Chemotaxonomy of spice plants

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The entire plant kingdom demonstrates an immense variation in forms, structures, colours, and flavours. This is the result of evolution over the course of time. The manifestation of a plant (phenotype) is determined by a combined action of environment and genetic ability (genotype). Different phenotypes belonging to a definite genotype are an expression of internal plasticity. In systematics these modifications must be recognized, but for natural classification genetically determined variation is used.

The main task of biological systematics is that of defining, delimiting, describing, and naming taxa, and their incorporation in a hierarchical classification expressing different levels of similarity. A *taxon* is a group of organisms—of any taxonomic rank—which can be recognized in nature as a unity of the total pattern of variation (mint, labiates).

As far as possible, biological classification should be natural, that is, based on the totality of the knowledge of taxa. At the same time botanical classification should reflect evolutionary lines within the taxa concerned (phylogenetic classification). Finally, botanical classification must follow internationally accepted rules concerning the systematical categories division, class, order, family, genus, species and subcategories) and nomenclature.

To delimit, describe, and classify taxa, biological systematics makes use of their attributes, which are called characters. A character may be defined as an attribute of a taxon that can be used to distinguish it from other taxa. These characters can have the so-called present-absent state (seedplants with seed and cryptogams without them), but they can also have a gradually changing state (dimensions of leaves) and consequently must be quantified.

One further feature of characters is worth noting: As mutation, recombination and selection are the operating forces of the process of evolution, characters may be affected by parallel, convergent, or divergent evolution. In evaluating characters and the relationships of taxa, attention should always be paid to the real meaning of similarities and dissimilarities (Hegnauer 1966).

Chemotaxonomy

Mutual delimitation and classification of taxa can be based on different types of characters. For historical and practical reasons morphological characters receive priority. However, to reach a system of natural classification this means incorporating as much information as possible and reflecting evolution with as much truth to nature as possible. Other characters, such as ecological, anatomical, karyological, and chemical have to be incorporated also. The use of systematic chemistry for plant classification is called *chemosystematics* or *chemotaxonomy*. The meaningful interpretation of the totality of taxonomic characters is a difficult task. However, it has contributed much to natural classification.

Metabolic products as chemical characters can show on the one hand a large uniformity for a large area (nucleic acids); on the other hand a multiple diversity (natural products). In this context, the division of metabolic products into primary and secondary metabolites can be handled to advantage. Both sets of characters, primary and secondary plant substances, can be useful in biological systematics. Most secondary metabolites are used for classification within complex species (experimental systematics) and for classification within genera, families, and orders. But certain secondary metabolites may be used for classification of large systems, such as angiosperms and the whole plant kingdom.

Primary metabolites are important for judgment of phylogeny (amino acid sequence of

