

# Importance of minor components in flavors and fragrances

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As recently as twenty years ago the isolation of natural substances and the elucidation of their structure had to be performed almost exclusively by chemical means. Large quantities of starting material were required, and time-consuming methods had to be applied. As a result, only the main constituents of essential oils were discovered. Thus, progress in this field was very slow.

The discovery of gas chromatography by James and Martin and the development of spectroscopic methods revolutionized the analysis of natural products. Suddenly it became possible to detect trace components. (By trace components I mean sensorily active compounds which are present in a complex mixture in amounts below 1%. This level is obviously chosen arbitrarily.)

This revolutionary development is particularly impressive in the field of flavors, if the number of products discovered in one year is taken as a measure. In 1967 not more than 750 aroma components were known, but today there are around 3,000. This means a quadruplication within the last 10 years. We observe the same tendency in the field of essential oils, and there is no sign when this curve is going to level off.

## Importance of trace components in flavors and fragrances

At this stage of accumulation of information, the natural products chemist has to ask himself what value trace components actually have in flavors and fragrances.

The most important feature of an aroma compound is its sensory contribution to the odor pattern of a complex mixture of natural substances. The relative importance of a constituent will be expressed in odor values which are defined as the ratio of the concentration of a constituent to its threshold concentration.

The odor value thus shows how much the actual concentration of a substance exceeds its threshold. Differences in odor values between constituents may therefore be taken as an index of odor effectiveness. Before such a calculation is discussed, the threshold concentrations of some important natural products can be seen in figure 1.

2-Isobutyl-3-methoxypyrazine (2), the character impact compound of bell peppers, in which it is present at a concentration of 2 ppb, exceeds its threshold value by 1,000 times. Although this

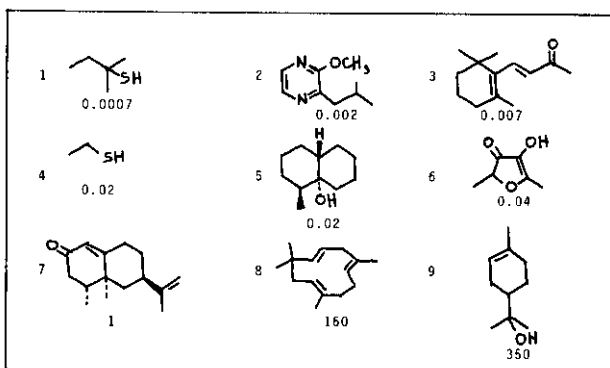


Figure 1. Odor threshold values in parts/10<sup>9</sup> (ppb) of water

is difficult to visualize, 0.002 ppb means that 1 g of this substance is present in 500,000 (metric) tons of water. The decalol derivative called geosmin (5), which emits a strong earthy-musty odor typical of freshly ploughed soil and which is found in several foodstuffs, also has an incredibly low threshold value. Finally, the well-known odorant  $\alpha$ -terpineol (9), identified in more than 150 essential oils, still has a low threshold although it exceeds that of geosmin by 17,500 times. These three examples make clear what minute amounts of a single chemical component are perceived by the human olfactory system. At the same time they show the immense differences in quantity, for which there has so far been no plausible scientific explanation.

## Bulgarian rose oil—an illustration

The problem inherent in trace components can be illustrated by Bulgarian rose oil. All the constituents of this essential oil present at concentrations of about 1% or more were known 20 years ago. However, although these 9 constituents account for 86% of the total oil (see Table I), mixing these individual components

Table I - 9 of the 39 constituents of Bulgarian rose oil, identified up to 1959

(-)-citronellol	38%
paraffins	16%
geraniol	14%
nerol	7%
$\beta$ -phenyl ethanol	3%
eugenol methyl ether	3%
linalool	2%
ethanol	2%
farnesol	1%
	86%

Table II - Odor units of 14 of 275 constituents of Bulgarian rose oil

Constituent	concentration %	threshold in ppb of water	$10^{-3} \times$ number of odor units	rel. % of odor units
(-)-citronellol	38	40	95.000	62
paraffins	16	--	0	0
geraniol	14	75	1.860	1.2
nerol	7	300	233	0.15
phenyl ethyl alcohol	2.8	750	37	0.024
eugenol methyl ether	2.4	820	29	0.019
eugenol	1.2	30	400	0.26
farnesol	1.2	20	600	0.39
linalool	1.4	6	2.300	1.5
(-)-rose oxide	0.46	0.5	9.200	6
(-)-carvone	0.41	50	82	0.05
rose furan	0.16	200	8	0.005
$\beta$ -damascenone	0.14	10*	140	0.09
$\beta$ -ionone	0.03	0.007	42.860	28

did not reproduce the odor pattern of rose oil at all. It was clear that the minor constituents, to a large extent, were responsible for the characteristic fragrance of rose oil.

