this is true for the simplest ones, therefore they are found in some essential oils (particularly those from Rutaceae).



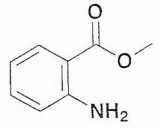
Eugenol



E-Anethole



Vanillin



Methyl anthranilate

C. Compounds of miscellaneous origins

These compounds arise from the conversion of non volatile constituents and often contribute to fruit flavors. They can be found in concretes and absolutes, which makes sense, given their mode of preparation. When the compounds are extractable by steam distillation, they can be found in essential oils.

Compounds arising from fatty acid degradation

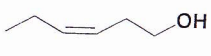
- The peroxidation of linoleic and αlinolenic acid induces their cleavage, the formation of C_9 or C_{12} acids, and the subsequent formation of low molecular weight alcohols, aldehydes, and esters, e.g., (3*Z*)-hexen-1-ol, (2*E*)-hexenals and their

acetate. Like methylketones, this type of derivative can also arise from a classical β oxidation mechanism. On the other hand, the hydroxylation of a double bond of a fatty acid is the only plausible pathway to γ and δlactones, e.g., the massoialactones of the bark of *Cryptocarya massoi* (Oken) Kosterm. of Irian Jaya (Lauraceae) and tuberolactone.

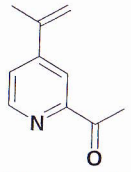
• Fatty acids are also the precursors of the jasmonic acids, their esters, and the δ jasmin-lactones: (–)-(*R*)-jasmin-lactone of jasmine (*Jasminum grandiflorum* L.) and (+)-(*S*)-jasmin-lactone of tuberose (*Polianthes tuberosa* L.). One of the proposed mechanisms for the formation of these compounds is analogous to that of prostaglandin biosynthesis in animals.

Compounds arising from terpene degradation

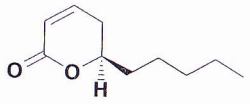
• C_{13}-*nor*isoprenoids. The principal C13-*nor*isoprenoids, the ionones, arise from the autoxidation of carotenes. They are widely distributed (e.g., violet) and commonly found in fruit flavors. Damascenones (rose, geranium) and damascones have a similar origin (allene-type carotenoids).



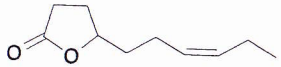
(3Z)-Hexen-1-ol



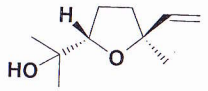
2-Acetyl-4-isopropenyl pyridine



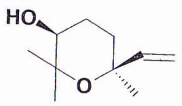
Massoialactone

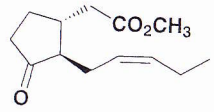


γ-Jasminlactone

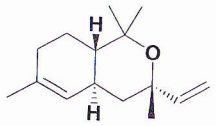


Linalol oxide(s)

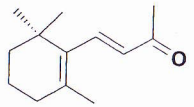




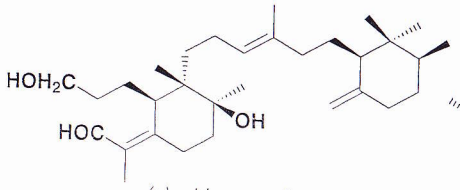
(–)-Methyl jasmonate



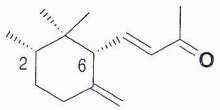
Cabreuva oxide "A"



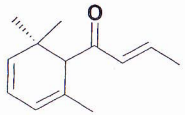
β-Ionone



(+)-γ-Irigermanal



(–)-(2S,6R)-cis-γ-Irone



β-Damascenone

• Irones. Irones are C_{14} ketones and are also degradation products. They are characteristic of the iris absolute (*Iris florentina* L., *Iris pallida* Lamk., *Iris germanica* L.); they are not preformed, instead they appear as the rhizome ages. They arise from the oxidation of bicyclic triterpenes, the iridals (iripallidal, iriflorentinal, irigermanal free or esterified by fatty acids).

Other compounds

Nitrogen- or sulfur-containing compounds are characteristic of roasted or grilled products, and are rarely found in essential oils. Some examples are the pyrazines and butenethioates of galbanum (*Ferula* spp.), and 2-acetyl-4-isopropenyl-pyridine and other pyridines in spearmint oil. (See also the sulfur-containing products formed upon bruising garlic or steam distilling mustard oil.)

In concretes, higher molecular weight constituents are not uncommon. They are extractible by solvents and not by steam distillation. They include homologs of the phenylpropanoids and diterpenes.

Comment

Glycosides of volatile substances

In 1993, twenty years after the isolation of a glucoside of linalool from rose petals, over 200 glycosides of volatile substances had been isolated and described. Their aglycones are generally identical to those found in essential oils: monoterpenes (e.g., linalool, αterpineol, menthol, thymol), sesquiterpenes (e.g., αcadinol, viridiflorol, αbisabolol), phenylpropanes and derivatives (e.g., eugenol, vanillin, 2-phenylethanol), aliphatic alcohols (e.g., hexenols, octenols), phthalides, but also many C_{13}-*nor*isoprenoids.

These compounds appear to be widely distributed: they are found in essential oil-containing plants (e.g., mint, rosemary, *Pinus* spp., cinnamon, celery) as well as plants devoid of essential oil. Research on fruit flavors and their origin has led to the finding of many glycosides of volatile substances in most flavors (e.g., *Prunus* spp. [e.g., peach, apricot], mango, pineapple, strawberry, grape).

5. VARIABILITY FACTORS OF ESSENTIAL OILS

Before enumerating succinctly the chief factors that can potentially influence the composition of essential oils, it seems useful, judging by the confusion which reigns over the naming of commercial products, to emphasize the importance of nomenclature.

It is not unusual to encounter, in the essential oil and concrete market, products for which the source organ (leaf, bark, or other), or the geographical origin, or even the botanical origin, are not specified with all of the desirable rigor. The same lack

published in so-called "scientific" reviews... To understand (which does not mean to excuse) shortcuts that create ambiguity, we must acknowledge that traditional appellations are sometimes equivocal.

For example, oregano or oregano oil are often discussed, but are they from Greek oregano (*Origanum vulgare* L. ssp. *viride* [Boiss.] Hayak), Spanish oregano (*Corydothymus capitatus* [L.] Hoff. and Link.), Mexican oregano (*Lippia graveolens* HBK), or Turkish oregano (*Origanum onites* L.)?

Another example is that of sandalwood, *Santalum album* L. (Sapindaceae), which has nothing to do with red sandalwood, *Amyris balsamifera* L. (Rutaceae), and is not the same as Australian sandalwood, *Santalum spicatum* (R. Br.) DC., or African sandalwood (*Colpoon compressum* Berg., *Osyris tenuifolia* Engl.).

We could list many more examples (thymes, cedars, citronellas, and more) which, like the examples of oregano and sandalwood, illustrate why the naming rules for these compounds must be respected *, and why the Latin binominal denomination (with the describer's name!) is the only one that does not suffer (theoretically!) from any ambiguity.

Occurrence of Chemotypes

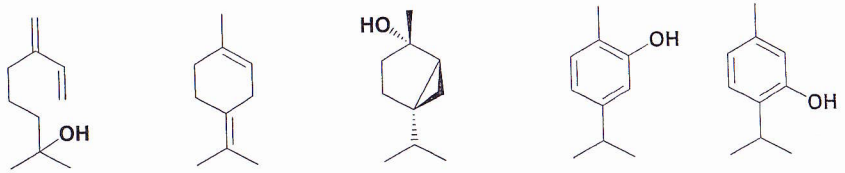
Chemotypes, also referred to as chemical breeds, are very common among plants containing essential oils. One of the best illustrations is thyme (*Thymus vulgaris* L.) from the western Mediterranean area. This species, which is morphologically homogeneous and has a stable karyotype, has seven different chemotypes: six in the arid lands or *garrigues* of the south of France (with either thymol, carvacrol, geraniol, linalool, αterpineol, or with both *trans*-4-thujanol and *cis*-8-myrcenol) and one in Spain (with cineole). The same phenomenon is observed for other *Thymus* species, but also for other Lamiaceae: thymol and carvacrol chemotypes have been shown in some species of *Thymbra, Satureja, Majorana, Origanum*, and *Corydothymus*. To give only one more example, there are three chemotypes of bay rum tree, i.e., *Pimenta racemosa* (Miller) J. Moore var. *racemosa*: a "clove" type rich (56 %) in eugenol and chavicol, a "lemon" type with

* Such rules exist: French standard NF T 75-002 specifies that the label must include, among other things, "the commercial designation of the essential oil, the (Latin) name of the plant, the part of the plant from which the oil was extracted, and the production technique or specific treatment (distillation or expression)".

French standard NF T 75-004 [1976], equivalent to ISO 3218 defines the naming rules to be followed for all possible parameters (chemotypes, clones, interspecies hybrids, various geographical origins, production site, and so on). The botanical name itself is standardized (French standard T 75-005 [1988]; see also the booklet on spice nomenclature, ISO 676 [1996]).

Examples of denominations are: essential oil of *Eucalyptus dives* Aiton - *rich in cineole*; essential oil of *leaves* of clove; essential oil of lemon of Italy - *obtained by expression*.

mostly geranial and neral (40+30%), and an "anis" type whose essential oil contains 48% methyleugenol and 32% estragole.



Myrcenol — Terpinolene — Sabinene trans-hydrate = (+)-4-Thuyanol — Carvacrol — Thymol

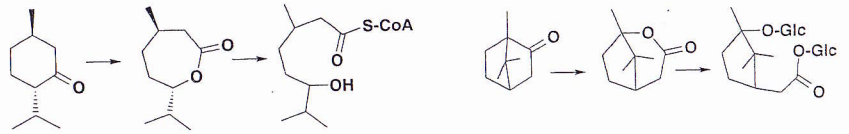
Influence of the Vegetative Cycle

For a given species, the proportions of the different constituents of an essential oil may vary greatly throughout its development. Thus, in the peppermint (*M.* x *piperita* L.), the decrease in (–)-menthone level observed during the vegetative cycle corresponds to a decrease in (–)-menthol and in (+)-neomenthol: the level of (–)-menthol (in the free state or esterified) increases, but that of (+)-neomenthol does not - quite the opposite - because it is converted to a water-soluble derivative, (–)-neomenthol glucoside *. Rather wide ranges are frequently observed in other species, including fennel, carrot, and coriander (the level of linalool is 50% higher in the ripe fruit than in the unripe fruit).

Influence of Environmental Factors

This is the impact of environmental factors and cultivation practices. The temperature, relative humidity, total duration of daylight, and wind patterns exert a direct influence, especially on species that possess superficial histological storage structures (e.g., glandular trichomes of Lamiaceae). When the localization is deeper, the oil quality is far more constant.

* Interestingly, this glucoside is transported into the rhizome, hydrolyzed, and reoxidized to (–)-menthone. In turn, this ketone is oxidized to a lactone, and this makes possible the opening of the ring and the initiation of a βoxidation-like process. Experiments with the tritiated lactone show that the carbon is reused (formation of tritiated fatty acids and phytosterols). The existence of this catabolism and of this *turn-over* drastically changes the classic view that the monoterpene accumulates as the end product of a passive process.



A similar phenomenon is known to occur in sage (*S. officinalis*): the decrease in the level of (+)-camphor which is observed when the leaves have reached their maximal size [illegible] to the formation of a lactone and to its subsequent solubilization by glycosylation.

A few examples will illustrate the influence of environmental factors:

- in the peppermint, long days and temperate nights lead to higher yields in oil, and to an increase in the menthofuran level. In contrast, cold nights favor the formation of menthol;
- in *Laurus nobilis*, the essential oil concentration of the leaves with a southern exposure is greater than that of the leaves with a northern exposure;
- in some *Citrus*, the higher the temperature, the higher the oil content. There are many, many more examples.

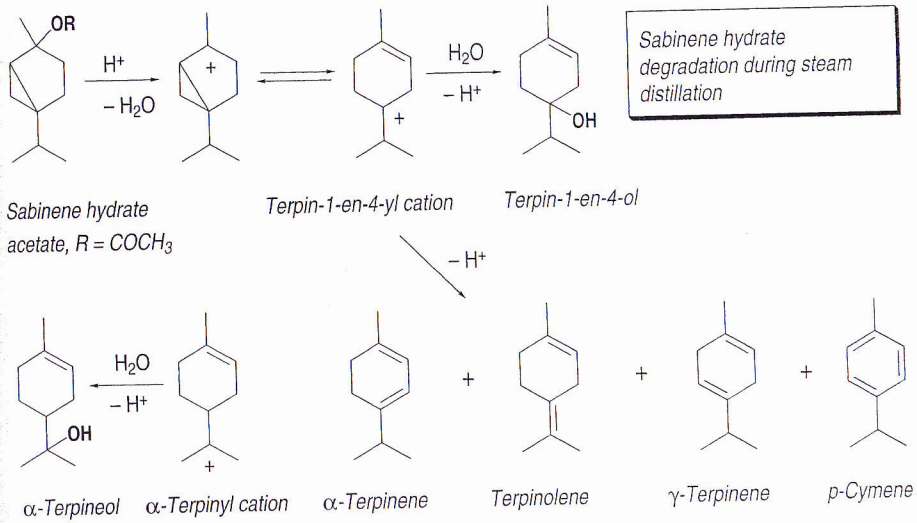
Cultivation practices are also a determining factor for the yield and the quality of the final product. Fertilization and the influence of variable amounts of N, P, and K have been studied for various species. Experience shows that there are no general rules applicable to all cases. Another fundamental element is the watering regimen. Again, it would not be wise to generalize anything.

Influence of the Preparation Method

The lability of the constituents of essential oils explains why the composition of the product obtained by steam distillation is most often different from that of the mixture of constituents initially found in the secretory organs of the vegetable. During the steam distillation, the water, the acidity, and the temperature may induce hydrolysis of the esters, and also rearrangements, isomerizations, racemizations, oxidations, and more. The behavior of sabinene *cis*-hydrate illustrates this instability. This compound and its acetate are by far the major constituents of the essential oil obtained from the flowering tops of marjoram (*Origanum majorana* L.) by crushing them in liquid nitrogen, then extracting them with pentane. In contrast, the steam distillation of the same stems yields a product which contains a large proportion of terpin-1-en-4-ol, alongside γ- and αterpinene, and other compounds, but in which the level of sabinene *cis*-hydrate acetate has become negligible. An experiment conducted with (synthetic) sabinene *cis*-hydrate and its acetate shows that they are destroyed simply by refluxing in water: after 30 minutes, only 10% of the acetate and 85% of the alcohol are left, while the following compounds appear: terpin-1-en-4-ol (major compound), γ- and α terpinene, *p*-cymene, limonene, terpinolene, and αterpineol. This helps to understand the wide range of variability which is found in the literature for the composition of this essential oil (practically 0-40% for sabinene hydrate, or terpin-1-en-4-ol, or both).

Experiments on tea tree oil (Myrtaceae, see p. 559) confirm the instability of the hydroxylated derivatives of sabinene and complete the example of marjoram. The older the leaves, the higher the initial concentration of sabinene *cis*-hydrate; the converse is true for terpin-1-en-4-ol. The experiments also show the determining influence of the pH of the medium on the composition of the final product. Using $H_2{}^{18}O$ shows that the formation of derivatives with a *p*-menthane skeleton involves the terpin-1-en-4-yl cation and, marginally, the αterpinyl cation.

Among the factors which influence the essential oil composition is the state of the starting material: in some Lamiaceae, storage for 24 hours is enough to ind...



detectable changes in composition—which may be desirable. Note that the distillation kinetics are not the same for all the constituents of an essential oil (e.g., hydrocarbons, alcohols, ketones): the composition of the distillate varies as a function of time. Hence the importance of studying, defining, and controlling all parameters, from cultivation to the preparation of the final product, in order to ensure its quality and reproducibility.

6. METHODS OF PRODUCTION

A. Of Essential Oils

By steam distillation

- Simple steam distillation consists in immersing the plant material to be treated (intact or crushed [turbodistillation]) directly in a still filled with water, which is then brought to a boil. The heterogeneous vapors are condensed on a cold surface, and the essential oil separates based on the difference in density and immiscibility. In one variation of this procedure, the plant material is crushed *in situ* (in a turbo-extractor).

- In saturated steam distillation, the plant does not come in contact with the water: the steam is injected through the plant material placed on perforated trays. To shorten the duration of the treatment, limit the alteration of the constituents of the essential oil, and conserve energy, it is possible to operate under moderate pressure (1 to 3 bar). The pressurization causes an increase in temperature, and the quality of the product may suffer.

Saturated steam distillation can also be conducted on-line, in automated set-ups. For the production of some essential oils (lavender, mint), mobile stills are used,

which are truck-size containers designed to be filled with the harvested plant material, then fitted between the steam generator and the condenser tank at the distillation facility.

- Hydrodiffusion consists of sending pulses of steam under very low pressure (0.02-0.15 bar) through the plant material, from top to bottom. The composition of the resulting products is qualitatively markedly different from that of the products obtained by the classic methods. The procedure saves time and energy.

By expression of Citrus *epicarps*

The principle of this method is quite simple: the rind is lacerated, and the contents of the ruptured secretory cavities are recovered by a physical process. The classic process consists in applying an abrasive action on the surface of the fruit in a flow of water. After eliminating the solid waste, the essential oil is separated from the aqueous phase by centrifugation. Other machines break the cavities by depression, and collect the essential oil directly; this approach precludes the degradations linked to the action of water. In fact, most facilities allow the simultaneous or sequential recovery of the fruit juice and of the essential oil, by collecting the latter with a spray of water after abrasion (scarification, puncture by pins) before or during the expression of the fruit juice. The enzymatic treatment of the residual water allows recycling, and markedly increases the final yield of essential oil. *Citrus* oils are also obtained directly from the fruit juices (by vacuum de-oiling).

Other methods

Novel technologies have emerged in the past few years. One example is steam distillation by microwaves under vacuum. In this procedure, the plant is heated selectively by microwave radiation in a chamber inside which the pressure is reduced sequentially: the essential oil is distilled away in the azeotrope formed with the water vapor from the plant itself (fresh plants require no added water). This method is fast, consumes little energy, and yields a product which is most often of a higher quality than the traditional steam distillation product (work time divided by 5 to 10 and lower temperature).

B. Of Concretes and Resinoids

Solvent Extraction

The extraction *per se* is generally preceded by a division of the drug: bruising the fresh, wilted, or semi-desiccated organs, chopping herbaceous drugs, pounding roots and rhizomes, or turning wood into chips or shavings. The procedure is

conducted in specialized facilities (static or moving extractors, with floating filter, Soxhlet-type, on-line extractors), and their description falls outside of the scope of this text (see chemical engineering texts).

The solvent selection is influenced by technical and economical parameters: selectivity (being a good solvent for the fragrant constituents); stability, in other words chemical inertness; boiling point: not too high, so that the solvent can be completely eliminated, but not too low, to limit losses, and control cost; and handling safety (if possible, non-toxic and non-flammable).

The solvents most often used—barring any specific restrictive regulations—are aliphatic hydrocarbons: petroleum ether, hexane, but also propane, or liquid butane. Although benzene is a good solvent, its toxicity limits its use more and more. Halogenated solvents are also used (chlorinated and fluorinated derivatives of methane and ethane), as well as ethanol, which is mostly used to obtain absolutes and washed resinoids. After extraction, the solvent is distilled. At the end of the procedure, the solvent contained in the plant material is recovered by steam injection.

Low boiling point solvents have an advantage in that using them precludes the degradations that could be induced by water at acidic pHs, although solvents of slighly higher boiling point, such as acetone or alcohols, allow various interesting modifications; also, when ethanol is used, traces of ethyl ether artefacts are, in some cases, beneficial for the olfactory qualities of the final product. The main drawback of solvent extraction is the lack of selectivity: many lipophilic substances may end up in the concretes (fixed oils, phospholipids, carotenoids, waxes, coumarins, and more), and render further purification necessary. Another drawback is the toxicity of the solvents, which leads to restrictive regulations regarding their use, and to the problem of the residues in the final product. (See, for example, about solvents used in food products and food ingredients manufacturing: the European regulation or *directive* 88/344/CEE of June 13, 1988, the decree or *arrêté* of November 19, 1990, and the amendments effective January 1, 1994 that apply *directive* 92/115/CEE of December 17, 1992.

Methods Using Oils and Fats

These procedures take advantage of the liposolubility of the fragrant components of plants in fats. In the technique referred to as "enfleurage", the plant material is placed in contact with the surface of the fat, and the extraction is achieved by cold *diffusion* into the fat, whereas the "digestion" technique is carried out with heat, by immersing the plant parts in the melted fat (this is also known in French as hot enfleurage or *enfleurage à chaud*). The resulting product is a floral pomade.

The classic procedure, quite ancient and now abandoned, consisted in placing the flowers onto glass plates coated with a thin layer of fat. The perfume exhaled by the flowers dissolved in the fat. Periodically, the flowers were eliminated, and were replaced by fresh flowers until the fat was saturated. Washing the fat yielded the absolute directly.

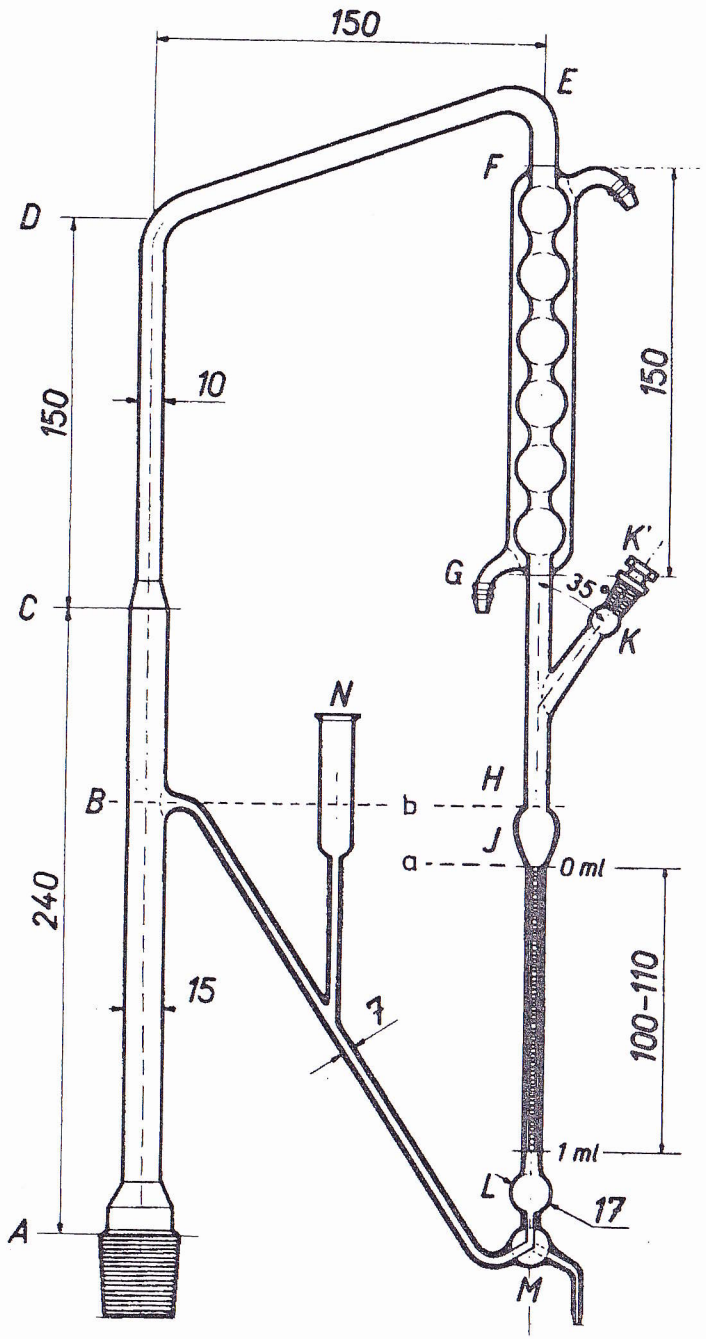
The extraction by a melted fat is still currently used, and is, in a sense, merely a specific example of solvent extraction.