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1.	Nutmeg	Seed kernel	Myristica fragrans Houtt.	Myristicaceae
2.	Mace	Seed aril	Myristica fragrans Houtt.	Myristicaceae
3.	Laurel	Leaves	Laurus nobilis L.	Lauraceae
4.	Cinnamon	Bark	Cinnamomum zeylanicum Nees.	Lauraceae
5.	Cassia	Bark	Cinnamomum cassia Blume	Lauraceae
6.	Star anise	Fruit	Illicium verum Hook.fil	Magnoliaceae
7.	Pepper (black)	Fruit	Piper nigrum L.	Piperaceae
8.	Pepper (white)	Fruit	Piper nigrum L.	Piperaceae
9.	Mustard (black)	Seed	Brassica nigra L.	Brassicaceae
10.	Mustard (white)	Seed	Sinapis alba L.	Brassicaceae
11.	Bitter almond	Seed	Prunus amygdalus L.	Rosaceae
12.	Fenugreek	Seed	Trigonella foenum-graecum L.	Fabaceae
13.	Allspice	Fruit	Pimenta officinalis Lindl.	Myrtaceae
14.	Clove	Flowerbuds	Eugenia caryophyllata Thunb.	Myrtaceae
15.	Dill	Fruit	Anethum graveolens L.	Apiaceae
16.	Celery	Fruit	Apium graveolens L.	Apiaceae
17.	Caraway	Fruit	Carum carvi L.	Apiaceae
18.	Coriander	Fruit	Coriandrum sativum L.	Apiaceae
19.	Cumin	Fruit	Cuminum cyminum L.	Apiaceae
20.	Fennel	Fruit	Foeniculum vulgare Miller	Apiaceae
21.	Anise	Fruit	Pimpinella anisum L.	Apiaceae
22.	Paprika	Fruit	Capsicum annuum L.	Solanaceae
23.	Chillies (Cayenne)	Fruit	Capsicum species	Solanaceae
24.	Peppermint	Leaves	Mentha piperita L.	Lamiaceae
25.	Spearmint	Leaves	Mentha spicata L.	Lamiaceae
26.	Sweet marjoram	Leaves/floral parts	Origanum majorana L.	Lamiaceae
27.	Origanum	Leaves/floral parts	Origanum vulgare var.	Lamiaceae
28.	Rosemary	Leaves	Rosmarinus officinalis L.	Lamiaceae
29.	Sage	Leaves	Salvia officinalis L.	Lamiaceae
30.	Thyme	Leaves	Thymus vulgaris L.	Lamiaceae
31.	Savory	Leaves	Satureia hortensis L.	Lamiaceae
32.	Sweet basil	Leaves	Ocimum basilicum L.	Lamiaceae
33.	Onion	Bulb	Allium cepa L.	Liliaceae
34.	Garlic	Bulb	Allium sativum L.	Liliaceae
35.	Saffron	Stigmas	Crocus sativus L.	Iridaceae
36.	Vanilla	Fruit	Vanilla planifolia Andr.	Orchidaceae
37.	Tahite-vanilla	Fruit	Vanilla pompona Schiede	Orchidaceae
38.	Turmeric	Rhizome	Curcuma longa L.	Zingiberaceae
39.	Ginger	Rhizome	Zingiber officinale Roscoe	Zingiberaceae
40.	Cardamom	Fruit	Elettaria cardamomum Maton	Zingiberaceae

Table I. Botanical origin of spices and herbs

cytochrome c) but sometimes also on the level of genera, species, and within species and local populations. Also, chemical characters can have a present-absent state, for example, chlorophylls occur in algae, mosses, and vascular plants but never in fungi. But it is important to realize that the demonstration of presence or absence depends on the sensitivity of the methods applied. In this framework it is also important to realize that biosynthesis and accumulation of secondary products are different processes. According to Hegnauer (1976), a combination of both is essential to make secondary metabolites taxonomically useful characters.

Spices

Spices are plant products with important economical, culinary, and pharmaceutical interest. In general, they have an ancient history. According to most handbooks of food chemistry and pharmacognosy (Schormüller 1961, Steinegger and Hänsel 1972) spices can be defined by the possession of a series of qualities.

- They are dried parts of plants
- They are highly aromatic or pungent
- They are added in small quantities to food or drink for the improvement of taste, flavour and appetite
- They exert physiological activity

Spices can originate from different parts of the plant: rhizomes (ginger, turmeric), leaves/herbs (laurel, mint), barks (cinnamon), flowers (clove, saffron), fruits (pepper, anise), seeds (mustard, nutmeg), or bulbs (onion, garlic) (Table I). The large number of spices that belong to the fruits is notable, indicating a relation between the storage of natural products and the maturity of the plant.

The physiological action of spices has a close relation to the medical application of these plant products. Added to food in general, spices will improve taste and flavour. However, they also stimulate gastric juice secretion and in this way improve appetite and digestion. An influence on bile secretion, heart activity and circulation is also possible (mustard is an example) and antispasmodic activity of essential oil constituents is rather common (eugenol esters, Wagner 1979).

A conserving and antioxidative action can sometimes be ascribed to spices, and it is accepted that this activity is one of the reasons for the use of spices in the past.

The 1978 Spice Decree of the Dutch Food and Drug Act mentioned thirty spices belonging to about the same number of plants. Most handbooks of food chemistry and spices (Parry 1962) discuss about forty to fifty plant products used as spices. Combining both sources, a list of forty of the most important spices can be constructed (Table I). Many other plants are potential spices; a number of other plants are used as spices in tropical countries. But the concept of spices is historically strongly determined, and the herbs and spices in Table I have been thoroughly investigated.