Among the 275 constituents known today, figure 2 illustrates a small selection of compounds which were discovered in our laboratories.

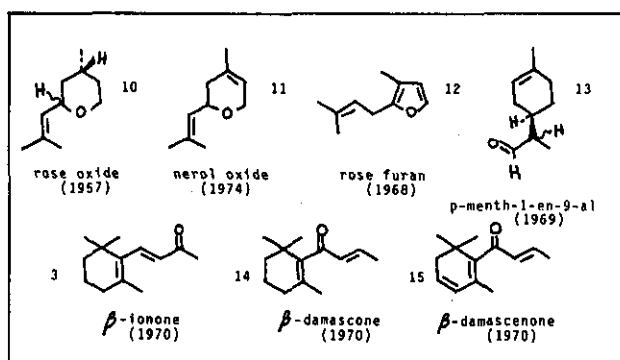


Figure 2. Monoterpenoid minor compounds of the odoriferous principle of Bulgarian rose oil (*Rosa damascena* Hill.). Representing 1% of the total essential oil

Although these constituents together amount to only about 1% of the essential oil, their sensory properties are decisive for the development of rose fragrance. Rose oxide and nerol oxide contribute to the geranium-like odor impression. Nerol oxide has been discovered independently by Dr. Tesseire's group in Grasse, although this compound had been synthesized in our laboratories 10 years earlier. Rose furan and the menthenal derivative, however, develop a characteristic citrus note.

These four monoterpene derivatives thus contribute highly to the flowery-green top note and the characteristic lifting effect in rose oil. In addition to these compounds  $\beta$ -damascenone, with its narcotic scent reminiscent of exotic flowers and its heavy fruity undertone, decisively participates in the so-called body note. It gives a high degree of diffusion to a composition. Although they differ chemically from each other

merely by the inversion of their allylic function in the side chain,  $\beta$ -ionone and  $\beta$ -damascenone have totally dissimilar sensory properties. While  $\beta$ -ionone is known to reproduce the typical scent of violets,  $\beta$ -damascenone has an odor related to that of  $\beta$ -damascenone.

Table II lists the quantitative contribution of some minor components in relation to the major rose constituents.

Although it is present in rose oil in a concentration 200 times lower than nerol,  $\beta$ -ionone has an odor value which is approximately 200 times higher, due to its extremely low threshold. Another substance with a remarkably high odor contribution is rose oxide. The last five minor components, constituting together only 1.2% of the total oil, possess half the odor value of all the main components put together.

### Damascenone and ionone sensory derivatives

Having thus demonstrated the importance of minor constituents, I should now like to discuss the chemistry of a selected group of carotenoid metabolites. To be more precise, I will talk about the occurrence and formation of important sensory derivatives of damascenone and ionone.

In the last few years,  $\beta$ -damascenone and three of its derivatives have proved to be fra-

\*Table II Note: Continuing the determinations of threshold values of minor components, Pickenhagen found that the actual threshold of  $\beta$ -damascenone is by a factor of  $10^3$  lower than reported here. The error is due to the fact that, at a certain level, which exceeds its threshold by a factor of around 1000,  $\beta$ -damascenone seems to have a fatiguing effect and stays in the mouth. Thus by determining the threshold by a double triangle test against water, with descending concentrations, tasters frequently indicate the product to be in the blank too. The now found threshold concentration is 0.009 ppb, and was determined using ascending concentrations. This value increases the number of odor units of  $\beta$ -damascenone to 156.000, which means that the value of this component is higher than the sum of the other compounds in this table.

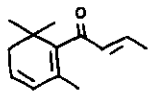
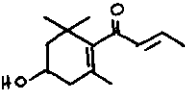
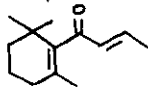
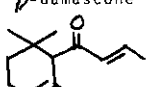
## Importance of minor components

grances of general importance. Their discovery in various essential oils and foodstuffs is indicated in Table III. As we shall see later, a chemical relation exists between  $\beta$ -damascenone and 3-hydroxy- $\beta$ -damascone. The hydroxy derivative presumably acts as a precursor of  $\beta$ -damascenone. Indeed, twice the quantity of its hydroxy compound is found in rose oil, whereas in several kinds of tobacco the proportion of 3-hydroxy- $\beta$ -damascone to  $\beta$ -damascenone reaches a ratio of 10:1. Damascone derivatives are not considered to be genuine metabolites, but rather metabolites of carotenoids like the ionones. 3-Hydroxy- $\beta$ -damascone can thus formally be conceived as a biodegradation product of zeaxanthene, and  $\beta$ -damascone as a bio-fragment of  $\beta$ -carotene.