Extraction by Supercritical Gases

Beyond its critical point, a fluid can have the density of a liquid and the viscosity of a gas, therefore it diffuses well through solids, and it is a good solvent. Although, in theory, several gases can be used, the focus is exclusively on carbon dioxide, and the reason for this becomes clear when considering its advantages: it is a natural product, chemically inert, non-flammable, perfectly non-toxic, easy to eliminate completely, selective, readily available, chemically unreactive, and inexpensive. The technological constraints are far from negligible (the critical point is at P = 73.8 bar and 31.1 °C), but there are many advantages: the ability to obtain extracts of composition very close to that of the natural products, the possibility of adjusting the selectivity, viscosity, and so on, by fine tuning the temperature and pressure (simultaneous extraction and fractionation), the absence of hydrolysis or rearrangement. These advantages explain why the method is currently spreading despite the high cost of the initial investment. Initially developed to decaffeinate coffees, prepare hops extracts (for the brewery industry), or to remove nicotine from tobacco, the method is now applicable to the preparation of spice extracts (ginger, paprika, celery), flavors (black tea, oak wood smoke), and plant oils, pure, free of the terpenes which add nothing to the smell and are oxidizable, or free of selected constituents, for example thujoneless wormwood oil. Extending the use of supercritical fluids to liquid-liquid extraction and chromatography expands the potential of this approach even further. In the near future, supercritical propane could be used.

Subsequent Treatments of the Oils

Sometimes, it is necessary to decolorize, neutralize, or rectify the oils obtained. Rectification, carried out dry or with a steam jet under vacuum, allows for the elimination of smelly or irritating products, and the obtention of a final product of desired "profile". The purpose of terpene removal is to eliminate terpenoid hydrocarbons. It is merely a specific example of rectification, and can be accomplished by other means: for example, by selective extraction of the oxygenated components of the oil by dilute alcohols, followed by distillation. Chromatographic techniques, especially gel filtration chromatography, permit a good separation of the essential oil from non-volatile lipophilic compounds, and even a prefractionation of mono- and sesquiterpenes.



Apparatus for the quantitation of essential oils in plant drugs
Dimensions in millimeters

European Pharmacopoeia

7. QUALITY CONTROL FOR DRUGS CONTAINING ESSENTIAL OILS AND QUALITY CONTROL FOR ESSENTIAL OILS

The quality control of drugs containing essential oils is codified by the various pharmacopoeias. The morphological and microscopic examinations are no departure from the ordinary, except for the fact that it is always possible to visualize essential oils *in situ* by using the appropriate lipophilic dyes. The drug assay normally comprises a preliminary analysis by TLC of the essential oil (obtained during the quantitation).

Quantitation of the Essential Oil. The main quantitative determination, other than residual water, is the quantitation of the essential oil. This quantitation is carried out by steam distillation in a special apparatus, which is described in the latest edition of the European Pharmacopoeia (2.8.12), and shown on the opposite page. The procedural steps are as follows: distillation of the drug in aqueous suspension, recovery of the distillate in the graduated tube which already contains a known amount of xylene, whose role is to retain the essential oil, phase separation by decanting, reading of the total volume of the organic phase, and calculation of the volume of essential oil by subtracting the known volume of xylene. The operating conditions (weight of the aliquot [5-50 g], rate of distillation [2-4 mL/min], boiling time [1.5 to 4 h]), are specified in each monograph. They depend on the nature, texture, and essential oil content of the drug. Laboratories that specialize in the analysis of this type of drugs also use apparatuses requiring a smaller sample and a shorter analysis time (e.g., simultaneous solvent extraction and steam distillation, Likens-Nickerson apparatus).

Quality Control of Essential Oils. Pharmacopoeias require different tests: evaluation of the miscibility with ethanol, physical measurements (refractive index, optical rotation, relative density, sometimes solidification temperature), determination of various indexes (acid, ester, carbonyl), and in some cases verification of the absence of fixed (fatty) oils and resinified essential oils, residue on evaporation, and so on. They also require an analysis of the essential oil by a chromatographic technique.

Although less powerful than GC, TLC of essential oils can be used routinely for quality control purposes (silica gel plates, solvent systems based on toluene or benzene, and chloroform, ethyl acetate, or both; detection by UV and by spraying various reagents: sulfuric acid followed by vanillin and heat, antimony chloride, and more). No matter what improvements are made (modified layers), TLC analysis remains clearly insufficient.

Gas Chromatography. Given the volatility of the constituents, the most suitable analytical method is GC. Its ease of implementation, fairly short analysis times, the reliability of the results and the automation potential make this a particularly

on packed columns, essential oil analysis is now commonly conducted on capillary columns, which provide better resolution in a shorter run time, and give more precise retention times. The use of typical gas chromatographic profiles as references is becoming generalized in Pharmacopoeias and standards. A "type chromatogram" is provided for information only, whereas a "chromatographic profile" is a standard to be met. The methods themselves are standardized (see Eur. Ph., 3rd Ed., 2.2.28; French standard NF T 75-400 and 401, equivalent to ISO 7359 and 7609).

Chromatographic Profile. The chromatographic profile is a list of constituents selected among those that are *representative* and *characteristic* of an essential oil, including a tolerance range for the concentration of each one, and in some cases, concentration ratios. A constituent is *representative* if it is found in all samples at a concentration whose frequency distribution is gaussian. A *characteristic* constituent is a representative constituent whose concentration—which may be zero—is a characteristic of a given oil (e.g., 10-epi-γ-eudesmol absent in geranium Bourbon, found in African geranium). The guidelines for developing a profile are the subject of an explanatory booklet (French standard NF T75-500 [1990]); implementation guidelines are also available (NF T 75-501 [1990]).

Other GC Techniques. Chiral analysis is possible by GC, which is important, considering the influence of chirality on odor, and the fact that botanical origin and enantiomer ratio are commonly linked. Chiral GC analysis is very useful for establishing product authenticity. It is now systematically carried out on stationary phases consisting of enantioselective cyclodextrin derivatives (alkylated or acylated).

The study of the volatile constituents of a plant can also be carried out by a method originally developed to study flavors: headspace GC, in other words the analysis of the gas phase in equilibrium with the sample placed in a gas-tight container. The method entails trapping the volatile substances either at very low temperature, or adsorbing them on a hydrophobic polymer, then releasing them (heat or other method of desorption), and analyzing them by GC. This technique allows, among other things, detection of the structural modifications induced by steam distillation.

The possibility of coupling chromatography with various types of spectrometers (*Fourier transform infra-red* = FTIR, MS) substantially increases the quantity and quality of the data. In GC-MS work, the computer-assisted comparison between the spectrum of the unknown peak and one or several reference libraries may lead to an identification (provided that the similarity between the two spectra, the unknown and the reference, is sufficient, and that the retention indexes are identical—in comparable operating conditions). GC-MS is a facile method, but to establish the structure of an unknown constituent, the best approach remains isolation and advanced spectral analysis (NMR).

For a more detailed study of essential oils, prefractionation may be helpful, whether it is chemical (e.g., separation of unsaturated compounds as adducts) or chromato-

deactivated supports). Various multidimensional techniques have been developed recently: two-dimensional GC, particularly for the analysis of chiral compounds (the first GC is used for prefractionation as a function of polarity of volatility, the second GC separates enantiomers on a chiral column); GC coupled with isotope ratio mass spectrometry (IRMS) to measure the $^{13}C/^{12}C$ isotope ratio. This is undoubtedly the most sophisticated method * to assess the authenticity of an essential oil, even if, as pointed out by Joulain [1994], it does not allow (*R*)-(–)-carvone from spearmint oil to be distinguished from (*R*)-(–)-carvone synthesized from (*R*)-(+)-limonene.

High Pressure Liquid Chromatography. Of little interest for volatile fractions, HPLC is an efficient way to check the authenticity of *Citrus* oils by analyzing their non-volatile constituents, or to quantitate herniarin (a coumarin) in lavender or tarragon oil. HPLC is a very good method for studying the non-volatile constituents of concretes and absolutes, or for prefractionation. Like GC, it can be coupled with mass spectrometry (LC-MS).

Electronic Noses. Electronic noses are novel and their use is spreading. This is essentially a way of obtaining an "objective" analysis of complex perfumes in a matter of minutes. The sample is analyzed by an array of sensors designed using concepts from semiconductor technology, and the data are treated by a computerized system of the neural network type. Once the numerical "fingerprint" of an odor is integrated and stored in memory, the system can use it as a reference for subsequent analyses. More cost-effective than sensory panels, capable of continuous operation without loss of "olfactive" sensitivity, this type of setup allows validation of starting materials and "on-line" process quality control (food technology, perfumery). For now, it is limited by the rapid deterioration of the sensors and the difficulties in standardizing them.

In some cases, one particular component (or group of components) must be quantitated. Age-old techniques can be used (e.g., cineole quantitation by determination of the crystallization temperature of a mixture of essential oil and *o*-cresol [French standard NF T 75-118, equivalent to ISO 1202]), in addition to chromatographic methods (e.g., safrole and isosafrole quantitation in nutmeg oil).

8. PHARMACOLOGICAL PROPERTIES OF ESSENTIAL OILS

First of all, the activity of an essential oil is sometimes confused with that of the plant from which it came. In reality, such an assumption is rarely justified: for

* See also the interesting case of differentiating balm oil from citronella oil from synthetic compounds: Hener, U., Faulhaber, S., Kreis, P. and Mosandl, A. (1995). On the Authenticity Evaluation of Balm Oil (*Melissa officinalis* L.), *Pharmazie*, **50**, 60-62. See also: Schultze, W., König, W.A., Hilkert, A. and Richter, R. (1995). Melissenöle – Untersuchungen zur Echtheit mittels enantioselektiver Gas-chromatographie und Isotopenverhältnis-Massenspektrometrie,

example, the rosemary oil is an antibacterial agent, whereas the plant infusion is traditionally used to treat miscellaneous digestive symptoms, based on antispasmodic and choleretic properties, which are probably linked to the presence of phenolics.

Secondly, although it is possible to study and describe the biological or pharmacological effects of a *pure* monoterpene, sesquiterpene, or alkylbenzene—there is abundant literature on this—it is difficult, if not impossible, to discuss the pharmacology, pharmacokinetics, or the metabolism of an essential oil, because it is a *mixture*. Thirdly, we must emphasize that the scope of the properties attributed to (and sometimes demonstrated experimentally for) essential oil drugs and essential oils *per se* is too broad to permit generalizations, and would be overly simplistic.

In spite of this, a few fundamental properties stand out:

• ***Antiseptic Activity***. This antiseptic activity is against various pathogenic bacteria, including strains that are usually resistant to antibiotics. Some essential oils are also active against fungi responsible for mycoses, and against yeasts (*Candida*). The doses required for activity are generally low, and those that are determined by *in vitro* experiments are directly transferable to external use, and all the more so to the use as a preservative. Savory, cinnamon, thyme, clove, lavender, and eucalyptus are among the most antiseptic essential oils. Compounds such as citral, geraniol, linalool, or thymol are 5.2, 7.1, 5, and 20 times more antiseptic than phenol, respectively.

• ***Spasmolytic and Sedative Properties***. A large number of essential oil drugs (mint, European vervain) are thought to be efficacious in decreasing or suppressing gastrointestinal spasms. Frequently, they stimulate gastric secretion, hence the adjectives "digestive" and "stomachic", and may play a role in all the possible consequences of this "eupepsia": improvement of certain insomnias and of miscellaneous psychosomatic disorders, decrease in "nervousness", and more. These miscellaneous favorable effects probably explain why these drugs find extensive uses in folk medicine, as well as in alternative medicines.

In vitro, many essential oils (angelica, sweet basil, chamomile, clove, balm, mint, thyme) have a marked spasmolytic activity on the isolated guinea pig ileum (and, to a lesser extent, on the tracheal chain of the same animal); in a few rare cases (anise, sweet fennel), on the contrary, an increase in the phasic contractions of this organ is observed. According to studies conducted with mint oil, it is possible that this activity is linked to an inhibition of calcium entry into the cells.

• ***Irritating Properties***. When used externally, products such as turpentine cause an increase in capillary blood flow, substantial rubefaction, a sensation of heat, and in some cases, a slight local anesthetic activity: these effects were sought in the past by using embrocations or liniments. Even today, there are many ointments, creams, or gels based on essential oils, and designed to relieve sprains, soreness, strains, and other joint or muscular pains.

When administered internally, essential oils are thought to trigger "irritation" processes at different levels. Thus, the oils of eucalyptus, pine, or niaouli are thought to stimulate mucus cells, and to increase the motility of the ciliated epithelium in the bronchia; other oils are thought to enhance the renal excretion of water by a direct local effect (juniper). Most claims of this type are not substantiated by pharmacology. Clinical trials are nonexistent. Other activities are attributed to essential oils (choleretic, healing, nervous sedative, and more); they will be described in the drug monographs.

9. TOXICITY OF ESSENTIAL OILS

This facet of the knowledge of essential oils has become more important, because the development of "therapies" such as aromatherapy (see 10. below), and the label of "natural product" attached to these products, have led to their abusive use. Unfortunately, some are quick to confuse essential oil plants and essential oils: this is a serious mistake, because the innocuous character of the plants is almost always well established, whereas the toxicity of many oils has been demonstrated. Self-prescription is dangerous, and is encouraged by the fact that many of these products are available over-the-counter; in some cases, this represents a complete disregard for regulations requiring that they be released only by a pharmacist, in order to ensure tight control over their identity and quality.

Acute Toxicity. As a general rule, the acute toxicity of essential oils by the oral route is low or very low: many of the oils currently used have an LD50 between 2 and 5 g/kg (e.g., anise, eucalyptus, clove) and for most of them, it is greater than 5 g/kg (e.g., chamomile, citronella, lavender, marjoram, vetiver). About 15 other oils have an LD50 between 1 and 2 g/kg: sweet basil, tarragon, hyssop (1.5 mL/kg), oregano, savory (1.37 g/kg), *Melaleuca*, sassafras (1.9 g/kg), or wintergreen (0.9-1.25 g/kg). The most toxic essential oils are those of boldo (0.13 g/kg; convulsions begins with 0.07 g/kg), chenopodium (0.25 g/kg), thuja (0.83 g/kg), pennyroyal (0.4 g/kg), and mustard oil (0.34 g/kg).

The same observations are true for essential oil constituents. Those with an LD50 <2g/kg are rare: thujones (≈ 0.2 g/kg), pulegone (0.47 g/kg), carvacrol (0.81 g/kg), and carvone (1.64 g/kg).

These data *[p. 508] were obtained in animals and are no more than relative indications. Clinical observations in humans show that acute intoxication is possible even if the LD50 is high: camphor (LD50 = 1.47 g/kg) was formerly responsible for many accidents (epileptiform convulsions) and for deaths, at least in young children. A review of the available literature shows that serious accidents—most of which involve young children—are due to a small number of essential oils, *ingested in large quantity*: clove (eugenol), eucalyptus (contradictory data), pennyroyal (pulegone, deadly), wintergreen (methyl salicylate, deadly), parsley (apiole). Also known is the neurotoxicity of thujone-containing essential oils (thuja, wormwood,

oils induce epileptiform and tetaniform episodes, and psychic and sensory disturbances which require hospitalization. The accidents that they have caused have led the French government to put in place restrictive legislation: the French law or *loi* n° 84-534 of June 30, 1984 completed Article L-512 of the French Public Health Code or *Code de la Santé Publique* in the following manner: "Shall be reserved for pharmacists [...] the retail sale, and any and all dispensations to consumers of the essential oils whose list is determined by decree, as well as their dilutions and preparations other than cosmetic or personal hygiene products, household products, foodstuffs, and beverages." A decree in 1986 (n° 86-778 on June 23, 1986) places on the list announced in the previous text the essential oils of wormwood, maritime wormwood, sagebrush, cedar, hyssop, sage, tansy, and thuja.

Nevertheless, serious accidents are rare. In 1994, poison control centers in the United States recorded 3,185 calls involving essential oils: of those, 1,086 and 72 described minor and moderate effects, respectively. Only 4 case reports of major intoxication and 1 death (wintergreen oil, 60 mL) were documented during that year [Litovitz, 1995].

Chronic Toxicity. The chronic toxicity of essential oils is not well known, at least not for uses such as aromatherapy, and not for any route of administration: potential side effects are rarely reported (they would have to be identified as such). In contrast, much experimental data has been accumulated while trying to evaluate the risks of using essential oils (especially their constituents and the plants containing them) as food flavors (or spices, or seasonings), in which case the doses ingested daily are mostly minute and, with rare exceptions, their safety has been established—*under normal conditions*.

The potential interactions between these products (at the doses commonly used in aromatherapy) and other medications are also not well known.

Skin Toxicity. Since essential oils are widely used in perfumery and in the cosmetics industry, extensive research has been conducted on their potential toxicity (acute, chronic) by topical application, and on their irritating (mustard, thyme), sensitizing (*Saussurea*, cinnamaldehyde), or phototoxic potential (angelica,

* The numbers listed above are merely examples. They are reproduced from compilations published since 1973 by D.L.J. Opdyke of the Research Institute for Fragrance Materials (RIFM) under the title: Monographs on Fragrance Raw Materials. They were first published as articles, then as supplements to the review Food and Cosmetics Toxicology: (**I**: 1974), **12**, 807-1016; (**II**: 1975), **13**, 681-924; (**III**: 1976), **14**, 659-894; (**IV**: 1978), **16**, 637-88; (**V**: 1979), **17**, 695-92. The series continued after the title of the review changed to *Food and Chemical Technology*; (**VI**: 1982, with C. Letizia), **20**, 633-852); (**VII**: 1988), **26**; (**VIII**: 1992), **30**, 1-138. See also Adams *et al.*, 1996 (see bibliography below).

On the status of plants, their extracts, or some of their constituents, see, among others, Conseil de l'Europe (1981). Substances aromatisantes et sources naturelles de matières aromatisantes, 3rd Ed., Maisonneuve, Moulins-lès-Metz. See also the recommendation of the *Code of Federal Regulations*, Title 21, US Government Printing Office, Washington, DC.

bergamot). The results have led international professional organizations to emit recommendations on their use and that of their constituents (maximum concentrations, exclusions, specific formulations [IFRA]).

Carcinogenicity. Several allyl- and propenylphenols are capable of inducing cancer in rodents: in rats, safrole (sassafras) induces the formation of hepatic tumors, βasarone (sweet flag) that of tumors of the small intestine, and in mice, estragole (sweet basil, tarragon) causes liver cancer. These phenylpropanic derivatives are hydroxylated at C-1' on the allylic residue (1'-hydroxysafrole, 1'-hydroxyestragole) by microsomal enzymes in the rodents' liver. Subsequently, the hydroxylated metabolites form highly electrophilic species—sulfuric esters—capable of establishing covalent bonds with nucleic acids and proteins. In the current state of knowledge, apiole, dill-apiole, eugenol, and myristicin are considered non-carcinogenic.

Evaluating the risks associated with these arenes is difficult because of the variations in metabolism between species or the disproportion between the doses administered in animal experiments and the doses that humans are likely to ingest daily. In animals, the average dose is 0.5-1% of the daily ration for 1 to 2 years (e.g., 550 mg/kg/day of anethole x 121 weeks), whereas in humans, the daily intake of propenylphenols is estimated to be between 60 and 70 µg, depending on the compound. The potential influence of the ingested dose on the metabolism must also be taken into account. For example, a 0.05-mg/kg dose of estragole administered to rats mostly gets demethoxylated, but if the dose is increased to 1 g/kg, up to 10% of it gets hydroxylated at C-1'. In humans, the same compound is converted to the 1'-hydroxy derivative (0.3% of a 100-µg dose). Under these conditions, exposure to the toxic metabolite (calculated on the basis of its urinary excretion) is 13 million times greater in rodents than in humans.

A systematic study of the hepatic carcinogenicity of alkenylbenzenes has shown that *Z*- and *E*-anethole, 3'-hydroxy-*E*-anethole, *Z*-isosafrole, and cinnamaldehyde do not induce the formation of tumors in the conditions in which asarone, hydroxylated derivatives of safrole, estragole, and to a lesser extent, elemicin do. Long-term (two-year) studies in rats have confirmed that anethole presents no significant risk to humans, even at the doses ingested by regular consumers of anise-flavored beverages. The differences in metabolism lend further support to the presumption of safety: in humans, anethole is preferentially degraded by oxidation of the lateral chain (*p*-methoxyhippuric and benzoic acids) whereas in rats, the lateral chain is mostly epoxidized.

10. USES OF DRUGS CONTAINING ESSENTIAL OILS

The following monographs will illustrate the main facets of the use of these drugs. Currently, they have uses in three main fields.

- ***Pharmacy***. The vast majority of drugs are used crude, especially for the preparation of infusions (mint, balm, European vervain, orange blossoms), and

under the form of simple galenicals. They are also used to prepare essential oils, some of which may have a therapeutic interest (particularly as external antiseptics), but the major part of which are designed for the aromatization of medications for the oral route.

Essential oils are also the underpinning of a specific therapy: "aromatherapy". This "alternative" medicine whose advocates can't seem to agree on a definition* will not be covered here. However, it is important to emphasize that most essential oil constituents are lipophilic, therefore they are rapidly absorbed by the pulmonary, cutaneous, or digestive route. Utter vigilance is in order when considering doses to be used, and the use of essential oils known to be potentially toxic must be proscribed. It is self-evident that great care must be taken when essential oils are administered orally and *a fortiori*, as mixtures, but caution is also necessary when they are used externally: some are harsh on mucous membranes or on the skin and should not be used without prior dilution in an appropriate solvent. Essential oils must be kept out of the reach of children and it would be best if they could be packaged properly and labeled correctly.

- ***Perfumery***. Perfumery is the main outlet for essential oils, concretes, absolutes, and other resinoids obtained from the drugs. Cosmetology and the hygiene product industry are also consumers, even though the cost of natural products is often high and leads to the selection, for mass production, of synthetic alternatives.

On the border between pharmacy and hygiene products, note the presence of essential oils in bath products ("soothing" or "relaxing" baths): they present the risk of percutaneous absorption of terpenoid constituents.

- ***Food technology***. Some drugs are used raw (herbs and spices), others are used as essential oils, resinoids, or oleoresins, dispersed, encapsulated, or complexed. Although in the past few decades, refrigeration has replaced spices to ensure the conservation of foods, the development of new culinary practices (prepared foods, frozen dinners), a taste for the exotic (and for marketing!), the flavor (or lack thereof!) of the products of an intensive agriculture, and other factors have led to a

* For some authors, aromatherapy is the use of odors and volatile substances to treat, alleviate, or prevent infections and disorders solely by means of inhalations [Buchnauer and Jirovetz, 1994], whereas for other authors, essential oils are to be applied to the skin after dilution in a vegetable oil (aromatherapic massage) [Tisserand and Balacs, 1995]. In France, essential oils are often administered *per os*... Since these substances are in part absorbed, they can have a pharmacological effect (which remains to be evaluated). In the past few years, different research projects have tried to substantiate the "psychodynamic" action of fragrances (aromatherapy and effects of scents). There is a large psychosomatic component to the effect. Some authors also evoke a hedonic mechanism (influence on behavior of the pleasure or displeasure linked to an odor) and a "semantic" mechanism (memory link between an odor and an exceptionally emotional situation). See, among others, Jellinek, J.S. (1997). Psychodynamic Odor Effects and their Mechanisms, *Perfum. Flavor.*, 22, (09-10), 29-41 and references therein.

rapid increase in the consumption of natural flavors. All of the sectors of food technology are consumers: alcoholic and non-alcoholic beverages, confectionery, dairy products, meat products, sauces, soups, snacks, bakery products, without forgetting animal foods.

Since the early 1980s, the use of natural food flavorings has been growing at the expense of the use of synthetic flavor mixtures. Like the market for products derived from fruits, the market for essential oils probably has more growth potential. The same is true for isolates (i.e., the pure substances isolated from essential oils): the popularity of what is "natural" will continue to make some isolates competitive with their synthetic analogs. As expected, fermentation and bioconversion products of natural origin are also increasingly successful.

Miscellaneous industries, mostly chemical, still use isolates in addition to synthetic products, for example pinenes from turpentine (see p. 582), sclareol (see p. 544), citral of *Litsea cubeba* from China (see p. 553), geraniol of palmarosa oil (*Cymbopogon martinii* [Roxb.] W.Watson, Poaceae), (+)- and (–)-linalool, (+)-citronellal, eugenol, and safrole. These are starting materials for the synthesis of the active principles of medicines, vitamins, and fragrances. One example is the use of safrole (extracted from Brazilian *Ocotea* or from *Cinnamomum* species from China) to synthesize the heliotropin used in perfumery or piperonyl butoxide, a pyrethrinoid synergist.