Constituents

The characteristic flavours of spices generally rest on the presence of essential oils, and in some cases on crystalline flavour substances, such as vanillin and coumarin, or on different pungent compounds such as piperine, capsaicine, or gingerol. Alkyl sulfides and cyanogenic compounds also occur. This makes it clear that the chemistry of essential oils will play an important role in the chemotaxonomy of spices.

Bearing in mind what has been said about the combination of morphological and chemical information, it is important to realize the morphological implications of essential oil production. Essential oils are excreted by glandular hairs (Labiatae, Compositae) or accumulated in the plant body in cell organelles or idioblasts (Lauraceae, Piperaceae, Zingiberaceae) or schizogenous or lysogenous cavities or ducts (Cinnamon, Umbelliferous fruits). Essential oils are very complex mixtures of aromatic, more or less lipophilic compounds. They can be isolated by pressing, extraction, or steam distillation.

The chemical components of essential oils are in the first place representatives of the terpenes: mono- and sesquiterpenes. The oxygen-containing derivatives (alcohols, aldehydes, ketones and esters) are especially responsible for the specific flavour.

Next to the terpenes, but biogenetically separated from them, are a number of phenylpropanes and other flavour-determining compounds that can be present: aldehydes, alcohols, ketones, phenols, phenolethers, organic acids, esters. Nitriles and mustard oils also occur. Many of these latter two classes of compounds are glycosidic bound and can be set free by steam distillation or enzymatic hydrolysis (for example, cyanogenic compounds, coumarin).

Biosynthesis and accumulation

The mere presence of certain compounds in the essential oil is not the only important factor. The biosynthetic pathways should also be taken into account. Because the biosynthesis of essential oil compounds is genetically controlled, it is possible in this way to connect genetic and chemical information. The importance of biogenetic pathways can be illustrated with the spice "bitter almond" (*Prunus amygdalus*), which is characterized by the cyanogenic compound amygdaline.

Cyanogenic compounds are derived from different amino acids, demonstrate a variety of structures, and are rather widely distributed in nature. According to Tjon Sie Fat (1979) some structurally similar cyanogenic compounds originate from different amino acids (prunasine and taxiphylline); otherwise chemically different cyanogenic compounds (taxiphyllene and triglochinine) can derive from the same amino acid (fig. 1). The former are examples of analogous compounds; the latter represent homologous metabolites.

Another important matter is the previously mentioned accumulation of secondary metabolites. There is experimental evidence that the biosynthetic potential of plant cells is much larger than is indicated by specific patterns of secondary metabolites. However, many secondary metabolites are toxic for the organism unless a mechanism is present that can eliminate this toxic activity. Only plant species that build up an efficient tolerance mechanism can accumulate secondary metabolites and are able to integrate a chemical character in the genom. Subsequent selection by ecological factors can result in polymorphism.

The difference between mass accumulation and mere presence can be well illustrated by eugenol, an important component of spices. Trace amounts of eugenol are extremely common. However, relatively few plants are able to store eugenol. It may be a main constituent in essential oils, and in this instance excretion in specialized cavities makes accumulation possible (Röst and Bos, 1979, recently demonstrated the presence of up to 50% cis-methyleugenol in the root oil of some chemotypes of *Acorus gramineus*). However, large amounts of eugenol are also present in some species of Geum, and in the young leaves of *Camellia sasanqua* Thunb. In these taxa accumulation is possible by glycolisation (Hegnauer 1978).

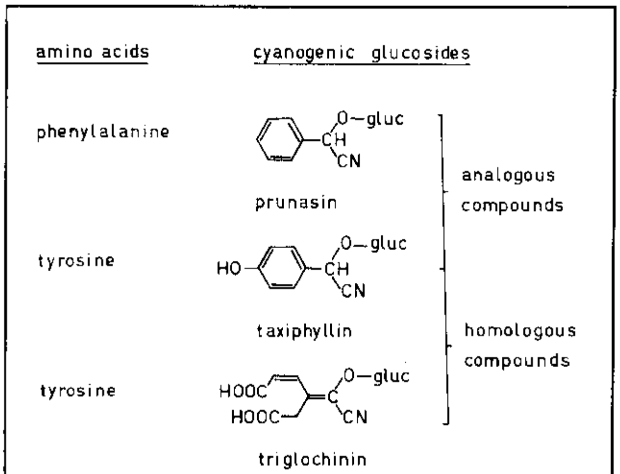


Figure 1. Cyanogenic glucosides; analogous and homologous compounds (Tjon Sie Fat, 1979)