$\beta$ -Damascenone (15) and 3-hydroxy- $\beta$ -damascone (17) are presumably formed from the acetylene diol 16 (fig. 3) in a biogenetic-type reaction in 70% yield in a proportion of 1:4. This acid-catalyzed transformation simultaneously shows the chemical relationship between the ionone series and the damascone series. We presume that the intermediate in this conversion is the allenic trien-diol 18 which, formally, yields 3-hydroxy- $\beta$ -damascone in a Meyer-Schuster-type reaction and  $\beta$ -damascenone

through cation 19. Isoe and coworkers presume grasshopper ketone (20) or its reduction product

Table III

	Bulgarian rose oil
$\beta$ -damascenone	various tobacco brands
	raspberries
	cooked Brambley's seedling apples
	various grape and wine varieties
	tea - coffee - beer
	buchu leaf oil
	Roman camomile
	Bulgarian rose oil
3-hydroxy- $\beta$ -damascone	various tobacco brands
	Bulgarian rose oil
$\beta$ -damascone	Burley tobacco
	tea
$\alpha$ -damascone	

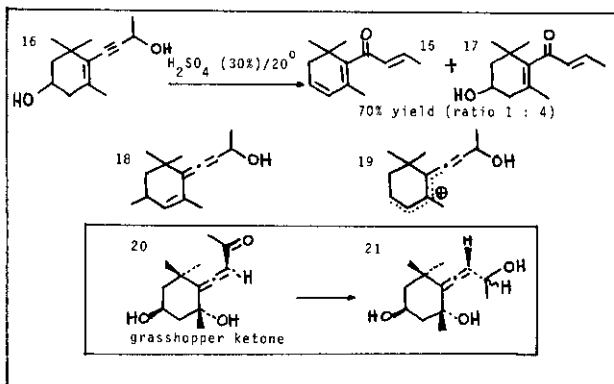


Figure 3. Biogenetic-type formation of  $\beta$ -damasconone (15) and 3-hydroxy- $\beta$ -damasconone (17)  
Ohloff, Rautenstrauch, Schulte-Elte, *Helv. 56*, 1503 (1973)

21 to be the precursor in the formation of damascenone in nature.

This presumption is supported by the fact that in 1976 Fujimori and coworkers found 3-hydroxy-7,8-dehydroionol (16) in Burley tobacco. As a precursor, one might postulate alloxanthin whose structural elements in the end group are also found in diol 16.

An analogous process involving a corresponding precursor without a hydroxyl group at the ring can be imagined for the formation of  $\beta$ -damascone in the plant (see fig. 4).

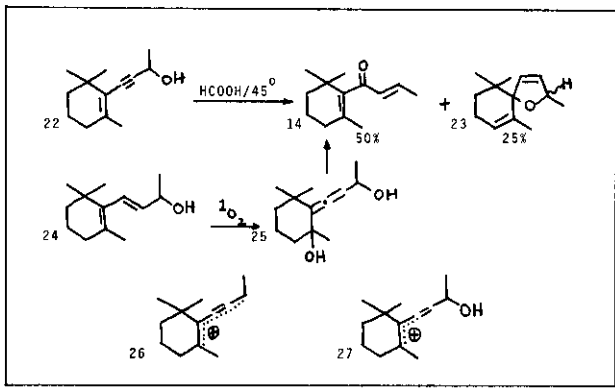


Figure 4. Biogenetic-type formation of  $\beta$ -damascone (14) and 7,8-dehydrotheaspriane (23)  
Ohloff, Rautenstrauch, Schulte-Elte, *Helv. 56*, 1503 (1973)  
Isoe, Katsumura, Sakan, *Helv. 56*, 1514 (1973)

Indeed, it was possible to convert dehydro- $\beta$ -ionol (22) into  $\beta$ -damascone (14) in an acid-catalyzed reaction, dehydrotheaspriane (23) being formed as a by-product. Isoe and coworkers were able to demonstrate the acid-catalyzed conversion of the allene diol 25 into  $\beta$ -damascone. This allene diol is a known photooxygenation production of  $\beta$ -ionol (24). The formation of  $\beta$ -damascone from the allene diol or dehydro- $\beta$ -ionol (22) presumably proceeds via the common cation 26, whereas dehydrotheaspriane might be imagined to be formed from intermediate 27.