Market for essential oil drugs and essential oils

The published figures do not allow one, except in a few cases, to grasp the extent of the pharmaceutical needs, because they do not distinguish the different consumers (pharmacy, food technology, perfumery). The major part of the 450 metric tons of peppermint consumed annually in France (1990 estimate) is directed toward consumption as an herbal tea, but the 50 metric tons of wild thyme, the 300 metric tons of rosemary, and the 1,200-1,400 metric tons of cloves chiefly have non-pharmaceutical outlets.

For essential oils, again the figures are aggregate (they take into account mixtures and compositions): French imports and exports (1995), 2.77 and 3.57 billion French francs, respectively. The estimated needs are sometimes lower than 500 kg/year (cinnamon leaf oil or savory oil); in most cases, they lie somewhere between 1 and 10 metric tons. The higher needs correspond to specific uses: mint oil (600-700 metric tons, including all grades) for food technology and hygiene products, clove leaf oil (500-800 t) chiefly for the obtention of isolates (eugenol) and for perfumery. For most of the species cultivated in France, the French production faces tough competition from countries with lower labor costs.

The top essential oils worldwide are by far the *Citrus* oils: sweet orange (26,000 t), lemon (2,200 t), and grapefruit (700 t). Other major products include

peppermint oil (3,700 t), *Mentha arvensis* oil (4,300 t), eucalyptus oil with cineole (3,300 t) or citronellal (2,000 t), citronella oil (*Cymbopogon* spp., 2,800 t), clove leaf oil (2,000 t), and coriander oil (700 t). In the early 1990s, 21 essential oils had a world market greater than 500 t. Other oils only have limited world outlets, for example matricaria (4.3 t), lovage (seed, 900 kg), costmary (100 kg) (1990 figures, rounded [Lawrence 1995]).

Storage

Evidently, the relative instability of the constituents of essential oils make their storage a challenge. The decomposition possibilities are numerous, easy to measure through indexes (peroxide, refraction), the determination of physical characteristics (viscosity, miscibility with alcohol, optical activity), and GC analysis. There are multiple risks: photoisomerization (*E* —> *Z*-anethole), photocyclization (citrals), oxidative cleavage of propenylphenols, peroxidation of hydrocarbons, and decomposition to ketones and alcohols (limonene), thermoisomerization (citrals), and more. Since degradation can alter properties or jeopardize product safety, it must be avoided by using small vials made of aluminum, stainless steel, or tinted antiactinic glass, nearly completely filled, tightly closed, sealed under a nitrogen or other inert atmosphere, and protected from heat and light. It is always possible to add antioxidants (depending on the intended application of the products). Regarding packaging, conditioning, and storage regulations, see French standard AFNOR NF T 75-001 [1996]; on labeling regulations, see NF 75-002 [1996].

11. CHIEF DRUGS CONTAINING ESSENTIAL OILS

The number of essential oil drugs that supply a substantial market is so large that we cannot plan to describe all of them, at least not in a text mainly devoted to plants with potential applications in pharmacy. Accordingly, we shall cover, sometimes succinctly, those drugs that are listed in the latest edition of the European or French Pharmacopoeia, or in the Annex I of the French Explanatory Note of 1998 regarding the marketing authorizations for plant-based medications, or in both documents. To those drugs, we will add a few more which deserve to be mentioned because of their potential toxicity (for example calamus), or for their aromatizing properties, often applied to pharmaceutical preparations (for example *Citrus* fruits). Non-pharmaceutical uses will be listed, but species with only non-pharmaceutical uses will not be included, even if they are used extensively, like the Poaceae (Java and Ceylon citronella, vetiver) or used on the sole basis of tradition (rose, geranium, jasmine, tuberose). Since a classification by indications would be fraught with difficulty (are all these drugs really medicinal plants?), we will use the botanical classification (families in alphabetical order).

A. Apiaceae Containing Essential Oils

Anise and fennel contain an essential oil in which phenylpropane derivatives predominate, particularly anethole. The acute toxicity of *E*-anethole is moderate (LD50 = 3.2 g/kg, rat, *per os*). The *Z* isomer is more toxic (LD50 = 0.24 g/kg, mouse, *per os*). The acceptable daily intake of *Z*-anethole for humans is set by international authorities at 2.5 mg/kg and estimates are that the average American consumes 60 µg of this type of product per day. Anethole consumption is far greater in France—250 mg/day for some consumers—and what is clear is that the abuse of anise-flavored alcoholic beverages is dangerous, but their toxicity is primarily that of the ethanol, present in massive quantities! (On propenylphenol toxicity, see 9. above). Most Apiaceae that contain essential oils are used primarily in food and food technology. Phytotherapists mostly attribute to them "digestive" virtues.

• ANISEED, *Pimpinella anisum* L.

The whole dried diakene is listed in the 3rd edition of the European Pharmacopoeia. It contains not less than 20 mL/kg essential oil. (On commercial categories and sources, see French standard NF V 32-168 [1984].)

The Plant, the Drug. Anise grows wild in the Near-East and is widely cultivated, among other places, on the Mediterranean rim (Spain, Balkans, Turkey, North Africa). The leaves are cordiform at the base and trifid at the top, with linear divisions; the leaves in between are compound with dentate lobes. The drug is a diakene, ovoid or pyriform, and yellowish-green (3-5 x 3 mm). The mericarps, attached by their apex to the end of the carpophore, have a convex dorsal side covered with verrucose hairs that can be seen with a magnifier.

Chemical Composition. The aniseed fruit contains polysaccharides, lipids (15-20%), flavonoids, a glucoside of *p*-hydroxybenzoic acid, and 20-30 mL/kg essential oil. The latter contains 80-95% *E*-anethole, alongside methylchavicol (= estragole), anisaldehyde, and the 2-methylbutyrate of 1-(*E*)-propenyl-2-hydroxy-5-methoxy-benzene [= pseudo-isoeugenyl-(2-methylbutyrate)]. Other derivatives (anisic acid, anisyl alcohol and ketone) may be found in partially oxidized essential oils.

Tests. The fruit is identified by morphological examination and by the microscopic characteristics of the powder: unicellular trichomes with a blunt apex, sometimes curved, and with a warty cuticle; secretory duct fragments. TLC analysis of a dichloromethane extract shows the presence of anethole and triglycerides. The assay includes a quantitation of the essential oil.

Pharmacological Properties and Uses. Like fennel, aniseed is reputed to be

PIMPINELLA ANSUM L.

anethole. In fact, the estrogenic activity of this compound is weak, and in addition, the compound could not be identified in the essential oil, even after storage under extreme conditions. Different animal experiments indicate that anethole is a spasmolytic and that it stimulates respiratory secretions and expectoration.

Long considered to be a galactagogue, expectorant, and carminative, anise fruit may now claim, in France, "digestive" indications by the oral route: traditionally used for the symptomatic treatment of gastrointestinal disturbances (epigastric bloating, impaired digestion, eructations, flatulence) and as an adjunctive treatment of the painful component of functional dyspepsia [French Expl. Note, 1998]. In Germany, where aniseed is mostly used in pediatrics—like fennel fruit—Commission E allows its use for gastrointestinal symptoms, but also, orally and topically (inhalation), for excess phlegm in the respiratory tract. The monograph includes a warning about the occasional risk of allergic reaction.

Conserving the drug is rather difficult: the concentration of essential oil decreases rapidly in storage (1% per month). The fruit must be stored in a tight container, protected from light.

Aniseed Oil. Aniseed oil (Eur. Ph., 3rd Ed.), must contain between 84 and 93% *E*-anethole and less than 0.5% of the *Z* isomer. Limits have also been established for the minor constituents: linalool (0.1-1.5%), estragole (0.5-6%), α-terpineol (0.1-1.5%), and anisaldehyde (0.1-3.5%). Curiously, the French Pharmacopoeia defines aniseed oil as "obtained by steam distillation of the ripe and dried fruits of *Pimpinella anisum* L. or *Illicium verum* Hook. *fil.*". There is no doubt that the compositions of the two essential oils are very similar, but to see the term aniseed applied to star anise is somewhat of a surprise. The text specifies that "the label must indicate whether the aniseed oil was obtained from *P. anisum* or *I. verum.*". The monograph includes the typical chromatograms (GC) of the two essential oils, which clearly show the differences in composition in minor products.

Aniseed oil is used in pharmaceutical technology as a flavor; it is an ingredient of benzoic opium tincture (better known as paregoric). In France, aniseed oil can be dispensed only by pharmacists and only by prescripion; the prescriptions must be recorded in the pharmacist's prescription log (Public Health Code or CSP, art. L 641). Anise-flavored oils (fennel, sar anise, aniseed) and semisynthetic products are used extensively in the liquor industry to manufacture beverages, alcoholic or not.

• Fennels, *Fœniculum* spp.

The European Pharmacopoeia (3rd Ed.) devotes two monographs to this genus: one to the dried fruit of *F. vulgare* (Miller) ssp. *vulgare* var. *vulgare*—the cultivated fennel—and the other to the dried fruit of *F. vulgare* ssp. *vulgare* var. *dulce* (Miller) hell. According to the Flora Europea, the Florence fennel—we eat the fleshy base of its stalks—is the var. *azoricum* (Miller) Thell. of the ssp. *vulgare.*

The Plant, The Drugs. A tall (2 m) perennial herb, fennel is easy to identify by its feathery leaves, divided into thread-like segments, its umbels of greenish-yellow flowers, and its characteristic odor. The species grows wild in the Mediterranean area and is widely cultivated (Egypt [main supplier to France in 1995], Turkey). The fruits are collected as soon as they turn yellow. The fruit of the sweet fennel is a diakene, almost cylindrical (3-12 x 3-4 mm), and surmounted by a large stylopod. The mericarps are often free and have prominent ribs. The drug smells strongly like anise, and has a penetrating and sweet taste.

Chemical Composition.

- Sweet fennel oil normally contains 80% and more of *E*-anethole, 1-5 (10)% methylchavicol (estragole), and less than 5% (+)-fenchone. Fennel fruits also contain furanocoumarins, including imperatorin, bergapten, and xanthotoxol.
- Bitter fennel oil contains 50-80% *E*-anethole, 3-20% estragole, and up to 24% (+)-fenchone.

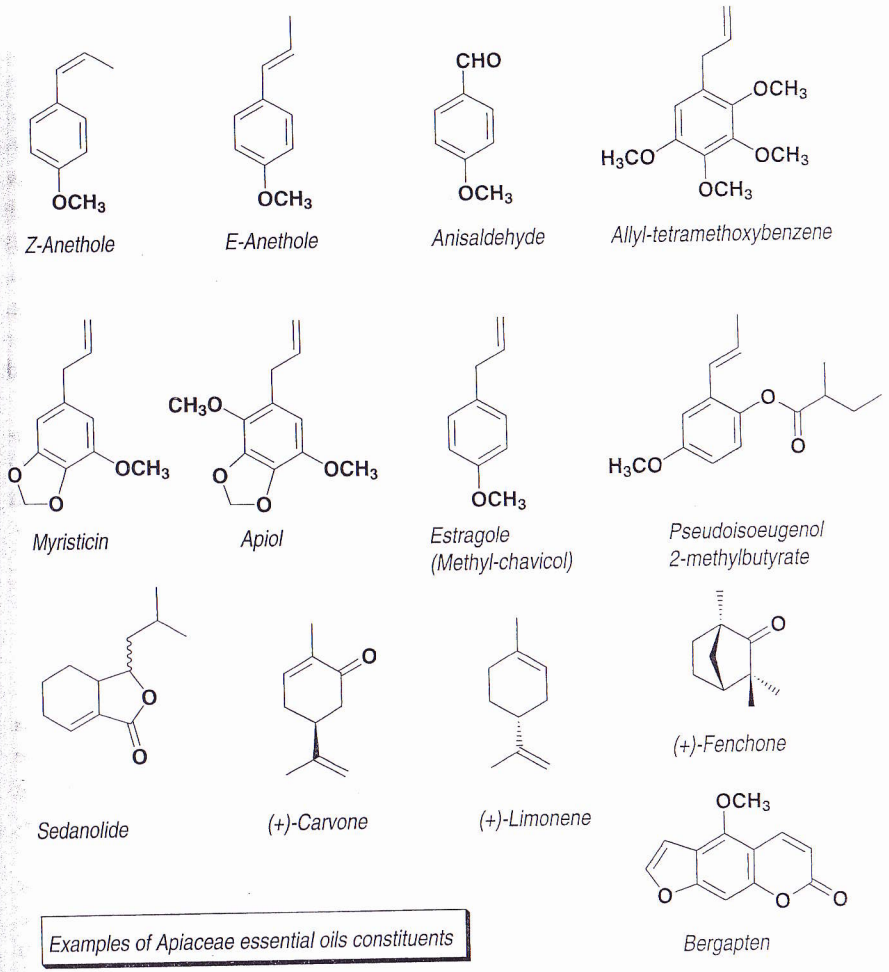
Tests. The drugs are identified by macro- and microscopic examination (pulverized fruit) and by TLC analysis of an extract in dichloromethane to characterize anethole. The assay for the two official drugs includes:

- a quantitation of the essential oil (bitter fennel, >40 mL/kg; sweet fennel, >20 mL/kg);
- a quantitation of anethole in the essential oil by GC. Its concentration is not less than 60% for bitter fennel, not less than 80% for sweet fennel;
- a quantitation of estragole and fenchone by the same method. Bitter fennel essential oil contains not less than 15% fenchone and not more than 5% estragole. Sweet fennel oil contains not more than 10% estragole and not more than 7.5% fenchone.

Uses. Fennel is an industrial source of *E*-anethole. In phytotherapy, the sweet fennel fruit may claim the same indications as the fruit of star anise or aniseed (traditionally used for the symptomatic treatment of gastrointestinal disturbances such as epigastric bloating, impaired digestion, eructations, flatulence, and as an adjunctive treatment of the painful component of functional dyspepsia). The sale of its essential oil is subject to the same restrictions. Fennel root is traditionally used to enhance urinary and digestive elimination functions, and to enhance the renal excretion of water [French Expl. Note, 1998].

Other Apiaceae Listed in Annex I of the French Explanatory Note of 1998

Four species provide drugs which may claim the same indications as aniseed or sweet fennel fruit: dill fruit, angelica root and fruit (see p. 272), caraway fruit seed, and coriander fruit. Parsley (root, leaf, fruit) and wild celery are also used in phytotherapy.



Examples of Apiaceae essential oils constituents

• DILL, *Anethum graveolens* L.

The fruit (Fr. Ph., 10th Ed.) consists of two mericarps that are easy to separate (3-4 x 2-3 x 1 mm), with three filiform yellowish dorsal ribs, and with lateral ribs enlarged into thin wings. The dried fruit contains not less than 25 mL/kg essential oil with five TLC bands, one of which corresponds to carvone.

The essential oil, in which (*S*)-(+)-carvone and (*R*)-(+)-limonene are major constituents, owes its characteristic odor to dill-ether (= dill-furan = [3*R*, 4*S*, 8*S*]-3,9-epoxy-*p*-menth-1-ene). The whole plant essential oil contains 20 to 50% (*S*)-(+)-α-phellandrene. The composition depends on the number of chromosomes. These products should not be confused with those prepared from a related species, *Anethum sowa* Roxb. or Indian dill. The essential oil of the fruit of Indian dill is characterized by dill-apiol (up to 30%).

• CARAWAY, *Carum carvi* L.

The fruit (Eur. Ph., 3rd Ed.) consists of free mericarps, elongated (3-6.5 x 1-1.5 mm), glabrous, practically falciform, and surmounted by two reflexed stylopods. Five pale yellow ribs are clearly visible on a brownish background. The dried fruit contains not less than 30 mL/kg essential oil. This Pharmacopoeia requirement is tighter than that of AFNOR for black caraway (i.e., with biennial fructification, >25 mL/kg); in the case of yellow caraway, the standard is even lower (> 15 mL/kg, NF V32-150 [1987]). TLC analysis of an extract in ethyl acetate shows, after visualization with anisaldehyde, one main spot corresponding to carvone and another spot corresponding to triglycerides.

The essential oil is mainly composed of (*S*)-(+)-carvone (50-55%) and (*R*)-(+)-limonene (35-45%). According to French standard NF T 75-347 (equivalent to ISO 8896), the concentration of carbonyl compounds ranges from 48 to 65% (determined by the free hydroxylamine method).

• CORIANDER, *Coriandrum sativum* L.

The coriander fruit (Fr. Ph., 10th Ed.) is globose (5 mm diameter). Its mericarps are not split; they are surmounted by the calix teeth and by a conical stylopod. The primary ribs are not very prominent. The dried fruit contains not less than 3 mL/kg of an essential oil which has three main TLC spots corresponding to geraniol, linalool, and geranyl acetate.

The fruit can contain up to 14 mL/kg essential oil with (+)-linalool as the major constituent (65-78%, ripe fruit), alongside camphor (4-6%), geranyl acetate (1-3.5%), γ-terpinene (2-7%) and other monoterpenoid hydrocarbons (see French standard NF T 75-205 [1991]). Reputed to be an antispasmodic, the fruit is mostly a highly prized spice (curries, bakery products, liquors), also widely used for its essential oil whose world production exceeded 700 t/year in the early 1990s. In the past few years, several European Union countries have initiated programs designed to evaluate the coriander fruit as a source of oil. Indeed, it contains 16-25% of an oil with petroselinic acid as the major constituent (80%), of potential industrial interest.

• WILD CELERY, *Apium graveolens* L.

This species, whose cultivated varieties are the edible celery, is used in France only for its root (a constituent of the former *sirop des cinq racines* or five-root syrup).

In the United Kingdom, it is the celery fruit that is used. It contains (BHP 1990) 20-30 mL/kg of an essential oil that includes mostly hydrocarbons (limonene, selinene, *p*-cymene, β-pinene), dihydrocarvone, α-terpineol, and phthalides (3-*n*-butylphthalide, sedanenolide). The celery fruit also contains flavonoids, phenolic acids, and numerous coumarins, isoprenylcoumarins, and furanocoumarins (bergapten and derivatives, free and as glycosides). The celery fruit is a diuretic in animals and a weak antispasmodic (phthalides). The phototoxicity of the furanocoumarins (upon UVA exposure) and the risk of allergic reaction are well documented for celery. The risk is real and has led the German Commission E to not recommend the therapeutic use of celery-based preparations (fruit, leaf, root), especially since their efficacy has not been substantiated.

• PARSLEY, *Petroselinum crispum* (Mill.) A.W.Hill.

The Drugs, Composition. The leaf is generally used, as well as the root and fruit.

- *Fruit.* The essential oil concentration in the fruit ranges from 20 to 60 mL/kg. The major component varies with the chemotype: apiol (60-80%), myristicin (55-75%), or 1-allyl-2,3,4,5-tetramethoxybenzene (50-60%).

- *Leaf.* The leaf contains a small quantity of essential oil (0.2-7 mL/kg). For curled parsley, the predominant compounds are myristicin, *p*-mentha-1,3,8-triene (major compound), and other hydrocarbons (limonene, β-phellandrene, myrcene, terpinolene, α-pinene, β-elemene); the apiol concentration is generally low (0-10%). The leaf also contains flavone glycosides, furanocoumarins (e.g., bergapten, oxypeucedanin, heraclenol), polyines, and phthalides.

- *Root.* The root contains phthalides, falcarinol, 3-7 mL/kg of apiol-containing essential oil, myristicin, and β-phellandrene.

Properties. According to tradition, parsley is a diuretic and an emmenagogue. Apiol—formerly official—has been used as an emmenagogue and, because it is presumably abortifacient at high doses, it has caused numerous accidents in Europe, until the late 1960s. Apiol intoxication can manifest itself by encephalopathy or, more often, by kidney damage, which can become fatal despite dialysis. Liver damage and at least one case of fatal hemolytic anemia have also been observed. Myristicin is known to have "hallucinogenic" properties (see nutmeg, p. 567) and an MAO inhibitor-type activity. It has also been shown to activate glutathione-*S*-transferase and to inhibit the induction of tumors by benzo[a]pyrene in mice.

Uses. Lacking pertinent pharmacological and/or clinical trials, parsley leaf may claim—in France—two indications: 1. orally, traditionally used for painful periods; 2. topically, traditionally used as an adjunct in the emollient and antipruriginous treatment of skin disorders, as a trophic protective agent for cracks, bruises, frostbite, and insect bites [French Expl. Note, 1998]. Parsley root and fruit are more traditionally used to enhance the renal elimination of water.

The German Commission E monograph emphasizes that the consumption of parsley fruit-based preparations can damage the renal epithelium and cause cardiac arrhythmia. It states that high doses of apiol are toxic, warns of its effect on the uterus, blood vessels, and smooth muscle, and recommends against using the essential oil. The fruit is used for gastrointestinal and urinary disorders, but since the basis for that use has not been demonstrated, it cannot be approved. The only indications allowed in Germany for parsley root are urinary elimination and the prevention of renal lithiasis (with a suitable fluid intake). Parsley root is contraindicated in pregnant women and in case of kidney inflammation.

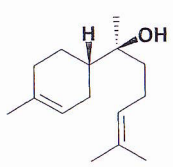
B. Asteraceae Containing Essential Oils

- **MATRICARIA,**
  Chamomilla recutita (L.) Rauschert

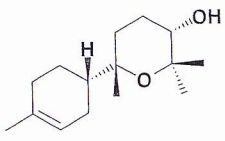
"Matricaria flower consists of the dried-flower heads of *Matricaria recutita* L. (= *C. recutita* [L.] Raushert)." (Eur. Ph., 3rd Ed.). Antispasmodic and anti-inflammatory properties are attributed to this drug, also known as German chamomile.

The Plant, the Drug. Matricaria is an annual herbaceous plant with very ramified stems bearing bipinnatisect leaves. The capitulums (10-17 mm) comprise 12-20 flowers with a large (10 mm), white, and tridentate ligula with four veins, and a large number of yellow, tubular, and synanthereous flowers, inserted on a conical and hollow receptacle without palea. The capitulum is surrounded by an involucre composed of one to three rows of lanceolate bracts with a scarious, brownish-gray edge. A common plant in the neglected fields of Europe, particularly abundant in Hungary and in Croatia, matricaria is also cultivated (selected varieties).

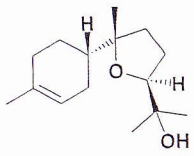
Chemical Composition. Together with an acidic mucilage, coumarins (umbelliferone, herniarin), phenolic acids, and sesquiterpenoid lactones, the drug contains an essential oil (3-15 mL/kg) and flavonoids. These are represented by flavone glycosides, including apigenin 7-glucoside and its 6"-acetylated derivative, which accumulate in the ligulate flowers up to a concentration of 8% of the dry weight. Note also the presence of luteolin glucosides, quercetin glycosides, and isorhamnetin (these are flavonols). In the dried drug, the glycosides are partially hydrolyzed, and the apigenin concentration may be high. The essential oil owes its blue color to chamazulene, which is often found at a high level (1-15%), and arises from the decomposition of a sesquiterpenoid lactone, matricin. It also contains spirononenoid dicycloethers with double and triple bonds, formed by the cyclization of polyalkynes, as well as several sesquiterpenes with a bisabolane skeleton: (–)-α-bisabolol, its A and B oxides, and the A oxide of (–)-bisabolone. These sesquiterpenes represent up to 50% of the essential oil, but their proportions vary as a function of the chemotype (at least four chemotypes are known).