Distribution of essential oils in the plant kingdom

Because essential oils are the main components of spices, it is interesting to know how they are distributed in nature. Very elaborate reviews of distribution of essential oil in plant kingdom have been given recently by Hegnauer (1978, 1979). It is beyond the scope of this article to reproduce these reviews extensively. They can be summarized by stating that:

- Essential oils occur in many red and brown algae and in some fungi
- Essential oils in Bryophyta are restricted to one class (Hepaticae)
- Within Pteridophyta only some true ferns have inner or external glandular hairs
- Most of the commercially available essential oils originate from seed-bearing plants

In *Gymnospermae* essential oils and balms are common within *Coniferopsida* (Cupressaceae, Pinaceae) and within *Taxopsida*, exclusively accumulated in schizogenous cavities. In *Angiospermae*, in particular in the Dicotyledons, essential oil is present in a scattered way but with certain concentrations in special taxa, such as *Myrtaceae, Rutales, Araliales, Lamiaceae*, and *Asteraceae*. There is also a distinct relation between essential oil synthesis and storage and the classification of the *Angiospermae*. In Monocotyledons synthesis of essential oil is rather rare, and accumulation happens almost exclusively in oil cells. With the exception of the Zingiberaceae species, essential oil synthesis and accumulation within Monocotyledons is, at best, a genus character.

To illustrate the relation between the presence of essential oil and the origin of spices, the most important essential oil-containing families are listed next to the forty spice plants belonging to these families. (See Table II; items within brackets are not essential oil families.) Most spices originate from true essential oil families, although some spices originate from non-essential oil families. Also, some true es-

Illiciaceae	1
Lauraceae	3
Myristicaceae	1
Piperaceae	1
(Brassicaceae)	2
(Rosaceae)	1
(Fabaceae)	1
Myrtaceae	2
Rutaceae	
Araliaceae	
Apiaceae	7
(Solanaceae)	2
Lamiaceae	9
Asteraceae	
(Liliaceae)	2
(Iridaceae)	1
(Orchidaceae)	2
Zingiberaceae	3
	40

Table II. Angiospermae: Essential oil families and origin of forty spices

sential oil families supply no spices (Rutaceae, Asteraceae).

Qualitative variation of essential oils

When using chemical compounds as taxonomic characters, insight into why and how variations occur in nature is necessary. In most cases, the composition of essential oil depends on the age of the plant and of the storage organ.

This can be illustrated with the labiatous spice plant mint (Mentha piperita). The composition of the essential oil of the leaves depends on the insertion height, and tends from menthone to menthyl acetate, according to the age of the leaf (Malingré 1966).

However, it is also possible that different parts of the plant synthesize and store their own characteristic and different essential oils.

For example, the essential oil of the stem and bark of the cinnamon tree (Cinnamonum zeylanicum) contains about 70% cinnamonic aldehyde, the oil of the leaves 60-90% eugenol, while the root oil is characterized by camphor.

Environmental factors can also influence the composition of the essential oil.

The influence of light on Mentha piperita is known. Long-day conditions result in menthol/menthyl acetate as the main constituents. In short daylight mainly menthofuran is synthesized and stored (Grahle and Höltzel 1963).

In nature individual plants usually live together in what can be called local populations. Within local populations, notable genetic variation can occur; this phenomenon is called genetic polymorphism. As metabolic products vary, the so-called chemotypes can be distinguished. Chemotypes occur frequently among essential oil plants, for example, Compositae, Labiatae, Lauraceae, Myrtaceae, Rutaceae and Apiaceae.

Different chemotypes within a local population are taxonomically meaningless. They can, however, be important for population genetics and for breeding for distinct chemical characters. Moreover, local chemical polymorphism can be the starting point of the evolution of chemical polytypism (chemical races).

Plant species consist of more or less extensive series of local populations. If the area of a species is large, different races will usually be formed by differences in conditions of life. In this way widely distributed species become polytypic and often comprise a varying number of morphologically and geographically delimited subspecies.