The spiroether 23 is formed quantitatively

## Importance of minor components

when the mono-dehydration product of diol 25, shown in figure 5 as compound 29, is allowed to react with phosphoric acid in pentane.

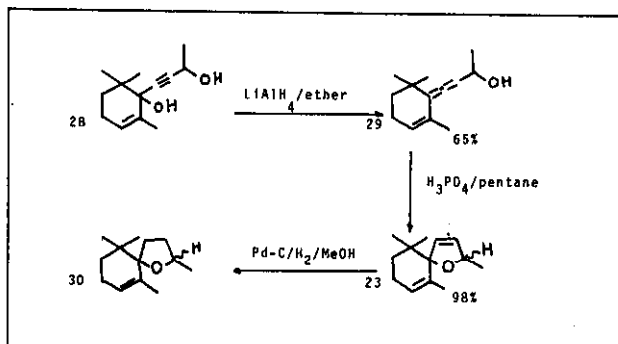


Figure 5. Formation of theaspirane (30), a minor constituent of black tea, raspberry, and yellow passion fruit

Schulte-Elte, Ohloff, 1977, unpublished

This trienol is formed from the acetylene diol (28) by reaction with lithium aluminium hydride in ether. The allene formation from  $\alpha$ -hydroxyacetylenes in a similar system has previously been reported by Weedon and co-workers. Dehydrotheaspirane, quantitatively formed by acid-catalyzed cyclization of trienol (29), can be converted by partial hydrogenation into theaspirane (30), an important trace component which I shall describe later in more detail.

In recent years, singlet oxygen has often been used for biomimetic model reactions. We have examined the photooxygenation of  $\beta$ -damascol (31) (see fig. 6). The attack of  $^1\text{O}_2$  on one or the other end of the endocyclic double bond resulted in the formation of either the epoxyketone (33) or an  $\alpha$ -hydroxy-hydroperoxide (34). The initially formed enolhydroperoxide (32) stabilizes in the form of the epoxyketone by intramolecular dehydration.  $\alpha$ -Hydroxy-hydroperoxide is a labile compound, since above room temperature or in the presence of protons even below  $0^\circ$  it undergoes fragmentation with formation of  $\alpha$ -methylene ketone (35) and crotonaldehyde (36):

When treated with HCl in dioxan at  $100^\circ$ , epoxy-dihydrodamascone (33) is converted

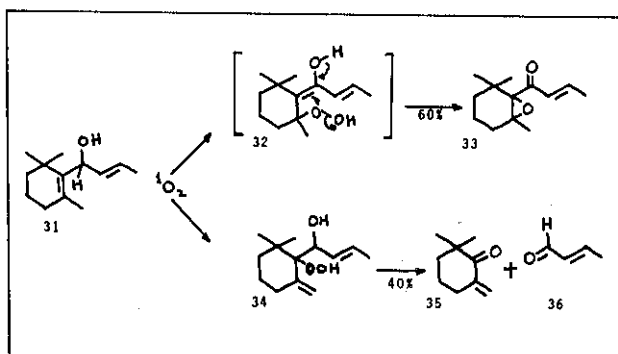


Figure 6. Dye-sensitized photooxygenation of  $\beta$ -damascol (31)

Schulte-Elte, Müller, Ohloff, Helv. 54, 1899 (1971)

quantitatively into  $\beta$ -damascenone (see fig. 7). At  $60^\circ$  the two double bond isomeric  $\alpha$ -hydroxyketones (37 and 38), which are in equilibrium, can be isolated. This method has been developed into an industrially feasible process for converting  $\beta$ -damascone via its monoepoxy compound into  $\beta$ -damascenone.