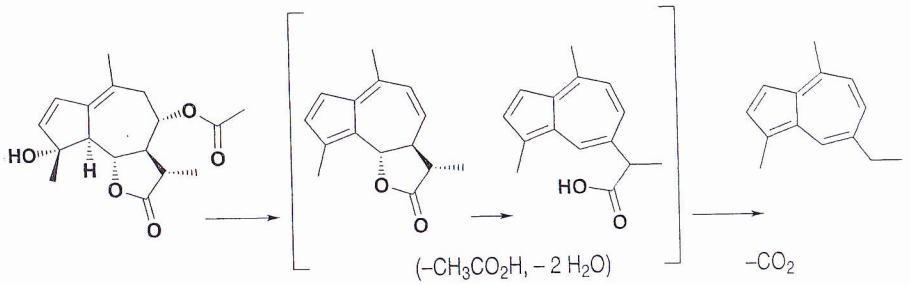
(–)-α-Bisabolol



(–)-α-Bisabolol oxide A



(–)-α-Bisabolol oxide B



Matricin: formation of chamazulen during steam distillation

Chamazulen

Pharmacological Properties. Matricaria is considered an anti-inflammatory. This activity may be due to chamazulene (an inhibitor of leukotriene synthesis) and to (–)-α-bisabolol, whose effects on different experimental models have clearly been shown (on carrageenin-induced edema and on induced arthritis). The hydroalcoholic extract of the capitulums is a spasmolytic (on guinea pig ileum). This activity may be due to the flavonoids (apigenin, more active than papaverine on the isolated guinea pig ileum), and also to one of the bicyclic ethers, and to (–)-α-bisabolol which, under similar experimental conditions, has an activity resembling that of papaverine. The essential oil of matricaria is an antibacterial and antifungal agent; it stimulates biliary secretion (cat, dog) and is thought to be hypotensive. Finally, (–)-α-bisabolol counteracts the gastric ulceration induced by various agents, including ethanol, stress, and indomethacin in the rat, *per os*. Observations in humans show that the drug is a sedative; they lend support to the use of topical preparations (anti-inflammatory, mild anesthetic, deodorant) as adjuncts to conventional treatment. Other trials indicate that matricaria extracts probably have wound healing and anti-eczema activities.

Tests. The drug is identified by its macroscopic characteristics and by the microscopic characteristics of the bracts, florets, ovules (tiny calcium oxalate crystals, epidermal cells with a wavy wall), stigmae with papillose apices, and triangular-rounded pollen grains with three pores. It is also identified by the color obtained upon addition of a phosphoric and acetic acid solution of dimethylaminobenzaldehyde to a dichloromethane extract (characterization of the proazulenes). The TLC analysis of the same extract shows the major constituents of the essential oil (examination under UV light and visualization by anisaldehyde). The drug must

contain not more than 25% (broken) flower pieces able to pass through a size 710 sieve; the essential oil level must be not less than 4 mL/kg and total ash not more than 13%.

Uses. In France [French Expl. Note, 1998], the drug and its preparations, which seem devoid of toxicity, are traditionally used by the oral route to treat the symptoms of digestive ailments (epigastric bloating, impaired digestion, eructations, flatulence), and to stimulate the appetite. Locally, the drug is an ingredient of preparations designed for the adjunctive emollient and itch-relieving treatment of skin disorders, and as a trophic protective agent for cracks, bruises, frostbite, and insect bites. These preparations may also be used as antalgics in diseases of the oral cavity, oropharynx, or both (collutoria, lozenges), and in the case of eye irritation or discomfort of various etiologies (for example eye strain, seawater or swimming pool water, or smoky atmospheres).

For the latter indication, consumers must be informed that these preparations must not be used in case of sharp pain in the eye, direct blow to the eye, wound, or when the irritation is accompanied by pus. If the symptoms get worse or persist beyond 48 hours, a physician must be consulted. Matricaria-based phytomedicines that are total drug powders must pass basic safety tests.

The German Commission E monograph contains virtually identical indications. Orally: gastrointestinal spasms and inflammation. Topically: inflammation of the skin and mucous membranes, bacterial infections (skin, mouth; gargles); anal and genital disorders (baths, washes); respiratory irritations (inhalations).

In cosmetology, matricaria is used in shampoos (to lighten hair color), and in suntan lotions. The essential oil is an ingredient of perfumes and soaps.

Comments

1 - The presence of lactones in matricaria-based preparations (medicinal, cosmetic, or other) may cause allergic reactions (contact dermatitis) in sensitive persons. These reactions to chamomile are uncommon, and the cases in which matricaria is formally incriminated seem exceptional (which is not true for species such as *A. cotula* L.). In exceedingly rare cases, major anaphylactic reactions after the ingestion of an infusion or exposure to pulverized plant dust have been described, including generalized urticaria, edema of the face and eyelids, edema of the pharynx, and obstruction of the respiratory tract.

2 - Synthetic azulenes—azulene (= bicyclodecapentaene) and guaiazulene (isopropyl-7-dimethyl-1,4-azulene)—are used in the composition of drug combinations designed to be anti-inflammatory: ointments, eye drops.

● **Roman Chamomile**, see flavonoid-containing drugs, p. 335.

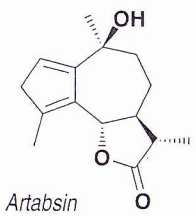
• WORMWOOD, *Artemisia absinthium* L.
MUGWORT, *Artemisia vulgaris* L.

These two perennial Asteraceae are both official (Fr. Ph., 10h Ed.). In both cases, the drug consists of the leaves and the flowering tops. In both cases, the inflorescences are racemes of small globulous capitulums of yellow flowers. Wormwood leaves are highly divided and have a characteristic silvery color; those of mugwort are dark green on the upper side and silvery on the underside.

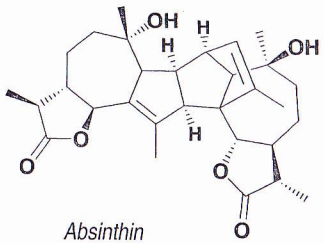
Wormwood, a very aromatic herbaceous plant, is common in the Mediterranean area. Mugwort is a very common species in all of Europe, where it colonizes piles of rubble and road shoulders.

Chemical Composition

• **Wormwood**. The drug concentration in essential oil reaches a maximum before blooming, and ranges from 2 to 6 mL/kg; the official drug must contain not less than 3 mL/kg and not more than 13 mL/kg. Older texts describe this essential oil as containing mostly thujones. In some cases this is true, but the systematic analysis of specimens from different sources has shown that there are many chemotypes: with *Z*-epoxy-α-ocimene (26-47%), with β-thujone (Italy), with sabinyl acetate or chrysanthemyl acetate (France) and intermediates. The plant also contains polyalkynes, flavonoids, and sesquiterpenoid lactones: absinthin (a dimer with a guaianolide skeleton), artabsin, matricin, and closely related derivatives.



Artabsin



Absinthin

• **Mugwort**. The concentration of the essential oil is low (1-2 mL/kg, >1 mL/kg for the official drug), and its composition is highly variable: camphor, borneol, vulgarin, and hydrocarbons are constants, but the thujones are only found in traces, or else absent. In fact, the French Pharmacopoeia requires verifying the absence of thujone in a hexane extract: in the prescribed conditions, it must not be detectable, and this distinguishes this species from another western European one with which it may be confused, namely *A. verlotorum* Lamotte. It is certain that the drug contains, among others, flavonoids and polyalkynes, on the other hand it appears that the sesquiterpenoid lactones (vulgarin, psilostachin) are not found in all of the batches analyzed.

Properties. The neurotoxicity of wormwood liquor, now known *p. 525 to be linked to thujone, had been recognized as early as the beginning of the [illegible]

century. It explains the characteristic presentation of absinthism, associating the symptoms of ordinary alcoholism—"superior" wormwood liquors were between 65 and 75 proof!— and epileptiform seizures. Wormwood and similar liquors were banned in France in 1915, clearing the way for anise-flavored substitutes! In 1959, the French legislation was completed in order to regulate the manufacture and sales of wormwood oils and similar products (essential oils of hyssop, aniseed, star anise, fennel, and one substance: anethole). A 1986 decree added these products to the list referred to in Article L 512 of the French Public Health Code or *Code de la Santé Publique*. The current European Union regulatory texts set limit concentrations for thujones in foods and beverages.

Uses. The drugs, especially wormwood, enjoy a reputation as old as it is flattering (wormwood is thought to be an anthelmintic, antibacterial, antipyretic, emmenagogue, and even schizonticide), but there are no reliable data on their pharmacology. In the absence of experimental or clinical data, the two drugs are traditionally used to stimulate the appetite. Mugwort is also traditionally used to relieve "painful periods" [French Expl. Note, 1998]. Phytomedicines based on wormwood or mugwort that are total drug powders, hydroalcoholic extracts of alcoholic titer greater than 30%, or tinctures must pass basic safety tests, and in the case of mugwort, a maximum concentration in active principle must appear on the label. In Germany, wormwood is used to stimulate the appetite and for dyspepsia. As far as mugwort is concerned, Commission E finds that the indications that are claimed have not been substantiated, therefore its therapeutic use cannot be approved.

• TARRAGON,
Artemisia dracunculus L.

Used as a seasoning for food, this herbaceous plant is mentioned in the French Explanatory Note of 1998. It contains an essential oil rich in estragole (68 to 80%), *cis* and *trans* ocimene (6-12%), and limonene (2-6%) (French standard NF T 75-351 [1987]).

The drug (non-flowering aerial parts) is traditionally used to treat the symptoms of various digestive ailments and as an adjunctive therapy for the painful component of functional dyspepsia [French Expl. Note, 1998]. The traditional indications are

* Is it that simple? After recalling that it takes 2.5 kg of wormwood (plant) to obtain 100 liters of wormwood liquor (at 0.4% essential oil), Max [1990] estimates the quantity of thujone in one glass of beverage at 2-4 mg. He emphasizes that this amount is insufficient to observe pharmacological effects; in addition, he feels that the claims that thujone is neurotoxic are not well substantiated. According to him, the green color of the liquor used to be adjusted with copper sulfate and its opalescence (after dilution) used to be "improved" with antimony chloride. Methanol, which is toxic, is said to have been found in the liquor (which contained other plants...). Max. B. (1990). This and That: Cheap Drinks and

similar to those of another Asteraceae, costmary or alecost (*Balsamita major* Desf. = *Chrysanthemum balsamita* [L.] Baill, non L. = *Tanacetum balsamita* L.), which is also used topically.

C. Lamiaceae Containing Essential Oils

Most of the species that we shall describe are known mainly for their various industrial applications (perfumes, liquors, confectionery products, cosmetics, detergents). Many are also better known as spices rather than medicinal plants (sweet basil, oregano, thyme). The chief indications that are accepted for these drugs are for the "treatment" of minor digestive disturbances, and for local use (dermatology, hygiene). In a small number of cases, the indications include minor sleeplessness (balm, lavender).

• Sweet Basil, *Ocimum basilicum* L.

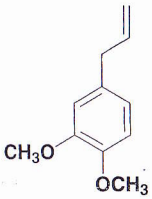
Although it is essentially known as a spice (Italian *pesto*, French *pistou*) and as a source of essential oil, sweet basil, which grows wild in Asia, and is now cultivated in the Mediterranean area and in the islands of the Indian Ocean, has reappeared in the French Pharmacopoeia as of January 1989 (dried leaf to contain not less than 2.5 mL/kg essential oil). Several chemotypes are known, the main one originating from the Réunion island, Comoro islands, and Madagascar. Its essential oil contains 65 to 85% estragol (= methylchavicol), alongside small amounts of cineole, fenchol, linalool, and methyleugenol. Another chemotype, characterisic of southern Europe and Egypt, produces an essential oil with high levels of linalool. Chemotypes with (*E*)-methylcinnamate and linalool are also known (Fiji and other tropical areas), as well as chemotypes with methyleugenol, and intermediates.

In mice, estragole administered *per os* is partially metabolized to 1'-hydroxy-estragole, which is carcinogenic (p. 509). Having considered the amounts of sweet basil-based (or essential oil-based) products normally consumed, and knowing that only a small fraction of the ingested estragole gets hydroxylated in humans, international authorities have not proposed any limits. In contrast, the use of sweet basil in aromatherapy (oral, massage) must be discouraged, at least in the case of the estragole chemotype.

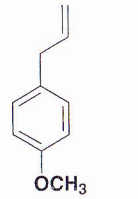
Sweet basil leaf is traditionally used orally: 1. for the symptomatic treatment of gastrointestinal disturbances (epigastric bloating, impaired digestion, eructations, flatulence); 2. as an adjunctive therapy for the painful component of functional dyspepsia [French Expl. Note, 1998]. The methylchavicol-type essential oil (French standard NF 75-357 [1991]: methylchavicol >75%) is used to manufacture some of the finest perfumes, whereas linalool-containing products (NF T 75-244 [1992]: linalool, 45-62%; eugenol, 2-15%; cineole, 2-9%) find outlets in food products and

• Calamint, *Calamintha sylvatica* Bromf.

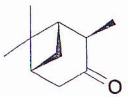
Calamint is not well known and not used much, yet it was listed in the French Explanatory Note of 1998. The official species, (*C. officinalis* Moench., Fr. Ph., 10h Ed.) was formerly classified by some within the genus *Satureja,* and would correspond, according to the Flora Europea, to ssp. *ascendens* (Jordan) P.W. Ball (= *C. officinalis* auct., non Moench.). According to the French forest flora, official calamint is *C. sylvatica* Bromf. (= *C. officinalis* Moench = *Satureja calamintha* (L.) Scheele). It is a perennial plant with glomerules of near-crimson flowers measuring less than 15-20 mm. The dried flowering tops of calamint produce a small amount of essential oil (not less than 6 mL/kg in the case of the official drug) containing neomenthol, pulegone, menthone, isomenthone, and other monoterpenes. Officially [French Expl. Note, 1998], the flowering tops of calamint are traditionally used: 1. for the symptomatic treatment of gastrointestinal disturbances (epigastric bloating, impaired digestion, eructations, flatulence); 2. as an adjunctive therapy for the painful component of functional dyspepsia. Around the Mediterranean, a related species is used in folk medicine, namely *C. nepeta* (L.) Savi which occurs as many different chemotypes, including chemotypes with pulegone, piperitenone, carvone, and cineole.



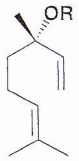
Methyleugenol



Estragol (Methylchavicol)



Pinocamphone (= 3-pinanone)



(+)-Linalool (R = H) *and (+)-Linalyl acetate* (R = $COCH_3$)

• Hyssop, *Hyssopus officinalis* L.

This polymorphous species (some distinguish four subspecies) is a small Mediterranean plant, common on piles of rocks and old walls.

The drug (Fr. Ph., 10th Ed.) consists of the leaves and flowering tops. The leaves are borne by a quadrangular stem, are lanceolate, almost sessile, and have a wafer-like surface. Upon desiccation, the edges curl toward the underside. The flowers have a blue or purplish corolla, and are grouped in axillary glomerules which form a compact and elongated spike; they have a larger lower lip, with a prominent middle lobe shaped like an upside-down heart. The drug fragments can be identified by their microscopic characteristics: unicellular thorn-shaped covering trichomes, multicellular hook-shaped or curved covering trichomes, and typical Lamiaceae

The drug contains phenolics (rosmarinic acid, flavonoids), di- and triterpenes (marrubiin, oleanolic acid), and essential oil (>3 and <10 mL/kg in the official drug). The main oil constituents are the ketones isopinocamphone (34.5-50%) and pinocamphone (= 3-pinanone, 5.5-17.5%), alongside mono- and sesquiterpenoid hydrocarbons (β-pinene, 13.5-23%), limonene (1-4%), and sabinene (2-3%) (French standard NF T 75-355 [1986]). In the absence of experimental data, the following properties are attributed to this drug: expectorant, antiseptic, and stimulant (essential oil).

The essential oil of hyssop is neurotoxic: pinocamphone and isopinocamphone are considered responsible for its epileptogenic activity; the neurotoxicity could be linked to the inhibitory action of the ketones on cell respiration (shown *in vitro*). Severe intoxications have been described, including some after ingestion of small doses: 2-3 drops (6-year old girl), 10 drops x 2 days (26-year old woman). Thus it is appalling to see that French books for the lay public are still being published that recommend using 3-4 drops, 3 times a day. However, in France, only pharmacists are authorized to dispense hyssop essential oil and its preparations (decree or *décret* 86-778 of June 23, 1986).

Hyssop-based phytopharmaceuticals are traditionally administered by the oral route for acute benign bronchial disease, and are administered locally to relieve nasal congestion in the common cold [French Expl. Note, 1998].

• LAVENDER, *Lavandula* spp.

Two species in this genus are used in perfumery and cosmetology: the true lavender (*L. angustifolia* Miller = *L. officinalis* Chaix = *L. vera* DC.), and the broad-leaved lavender, also known as *grande lavande* or *lavande aspic* (*L. latifolia* (L.*f.*) Medikus = *L. spica* auct., non L.). The *lavandins* are hybrids of the two species listed above (*L.* x *intermedia* Emeric ex Loisel = *L. hybrida* Reverchon ex Briq.), and are also used to produce the essential oil. *Lavandula stoechas* L. (fenchone and camphor-containing essential oil) is no longer used.

• **True lavender.** The French Pharmacopoeia (10th Ed.) devotes a monograph to the true lavender: the drug consists of the dried flowers, and must contain not less than 8 mL/kg essential oil.

The Plant, the Drug. True lavender is a shrub growing in the low mountains (800-1,800 m or 2,600-6,000 ft) of the Mediterranean basin. It has silvery green leaves that are very narrow and are curled on the edges, and flowers grouped in biparous cymes on short peduncles. The calyx is bluish-gray and the corolla, bilabiate with the upper lip bifid and the lower lip trilobate, is blue. The pulverized drug is characterized by numerous bifurcate covering trichomes divided once or several times.

Pharmacological Properties and Uses. The dried flower has not undergone any pharmacological experiments in animals. *In vitro*, lavender oil has moderate antibacterial activity; *per os* and in the mouse, it has a depressant activity.

The drug may be used in the composition of phytomedicines [French Expl. Note, 1998] with the following indications: 1. topically, traditionally used to treat minor wounds after thorough cleansing; for sunburns, superficial burns of limited area, and diaper rash; to relieve nasal congestion in the common cold; as a mouthwash for oral hygiene; 2. orally, it is traditionally used to treat the symptoms of neurotonic disorders in adults and children, especially in case of minor sleeplessness. If the phytomedicines are total drug powders, hydroalcoholic extracts of alcoholic titer >30%, or tinctures, they must, like the sages (p. 540), pass (basic) safety tests. The same type of indications—for the oral route—are allowed in Germany where lavender is also used in baths to improve functional circulatory problems.

Essential Oil. The composition of the essential oil varies depending on many factors, particularly the mode of cultivation (spontaneous or Mailette and Matterone cloned lavenders) and the environmental conditions. To fulfill the requirements of the French Pharmacopoeia, lavender oil must contain 25-38% linalool, 25-45% linalyl acetate, 0.1-0.5% limonene, 0.3-1.5% cineole, 0.2-0.5% camphor, and 0.3-1% α-terpineol (determined by GC). The French standard NF T 75-301 [1992] (for lavender oil from France) echoes these specifications in great part, and completes them, especially with regard to the β-ocimenes (*cis*, 4-10%; *trans*, 1.5-6%), terpin-1-en-4-ol (2-6%), and octan-3-one (not more than 2%); minimal levels of specific compounds (lavandulol and its acetate) are also required (0.3 and 2%, respectively). In 1981, the French government specified the requirements that a lavender oil must meet before it can claim the "*appellation d'origine*" "*de Haute-Provence*" (decree or *décret* of December 14, 1992).

• ***Lavande aspic.*** *Lavande aspic* grows wild in the same areas as true lavender, but at lower altitudes, and is cultivated in Spain. It produces an essential oil that is particularly rich in cineole (30-40%) and in camphor (15%), and therefore less prized. Like the previous oil, *lavande aspic* oil is listed in the French Pharmacopoeia (it is also subject to a French standard, AFNOR NF T 75-201 [1992]). The ranges of tolerance specified for its chromatographic profile are as follows: limonene (0.5-3%), cineole (20-35%), camphor (8-20%), linalool (25-50%), linalyl acetate (<3%), and α-terpineol (0.5-3%) (Fr. Ph.). The AFNOR standard differs from that of the Pharmacopoeia only by the upper limit for linalyl acetate (<1%).

• ***Lavandin.*** The composition of lavandin oils is intermediate beween those of *lavande aspic* and true lavender. The *lavandin "Abrial", "Grosso",* and *"Super"* oils are subject to standards which, as above, include the description of their chromatographic profile (AFNOR NF T 75-303 and 304). *Lavandin "Grosso"* oil is official, and as far as the principal constituents are concerned, the French Pharmacopoeia specifications are identical to those of the AFNOR standard:

acetate (28-38%), and α-terpineol (0.5-1%). The standard takes into account other minor constituents: lavandulol (0.2-0.8%), lavandulyl acetate (1.5-3%), borneol (1.5-3%), terpin-1-en-4-ol (1.5-5%), and *cis*- and *trans*-β-ocimenes (0.5-1.5 and <1%).

France contributes 90% of the world production of lavandin oil (1,100 t in 1995, including 850 t of "Grosso", decreasing to 980 t in 1996). For comparison, the production of true lavender oil was about 39 t in 1995 (13 t of "cloned", 26 t of "*fine*", and constantly decreasing). For low-end preparations, lavandin oils cannot compete with synthetic products (linalool, linalyl acetate).

• Sweet Marjoram, *Origanum majorana* L.

The term marjoram is ambiguous: normally it designates sweet marjoram (*O. majorana* L. = *Majorana hortensis* Mœnch.), but "wild marjoram" designates *O. vulgare* L. and "wild Spanish marjoram" is wild Spanish thyme (*T. mastichiana* L.). Originally from the orient, marjoram is widespread in all of the Mediterranean basin (production: Egypt). It contains 7-30 mL/kg essential oil with terpin-1-en-4-ol, α-terpineol, sabinene hydrates, and linalool (see p. 496). In the absence of relevant pharmacological or clinical studies, the leaves and flowering tops are traditionally used orally [French Expl. Note, 1998], 1. for the symptomatic treatment of gastrointestinal disturbances (epigastric bloating, impaired digestion, eructations, flatulence); 2. to treat acute benign bronchial disease. Marjoram is used locally to relieve the symptoms of the common cold, such as nasal congestion, and in mouthwashes for oral hygiene.

• Balm, *Melissa officinalis* L.

The dried leaves of balm (Fr. Ph., 10th Ed.) must contain not less than 5% total hydroxycinnamic derivatives expressed as rosmarinic acid.

The Plant, the Drug. Balm is a perennial subshrub which grows in tufts. The stems are erect, and bear opposite leaves with a wafer-like surface, which are rough to the touch, and release a lemony odor when crushed. The flowers are irregular, white or pinkish, and are whorled in axillary groups that are spaced far apart. Probably originally indigenous to Turkey, balm grows wild in the woods, along the edges of country roads, in hedges, in other cool places, and is also cultivated * (p.531). The drug consists of the oval and cordate leaves. The blades bulge between the branches of the network formed by the veins, are roughly dentate or crenate, and are lighter on the underside.

Chemical Composition. Despite its low concentration (0.5 mL/kg), the essential oil has received most of the attention; nevertheless, other constituents have been isolated from balm, including triterpenes and phenolic acids derived from caffeic acid, dimers such as rosmarinic acid, and trimers such as malitric acids A and B; flavonoids (quercitrin, rhamnocitrin, and the 7-glucosides of apigenin and luteolin); and glycosides of monoterpenes and of aromatic alcohols. Balm oil is characterized by monoterpenoid aldehydes: citrals (geranial + neral) in very variable quantities, but in a constant ratio (4/3), (*R*)-(+)-citronellal (found in substantial amounts in some batches of German origin), alongside methylheptenone (a degradation product of citral), geranyl acetate, β-caryophyllene, β-caryophyllene oxide, germacrene D, and several dozen other compounds, mainly terpenoids.

Tests. The identification of he drug, which is often in fragments, entails a microscopic examination (conical, short, and straight covering trichomes with a thick wall); pluricellular uniseriate covering trichomes with thick and bumpy cuticle; octocellular glandular trichomes). Phenolic acids can be extracted with methanol and characterized by TLC with conventional visualization with aminoethanol diphenylborate and PEG400. The colorimetric quantitation of the hydroxycinnamic acids is carried out on a hydroalcoholic (50:50) extract of the drug.

Pharmacological Properties. Balm oil is an antibacterial and antifungal agent, and is also responsible for the spasmolytic properties that are recognized for the drug. The hydroalcoholic extract is a CNS sedative (in the mouse), but according to recent studies, the essential oil does not appear to play a role in this activity; the same extract potentiates the sleep-inducing effect of pentobarbital. Several activities have been shown for the aqueous extract: antithyroid activity linked to the oxidation products of the phenolic acids, antigonadotrophic activity, and most of all, antiviral activity. This latter activity, shown on various viruses (herpes, vaccinia), could be due to the phenolic acids, or their derivatives, or both, and to their interaction with viral proteins. Other studies have shown a real activity of acyclic monoterpenes on the hydroxymethylglutaryl coenzyme A in rats.