Similarly, chemical races (chemodemes) are not individual plants, but geographically bound and chemically characterized populations of a species. Many chemical races occur in nature, but it is difficult and time consuming to substantiate that a given pattern of infraspecific chemical variation represents chemical polytypism (genetically fixed chemical differences between local populations). If these chemical races are the result of plant breeding in the past, they should be considered to represent chemocultivars (clove); in fact, many of the economically important essential oil plants, such as dill, fennel, marjoram and basil, are the ultimate result of breeding for flavour.

Many investigations of chemical races of essential oil plants are reported in the literature (Stahl 1971). A very illustrative demonstration of chemodemes is given by Adzet and coworkers (1977) by their investigation of thyme.

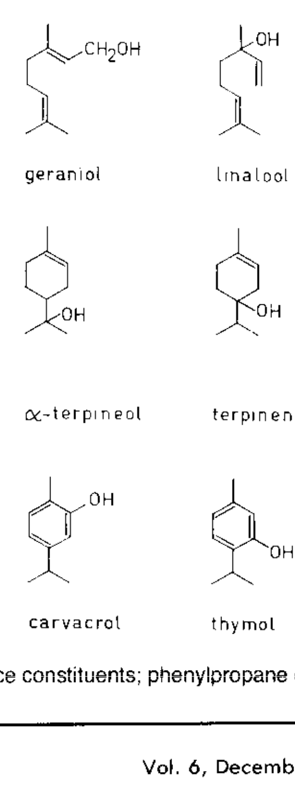
The West-Mediterranean species Thymus vulgaris L. is fairly homogeneous from the cytological (2n=30) and morphological point of view. But with regard to the essential oil composition Thymus vulgaris is very polymorpheus. Differentiation within this species seems mainly to proceed at the chemical level (see figure); geraniol, linalool, α-terpineol, terpinene-4-ol, carvacrol and thymol are main constituents. Moreover, there is a clearcut correlation between chemotypes and habitats; the two phenolic types being restricted to the hot and dry Carrigues. All information available indicates that chemodemes of Thymus vulgaris do present predominantly climatic ecotypes.

Chemical characters of spices in plant systematics

Comparing chemical characters of taxa of different levels means entering the field of plant systematics or taxonomy, in this case chemotaxonomy: the use of chemical variation for the classification of taxa. For a correct judgement of chemical variation, insight into different causes of variation is necessary. In the case of essential oils a distinction has to be made between the essential oil as such and its separate constituents.

Essential oils

The presence or absence of essential oil, according to the biological definition, can be very important systematically. It depends on the taxa concerned at which level this character can be used.



Spice constituents; phenylpropane derivatives

In Bryophytes the class Hepaticae and within Gymnospermae the class Coniferopsida are characterized by the presence of essential oil. In both cases essential oil represents a class character. Within Angiospermae essential oil also can be used as a chemical character for classifying taxa. Almost all Magnoliidae are aromatic plants and the presence of oil cells is one of their outstanding attributes. In fact Piperaceae and other families with deviating flower morphology are incorporated into this subclass on the base of this attribute. Essential oil synthesis and storage is a family character of several families, among others the Myrtaceae, Rutaceae and Apiaceae. Actually the presence of essential oils favours reclassification of the taxa of the Aralianae

In particular, Labiates (Lamiaceae) are known as a family of strongly aromatic plants, but according to Hegnauer and Kooiman (1978), Labiates may be divided into an oil-poor group which often contains iridoid glycosides (relation with oil-poor Verbenaceae) and an oil-rich group without iridoid glycosides and comprising many spices such as mint, rosemary, and thyme.

Finally, within the Monocotyledonae, Zingiberaceae is the only family characterized by the presence of essential oil in oil cells and by supplying different spices, such as ginger, turmeric, and cardamom.

Essential oil constituents

In general, the main essential oil components, such as phenylpropanes and mono- and sesquiterpenoids, can be helpful for the analysis of variation or the study of microevolution, but hardly can be used for taxonomic purposes. Even rather complicated sesquiterpenoids such as humulene and caryophyllene are ubiquitous in essential oils, and can only be used for characterizing chemodemes, species, or genera, not for characterizing higher taxa.