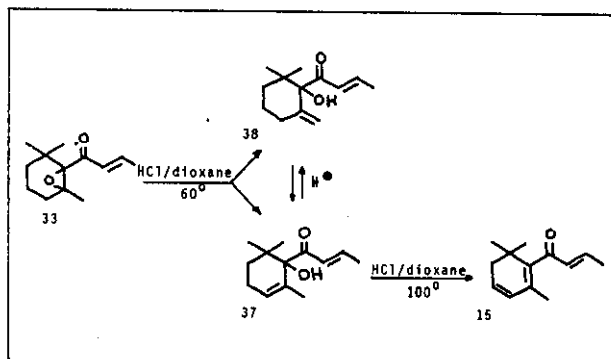


Figure 7. Acid-catalyzed formation of  $\beta$ -damascenone (15) from 5,6-epoxy-dihydro-damascone (33)

Schulte-Elte, Müller, Ohloff, Helv. 54, 1899 (1971)

### Damascones in natural products

The chemistry of the damascones, which has been developed with particular intensity at M.I.T. in Boston under the direction of Prof. Büchi and in Geneva, in our laboratories, has led to some interesting results. I should like to discuss here two aspects of this chemistry in relation to products found in nature.

At room temperature and in the presence of 85% phosphoric acid,  $\beta$ -damascone and  $\beta$ -damascenone are converted into their bicyclic isomers (39 and 42) (see fig. 8). This conversion involves a Nazarov-type cyclization reaction which proceeds by a conrotatory electrocyclic process, in the first case via the cyclopentadienyl cation (40) and in the second case via

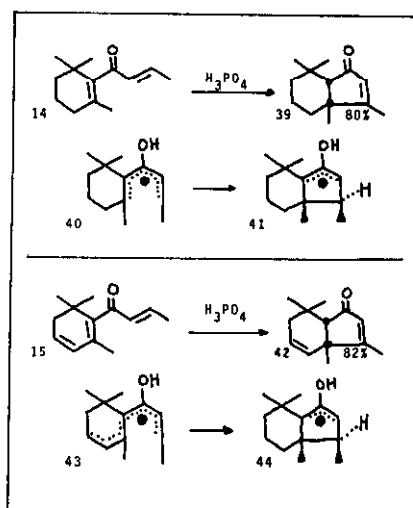


Figure 8.  $\text{H}_3\text{PO}_4$ -catalyzed cyclization of

$\beta$ -damascone (14) and  $\beta$ -damascenone (15)

Ohloff, Schulte-Elte, Demole, Helv. 54, 2913 (1971)

the cycloheptatrienyl cation (43). This conversion gained importance after we had found the bicyclic ketone as a minor constituent, in addition to  $\beta$ -damascone, in Ceylon tea.

If  $\beta$ -damascenone is subjected to the action of strong acids such as p-toluenesulfonic acid at 180°, the cyclization reaction takes another course (see fig. 9).

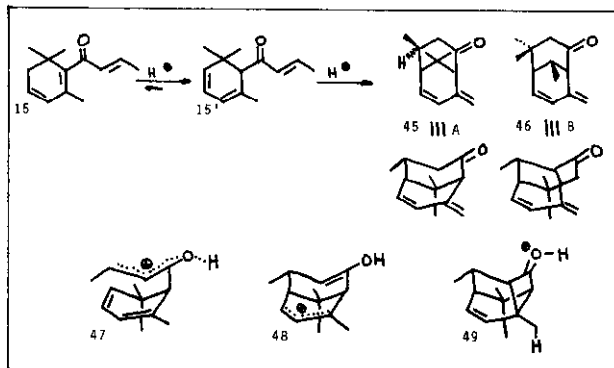


Figure 9. Biogenetic-type formation of bicyclodamasconone A and B (45 and 46 resp.) through p-TsOH-catalyzed cyclization of  $\alpha$ -damascenone (15')

Ohloff, Schulte-Elte, Demole, Helv. 54, 2913 (1971)  
Demole, Enggist, Helv. 59, 1938 (1976)

The reaction mainly results in the formation of two ketones called bicyclodamasconones A and B (45 and 46) in a yield of more than 80%. The

cyclization reaction is preceded by a [1,5]-H-shift from  $\beta$ -damascenone (15) to  $\alpha$ -damascenone (15'). The formation of the bicyclodamasconones from  $\alpha$ -damascenone can be rationalized via the hydroxyallyl cation (47) leading successively to the bicyclic isomeric cation (48) and to the protonated cyclobutanone (49). Proton elimination and cleavage of either one or the other C-C-bond of the cyclobutanone ring in 49 would furnish bicyclodamasconones A and B respectively. Both bicyclic ketones 45 and 46 were found last year as minor components in aged Virginia tobacco.