In humans, interesting results have been observed in the local treatment of herpes: using a balm extract-based ointment shortened the duration of the disorder and delayed recurrent episodes. A fairly old study, which was not confirmed by subsequent work, showed the benefits of balm in neurovegetative dystonia.

Uses. Currently, balm-based phytomedicines for oral use may claim three indications [French Expl. Note, 1998]: traditionally used: 1. for the symptomatic treatment of gastrointestinal disturbances (epigastric bloating, impaired digestion, eructations, flatulence); 2. as an adjunctive therapy for the painful component of

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* Generally, only one of the two subspecies described by the Flora Europaea is cultivated: ssp. *officinalis* ($2n = 32$). The *altissima* (Sibthr. and Smith) Arcang. ($2n = 64$) subspecies produces an essential oil of clearly different composition (including β-cubebene, terpinolene, 3-carene, γ-terpinene, β-caryophyllene, and T-muurolol) with a green and woody note. Recently, the composition of another subspecies [ssp. *inodora* (Bornm.) Bornm.] has been published, and includes citral (geranial + neral), β-cubebene, β-caryophyllene, and α-cadinol.

functional dyspepsia; 3. for the symptomatic treatment of neurotonic disorders in the adult and in the child, for example in case of minor sleeplessness. In Germany, balm has similar uses.

Note that balm oil-based commercial products are likely to be substituted by or adulterated with cheaper products: citronnella or *lemon grass* oil (see note *, p. 505).

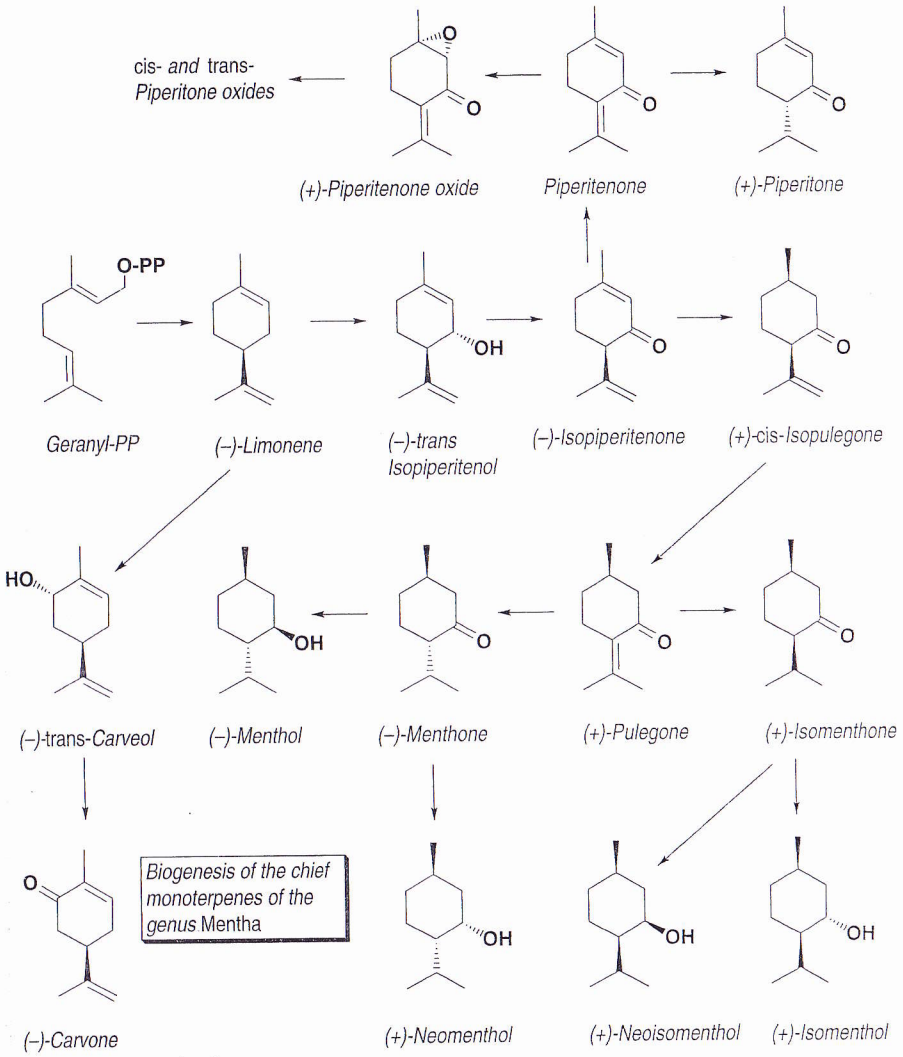
• PEPPERMINT, *Mentha* x *piperita* L.

The taxonomy of the *Mentha* genus is greatly complicated by the common occurrence of hybrid species, polyploid species, and the abundance of morphological variations: there are numerous forms, and from one to the next, the plants display gradual changes. The species listed in the 3rd edition of the European Pharmacopoeia for its dried leaves is a hybrid of *M. aquatica* L. and *M. spicata* L.

The Plant, the Drug. An extremely hardy perennial plant that propagates by stolons, peppermint is characterized by quadrangular stems most often purplish, by entire opposite-decussate leaves which are ovate-acuminate and dentate, and by inflorescences composed of slightly bilabiate, purple flowers grouped into very tight spikes. Different varieties are cultivated: Mitcham peppermint (*rubescens* form of the *officinalis* variety), white mint (*palescens* form of the same variety), and, more rarely, the *rubescens* form of the *sylvestris* variety. Multiplication is achieved exclusively by the vegetative mode. The development of the plant requires a temperate climate, a sufficient water supply, and enough sunlight. The harvest is mechanized, and takes place at the beginning of the blooming period.

Drug identification is easy: characteristic odor, leaf blade (3-9 x 1-3 cm) acuminate, serrate, asymmetric at the base, lateral veins at a 45° angle, and principal vein prominent on the underside. Under the microscope, the drug has glandular trichomes with unicellular stalks and with heads comprising either one cell (15-25 µm diameter), or else eight radiating cells; in the latter case, the head (55-70-µm diameter) is swollen and oval, and the essential oil accumulates under its cuticle.

Chemical Composition. The peppermint leaf contains many compounds: triterpenes, carotenoids, phenolic acids (up to 7%), and flavonoids; the latter are represented by lipophilic polysubstituted flavones (aglycones) and by particularly abundant flavone and flavonol glycosides: over 17% in some clones, always with eriocitrin (eriodictyol 7-*O*-rutinoside) as the major compound. The essential oil represents from 10 to 30 mL/kg of the weight of the dried drug. Its composition varies as a function of multiple factors, intrinsic and extrinsic, including the cultivation conditions, climatic variations, and harvest time. The chief constituent is always (–)-menthol (30-40%, sometimes more than 50%). It occurs alongside (–)-menthone (15-25% in the case of Mitcham peppermint, half as much in the case of the white peppermint), (–)-menthyl acetate, (–)-menthofuran (sometimes absent, it can also represent up to 10% of the essential oil), (+)-isomenthone, (+)-pulegone,



Biogenesis of the chief monoterpenes of the genus Mentha

(+)-neomenthol, (–)-piperitone, hydrocarbons, and other compounds. (+)-Pulegone is found in the young leaves, but disappears rapidly later on. The decrease in the level of (–)-menthone and the increase in (–)-menthol observed during the vegetative cycle correspond to a reduction of the ketone to (–)-menthol and (+)-neomenthol. The latter is converted to a water-soluble glycoside, which is transported into the roots (see the comment on this topic on p. 495).

Tests. Peppermint identification includes, in addition to the microscopic examination of the leaf powder, a TLC analysis of a dichloromethane extract, to show the presence of the chief constituents (e.g., menthol, menthyl acetate, menthone), and to verify the absence of bands corresponding to pulegone.

- verifying the absence of foreign elements: foreign plant parts (stems of diameter <1 mm: <5%), foreign matter (<2%), leaves with brown spots from *Puccinia menthae* (<8%);
- total ash (<15%) and ash insoluble in HCl (<1.5%);
- water (<11%);
- quantitation of the essential oil: >9 mL/kg (cut drug) or >12 mL/kg (entire drug).

Pharmacological Properties and Uses. The pharmacology of the drug has not been explored much. Regarding the essential oil, note that it suppresses the morphine-induced contraction of the sphincter of Oddi (guinea pig, IV), and that it is, exactly like menthol, a spasmolytic *in vitro*: it acts by non-competitive inhibition of the contractions of the smooth intestinal muscle induced by various agonists or by depolarization (guinea pig ileum, rabbit jejunum, and other models), and does so by reducing the calcium influx (a more complex activity is suspected, but has yet to be elucidated). Several authors have pointed out the benefits of peppermint oil in the treatment of colon spasms in humans (particularly the forms with enteric coating). It is used for this purpose in the United Kingdom, even though the methodology of the initial clinical trials was criticized, and even though a later trial failed to show that peppermint oil was superior to a placebo. The use of peppermint oil as an antispasmodic during colonoscopy or rectal barium sulfate administration for X-ray visualization decreases the frequency of spasms and decreases the need to administer spasmolytics by the IV route. Menthol glucuronate might be a pro-drug of interest to release menthol in the colon. A combination of menthol and ursodeoxycholic acid is thought to have a beneficial effect on biliary lithiasis.

Menthol has been presented as a nasal decongestant for over a century: empirically, it has been abundantly demonstrated that this reflects a purely subjective sensation, linked to the cool sensation thought to be due to the stimulation of the nasal cavity thermoreceptors. Menthol vapors inhibit respiration (like cold air) and may cause very transient apnea in very young children. The risk is minimal, yet the direct application of peppermint oil or menthol on the nasal mucosa of very young children is discouraged. Application of menthol on the skin can induce a cold sensation, but menthol is neither antipruriginous nor analgesic. In animal experiments (guinea pig), menthol counteracts the bronchoconstriction induced by capsaicin. A mixture of menthol and eucalypus oil (75:25), if inhaled, soothes the cough induced by a citric acid aerosol.

It has been shown, in the case of herbal teas prepared from peppermint leaves, that a large proportion of the volatile substances is lost during the process of infusion. The activity of this "pleasant and relaxing beverage" in humans, which remains to be demonstrated, may be due in part to the phenolic substances (flavonoids, see thyme).

Uses. The drug is devoid of toxicity, and is traditionally used—in France—orally for the symptomatic treatment of functional dyspepsia, including epigastric bloating, impaired digestion, eructations, and flatulence. It is also traditionally used orally:

enhance urinary and digestive elimination functions; 3. in functional dyspepsia when it is attributed to a hepatic origin. Topically, it is traditionally used to relieve nasal congestion in the common cold; as an antalgic for diseases of the mouth, pharynx, or both (collutoria, lozenges); in mouthwashes for oral hygiene; as an adjunctive, emollient, and itch-relieving treatment for skin disorders, as a trophic protective agent for cracks, bruises, frostbite, and insect bites [French Expl. Note, 1998].

The German Commission E monograph specifies that peppermint is used for gastrointestinal, gallbladder, and biliary tract spasms. For lithiasis, it should be used only with medical advice. There are no known side effects or drug interactions.

Mint Oils

• Peppermint Oil

The market is dominated by the United States, which produced, in 1992, about 3,200 t of essential oil from 43,000 hectares (= 106,210 acres) planted with selected clones (Idaho, Oregon, Indiana, Wisconsin, Washington). The flowering tops, cut mechanically and partially dried, are collected into truck-size containers which fit directly between the steam generator and the condenser tank of the distillery facilities.

To be official (Eur. Ph., 3rd Ed.), peppermint oil must pass the Pharmacopoeial tests:

- TLC (visualization by anisaldehyde) allows the identification of the essential oil by showing the principal constituents; minor ketones, if present, are visualized;
- the assay includes the determination of the acid value (<1.4), refractive index (1.457 to 1.467), and specific optical rotation (– 10° to – 30°). The product must pass tests for fixed (fatty) oils and resinified essential oils;
- finally, the chromatographic profile must be determined: the essential oil must contain 30-55% menthol, 14-32% menthone, 1-9% menthofuran, 2.8-10% menthyl acetate, not more than 4% pulegone, and not more than 1% carvone. It must also contain 1-5% limonene, 3.5-14% cineole, and 1.5-10% isomenthone; the cineole (%)/limonene (%) ratio must be more than 2.

The French standardization organization AFNOR has proposed a standard for peppermint oil. This standard (NF T 75-210 [1982], equivalent to ISO 656) does not describe a chromatographic profile, but it specifies tolerance ranges for the ester value, the ester value following acetylation, and the carbonyl value, all as a function of the geographical origin (France, Italy, United Kingdom, or United States).

Peppermint oil is probably not completely devoid of toxicity: administration of high doses (40-100 mg/kg) to rats induces histopathological changes in the brain. Menthone, like pulegone, produces similar effects. Menthol itself induces no serious adverse effects (rat, 800 mg/day x 28 days).

Peppermint oil is used as a flavor in medications, as well as in parapharmaceutical,

sodas, concentrated syrups, confectionery (candy, chewing gums, chocolates), and other products. The tobacco and perfume industries are also consumers.

• Spearmint Oil

Spearmint or ordinary garden mint is a species close to the latter one, cultivated in the same states of the United States (Oregon, Idaho, Washington). The steam distillation is generally preceded by partial drying for 24 hours.

According to the French Pharmacopoeia, spearmint oil is "obtained by steam distillation of the aerial parts recently harvested from *M. spicata* L. The essential oil obtained from *M. cardiaca* Gérard also satisfies the requirements of the monograph *." French standard NF T 75-245 [1986] specifies that only those varieties or descendants from hybrids of *M. spicata* that yield an oil rich in carvone qualify as sources of spearmint oil. The latest edition of the standard lists the chief constituents of the essential oil, but does not define tolerance ranges. The official essential oil must contain from 55 to 67% carvone and from 2 to 25% limonene. The concentration of the other constituents (menthone, isomenthone, menthol, menthofuran, menthyl acetate, and cineole) must be less then 2%; the level of pulegone must be not more than 0.5%. The level of nitrogen-containing products is twice that of the essential oil of *M.* x *piperita* (10 ppm, including 1/3 2-acetyl-4-isopropenylpyridine).

• Cornmint oil

Cornmint oil is the partially dementholated essential oil of *Mentha arvensis*, and is official. It is obtained from the "flowering tops of *M. arvensis* L. var. *piperascens* Holmes and var. *glabrata* Holmes. It is partially dementholated by freezing." *Mentha arvensis* can be cultivated in subtropical climates. The world production [1994] ** of *M. arvensis* oil is from China (3000 t, a 50% increase in 4 years), India (2000 t), North Korea (300 t); the traditional production from Paraguay has become marginal (60 t versus 800 t in 1990). A very slow cooling of the essential oil (2°C/day from 35 to 5°C) induces the crystallization of part of the menthol that it contains. The average composition of the essential oil after menthol removal is outlined by the chromatographic profile chosen by the French Pharmacopoeia: menthol (30-45%), menthone (17-35%), isomenthone (5-13%), menthyl acetate (2-

* The same edition of the French Pharmacopoeia defines "*menthe verte*" as "consisting of the dried leaf of *M. viridis* L. or *M. spicata* L. It contains not less than 10 mL/kg essential oil". It is identified by the microscopic characteristics of the powder (trichome morphology) and by TLC analysis of the essential oil (carvone). [Fr. Ph., 10th Ed., add. number 38, decree or *arrêté* of August 25, 1997].

** Figures cited by Joulain, D. (1994). Successes and Failures of Essential Oils. A Historical Overview, *in* "Plantes aromatiques et médicinales. 4es Rencontres techniques et économiques". (Verlet, N., Ed.), p. 4-11, CFFPPA-CERDEPAM, Nyons.

7%), and limonene (1.5-7%); the level of the other constituents must remain low (menthofuran <1%, cineole and pulegone <1.5%, and carvone <2%); the cineole (%)/limonene (%) ratio must be less than 1.

The French standard NF T 75-306 [1985] defines this essential oil as obtained from the "*piperascens* Malinvaud variety, cultivated in Brazil and in China". The standard refers to two chromatographic profiles (China and Brazil); in fact the limits described by the French Pharmacopoeia correspond to the extremes observed for each of the sources; for example, in the case of limonene, the standard requires 1.5 to 4% in the variety from China, 5 to 7% in the variety from Brazil; menthol: Brazil, 30-40%, China 35-45%, and so on. The proposed upper limit is 32% for menthone (Brazil) and 1% for pulegone (for either origin). The standard mentions explicitly that carvone should not be found in *M. arvensis*; finally, it includes in the chromatographic profile neomenthol (3-4% [Brazil] and 4-7% [China]) and piperitone (2.5-3.5% [Brazil] and 0.5-1% [China]).

Comment: the use of mint oils to flavor foods and beverages can introduce pulegone. French regulations set the limit pulegone concentrations at 25 mg/kg in foods, 100 mg/kg in beverages, 250 mg/kg in mint-flavored beverages, and 350 mg/kg in mint candy.

• Menthol

Only one of the stereoisomers of menthol is used, (–)-(1*R*,3*R*,4*S*) menthol, easier to refer to as (–)-menthol, and sometimes called levorotatory menthol. The other optically active isomers do not have the same organoleptic characteristics: only the racemate induces—but to a lesser extent—the cool taste characteristic of natural (–)-menthol. Although it is the chief constituent of peppermint oil, menthol is generally not extracted from it. Several approaches are in use to obtain this monoterpenoid alcohol.

- **Mentholated essential oils**. Menthol is crystallized by freezing the essential oil of *M. arvensis*, which remains a competitive starting material, especially since there is a market (toothpastes, chewing gums) for the dementholated oil. An additional quantity of menthol can be recovered by saponifying menthyl acetate, and by hydrogenating (–)-menthone (formation of (–)-menthol and (+)-neomenthol).

- **Semisyntheses**. Most often their interest is purely academic: double reduction of pulegone, cyclization of (+)-citronellal to (–)-isopulegol followed by reduction, and so on. Only those procedures developed from pinenes are exploited in the industry; some countries are thought to use 3-carene.

- **Total synthesis**. Several routes have been described. One of the more conventional ones goes through thymol, the product of alkylation of *m*-cresol by propylene or by iso-propanol. Racemic menthol is separated by distillation and the

enantiomers are resolved by crystallization as benzoates, or by biotechnological methods.

Menthol is largely consumed by the tobacco industry (chiefly in the United States). In pharmacy, it is an ingredient of itch-relieving creams and of preparations designed to decongest the upper respiratory tract in case of rhinitis; it is also an aroma. It is incorporated in oral hygiene products and shaving products. Some topical preparations also contain the esters of a pyrolidinic acid: they are slowly hydrolyzed on the skin and provide a lasting cool sensation (deodorants, after-shaves). Food technology, especially confectionery, finds many uses for menthol (Acceptable Daily Intake: ADI = 0-0.2 mg/kg).

• Oregano, *Origanum vulgare* L.

This perennial species, sometimes mistaken for other species in the genus, is common in France. It produces an essential oil generally rich in thymol, or carvacrol, or both. The market is largely supplied by Greece (gathering and cultivation) which exports toward the United States (for use in foods, e.g., pizzas). Several species in this genus of complex taxonomy are used as spices around the Mediterranean basin.

Phytopharmaceutical products based on the flowering tops of oregano and designed for oral use may claim therapeutic indications identical to those of marjoram (see p. 530); topically they are traditionally used: 1. as an adjunct in the emollient and antipruriginous treatment of skin disorders, as a trophic protective agent for cracks, bruises, frostbite, and insect bites; 2. as antalgic lozenges and collutoria for diseases of the oral cavity, pharynx, or both; 3. to relieve nasal congestion in the common cold.

• Rosemary, *Rosmarinus officinalis* L.

The flowering tops of rosemary, in addition to their cholagogue and choleretic properties (see p. 249), contain 10 to 25 mL/kg of an essential oil in which the principal constituents are camphor (15–25%), cineole (15-50%), α-pinene (10-25%), and borneol, free and esterified.

Since the composition of the essential oil depends on several factors, including source, the French Pharmacopoeia describes two products: the Spanish type, and the Moroccan and Tunisian type. Obtained by steam distillation from native plants, the two essential oils differ slightly by their physical characteristics and mostly by their compositions: cineole represents 38-55% of the Moroccan and Tunisian-type oil, but only 16-25% of the Spanish-type oil; the Spanish type is characterized by a [illegible] of hydrocarbons (18-26% α-pinène and 8-12% camphene, whereas

the Morocco and Tunisian type contains only 9-14% and 2.5-6%, respectively). The Moroccan and Tunisian type can contain little camphor (5-15%) whereas the Spanish type always contains a fair amount (13-18.5%). For the other constituents, the differences are less marked (see chromatographic profiles, Fr. Ph., identical to those of French standard NF T 75-214 [1994]).

Rosemary oil, like mint oil and sage oil, is a spasmolytic. Its CNS toxicity is thought to be non-negligible.

• Winter Savory, *Satureja montana* L.

According to the French Pharmacopoeia (10th Ed.), the drug—not to be confused with summer savory, *S. hortensis* L.—consists of the flowering tops of this subshrub of southern Europe (west of the Balkans) and north Africa (several subspecies are known). The multiple branches bear opposite leaves, ciliated on the edges, and lanceolate. The flowers, white or spotted with pink, form long unilateral racemes of small glomerules. Winter savory oil (>7 mL/kg in the official drug) is rich in carvacrol (up to 80% in some specimens), or, in some cases, in thymol; among hydrocarbons, *p*-cymene is always the most abundant (up to 25%). The composition is highly dependent on the harvest time, but independent from the subspecies.

Winter savory oil is a strong antiseptic *in vitro*, and this leads some prescribers to use it to treat infectious diseases of the respiratory or urinary tract. The drug is traditionally used orally to treat the symptoms of functional dyspepsia such as epigastric bloating, impaired digestion, eructations, and flatulence [French Expl. Note, 1998]. Topically, the Note authorizes the following claims: traditionally used to treat minor wounds after thorough cleansing; to relieve nasal congestion in the common cold; in mouthwashes for oral hygiene.

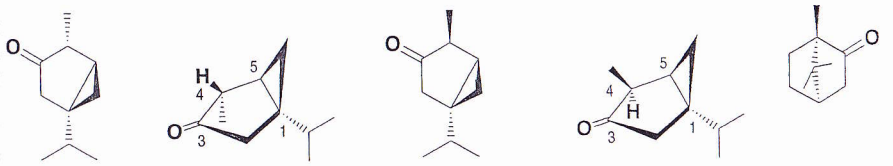
• Sage, *Salvia* spp.

According to the latest edition of the French Pharmacopoeia, the drug consists of the dried leaves of *Salvia officinalis* L. The French Explanatory Note of 1998, mentions three sages: *S. officinalis* L. (sage), *S. sclarea* L. (clary), and *S. lavandulifolia* Vahl., and the drug consists of the dried leaves in all three cases. The essential oil of *S. lavandulifolia* was added to the French Pharmacopoeia in January 1991. Acclimatized all around the Mediterranean rim, sage is cultivated and largely used as a spice.

The Plant. A very branchy subshrub, sage is characterized by oblong, lanceolate, and greenish-gray leaves with a granular surface (lower, petiolate leaves, 3.8 x 3.4 cm; higher leaves, smaller and smaller and [illegible]

clearly bilabiate and falsely whorled in groups of three at the apex of the stems. All of the leaves are denticulate, rough to the touch, and pubescent on both sides.

Chemical Composition. Sage leaf is rich (1-3%) in flavonoids which, like in many Lamiaceae, are flavones substituted at C-6 (e.g., 6-hydroxylated and 6-methoxylated derivatives of luteolin, of apigenin, and of their 7-*O*-methylated derivatives). It also contains many triterpenes derived from ursane (chiefly ursolic acid), oleanane (oleanolic acid and derivatives hydroxylated at C-2), as well as diterpenes (carnosol, rosmanol, epirosmanol, carnosic acid, methyl carnosate, lactone of the C-12 methyl ether of carnosic acid, rosmadial), and phenolic acids (rosmarinic acid).