Exceptions to this rule can be found if patterns of compounds are used instead of individual compounds, or if unusual types of compounds are taken into account and at the same time recognizable tendencies are evaluated. This can be illustrated by some examples.

The essential oil spectra of Hepaticae have a typical sesquiterpenoid character, while the leaf oils of Coniferopsida are characterized by diterpene hydrocarbons (phyllocladene, hibaene, kaurene) and within the Coniferopsida the Cupressaceae are distinct by the presence of isoprenoid tropolone derivatives. Within the Lauraceae furano-sesquiterpenes (germacrene-eudesmane skeleton) can be used for chemotaxonomical purpose. Furthermore, even the presence of benzyl benzoate is ex-

ceptional as constituent in healthy wood (Aniba species) or in the leaves of some native forms of Cinnamomum zeylanicum Blume, indicating relationships with the Annonaceae (Gottlieb 1972).

Most essential oil components of Umbelliferae (Apiaceae) are by no means typical of the family. Nevertheless biosynthetic tendencies are recognizable at the family level, such as ferulol derivatives and polyacetylenes like falcarinone. The latter even indicate relationships between Umbelliferae and Compositae (Hegnauer 1978).

Another tendency can be recognized at different species of the Lamiaceae; reduction of oxygenated monoterpenes to alcohols and their subsequent esterification with increasing maturity of the plant (1,2-epoxymenthyl acetate in Mentha suaveolens as analogue of menthyl acetate in Mentha piperita (Hendriks 1970).

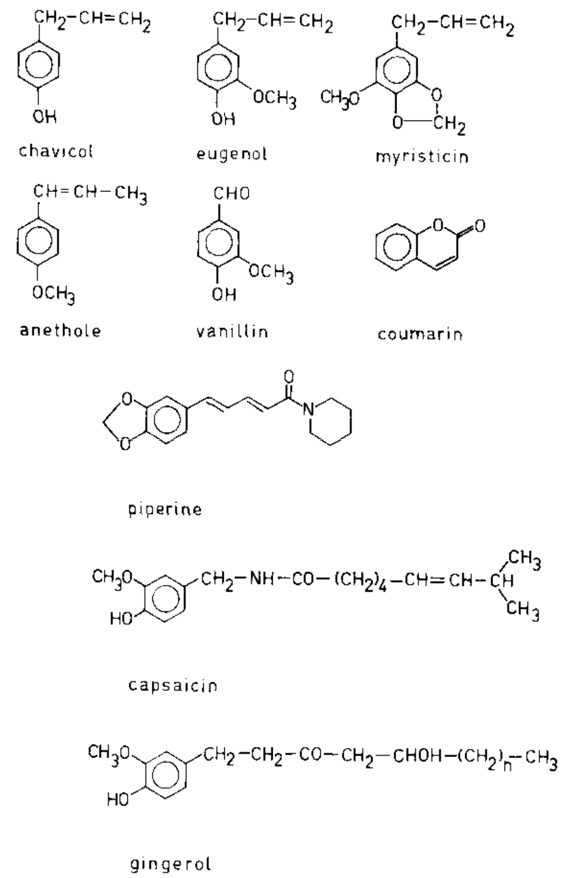


Figure 2. Chemotypes of Thymus vulgaris L. (Adzet a.o. 1977)

Nonvolatile constituents of spices

Not all substances responsible for the flavour of spices appear in the essential oil, depending on the isolation method applied. Many natural substances have limited volatility and are instable in steam, including representatives of terpenoids and phenylpropanes. This depends on molecular weight, structure, and substitution. The acetylenes also belong to this group of natural substances, which indicates close phytochemical relations between Araliaceae, Apiaceae, and Asteraceae. The same applies to the prenylated coumarins (the furanocoumarins are biogenetically included), which together with other characters suggest close relations between Rutaceae, Umbelliferae and Compositae. It is notable how many flavour substances and pungent compounds of spices can be derived of C_6-C_3 acids (e.g. chavicol, estragole, coumarin, eugenol, and myristicin). See figure 2.

Finally, it can be concluded that many of the bitter compounds, another important aspect of spices, belong to the terpenoids, are more or less cyclisized, and sometimes have unusual structures.

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