*Ionones—edulans and theaspiranes in natural products*

Let me now turn to a group of cyclic ethers related to the ionones and which have recently attracted the attention of flavor chemists. Compounds 50 to 54 belong to the edulan group and comprise tetrahydro- and hexahydrobenzopyran derivatives (see fig. 10). Compounds of the theaspirane series (55 to 58) contain a tetrahydrofuran ring in a spirocyclic arrangement. trans- and cis-edulans (50 and 51), originally named Edulan I and II, as well as their dihydro derivatives (53 and 54) were considered by Whitfield and Murray to be important trace components because of their attractive rose-like aroma. Edulan I is considered to contribute significantly to the flavor of the purple passion fruit; it is mainly present in the desired aroma of the fresh juice. The corresponding keto compound (52), which was discovered by Demole, appears to be an important component of Burley tobacco and is reported to have an oriental-type tobacco note.

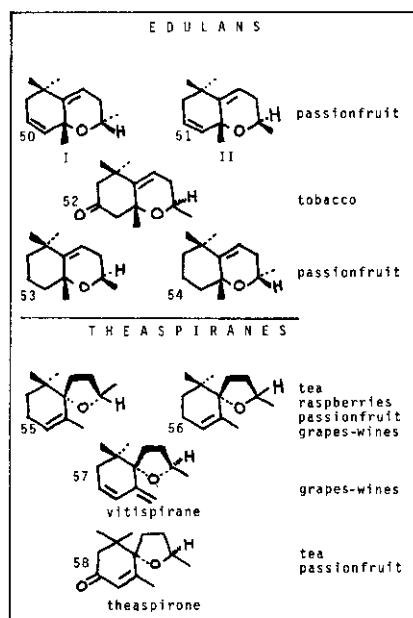


Figure 10

The diastereoisomeric theaspiranes (55 and 56) were recently found to be important minor constituents in tea, raspberries and passion fruit. Nine years ago Ina and coworkers discovered the corresponding carbonyl compound (58) in tea and called it theaspirone. Our investigations have shown that theaspirone is also present in passion fruit. Two months ago, Williams and coworkers in Australia described the identification of a dehydrotheaspirane (57) with conjugated double bonds, called vitispirane, isolated from grape juice as well as table and fortified wines.

Let us now turn to the chemistry of these interesting components which, despite their low concentration, certainly have an impact on different aromas.

In figure 11 the elegant synthesis of the diastereoisomeric edulans of Cookson and coworkers can be seen. As an easily available starting product they used the diol (59) which shows the structural elements of  $\beta$ -damascenol as well as those of dehydroionol. In the presence of Lewis acids, this diol was converted in a high yield into a mixture of the diastereoisomeric edulans, presumably via a carbonium ion (60) as an intermediate.

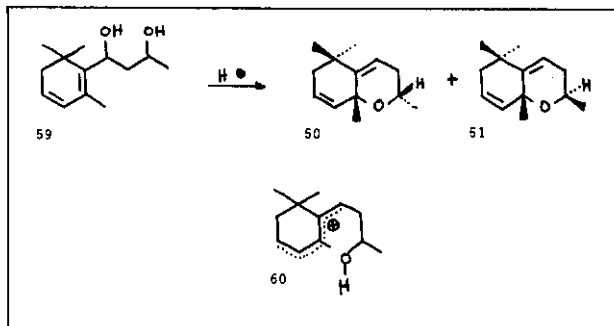


Figure 11. Acid catalyzed formation of edulans I and II (50 and 51)

Adams, Bhatnagar, Cookson, J.C.S. Perkin I, 1975, 1736

It might logically be expected that an analogous reaction starting from a diol with a hydrogenated disubstituted double bond would lead to the diastereoisomeric dihydroedulans. But, as

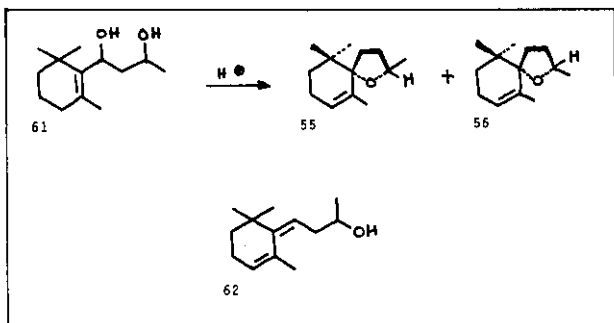


Figure 12. Acid-catalyzed formation of diastereoisomeric theaspiranes 55 and 56

Schulte-Elte, Ohloff, 1977, unpublished

indicated in figure 12, this is not so; the diastereoisomeric theaspiranes are produced exclusively. The assumption that retro-ionol (62) is formed as an intermediate by dehydration of the allylic hydroxyl group in 61 is highly probable (see fig. 13).