α-Thuyone = (–)-(1S,4R,5R)-thuyan-3-one] β-Thuyone = (+)-(1S,4S,5R)-thuyan-3-one (+)-Camphor

Sage oil (8-25 mL/kg) is characterized by camphor, cineole, and bicyclic monoterpenoid ketones: the thujones (α-thujone [= (–)-(1*S*,4*R*,5*R*)-thujan-3-one] and β-thujone [= (+)-(1*S*,4*S*,5*R*)-thujan-3-one]). These may represent up to 60% of the essential oil, with α-thujone almost always accounting for the major part. The composition varies as a function of many factors. The profile defined by standard ISO 9909 [1999]for official sage oil is α-thujone (18-43%), β-thujone (3-8.5%), camphor (4.5-24.5%), cineole (5.5-13%), humulene (0-12%), α-pinene (1-6.5%), camphene (1.5-7%), limonene (0.5-3%), linalool, free and esterified (1% maximum), and bornyl acetate (2.5% maximum).

Tests. Sage leaf is identified in part by its microscopic characteristics: covering trichomes, swollen at the base, with a tapered apex, three cells, and thick walls, found on both epiderms, and visible in the powder; numerous glandular trichomes, sessile, and octocellular. The identification is completed by observing the red color developed by a hexane extract in the presence of sodium hydroxide (thujones). The assay includes a quantitation of the essential oil (required concentration 20-30 mL/kg) and a TLC analysis.

Pharmacological Properties. Sage enjoys a reputation for being a panacea * (p. 543). Although experimental work does not confirm all of the virtues that are attributed to it, it does prove the antispasmodic properties: small doses of essential oil inhibit the isolated guinea pig ileum contractions induced by electrical stimulation. The hydroalcoholic extract also displays spasmolytic properties against the spasms induced by acetylcholine or serotonin (possible role of polymethoxylated flavones? See thyme, p. 545). The antioxidant properties have long been applied to food

preparation, have also been demonstrated, and are linked to the presence of diterpenes.

In contrast to sage, its aqueous preparations, and its hydroalcoholic extracts, which seem to have little toxicity, the essential oil is neurotoxic: its ingestion causes convulsions preceded by hypersalivation and vomiting, and interrupted by periods of obnubilation, hyporeflexia, and hypotonia. The action is of central origin and seems linked to the ketones (thujones), and, to a lesser extent, to camphor.

Uses. Sage-based phytomedicines—official sage, clary, *Salvia lavandulifolia*—for oral administration may only claim one indication in France: traditionally used to treat the sympoms of gastrointestinal disturbances (epigastric bloating, impaired digestion, eructations, flatulence). Topically, all three species are traditionally used in mouthwashes for oral hygiene. In addition, clary may be used for minor wounds after thorough cleansing [French Expl. Note, 1998]. To apply for a marketing authorization from the French government, the total drug powders, hydroalcoholic extracts (titer >30%), and tinctures of any of the three drugs are required to pass basic safety tests (acute toxicity, chronic toxicity over 4 weeks); for official sage, a concentration limit must be stated for the active constituent. In any event, the prolonged use of these species for medicinal purposes, especially that of official sage, is not recommended.

In Germany, only one species is recognized as official, namely *Salvia officinalis* subsp. *minor* (Gmelin) Gams (= *S. officinalis s.s.*) and subsp. *major* (Garsault) Gams (= *S. tomentosa* Miller); in contrast, *S. lavandulifolia* Vahl. is not. Sage leaf is used orally for gastrointestinal problems and for excessive perspiration. Topically, it is proposed for inflammation of the mucous membranes of the mouth and throat. Furthermore, Commission E issues a reminder that alcoholic sage extracts and sage oil are contraindicated in pregnant women and that their prolonged use may cause epileptiform convulsions.

The dispensation of sage oil and of preparations that contain it is regulated (see hyssop above, p. 528).

Although thujones may not be added as such to food products, their presence in foods is permitted when it results from the addition of flavors of natural origin. The European legislation allows up to 0.5 mg/kg in foods and beverages, 5 and 10 mg/kg in alcoholic beverages with up to 25% or more than 25% alcohol, respectively, 25 mg/kg in foods containing sage-based preparations, and 35 mg/kg in bitter beverages.

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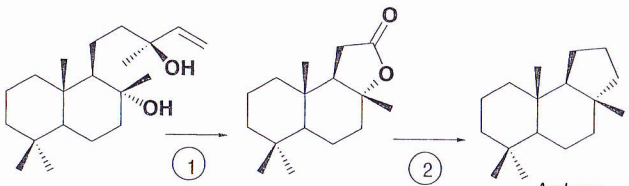
* Spanish authors have emphasized that the sage of Greek antiquity was probably a species commonly found in Greece, *Salvia fruticosa* Miller (= *S. triloba* L. *f.*) rather than *S. officinalis* L., as suggested by hasty translations [Rivera, D., Obón, C. and Cano, F. (1994). The Botany, History and Traditional Uses of Three-lobed Sage (*Salvia fruticosa* Miller) (Labiatae), *Econ. Bot.*, **48**, 190-195]. The essential oil of this Greek sage is characterized by a high concentration of cineole (up to 75%) and a low concentration of ketones (camphor and thujones) in most specimens.

• **The essential oil of *Salvia lavandulifolia*** has a very different composition: it contains no or almost no thujones (Fr. Ph. and French standard NF T 75-218 [1989]: <0.5%). Camphor and cineole are the most abundant elements: the chromatographic profile selected by the French Pharmacopoeia and standard gives, for these two compounds, the following tolerance range: camphor (11-36%) and cineole (11-25%). The profiles also covers α-pinene (4-11%), sabinene (0.1-3%), limonene (2-5%), linalool (0.5-9%), borneol (1-8%), linalyl acetate (< 5 %), and terpin-1-en-4-ol (< 2%). Most commercial samples contain sabinyl acetate, which is toxic; a systematic quantitation of it seems indicated.

• **Clary sage oil** is rich in (–)-linalool (10-20%) and (–)-linalyl acetate (45-75%), which occur alongside (–)-germacrene D and (–)-caryophyllene. The essential oil is either "traditional clary oil" (i.e., from steam distillation of the dried flowering tops) or "oil of clary crushed green" (*de sauge sclarée broyée en vert*). The chromatographic profiles of these two products, (NF T 75-255 [1992]) take into account the difference in composition that results from different preparation methods: linalool, 6.5-13.5% (traditional) or 13-24% (*en vert*); linalyl acetate, 62-78% (traditional) or 56-70.5% (*en vert*), and so on.

Amber oxide preparation:
1- chromic oxidation
2- reduction by a hydride



Sclareol — *Sclareolide* — *Ambrox (Amber oxide)*

The concrete essence contains mainly (–)-sclareol (70%) and its C-13 epimer. Sclareol is a diterpene concentrated in the calix (0.9-1.7%) and a minor compound in the steam distillation product (2%). It is a raw material for the perfume industry (a fixative), for tobacco flavoring, and for the semisynthesis of amber oxide *via* scareolide (including biotransformation steps or not). Clary oil comes mainly from Russia and other former Soviet republics (40% decrease in production between 1984 and 1994) and from the United States (increasing production). It is also produced in France, China, and Bulgaria.

• **The essential oil of *S. triloba* L. *f.*** (*S. fruticosa* Mill.) chiefly contains cineole (60%), camphor, borneol, terpineol, and about 7% thujones. This species of strong and pungent odor is listed in some pharmacopoeias (e.g., DAB 10, Helv. VII). It is characterized by a leaf blade with one or two small lateral lobes at the base, shaped approximately like ears. It contains tannins, flavonoids, and diterpenes.

• WILD THYME, *Thymus serpyllum* L.

Wild thyme is a small perennial herb with slender creeping stems, and with small glomerules of very fragrant white, pink, or purple flowers, is quite common in western Europe. The plant contains 1-6 mL/kg of an essential oil of highly variable composition (thymol, carvacrol, linalool, and more); the composition is all the more variable because *T. serpyllum* is not always distinguished from *T. pulegioides* L. and because many subspecies are known.

In the absence of relevant pharmacological or clinical studies, wild thyme leaf and flowering tops are traditionally used [French Expl. Note, 1998] orally, 1. for the symptomatic treatment of gastrointestinal disturbances (epigastric bloating, impaired digestion, eructations, flatulence); 2. for the symptomatic treatment of cough. Topically, both drugs are traditionally used to relieve nasal congestion in the common cold; to treat minor wounds after thorough cleansing; in antalgic lozenges and collutoria for disorders of the mouth, pharynx, or both, and in mouthwashes for oral hygiene. In Germany, its spasmolytic and antimicrobial effects are applied to the treatment of respiratory catarrh (orally).

• THYME, *Thymus vulgaris* L., *T. zygis* L.

It is unlikely that the constituents of the essential oil are responsible alone for the activities traditionally attributed to the drug, namely the "whole leaves and flowers separated from the previously dried stems of *Thymus vulgaris* L. or *Thymus zygis* L. or a mixture of both species" (Eur. Ph., 3rd Ed.). "It contains not less than 12 mL/kg of essential oil and not less than 5 mL/kg of volatile phenols, expressed as thymol [...], both calculated with reference to the anhydrous drug."

The Plant, the Drug. Thyme (*T. vulgaris*) is a subshrub with lignified and twisted stems. These are whitish and bear opposite, lanceolate or linear leaves (4-12 x 3 mm), curled downward on the edges (revolute margin). The upper side is marked by a depressed midrib; both sides are covered with a gray to gray-green indumentum. The flowers have a pubescent calyx and a bilabiate, pinkish or whitish corolla, which turns brownish upon drying; the flowers are gathered in ovoid glomerules. The leaf of *T. zygis* is smaller (4.7-6.5 x 0.4-1.2 mm) than that of *T. vulgaris*; it is linear to acicular, of the same color on both sides, and has long white hairs on the edges of the blade. The flowers of the two species are very similar. Microscopic examination of the cut and powder shows dodecacellular glandular trichomes, and covering, uni-, bi- and tricellular verrucose trichomes, most often bent (*T. vulgaris*) or more or less erect (*T. zygis*).

Chemical Composition. The essential oil content of the drug ranges from 5 to

with half a dozen of these having been described in the south of France alone. This explains the French Pharmacopoeial requirement for verification, by TLC, that the phenols—thymol and carvacrol—dominate over linalool. The drug contains apigenin, luteolin, and 6-hydroxyluteolin glycosides, as well as di-, tri- and tetramethoxylated flavones, all substituted at C-6 (5,4'-dihydroxy-6,7,3'-trimethoxyflavone (cirsilineol) and its 8-methoxylated derivative, 5,6,4'-trihydroxy-7,8,3'-trimethoxyflavone (thymonin), 5,4'-di-hydroxy-6,7,8-trimethoxyflavone, 5,4'-dihydroxy-6,7-dimethoxyflavone). Other constituents have been characterized: triterpenes, phenolic acids, saccharides, biphenyls, and more.

Tests. Thyme is identified by its morphology, the microscopic characteristics of its powder, and by TLC analysis of a dichloromethane extract. The assay includes a verification of the absence of foreign matter (stems of diameter >1 mm and length >15 mm, <10%) and of the absence of wild thyme leaves, which have long hairs at the base. Phenol quantitation is carried out on the essential oil, by dilution with water and ethanol followed by reaction with aminopyrazolone and potassium ferricyanide in the presence of ammonia. The reaction medium is extracted with dichloromethane and the absorbance of the organic phase is measured.

Pharmacological Activity. Thyme essential oil is rich in phenols, and it has antibacterial and antifungal properties readily shown *in vitro*; all chemotypes are active, but the bactericidal activity is strongest for the thymol—and carvacrol—containing types.

The spasmolytic activity of thyme is most often attributed to the essential oil phenols. However, Lemli and Van den Broucke have shown that although phenols do tend to prevent the contractions induced, in the ileum and the trachea of the guinea pig, by histamine, acetylcholine, or other reagents, their concentration in the aqueous preparations of the drug is insufficient to account for the activity. These authors showed that the spasmolytic activity of these preparations is linked to the presence of polymethoxyflavones. The antioxidant properties of the drug are due to flavones and to a biphenyl-type constituent far more efficacious than BHT (butylhydroxytoluene): at a 1µM concentration, it inhibits the lipid peroxidation induced *in vitro* in mitochondria and microsomes; it also partially inhibits the production of the superoxide anion.

Uses. Thyme leaf and flowering tops are traditionally used [French Expl. Note, 1998] orally: 1. for the symptomatic treatment of gastrointestinal disturbances such as epigastric bloating, impaired digestion, eructations, and flatulence; 2. for the symptomatic treatment of cough. Topically, they are traditionally used to relieve nasal congestion in the common cold; for the treatment of minor wounds after thorough cleansing; in antalgic lozenges and collutoria for disorders of the mouth, pharynx, or both; and in mouthwashes for oral hygiene.

In Germany, thyme is considered to be a bronchospasmolytic, expectorant, and antibacterial (Commission E). It is used for catarrh of the upper respiratory tract and for the symptoms of bronchitis.

Both the essential oil and thymol are ingredients of various proprietary drugs: antiseptic and healing ointments, syrups for the treatment of respiratory disorders, and preparations for inhalation. The essential oil is widely used as an antiseptic in aromatherapy. Thymol, an external and intestinal antiseptic, as well as an antifungal and anthelminthic agent, is used in the composition of products designed for buccal antisepsis and for the treatment of cutaneous irritation. Diodothymol is used for dental antisepsis.

The ***essential oil*** "of thyme, containing thymol (*Thymus zygis* [Loefl.] L.), Spain type" is the subject of a French standard (NF T 75-349 [1993]). Its chromatographic profile covers 13 constituents, including thymol (36-55%), *p*-cymene (15-28%) linalool (4-6.2%), and γ-terpinene (5-10.3%).

Oher Lamiaceae

• PENNYROYAL

Pennyroyal is *Hedeoma pulegioides* (L.) Pers., a New World species which gives "American" pennyroyal oil. The name pennyroyal is also used for *Mentha pulegium* L., which gives "European" pennyroyal oil. Neither species is official in France; nor are they listed in the French Explantory Note of 1998. European pennyroyal oil is subject to a French standard (NF T 75-233 [1980], equivalent to ISO 3714).

Like the essential oil of *M. pulegium* (5-10 mL/kg), that of *H. pulegioides* (10-20 mL/kg) is characterized by its major constituent, (+)-pulegone (70-90%), which occurs alongside monoterpenoid ketones: isomenthone, menthone, and piperitenone. Like European pennyroyal, American pennyroyal enjoys a reputation for being an antispasmodic, a digestion-stimulating agent, and externally, an antiseptic and repellent (hence the vernacular name *tick-weed* and the species name *pulegium*, from the latin *pulex* = flea).

Although the properties traditionally attributed to these drugs have not undergone any pharmacological investigation, the toxicity of the essential oil is known: it is periodically responsible for fatal accidents, often the consequence of abortion attempts (the essential oil is reputed to be emmenagogue and abortifacient). The toxicity is essentially hepatic, and it is due to pulegone (LD_{50} = 0.47 g/kg, rat, *per os*). Although the exact toxic mechanism has not been elucidated, it is known that the ketone is oxidized in the liver to metabolites (menthofuran and others) responsible for tissue necrosis.

It is generally accepted that drinking infusions of the leafy stems of either pennyroyal species is without side effects. However, two case reports of acute liver damage were described recently in California (1996), in two infants (6- and 8-

- **PATCHOULI,**
  Pogostemon cablin (Blanco) Benth.

Widely cultivated in southeast Asia, patchouli is a herbaceous species used for its leaves. Distillation after drying gives 1.5-2.5% essential oil. This oil is rich in sesquiterpenes (e.g., patchouli alcohol [30-40%], norpatchoulenol), and has outlets only in the perfume, cosmetics, and soap industries. The resinoid is also marketed (the yield can reach 5%). Other species in the genus give essential oils of inferior quality (e.g., *P. heyneanus* Benth. from India).

D. Lauraceae Containing Essential Oils

- **CEYLON CINNAMON,**
  Cinnamomum verum J. Presl

The barks of several species in the genus *Cinnamomum* Schaeffer constitute the various cinnamons, which are treasured spices. Those known as Ceylon cinnamon are listed in the 3rd edition of the European Pharmacopoeia, which specifies: "dried bark, freed from the outer cork and from the underlying parenchyma, from the shoots growing on the cut stumps of *C. zeylanicum* Nees".

The Plant, the Drug. The Ceylon cinnamon (still often referred to as *C. zeylanicum* Blume) is a small tree with indeciduous leaves, and thick and rough bark. Originally from India and introduced in the islands of the Indian Ocean and in southeast Asia, cinnamon is mainly cultivated in Sri Lanka.

On the spice market, Sri Lanka cinnamon, Seychelles cinnamon, or Madagascar cinnamon may appear as whole quills (scraped epidermis of the inner bark), quillings, featherings, chips, or powder. The French Pharmacopoeia provides the description of the whole quills: outer surface, smooth and finely striated, inner surface, darker, odor, aromatic, taste, warm and very refined. Under the microscope, a transverse section of the bark reveals, among other characteristics, the absence of suber (since the bark has been scraped), and the presence in the phloem parenchyma of large secretory cells as well as bundled or isolated fibers; these are long, have a regular lumen, and are clearly visible in the powder. A TLC analysis of a dichloromethane extract completes the identification. The essential oil concentration is not less than 12 mL/kg (for specifications for the different commercial products, see French standard V 32-115, equivalent to ISO 6539 [1998]).

Chemical Composition. Cinnamon bark contains starch, polycyclic diterpenes, and proanthocyanidinoid oligomers. The essential oil (5-20 mL/kg) is composed in major part of phenylpropane derivatives: (*E*)-cinnamaldehyde (65-80%), eugenol (up to 10%), and cinnamyl acetate. It also contains a large number of mono- and sesquiterpenes.

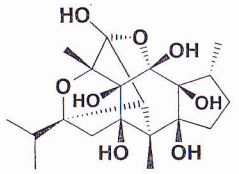
Pharmacological Properties and Uses. Although the activity of cinnamon—traditionally described as an "aromatic stimulant"—has barely been studied, the potent antibacterial and antifungal activity of the essential oil has been demonstrated *in vitro*. Cinnamaldehyde has also been the topic of experimental studies: a CNS sedative in the mouse (IP), a respiratory and myocardial stimulant in the dog (IV), it is also an antibacterial agent. Its hypotensive activity (dog, guinea pig) is thought to be due to a peripheral vasodilating effect. It decreases gastric and intestinal motility (rodents). In fact, most of these activities are observed only with high doses administered parenterally.

In France, the drug is traditionally used orally: 1. to treat the symptoms of gastrointestinal problems (epigastric bloating, impaired digestion, eructations, flatulence); 2. for functional asthenia; and 3. to facilitate weight gain [French Expl. Note, 1998]. In Germany, it is used for the same type of indications (lack of appetite, dyspepsia) but package inserts must mention contraindications: pregnancy and stomach ulcer.

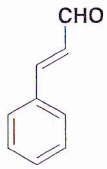
Cinnamon bark is used to prepare cinnamon tincture (Fr. Ph., 10th Ed.). Some practitioners use cinnamon oil *per os* for urinary infections, because a urinary bacteriostatic activity has been demonstrated.

Cinnamon and its oil are mostly used in food technology. Cinnamon oil and cinnamaldehyde, which are found in food and hygiene products, are both irritating for the skin and mucous membranes. They commonly induce allergic reactions such as urticaria or edema of the face and lips. Cross-reactions are frequently observed with subjects allergic to Peruvian balsam.

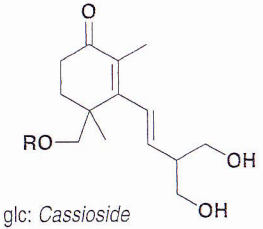
For miscellaneous products (perfumes, flavors), cinnamon leaf oil can be used. It contains, depending on the source, 70-95% phenols (eugenol) and 4-7% carbonyl compounds (French standard NF T 75-211, equivalent to ISO 3524). Cinnamon root bark oil is also available (60% camphor, 15% cineole).



Cinncassiol E



Cinnamic aldehyde



R = glc: *Cassioside*

- **CASSIA CINNAMON,**
  Cinnamomum aromaticum Nees, (= *C. cassia* Nees ex Blume)

Cassia cinnamon bark is not official, but it is listed in the French Explanatory Note of 1998, in the section regarding requests for government authorization to market plant-based medications; it may claim the same indications as those of the Ceylon cinnamon.

The Drug. This species is cultivated in the southwest of China (Guangxi and Guandong provinces); it is the source of a drug which, most often, still has a part of its suber and cortical parenchyma. Yellowish fibers, shorter and wider, and starch in greater abundance and in larger grains constitute the significant differences between cassia cinnamon and Sri Lanka cinnamon.

Chemical Composition. By steam distillation, cassia cinnamon bark yields 20 mL/kg of an essential oil containing 90% *E*-cinnamaldehyde and very little eugenol. Several diterpenes, free and glycosylated, have been described: cincassiols (A, B, $C_{1\text{-}3}$, $D_{1\text{-}3}$, E), cinnzelanine, and more. In addition, the drug contains phenylpropanoid derivatives, furanofuranoid lignans, polysaccharides, mono- and sesquiterpenoid glycosides (cassioside, cinnamoside), numerous flavonoid derivatives, particularly proanthocyanidins (A-2, B-1,2,5,7, C-1), and oligomers with four to six units, the cinnamtannins.

Pharmacological Properties and Uses. The anti-ulcer properties of the aqueous extract of the drug are attributed to 3-(2-hydroxyphenyl)-propanoic acid and to its glucoside, which turn out to be have a protective effect on cells (inhibition of the ulceration induced by phenylbutazone, serotonin, or ethanol, 40 µg/kg, rat, IP). Another study attributes the anti-ulcer activity to cassioside, to cinnamoside, and to a glycosidic derivative of trimethoxyphenol, all on the basis of a fractionation monitored by a pharmacological assay.

Despite noticeable differences in composition, the French Explanatory Note of 1998 allows cassia cinnamon-based phytomedicines to claim the same indications as those based on Ceylon cinnamon.

A traditional drug in Chinese medicine, which attributes to it stomachic virtues and a soothing effect on abdominal pains, cassia cinnamon is mostly used in food technology. The French standardizing organization AFNOR groups under a unique standard (NF V 32-116, equivalent to ISO 6538 [1998]) the Chinese cinnamon (*C. aromaticum*) and products less often found on the market, the Saigon cinnamon (*C. loureirii* Nees) and the Java cinnamon (*C. burmanii* [Nees] Blume).

Cinnamon oils are also used, especially cassia cinnamon oil (ISO 3216 [1997]), rich in aldehydes, including cinnamaldehyde (70-88%), (*E*)-*o*-methoxycinnamaldehyde (3-15%), benzaldehyde (0.5-2%), salicylaldehyde (0.2-1%), cinnamyl acetate (0-6%), eugenol (< 0.5%), coumarin (1.5-4%).

• Camphor Tree, *Cinnamomum camphora* (L.) J.Presl.

The camphor tree is a tall Asian tree (Taiwan, China) which has long been exploited for the production of (+)-(1*R*)-camphor, obtained by cooling and distilling the essential oil found in the wood (Hon-sho oil). Racemic camphor, a cardiac and respiratory analeptic, is easy to synthesize. After camphor has crystallized and has been filtered out, the essential oil (camphor oil) can be distilled into three fractions:

light, medium (80% safrole), and heavy (sesquiterpene rich). These three fractions are known as white, brown, and blue camphor oil, respectively. Some *C. camphora* chemotypes produce a leaf essential oil referred to as "ho oil of Taiwan" with (–)-linalool as the major constituent (75-85%); this industrial source represents tough competition for the more traditional sources such as Brazilian rosewood (annual production, China: 800 t [1994]).

Other Chinese *Cinnamomum* are exploited to produce Chinese sassafras (*C. porrectum* [Roxb.] Kosterm., *C. rigidissimum* H. H. Chang), a source of safrole that also competes with the Brazilian *Ocotea*.

• Sassafras, *Sassafras albidum* (Nutt.) Nees

The root bark of this North American deciduous tree yields 50-100 mL/kg of essential oil. Sassafras oil contains over 80% safrole, other phenylpropane derivatives, and hydrocarbons (α-pinene, phellandrenes). In addition, the drug contains isoquinoline alkaloids and lignans.

In the eastern United States, the drug has long been considered to be a carminative, diuretic, and antiseptic. It was formerly used—and it is believed that it is still used on occasion—to treat rheumatism or cutaneous eruptions. Sassafras bark and sassafras oil contain safrole, a known carcinogen in rodents (see p. 509), therefore any and all use must be proscribed.