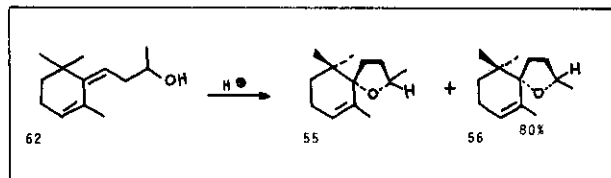


Figure 13. Acid-catalyzed formation of theaspiranes 55 and 56 from retro-ionol (62)

Schulte-Elte, Ohloff, 1977, unpublished

retro-Ionol, which can be obtained in a simple manner either from the known retro-ionone or from ionone enol acetate by reduction with sodium boron hydride, indeed furnishes in a yield of about 80% the mixture of spiroethers 55 and 56 in the presence of Lewis acids. Careful distillation allows the diastereoisomeric theaspiranes to be separated.

Figure 14 shows the epoxy and hydroxy compounds derived from the diastereoisomeric theaspiranes.\* In the presence of buffered peracetic acid the two theaspiranes are con-

verted in separate operations into the two pairs of diastereoisomeric 1,2-epoxides (63-66). Stereospecific reduction of the latter leads to the corresponding hydroxyspiroethers (67-70).

Nature appears to operate with an even higher stereospecificity than what can be achieved in the laboratory, since in tea flavor we find the main products of the epoxidation of theaspiranes exclusively.

The 1,2-epoxy derivatives (64 and 65) both develop a pleasant woody odor impression. The camphor-like undertone is more intense in the

trans-isomer. The hydroxy-compound (68) of the cis-series has a high odor effectiveness and represents the earthy note of patchouli. Hydroxytheaspirane (69) of the trans-series possesses the same odor character to a considerably lesser extent. Although the six compounds amount to hardly 1% of the volatile constituents of black tea, they still furnish an important contribution to its characteristic flavor, especially due to the extremely low threshold concentration of hydroxy-compound 68 in the range of 0.2 ppb in water.

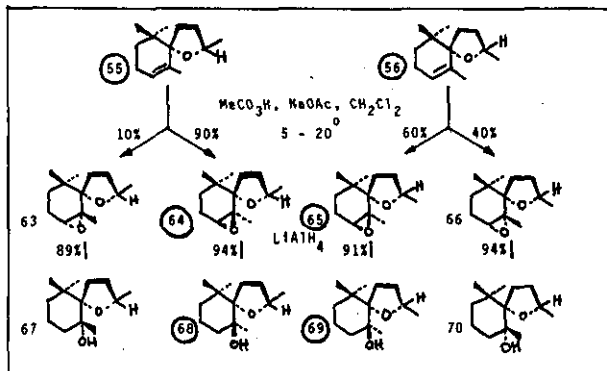


Figure 14. Formation of theaspirane derivatives, naturally occurring in black tea aroma (circled)\*  
Skorianetz, Renold, Ohloff, Schulte-Elte, U.S. Pat. 4,014,905 (1977)

**Stereoisomeric megastigmatrienones, megastigmadienone**

In a final example I should like to discuss a special group of carotenoid metabolites related to the ionone series. However, their allylic carbonyl function is not positioned in the side chain but in the ring. The four stereoisomeric megastigmatrienones (71-74) (see fig. 15) belong qualitatively and quantitatively to the key flavoring components of Burley tobacco, and as such amount to about 10% of its volatiles.

\*The formulas whose numbers are circled correspond to all compounds isolated from tea.



One of the latest discoveries in this field relates to the megastigmadienone (75). We were able to isolate this compound in trace quantities from Virginia tobacco and passion fruit. This ketone (75) possesses a very impressive fine flowery-fruity odor with a tobacco-like note.

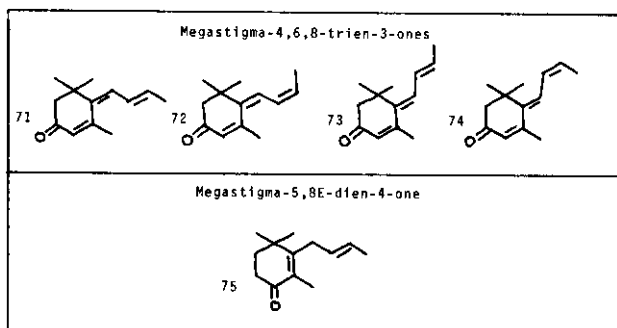


Figure 15. Key flavoring components of Burley tobacco (71-74) and a minor component of Virginia tobacco and passion fruit (75)

Aasen, Kimland, Almquist, Enzell, Acta Chem. Scand. 26, 2573 (1972)  
 Roberts, Rhode, Tobacco Science 16, 107 (1972)  
 Demole, Berthet, Helv. 55, 1866 (1972)  
 Demole, Winter, Schulte-Elte, Ohloff, unpublished

While the formation of the megastigma-trienones can be explained by the dehydration of 3-keto-ionol, a known carotenoid metabolite, direct access to the new dienone (75) is hardly

conceivable. Access to this ketone is connected with the ionone series as well as the damascone series (see fig. 16). Indeed, when treated with sulfuric acid in dioxane, the epoxides of  $\beta$ -ionol (76) and  $\beta$ -damascol (77) are both converted into megastigmadienone in about 10 to 30%.

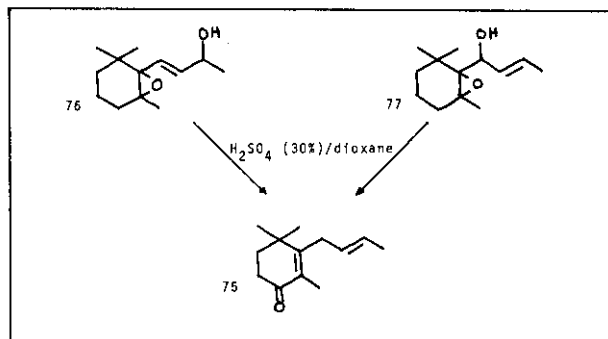


Figure 16. Acid-catalyzed transformation of 5,6-epoxides of  $\beta$ -ionol (76) and  $\beta$ -damascol (77) into megastigma-5,8E-dien-4-one (75)

In this transformation, which proceeds by dehydration, the two tert.-sec. allyldiols (78 and 80) are first formed by deprotonation at carbon atom 4 of the protonated epoxy group (see fig. 17). Both these diols exist in an acid-catalyzed equilibrium with the di-secondary allyldiols (79

and 81). Finally the di-secondary allyldiols are converted by dehydration into megastigmadienone. This reaction takes place with about 70% yield if the di-secondary allyldiols (78 and 81) are isolated, then used as the starting products. In this case, too, the deprotonation takes place at carbon atom 4 of the di-secondary allyl alcohols with elimination of their hydroxyl groups in the side chain, these alcohols forming vinylogous pinacolin systems. In the stabilization of the enol (82) acting as the common intermediate the second double bond in the side chain is not involved, so that it stays in its original position in the resulting megastigmadienone.

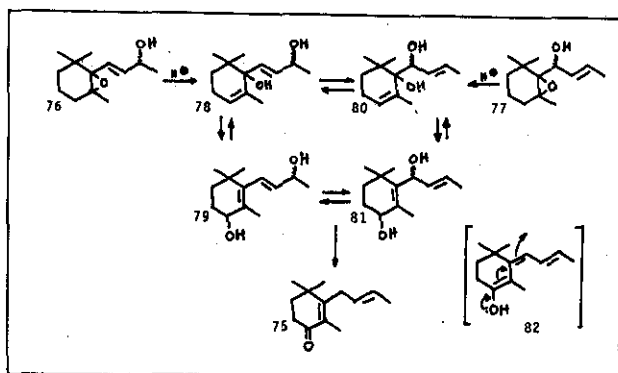


Figure 17. Course of the biogenetic-type formation of megastigma-5,8E-dien-4-one (75)

### Conclusion

This discussion was intended to demonstrate, by use of a relatively uniform group of compounds, that minor constituents can exert a considerable impact on flavors and fragrances. In order to determine which of the many compounds constituting an aroma are critical for the characteristic flavor and which are superfluous,

we have to apply the methods of sensory analysis. The organoleptic importance of a constituent can be expressed by its odor value. Threshold measurements and the systematic application of the recombination technique, coupled with profile studies and other psychophysical methods by skilled panel members, may provide a certain security in the evaluation of the qualitative and quantitative contribution of a given aroma compound.

The examples were selected to show how important knowledge of the chemistry of the precursors of minor components is. Their formation in the substrate can be stimulated by biomimetic approaches, and the application of reaction-mechanistic principles may allow predictions as to their occurrence.

My intention was to indicate what direction the analysis of flavors should follow in the future. The instrumentalist by himself will not achieve much more unless he is assisted by the biochemist, the organic chemist, and the neurophysiologist, not to mention the perfumer and the food technologist.

### Acknowledgments

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