Sassafras oil was widely used as a flavor before it was banned in most countries. Occasionally, and illicitly, it has been used to obtain 3,4-methylenedioxy-methamphetamine (MDMA, Ecstasy). In Europe, the final safrole concentration in cosmetic products must be < 0.01%. For the residual safrole concentrations in products to be ingested, see "nutmeg", p. 568.

The use of sassafras leaf, leaf extract, or safrole-free root bark extract (extraction with dilute alcohol, dilution, extraction of the oily fraction) remains possible.

• Laurel, *Laurus nobilis* L.

The bay laurel or noble laurel is a dioecious tree native to western Europe. Its leaves are alternate, coriaceous, slightly undulate. By steam distillation, bay leaves yield about 10-30 mL/kg of an essential oil in which cineole is always the major compound (25-60%) but whose composition depends on its geographical origin. In addition, the leaf contains sesquiterpenoid lactones (e.g., costunolide, laurenobiolide, artemorin) and isoquinoline alkaloids (reticuline, aporphinoids) very close to those found in boldo. The fruit is the blackish bay berry with its oily mesocarp, which gives bay butter, a solid at ambient temperature. Bay leaf is traditionally used orally to treat the symptoms of gastrointestinal problems such as epigastric bloating, impaired digestion, eructations, and flatulence.

• Other Lauraceae Used for Their Essential Oils

Litsea cubeba (Lour.) Pers. The essential oil of the fruit of this small tropical tree is responsible for its lemony odor. The essential oil is characterized by a high concentration of citral (geranial + neral, up to 85%); world production: about 1,400 t in 1994, mostly from China.

Ocotea pretiosa (Nees) Mez. The wood of this tall tree contains an essential oil with safrole as the major constituent (90%), which explains why it is often referred to as Brazilian sassafras (French standard NF T 75-229, equivalent to ISO 590)*. It is produced by Brazil in decreasing quantities and there are proponents of substitutes from sustainable sources (leaves of various species of the genus *Piper*).

Aniba rosaeodora A. Ducke var. *amazonica* and *A. parviflora* (Meissner) Mez. or Brazilian rosewood. Like the previous tree, it is exploited for its wood. Wood shavings are steam distilled to obtain an essential oil which contains 75-95% linalool and 3-6% α-terpineol (French standard NF T 75-227 [1988]).

E. Myrtaceae containing essential oils

Myrtaceae—a family comprising 3,800 species, including nearly 700 *Eucalyptus* and 500 *Syzygium*—are characterized by oil glands in their tissues. Highly prized in perfumery, food technology, and the fine chemicals industry, they often have antiseptic properties that are due to their essential oil and are exploited by the pharmaceutical industry.

• Clove, *Syzygium aromaticum* (L.) Merr. & Perry (= *Eugenia caryophyllus* [Sprengel] Bullock & Harrison)

An oriental spice with a rich history, cloves are widely used in culinary art. The 3rd edition of the European Pharmacopoeia specifies that the drug consists of the whole flower bud [...] dried until reddish-brown. "It contains not less than 150 mL/kg of essential oil".

The Plant, the Drug. The tree bears nondeciduous leaves. The flowers are tetramerous, pinkish-white, and grouped in small, compact, and ramified cymes. Originally from the Molucca Islands, cloves are traditionally cultivated in Tanzania (Zanzibar), in Madagascar, and in areas of Indonesia that have kept expanding over the last thirty years. Although the Indonesian production is essentially consumed

* Also known is the "Australian sassafras", *Doryphora sassafras* Endl. (Monimiaceae). According to Lawrence (1995), the world production of Brazilian sassafras oil and Chinese sassafras oil was 1,000 t and 750 t, respectively, in 1990.

locally (cigarettes), the world production is such that the price of the drug has decreased considerably in the late 1980s. The flower buds are collected manually, then dried. The floral peduncles ("clove stalks") are separated and directed toward distillation ("clove stem oil"). Although the flower buds are used in part for the obtention of oil ("clove oil"), the leaves are the main source of oil ("clove leaf oil").

The drug is very easy to identify: characteristic aromatic odor, pungent taste, and typical morphology. As suggested by the French name "clove nail" or *clou de girofle*, the flower bud is shaped roughly like a nail. It has a quadrangular part called hypanthium (length 10-12 mm x diameter 2-3 mm), which corresponds to the inferior ovary, and a globulous head (diameter 4-6 mm), surrounded by the four divergent lobes of the sepals, and consisting of four imbricated petals wrapped around numerous incurved stamens. The pulverized drug is characterized by fragments of parenchyma containing large secretory cavities, cluster crystals of calcium oxalate, many triangular pollen grains, and short fibers with thick, lignified, and slightly punctate walls.

Chemical Composition. Only clove oil has been the focus of attention. It occurs in exceptionally high concentrations: 150-180 mL/kg, 200 mL/kg in some specimens. Its composition is characterized by a propenylphenol which is by far the major constituent: eugenol. Found mostly in the free state, and in part as eugenyl acetate, its concentration fluctuates between 70 and 85%. Eugenol occurs alongside several dozen aliphatic, aromatic, and heterocyclic terpenoid compounds, as well as about 10% β-caryophyllene. The flower bud also contains chromone glycosides.

Tests. The assay includes the quantitation of the oil, the characterization of eugenol, its ester, and caryophyllene (TLC) in a dichloromethane extract, and the verification of the absence of foreign elements: the peduncle, petiole, and fruit (known as mother clove) content must be not more than 4%, that of spoiled cloves must be less than 2%, and foreign matter must be less than 0.5%. (On commercial categories, see French standard NF V 32-105 [1981], equivalent to ISO 2254).

Clove oil (Eur. Ph., 3rd Ed.) must contain 75-88% eugenol, 5-14% β-caryophyllene, and 4-15% eugenyl acetate (French standard NF T 75-207 [1991]). These specifications differ markedly from those of the French standard, NF ISO 3142 [1997].

Uses. Clove-based phytomedicines may be used locally for the following indications [French Expl. Note, 1998]: 1. to treat minor wounds after thorough cleansing; 2. as an antalgic (headaches, toothaches); 3. as an antalgic in diseases of the mouth, pharynx, or both (collutoria, lozenges); 4. in mouthwashes for oral hygiene. Internally, cloves are traditionally used to treat the symptoms of gastrointestinal problems such as epigastric bloating, impaired digestion, eructations, and flatulence. In Germany, clove- or clove oil-based preparations are used in mouthwashes for inflammation of the mouth or throat.

Cloves are seldom used in pharmacy. They are a prized spice (curry), and in Indonesia, they are consumed extensively as "kretek" cigarettes.

Properties and Uses of Clove Oil and Eugenol. Clove oil eugenol is a potent inhibitor of platelet aggregation (see nutmeg, p. 568). For a long time, dentists have used eugenol by the intracanal route, but the fact that this product can induce histological damage upon direct contact with living tissue has led some dentists to abandon its use. Eugenol has interesting local anesthetic properties (it inhibits nerve conduction) and it is also an anti-inflammatory (an inhibitor of prostaglandin synthesis and of the chemotaxis of leukocytes). It is bactericidal at low concentrations (10^{-2}-10^{-3} mM/L). The form normally used is the *zinc oxide and eugenol* (= ZOE) paste, a dressing in dentistry, in other words the chelate formed by two eugenol molecules and one zinc atom. It is sometimes combined with benzoin tincture to adjust the time it takes for the paste to harden. Eugenol also is used in the formulation of mouthwashes and ointments.

Systemically and at high doses (0.5 mL/kg), clove oil is toxic, especially in young children in which it causes CNS depression, hepatic necrosis, convulsions, and/or major hemostatic abnormalities. Eugenol is rapidly metabolized and excreted, mostly conjugated, epoxidized only to a small extent, and it is not carcinogenic. It is caustic for the skin and mucous membranes.

Clove leaf oil is an industrial source of eugenol (80-92%, French standard NF ISO 3141[1997]), which is a starting material for semisynthesis, for example for vanillin. Eugenol can also be isolated from clove stem oil (83-92%, NF 3143 [1997]), which is obtained by steam distillation.

• EUCALYPTUS, *Eucalyptus globulus* Labill.

With the exception of Papua New Guinea and a few nearby islands, the genus *Eucalyptus* is native only to Australia (not New Caledonia or New Zealand). Because eucalyptus trees grow fast, produce quality hardwood, and drain wetlands, they were introduced in all of the areas of the globe that have a favorable climate (from 45°N to 45°S); in some of those regions, eucalyptus trees now account for a significant part of the vegetation.

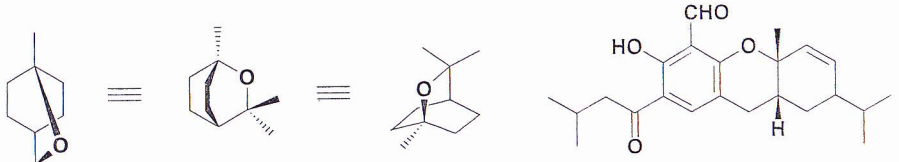
One of the assets of this genus is the diversity in the composition of the leaf essential oil, made even greater by the frequent occurrence of chemotypes. Thus the following essential oil types are known, and in some cases exploited: with piperitone and phellandrene (40-55%+20-30%, *E. dives* Schauer Type, *E. elata* Dehnh.), with phellandrene (60-80%, *E. dives* Schauer var. A), with phellandrene and cineole (40+50%, *E. radiata* Sieb. ex DC.), with geranyl acetate (45-55%, *E. macarthurii* D. & M.Camden), with citronellal (65-85%, *E. citriodora* Hook., see French standard NF ISO 3044 [1997]); with citral (up to 63%, *E. staigerana* F. Muell. ex Bailey) and of course, with cineole. According to Lawrence [1995], the production of cineole-type essential oil was about 3,300 t in 1990; in the same year, that of the citronellal type exceeded 2,000 t.

SYZYGIUM AROMATICUM (L.) Merr. & L.M. Perry

The dried eucalyptus leaf is listed in the 10th edition of the French Pharmacopoeia and the essential oil is listed in the 3rd edition of the European Pharmacopoeia. The official dried leaf is that of *E. globulus* only, but the essential oil may be "obtained by steam distillation followed by rectification, of the fresh leaves or the fresh terminal twigs of [...] *E. globulus* Labillardière, *E. fruticetorum* F. von Mueller (= *E. polybractea* R.T. Baker), *E. smithii* R.T. Baker [...and it] must contain not less than 70% 1,8-cineole (eucalyptol)".

The Plant, the Drug. The eucalyptus is a very large tree with a trunk that exfoliates into long shreds, and its leaves are characterized by a striking dimorphism: the leaves on young shoots are opposite, sessile, with a rounded blade, waxy, and bluish-green; the leaves on older branches are dangling, alternate, shortly petiolate, with a scythe-shaped blade, coriaceous, and grayish-green. The flower bud has four rough and waxy sepals, fused into a four-sided urn with a "lid" formed by the four fused petals. This lid opens when it becomes detached upon anthesis, and numerous stamens appear. The species grows wild in Australia, and has been introduced in many other areas of the globe, including in Europe (Mediterranean area) where it is cultivated (Portugal, Spain). The French Pharmacopoeia describes the leaf as "long and scythe-shaped" and "with a coriaceous blade", which effectively excludes the leaves of young shoots. The identification is extremely easy and may be completed by a microscopic examination of the leaf cut, which shows a blade with centric mesophyll (it is normally asymmetrical in the leaves of young shoots), a vein with three ligneous phloem bundles (one of them arched, the other two in the reverse orientation), and in all of the tissues, large schizogenous cavities.

Chemical Composition. The essential oil concentration ranges from 5 to 35 mL/kg. The major constituent (70-80%) is 1,8-cineole (= eucalyptol); the other constituents are chiefly terpenoids. The leaf also comprises a dozen oxygen-containing heterocycles with a mono- or sesquiterpenoid structure, including an acylphloro-glucinol moiety—the euglobals and macrocarpals—as well as phenolics, common phenolic acids, and flavonoids (rutin, hyperin, and methylated flavones in the epicuticular wax).



1,8-Cineole *Euglobal IIc*

Tests. They include quantitation of the essential oil (>20 mL/kg), TLC to verify the absence of citronellal, and a search for foreign elements (<2%). The assay for the essential oil is conventional, and includes refractive index, specific gravity, optical rotation, a search for other *Eucalyptus* (absence of citronellal by TLC), an

quantitation of cineole (by determination of the solidification temperature of a mixture of essential oil and melted cresol; the cineole content must be not less than 70% w/w [Eur. Ph., 2.8.11]). Cineole quantitation can also be carried out by GC according to the protocol of the French standard (NF T75-404 [1986]). Since 1993, the French standard organization AFNOR has included under standard NF T 75-225 the whole (raw) essential oils obtained from crushed green leaves, as well as essential oils rectified under vacuum, namely "70-85" and "80-85". These four products contain not less than 48, 58, 80, and 80% cineole, respectively. The raw oil of crushed leaves is characterized by 6-10% aromadendrene. Like globulol and *trans*-pinocarveol, aromadendrene disappears from the rectified products. The raw oils contain from 10 to 20-22% α-pinene. The limonene and *p*-cymene concentrations can double upon rectification.

Pharmacological Activity. Eucalyptus oil has antiseptic properties which have been established unambiguously *in vitro* on many germs. Cineole is readily absorbed by the digestive route, as well as by the cutaneous or rectal route, and is eliminated by pulmonary or renal excretion. It is widely accepted that eucalyptus oil (0.05-0.2 mL/day) has expectorant and mucolytic properties, and stimulates the bronchial epithelium. Eucalyptus oil, like menthol, is believed to "decongest" the upper respiratory tract in case of a common cold; the action of these products probably boils down to a stimulation of the receptors normally stimulated by the incoming nasal air flow (hence the *sensation* of easier breathing). At high doses, eucalyptus oil is neurotoxic (LD50 = 1.7 mL/kg, rat, IP). Cineole is an epileptogenic agent as a result of an inhibition of oxygen consumption and of ionic gradients within the brain tissues. It also induces hepatic microsomal enzymes (risk of drug interaction, not well-known). In humans, the ingestion of 10 to 30 mL of essential oil can be fatal, but the published data are contradictory. Lower doses may cause gastrointestinal distress (vomiting), an altered mental status, and in some cases, breathing difficulties. *In vitro*, the leaf hydroalcoholic extract is an antibacterial agent, particularly against cariogenic bacteria in the mouth; this activity is due to the macrocarpals.

Uses. Despite the absence of clinical trials to demonstrate the undeniable therapeutical interest of eucalyptus oil and cineole, both products are ingredients of many proprietary drugs because of their antiseptic and "decongestant" activity: syrups, lozenges, nasal drops, preparations for inhalation (these are proposed to treat the symptoms of ordinary respiratory disorders, such as non-productive, hacking coughs). In preparations for external use, cineole can be used to facilitate the transcutaneous absorption of other substances.

Phytopharmaceuticals based on eucalyptus leaves are traditionally used to treat acute benign bronchial disease (oral route and local use), and locally, to relieve nasal congestion in the common cold [French Expl. Note, 1998]. Eucalyptus is also used in Germany for catarrh of the upper respiratory tract and for bronchitis. There, package inserts must list: 1. the contraindications (no use *per os* in case of gastro-

(rare) side effects (vomiting, diarrhea); 3. a warning not to use eucalyptus in children under the age of two. The recommendation to drink the infusion slowly is based on the notion that the tannins in the drug exert an astringent effect on the inflamed mucous membranes of the throat.

• Tea Tree, *Melaleuca alternifolia* Cheel

The name "tea tree" causes much confusion. Confusion with tea (*Camellia sinensis* [L.] Kuntze) of course, but also with other Myrtaceae, because in Australia, the term "tea tree" is used for other species of the genus *Melaleuca* L., as well as for species of a closely related genus, *Leptospermum* Forster & Forster *f.* The tea tree is native to the northeast of New South Wales where its leaves are harvested to produce essential oil. (According to Weiss [1997], the production was 200 t in 1995). The major constituents are generally terpin-1-en-4-ol (see p. 496) and hydrocarbons (terpinenes), but some clones produce an essential oil in which the cineole concentration can reach 60%. To meet French standard NF T 75-358 [1991], the essential oil must contain not more than 15% cineole and not less than 30% terpin-1-en-4-ol. The chromatographic profile takes into account 11 other compounds (e.g., γ-terpinene, 10-28%, *p*-cymene 0.5-2%, α-terpinene, 5-13%).

The antibacterial reputation of the essential oil is consistent with *in vitro* tests that show its activity, as well as that of terpinen-4-ol and other constituents, against various strains (*Staphylococcus aureus*, *Escherichia coli*) but also against *Candida albicans* (MIC of the most active constituents: 0.06-0.5%, *v/v*) or against *Aspergillus niger* or *Trichophyton mentagrophytes*. Comparative tests have shown its potential benefits in the treatment of acne (*vs.* benzoylperoxide) and onychomycosis (*vs.* clotrimazole). It has also been tested for vaginal infections.

It is because of its antiseptic properties that the use of tea tree oil spread during the 1920s. After a period of decline, like many other natural products, it is now increasingly popular. It is now an ingredient, and not only in Australia, of gels, creams, lotions, and shampoos for human and animal use, foot care products, soaps, toothpastes, insect repellents, and air fresheners. It is frequently used in phytotherapy and can induce cutaneous irritation in very rare cases.

• Niaouli, *Melaleuca quinquenervia* (Cav.) S.T. Blake

Niaouli is a small tree native to the Moluccas and a species with persistent leaves. It grows in Australia, southeast Asia, and New Caledonia where it is widespread in all the savannas, and in Madagascar. The leaf essential oil—found at a concentration of about 15 mL/kg in trees from New Caledonia, which was the principal producer for a long time—contains cineole as the most common major

Madagascar trees: nerolidol, viridiflorol, and cineole/viridiflorol types. Niaouli oil, known as petroleum jelly after purification by treatment with lead oxide (see Fr. Ph., 8th Ed.), is an antiseptic; used for a long time in otorhinolaryngology, it is still an ingredient of combinations proposed for the adjunctive treatment of rhinitis and bronchial infections (e.g., oily preparations for the nasal route, mixtures for inhalations, suppositories).

• Cajeput, *Melaleuca cajuputi* Powell

Cajeput is a tree that grows in Australia, India, and southeast Asia. The leaf essential oil (5-25 mL/kg) contains, depending on the source, up to 65% cineole. Cajeput oil, which is traditionally used in southeast Asia, China, and Indonesia to treat infected cutaneous lesions and by inhalation for respiratory tract disorders, is an antibacterial *in vitro*. It is sometimes used in aromatherapy and it is an ingredient of antipruriginous ointments.

• Other Myrtaceae

The essential oil of myrtle (*M. communis* L.), seldom used in pharmacy, contains myrtenyl acetate, α-pinene, cineole, myrtenol, linalol, and methyleugenol, among others. Other Myrtaceae have culinary uses, for example allspice or Jamaica pepper, *Pimenta dioica* (L.) Merr., from the Caribbean Islands and from Central America: its fruit, dried when almost ripe, is rich (30-50 mL/kg) in an essential oil with eugenol (80%), methyleugenol, cineole, chavicol, and caryophyllene. The leaves of a closely related species, *Pimenta racemosa* (Mill.) J. Moore, are also of interest. They produce bay oil which is less rich in eugenol (50%), but contains 20% chavicol and aliphatic derivatives (octanone, octenol); it is said to be an antiseptic and has outlets in perfumery and cosmetic formulation.

F. Rutaceae Containing Essential Oils

Most Rutaceae elaborate essential oils in schizolysogenous pockets characteristic of the family. Some Rutaceae essential oils have limited uses, for example those of *Skimmia laureola* (DC.) Decne. (with linalool and linalyl acetate) or balsam amyris, *Amyris balsamifera* L. (Haiti candlewood). This is also true for the absolute of *Boronia megastigma* Nees ex Bartling, a species cultivated in Tasmania and New Zealand: the absolute is rich in ionones and it is used as a flavor in food products. Other genera within the Rutaceae are among the leaders in the world market for essential oils (*Citrus* spp.). Some species are growing in importance (*Clausena*, source of anethole). The number of species celebrated for their medicinal virtues is small:

• Bitter Orange Tree, *C. aurantium* L. ssp. *aurantium*

The bitter orange tree is a small tree cultivated mainly in the Mediterranean area (Seville or Bigarade orange). Its leaves have an oval and coriaceous blade which is articulated onto a winged petiole. The 10th edition of the French Pharmacopoeia devotes three monographs to the orange species (under *C. aurantium* L., spp. *amara* Engl.):

- the dried flower, collected before it blooms;
- the bitter orange peel, in other words the pericarp of the ripe or nearly ripe fruit;
- the dried leaf.

The Drugs. The flowers have a sweet fragrance, and are white or yellowish-white; they have a cupuliform, waxy calyx, five thick petals, and about 20 stamens fused at the base by their filaments. They contain many flavonoids, identifiable by TLC (naringin, neoeriocitrin) for which the French Pharmacopoeia requires quantitation by colorimetry on an ethanolic extract (magnesium and hydrochloric acid, see p. 319): the drug must contain not less than 0.8%.

Orange peel consists of spiral-shaped ribbons, fragments of ribbon, or fusiform quarters. Hard and brittle, brownish-green (external flavido) and white (internal albedo), it tastes very bitter and spicy. It can be identified by showing that it contains flavanones. TLC analysis of a methanol extract (flavanoids) and a quantitation of the essential oil (<20 mL/kg) are the main determinations required by the assay.

The leaf, large and oval (8 x 4 cm), has an articulated and more or less winged petiole. The leaf blade is punctuated by schizolysogenous pockets that are clearly visible in the leaf section under the microscope. The assay is similar to that of the flower (total flavonoids > 0.8%).

Pharmacological Properties and Uses. In the absence of pharmacological experiments and clinical data, and based on tradition, bitter orange peel is used orally to stimulate the appetite and facilitate weight gain. In Germany, where the indications are similar, gastric ulcer is a contraindication. Also based on tradition, the leaf and flower of bitter orange ***and of sweet orange*** are used—generally in infusion— to treat the symptoms of neurotonic disorders in adults and children, especially in case of minor sleeplessness [French Expl. Note, 1998]. The fresh bitter orange tree flowers are used to prepare (distilled) orange flower water, and the pericarp is the starting material for bitter orange syrup and bitter orange tincture (Fr. Ph.), which are used as flavors in medications for the oral route. Orange flower water is to be obtained from "fresh flower buds"; it contains 0.01-0.04% linalool (GC) and antimicrobial agents may be added as needed.

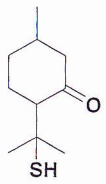
Comment: the presence of many flavonoids in the ***sweet orange*** peel justifies the following indications for this drug: traditionally used to treat the symptoms of cutaneous capillary fragility, such as ecchymoses and petechiae [French Expl. Note,

• BUCHU, *Agathosma* (= *Barosma*) spp.

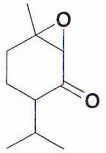
According to the 10th edition of the French Pharmacopoeia, the drug consists of the dried leaf of *Barosma betulina* (Berg.) Bartl. and Wendl. (= short or round buchu), *B. crenulata* (L.) Hook (= oval buchu), and *B. serratifolia* Willd. (long buchu). The Pharmacopoeia describes the leaves of the three species and specifies that "druggists' specimens are most often mixtures of official species".

This definition calls for a few comments. In addition to the fact that botanists favor *Agathosma* over *Barosma*, the most recent publications refer to only two species as sources of "buchu", namely round buchu, *A. betulina* (Bergius) Pill. and oval buchu, *A. crenulata* (L.) Pill.: they specify that spontaneous hybridization between species growing in the same geographical areas is very common, as are intermediate leaf shapes. However, Wichtl also refers to another buchu, namely long buchu, *A. serratifolia* (Cur.) Spreeth. All of these species are small shrubs widespread at high altitudes around Cape Town (South Africa).

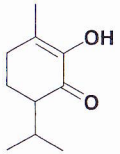
Chemical Composition. The leaves of round buchu contain flavonoids (diosmin and other glycosides), mucilage, and an essential oil (10-20 mL/kg) containing primarily ketones with a *p*-menthane skeleton: (–)-isomenthone, (+)-menthone, and less than 4.5% (–)-pulegone. Buchu oil also contains bifunctional derivatives, namely diosphenols (they might arise from the degradation of a piperitone epoxide). Its specific odor is due to sulfur-containing compounds: *cis*-(1*S*,4*R*)- and *trans*-(1*S*,4*S*)-3-oxo-*p*-menthane-8-thiol, and their acetylated derivatives. A recent study of 64 specimens showed that there are two chemotypes: the first one is rich in diosphenol (>10%) and ψ-diosphenol (>12%), and poor in isomenthone (<29%); in the second chemotype, which is of limited geographical distribution, the proportions of these compounds are reversed (diosphenol<0.14%, ψ-diosphenol<0.16%, isomenthone ≥ 31%). The essential oil of *A. crenulata* contains no diosphenols (<0.05%) but it can contain up to 70% pulegone.



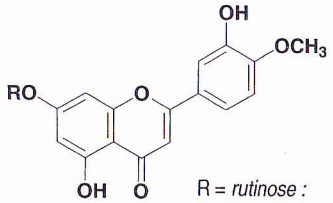
p-Menthane-8-thiol-3-one



Piperitone oxide



Diosphenol



R = *rutinose* : *Diosmin*

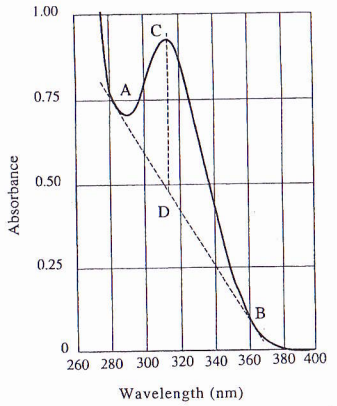
Tests. The French Pharmacopoeia requires buchu identification by the microscopic characteristics (fragments of epidermis, diosmin sphaerocrystals which turn yellow in KOH solution), and by characterization of the flavonoids (cyanidin reaction). The drug must contain not less than 13 mL/kg essential oil. It is possible (but not required by the Pharmacopoeia), to differentiate *A. betulina* from *A. crenulata* by TLC or GC analysis, by the presence or absence of diosphenols.

Pharmacological Properties and Uses. Buchu is thought to be a urinary antiseptic, despite the fact that the only tests carried out *in vitro* on the common germs of urinary infections have shown the leaf extracts activity to be negligible. The drug has not been studied in animals. It is traditionally used orally to enhance the renal elimination of water and as an adjunctive treatment to increase diuresis in benign urinary disorders [French Expl. Note, 1998]. The *British Herbal Compendium* lists pregnancy as a contraindication. Because buchu oil is rich in pulegone, its use in aromatherapy must be discouraged.

• ESSENTIAL OILS OF *CITRUS*

The various species in this genus elaborate and store essential oils in schizolysogenous pockets located in the external part of the mesocarp of the fruit (flavido). It is this favorable location that allows direct recovery of the oils by "expression". These oils may be used as flavors for medications and to formulate parapharmaceutical products. They are used primarily in food technology and perfumery.

Some of these essential oils are listed in the 10th edition of the French Pharmacopoeia, for example bergamot oil and mandarin oil. Others, such as lemon oil, appear in the European Pharmacopoeia (3rd Ed.). The essential oils of the *Citrus* species most widely used are subject to the French AFNOR standards, but to date, their chromatographic profiles have not been defined.



(Scheme: European Pharmacopoeia, 3rd edition)

A characteristic element of the *Citrus* oils obtained by expression is the presence of non-volatile compounds: their concentration is generally lower than 5%, but can exceed 10% (lime). The assay for *Citrus* oils includes the customary determinations (optical rotation, refractive index, acid value, and more), as well as a measurement of the carbonyl value, and a determination of the "CD" value by spectrometric analysis in the UV. This CD value provides information on the level of carbonyl compounds. The CD segment is that which connects point C at the absorption maximum to point D, namely the intersect of the projection of C onto the x-axis (wavelength) and of the tangent common to both portions of the spectrum (on both sides of the peak) (French standard NF T 75-122 [1982], equivalent to ISO 4735).

• *Bergamot Oil*

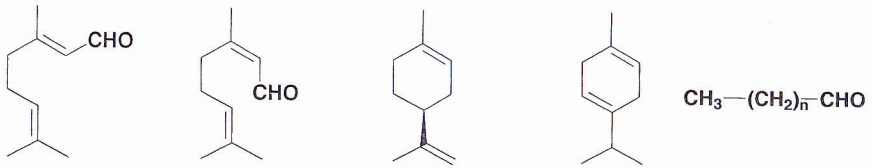
This essential oil is extracted without heating, by mechanical processes, from the fresh pericarp of the fruit of *C. aurantium* L. ssp. *bergamia* (Wight and Arnott) Engler, a species cultivated in Calabria (90 t in 1997) and in the Ivory Coast. Its composition is clearly different from that of other *Citrus* oils.

The chromatographic profiles chosen by the French Pharmacopoeia and by the French Standard NF T 75-215 are barely different. The Pharmacopoeia describes (in %): β-pinene (5-9.5), limonene (33-42), γ-terpinene (6-10.5), linalool (7-15), linalyl acetate (22-33), and geranial (<0.5). The evaporation residue is substantial: 4.5 to 6.5%. The bergapten concentration, estimated by HPLC, must fall between 0.2 and 0.45% (standard), or between 0.15 and 0.35% (Pharmacopoeia).

The chief consumers of bergamot oil (whole or for some uses, freed from bergapten) are the perfumery industry (colognes), and the cosmetology industry; regarding the therapeutical applications of photodynamic sensitizers, see the chapter on "Coumarins". The phototoxicity of bergamot oil has led the relevant international organizations to recommend a maximum concentration of 2% (75 ppm of bergapten) in perfumery products, whenever their use might be followed by exposure to sunlight.

- ***Sweet Orange Oil***

The pericarps of the different cultivars of the sweet orange tree (*Citrus sinensis* [L.] Pers. = *C. aurantium* L. var. *dulcis* Pers.) produce an essential oil comprising monoterpenoid hydrocarbons almost exclusively (limonene 93.5-96.5%, β-myrcene 1.5-2%). Alongside small quantities of aliphatic aldehydes (e.g., decanal<1.5%), monoterpenoid aldehydes (citrals<0.5%), and linalool (0.4-1%), it contains several dozen terpenoid and aliphatic compounds in trace amounts. The French standard includes in its definition the sweet orange oil "*type Guinée*", from the *limo-viridis* A. Chevalier and *djalonis* A. Chevalier varieties of *C. aurantium* L. Some sweet orange oils are high in carotenoids (1000 ppm), therefore they are prized in the beverage industry. ISO has proposed a spectrophotometric method for the quantitation of these coloring agents (NF ISO 9910 [1993]).



Geranial (citral A) — *Neral (citral B)* — *(–)-Limonene* — *γ-Terpinene* — n = 6: *Octanal*; n = 8: *Decanal*

- ***Bitter Orange Oil***

The fresh pericarp of the bitter orange, also called Bigarade orange (*C. aurantium* L. ssp. *aurantium*), produces, by expression, an essential oil fairly similar to that of the sweet orange, although less rich in carbonyl compounds: 96-98% limonene and other hydrocarbons (e.g., myrcene, α-pinene), 0.4-0.5% aliphatic aldehydes, and approximately 0.1% monoterpenoid aldehydes (French standard NF T 75-334 specifies 0.5-2.9% carbonyl compounds expressed as decanal).

- ***Bitter-Orange-Flower Oil***

Bitter-orange-flower oil is rich in linalool (28-44%), linalyl acetate (3-15%), limonene (9-18%), and β-pinene (7-17%). It also contains geranyl acetate (1-5%)

and neryl acetate (< 2.5%), *trans*-nerolidol (1-5%) and *trans-trans*-farnesol (1-4%) (see French standard NF T 75-202 [1995]). It was recently added to the European Pharmacopoeia (3rd Ed., add. 1998) with a slightly different chromatographic profile, particularly for linalool (18-42%), linalyl acetate (3-16%), and *trans*-nerolidol (1-9%).

- ***Lemon Oil***

This essential oil, prepared from the pericarps of *C. limon* (L.) Burm. *f.*, is a little less rich in monoterpenoid hydrocarbons (92-85%) than bitter orange oil, and the limonene level fluctuates between 60 and 75%; this monocyclic hydrocarbon occurs alongside 8-12% β-pinene and 8-10% γ-terpinene (average values). Note the presence of aliphatic aldehydes (0.2-0.5%, including nonanal and octanal) and monoterpenoid aldehydes (2-3%, including geranial, neral, and citronellal). The European Pharmacopoeia requires TLC analysis for the purposes of identification and verification of the absence of falsifications, and a determination of the carbonyl value (hydroxylamine method), which must fall within 2.2 and 4.5%, expressed as citral. The non-volatile residue must be between 1.8 and 3.6%. Measuring the specific rotation allows detection of adulteration by other essential oils. The French standard NF T 75-335 [1995] describes the specifications for this oil for three distinct origins: "Italy", "Spain", and "other origins", with minor differences between the chromatographic profiles.

- ***Mandarin Orange Oils***

Like lemon oil and lime oil (*C. aurantiifolia* [Christm.] Swingle, see French standard NF ISO 3519 [1997]), the (European) mandarin orange oil (*C. reticulata* Blanco) is characterized by a fairly low level of limonene (65-75%), and a high level of γ-terpinene (10-20%). The β-pinene level is low (1-3%). The 10th edition of the French Pharmacopoeia describes, for the "Italy"-type oil, the following chromatographic profile (in %): α-pinene (2-3), β-pinene (1.2-2), myrcene (1.5-2), limonene (65-75), γ-terpinene (16-22), *p*-cymene (<0.5), terpinolene (<1), and methyl *N*-methylanthranilate (0.3-0.6). The American variety, also known as tangerine, produces an oil consisting of nearly 90% limonene. In the chromatographic profile, French standard NF ISO 3528 [1997] does not include terpinolene nor *p*-cymene, but it specifies that the α-sinensal concentration must fall between 0.1 and 0.5%.

- ***Grapefruit Oil***

Grapefruit oil is from grapefruit or pomelo (*C. paradisi* Macfad.) and contains 96-97% monoterpenoid hydrocarbons (e.g., limonene, myrcene). The level of aliphatic aldehydes is low (0.6% including octanal and decanal). The total concentration in carbonyl compounds is 1-1.5%, including a non-trivial amount of nootkatone (0.15-0.30%).

- ***Petitgrain Oils*** (lemon tree, bitter orange tree, mandarin orange tree).

The term petitgrain oils designates the essential oils obtained by distillation of the leaves, small twigs and small unripe fruits of the species considered. The

MYRISTICA FRAGRANS Houtt.

composition of these essential oils is very different from that of the oils produced by the expression of the pericarps. Examples are: 1. lemon petitgrain oil (Italy), with carbonyl compounds (14-33%); 2. Bigarade and bergamot petitgrain oil, which contain linalyl acetate, linalool, and limonene; 3. mandarin orange petitgrain oil, which contains, as the major component, methyl *N*-methylanthranilate (45-63%); 4. Paraguay petitgrain oil with linalyl acetate (40-55%) and linalool (15-30%). All of these oils are subject to French AFNOR standards (NF T 75-236 to 239 [1987] and NF T 75-243 [1995]).

G. Other Drugs Containing Essential Oils

- **NUTMEG,**
  Myristica fragrans Houtt., Myristicaceae

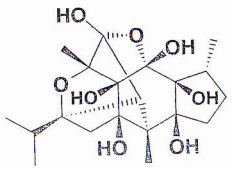
This tree (10-20 m tall) with indeciduous leaves produces drupaceous pale yellow fruits, which open at maturity by two valves. Out comes a unique seed with lignified tegument, ovoid, surrounded by an orangy-red, laciniate, and fleshy aril—the mace. The commercial drug—the nutmeg—corresponds to the seed reduced to the kernel (the "almond of the dried ripe fruit of the nutmeg tree" according to French standard NF V 32-125, after elimination of the husk and ligneous shell).

Originally from Amboine island (one of the Molucca Islands), the nutmeg tree was introduced on Mauritius, then in Malaysia, and later in Ceylon, Sumatra, and in the Caribbean Islands. Indonesia is currently the chief producer, together with Grenada (Windward Islands).

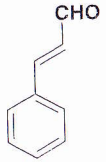
Chemical Composition. The nutmeg is mostly known for its essential oil. Found in a range of 50 to 150 mL/kg, it is mainly composed of terpenoid hydrocarbons: sabinene (14-29%), α-pinene (15-28%), β-pinene (13-18%), limonene (2-7%), γ-terpinene (2-6%), Δ^3-carene (0.5-2%), along with a fairly small amount of alkenylbenzenes—myristicin (5-12%), safrole (1-2.5%)—but also terpin-1-en-4-ol (2-6%) (concentrations expressed for the official nutmeg oil, Indonesia type, Fr. Ph., 10th Ed., see also French standard NF T 75-250 [1993]).

The chemistry of mace is dominated by the presence of phenylpropanoid compounds:

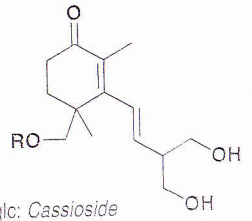
- phenolics (dehydrodiisoeugenol and its 5'-methyl ether);
- neolignans, of the bis-arylpropanoid type, and either acyclic or benzo-



Cinncassiol E



Cinnamic



R = glc: *Cassioside*

furanoid, with some arising from a coupling involving propenylbenzenes of the myristicin type (fragransols, myristicanols);

- lignans: 2,5-bis-aryl-3,4-dimethyltetrahydrofuranoid (fragransins) and diarylnonanoid (malabaricone C).

The level of essential oil in mace varies depending on the geographical origin from 8% (Grenada) to 12-13% (Indonesia); it is composed in major part of terpenoid hydrocarbons.

Pharmacological Activity. The volatile fraction has an effect against platelet aggregation, which is linked to eugenol and isoeugenol, despite their low concentrations. Eugenol, whose antiaggregating activity is comparable to that of indomethacin, also has antibacterial properties, and in the rat, it inhibits intestinal transit and intestinal secretions. Mace (methanol extract) displays anti-inflammatory activity. Eugenol and isoeugenol are inhibitors of cyclo-oxygenase, and inhibit prostaglandin synthesis in different tissues, including the mucosa of the colon.

The "hallucinogenic" properties of nutmeg have also been known for a long time; they have been observed in different contexts (inmates, adolescents). Some authors emphasize that the hallucinogenic effects are only the manifestation of a general toxicity; furthermore, human experiments show that the symptoms indicative of hallucinations are only observed in some of the research subjects. Therefore, nutmeg cannot be considered to be a hallucinogen: the most recent ethnobotanical investigations in Indonesia have found no trace of such use. The psychotropic activity of nutmeg (euphoria, hallucinations) seems linked to myristicin and closely related products: some authors do not exclude that these phenylpropanoids are transformed, by transamination within the human body, into amines such as *3-methoxy-4,5-methylenedioxyamphetamine* (= MMDA). Multiple side effects explain why this particular use is only marginal. Several cases of intoxication following ingestion of high doses of nutmeg (5-15 g) have been reported. The symptoms resemble those of the intoxication by atropine, except for the ocular signs: in that case a myosis is observed. The range of effects observed in the intoxicated patients would be due to the range of concentrations in the active ingredient, itself linked to the length of storage.

Uses. Still highly prized as a spice in the areas where it originates, and recommended as a medicinal plant for many indications, nutmeg is traditionally used in Chinese medicine as a stomachic and an antidiarrheal. Ayurvedic medicine attributes to mace digestive, carminative, and expectorant properties. Nutmeg is virtually no longer used in western medicine. It is used in foods, therefore these can contain safrole. In the particular case where the food product contains mace or nutmeg, the legal limit is 15 mg/kg whereas, as a general rule, the limit concentration for safrole and isosafrole in food products is 1 mg/kg (for alcoholic beverages of alcoholic titer <25% or >25% v/v, the limit is <2 mg/kg or <5 mg/kg, respectively).

• LEMON VERBENA, *Aloysia triphylla* (L'Hérit.) Britt. (= *Lippia citriodora* H.B. & K.), Verbenaceae

The dried leaf (Fr. Ph., 10th Ed.) has long been sold over-the-counter, like mint leaves and flowering tops, or linden inflorescences. Like these, it is widely used to prepare infusions, which some refer to—with some justification—as comforting health beverages. The drug consists of unifoliolate leaves with a narrow, lanceolate, acute, and undulate blade. The edges of the blade curl up during desiccation. Upon bruising, the drug gives off a pleasant odor, reminiscent of that of lemon. The powdered drug can be identified by microscopic examination, which shows short and thick hairs with cystoliths and a large number of anomocytic stomata. The drug assay includes quantitation of the essential oil (>0.4% v/w) and TLC analysis (to show the presence of citral).

The drug also contains flavonoids, chiefly 6-hydroxylated flavones and their methyl ethers (salvigenin, eupafolin, hispidulin, and more).

Lemon verbena is traditionally used orally to treat the symptoms of various digestive ailments, such as epigastric bloating, impaired digestion, eructations, and flatulence, and of neurotonic disorders in adults and children, especially in case of minor sleeplessness. Marketing a total drug powder-based medicine in France requires basic safety testing [French Expl. Note, 1998].

A study published in 1990 showed that verbena infusions have no sedative or anxiolytic activity in humans.

• STAR ANISE, *Illicium verum* Hook. *f.*, Illiciaceae

A small evergreen tree indigenous to the south of China and the north of Vietnam, the star anise (Eur. Ph., 3rd Ed.) is prized for its fruits. The star anise fruit is normally composed of 6-11 carina-shaped, reddish-brown, rough, often unevenly developed follicles, arranged in a star shape around a central, truncated pedicel. Each follicle opens at the upper edge by a slit through which one shiny brown seed can be seen. The pericarp has a marked aromatic odor and a warm, sweet, and anise-flavored taste.

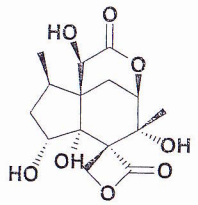
Chemical Composition. The drug is known to contain 5 to 9% essential oil. The chief constituent, by far (80-90%), is *E*-anethole. It occurs alongside methylchavicol (= estragole), anisaldehyde, and terpenoids (limonene, linalool, sesquiterpenoids hydrocarbons). The fruit also contains caffeic acids, shikimic acid (up to 8.5%), flavonoids, tannins, triglycerides, and sesquiterpenoid lactones that are convulsant but occur in very small quantities: 1 and 1.5 ppm of veranisatins A and B, respectively.

Tests. Star anise is easy to identify. The absence of *Illicium anisatum* must be verified: 1. macroscopically, by the absence of follicles with sharp points curved

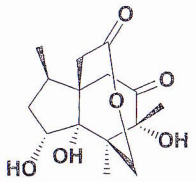
upward and of straight peduncles with a depression at the end; 2. by TLC analysis, the xylene solution resulting from the essential oil quantitation must have no band corresponding to myristicin. The drug must contain not less than 70 mL/kg essential oil.

Pharmacological Properties and Uses. Traditionally, the drug is thought to have antispasmodic and carminative properties, and to inhibit intestinal fermentations: in reality, pharmacological experiments (guinea pig ileum) show that anise-flavored oils increase the basal tone and the contractions of the smooth intestinal muscles. The star anise fruit is traditionally used orally in the symptomatic treatment of digestive disturbances such as epigastric bloating, impaired digestion, eructations, and flatulence; it is also traditionally used as an adjunctive therapy for the painful component of functional dyspepsia. The German Commission E monograph describes star anise as a gastrointestinal spasmolytic and broncho-secretolytic, which justifies its use for catarrh of the respiratory tract and dyspepsia..

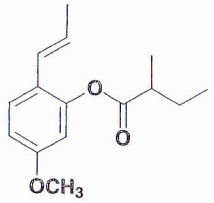
In France, the sales of star anise oil are regulated: it can only be obtained with a medical prescription, from pharmacists, and with proper recording of these prescriptions in the corresponding register (French Public Health Code or *Code de la Santé Publique*, article L 641). According to the French AFNOR standard [1995], the essential oil of "star anise- China type" is obtained by steam distillation of the fruits, leaves, and petioles. The concentration of (*E*)-anethole must be 86-93% and that of its *Z* isomer must be <1%; for other constituents, the specifications are slightly different from those of the Pharmacopoeia (which only lists the dried fruit oil, see p. 515): estragole (0.6-6%), anisaldehyde (0.1-0.5%), linalool (0.2-0.5%), α-terpineol (<0.3%), and more (see French standard NF T 75-252).



Anisatin



Pseudo-anisatin



2-Hydroxyanethole 2-methylbutyrate

• BASTARD STAR ANISE, *Illicium anisatum* L. = *I. religiosum* Sieb. & Zucc., Illiciaceae

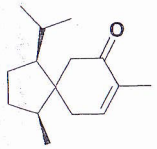
Official star anise is sometimes confused with the fruit of the bastard star anise. In the latter, also known as shikimi, the fruit and the seeds contain sesquiterpenoid lactones (anisatin, neoanisatin, pseudoanisatin, and related compounds) which impart convulsive properties to the drug. The essential oil, less abundant (2.5-10 mL/kg), is chiefly composed of terpenes (60-80%); phenylpropanoids are represented by safrole, methyleugenol, and in small amounts, by myristicin.

Morphologically, the fruit of the bastard star anise is smaller and the follicles form an irregular star. Differentiating the two species is not easy (the camphor-like odor and microscopic characteristics are very close): yet it is possible, even on a pulverized drug, by TLC, and based on the presence (bastard star anise) or the absence (star anise) of myristicin. This distinction is also possible by direct mass spectrometry of the drug.

• CALAMUS (SWEET-FLAG), *Acorus calamus* L., Araceae

This species is neither described in the latest edition of the French Pharmacopoeia, nor is it listed in the French Explanatory Note of 1998; however, it receives attention because of its potential toxicity.

The Plant, the Drug. This plant, originally indigenous to Asia, is fairly common on the edges of ponds and swamps in Europe and in eastern North America. Also known as sweet flag, it is perennial by a rhizome, and characterized by leaves that are reddish at the base, as well as flowers gathered along a fleshy axis, into a compact spike, wrapped into a very large spathe. The species is polytypical and includes several varieties: var. *americanus* (Raf.) Wulff (2*n*, America), var. *calamus* L. (3*n*, Europe, sterile), and var. *angustata* Bess. (4*n*, India, "Jammu" variety); there may also be an Indian, hexaploid variety.



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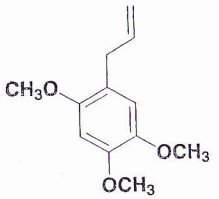
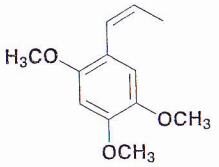
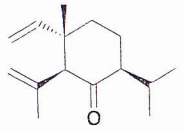


γ-Asarone



β-Asarone (cis-isoasarone)



Shyobunone

Chemical Composition. The drug (i.e., the rhizome) produces an essential oil ranging in concentration from 20 to 90 mL/kg. The essential oil of the European variety contains mono- and sesquiterpenoid derivatives (camphene, *p*-cymene, β-gurjunene, α-selinene, δ-cadi-nene, linalol, α-terpineol, α-cadinol, acorenone, calamendiol, isoshyobunone, and more), and phenylpropanoids at levels that rarely exceed 10% and which are chiefly represented by β-asarone (*Z*-isoasarone). This compound is absent from the essential oil of the American variety, which is characterized by shyobunone and acorone. β-asarone is dominant in the oil from the Indian variety: up to 96%. The triploid oil contains acorenone and the tetraploid oil contains γ-asarone.

Pharmacological Properties. The pharmacological properties of the essential oil (spasmolytic) and of β-asarone (CNS sedative) have been the subject of multiple studies. In addition, the toxicity of β-asarone has been demonstrated: long-term administration of Indian calamus oil to rats induces duodenal tumors. In mice, pure β-asarone is a hepatocarcinogen.

Uses. A drug reputed since ancient times for its "stimulant digestive" virtues, calamus is mostly used in food technology: although calamus and its products are banned in the United States, they are authorized in Europe (highest acceptable concentration in foods and beverages, 0.1 mg/kg; in alcoholic beverages and cracker seasoning, 1 mg/kg). Perfumery and cosmetology use the essential oil, whereas food technology mostly uses the hydroalcoholic extract. This extract is prepared from the European variety by maceration in 60% alcohol, and only contains traces of β-asarone. In spite of this, and considering the acceptable limits, a strict selection of the varieties to be used and rigorous control of the starting material are in order.

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