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THERMOANALYSIS SUPPORTED BY PRINCIPAL COMPONENT ANALYSIS OF ESSENTIAL OIL SAMPLES

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ABSTRACT

As exemplified by the analysis of 38 essential oil samples from 15 different species, the usefulness of thermoanalytical techniques in quality control has been estimated. The data were processed using principal component analysis. The results indicate that this multivariate statistical method greatly assists the analyst in assessment of the quality of essential oils, particularly via thermal analysis. Some preliminary suggestions concerning selection of the thermoanalytical data are made.

INTRODUCTION

Essential oils are complex mixtures of flavor and fragrance substances originating in plants. They have found comprehensive commercial application in the pharmaceutical, food and cosmetics industries. From the chemical viewpoint the majority of essential oils are mixtures of mono- and sesquiterpenoids, containing only minor amounts of compounds belonging to other classes. A second rather large group of essential oils consists mainly of aromatic compounds, including phenols [1]. The quality and identity control of essential oils includes sensory and chemical analysis. Commonly, the oils are characterized by physical constants, such as density, refractive index and optical rotation. These investigations are increasingly supplemented by modern instrumental techniques, among which gas chromatography plays the most important role [2–4]. Chemical analysis of essential oils is generally a difficult task because of their variability, complexity and wide range of possible impurities; hence the need for extension of analytical methods available.

Thermoanalytical methods are widely used in analysis of complex mixtures of natural origin. Although they have not been used in essential oil chemistry, they are successfully applied in quality control of vegetable oils. Hannewijk and Haighton [5] showed that the DTA curves are very useful in studying the melting behavior of various stabilized and unstabilized natural

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oils. Hassel [6] used the DTA method to evaluate the relative oxidative stabilities of vegetable oils. This method offers the advantages of a short quality control time and of requiring limited equipment and operator expertise. Moreover, Nieschlag et al. [7] proved that the TG method provides data which can be useful for estimating the relative keeping qualities of different samples of the same oil. Complicated chemical composition and the heat transformations occurring under an atmosphere of air, makes classical interpretation of the results of thermal decomposition of natural products difficult. Based on analysis of the shape of the TG and DTG curves, the characteristic parameters for these products can be indicated, such as the temperatures of the beginning and end of successive mass losses [8].

The many parameters which have to be considered produce a multivariate problem. Hence this work is an attempt to resolve these problems using principal component analysis (PCA) [9,10]. PCA provides an approximation of a data matrix (**X**) in terms of the product of two small matrices **T** and **P**. These matrices capture the essential data patterns of **X**. Plotting the columns of **T** gives a picture of the dominant object patterns of **X**. Similarly, plotting the rows of **P** shows the complementary variable patterns.

Recently [10], principal component analysis was used for processing the thermoanalytical data in order to control the quality of edible fish oils. In this work we intend to initiate a systematic study on the possibility of application of thermoanalysis supported by the PCA method in evaluation of essential oils and other complex mixtures of natural origin.

EXPERIMENTAL

Samples for testing

The essential oils were prepared from the following species of plants; *Mentha piperita* L. (peppermint), *Mentha spicata* var. *crispa* (spearmint), *Salvia officinalis* L. (sage), *Salvia sclarea* L. (clary sage), *Thymus vulgaris* L. (thyme), *Majorana hortensis* Munch. (sweet marjoram), *Pinus sylvestris* L. (pine needles), *Abies alba* Mill. (silver fir), *Valeriana officinalis* L. (valerian), *Foeniculum capillaceum* Gilib. (fennel), *Anethum graveolens* L. (dill), *Carum carvi* L. (caraway), *Coriandrum sativum* L. (coriander), *Archangelica officinalis* Hoffm. (angelica root) and *Daucus carota* L. (carrot seed). All plants except *Pinus sylvestris* and *Abies alba* were cultivated on plantations in central Poland. Plant material of the two remaining species was collected from the forests of various parts of Poland. The essential oils were obtained by the steam distillation process in the Herbapol enterprise. A full list of the investigated oils is given in Table 1. Roman numbers following the oil name denote different production outputs of the same oil.

TABLE 1

Results of thermogravimetric analysis of essential oil samples

Sample	Temperatures of successive mass losses (°C)							h (mm)	T_m (°C)
	T_5	T_{30}	T_{70}	T_{80}	T_{90}	T_{95}	T_{100}		
1 Peppermint I	90	150	180	185	190	195	550	185	180
2 Peppermint II	80	135	160	165	170	175	480	185	165
3 Peppermint III	80	140	170	175	180	185	500	195	180
4 Spearmint I	95	150	175	180	185	270	535	170	175
5 Spearmint II	90	140	170	180	180	185	495	170	175
6 Sage I	85	135	175	180	190	200	475	139	175
7 Sage II	85	130	180	220	280	420	535	86	150
8 Clary sage I	100	145	200	240	320	445	540	102	160
9 Clary sage II	100	145	195	235	335	455	515	83	155
10 Thyme I	80	130	175	185	200	205	480	88	140
11 Thyme II	70	120	150	160	170	180	470	120	140
12 Dill	80	125	160	170	175	185	515	116	155
13 Fennel I	80	130	170	180	230	270	475	104	160
14 Fennel II	80	130	170	180	190	195	480	167	190
15 Fennel III	100	165	240	265	295	305	535	56	250
16 Caraway I	60	115	160	170	180	190	525	110	160
17 Caraway II	80	125	165	175	185	190	500	114	190
18 Caraway III	80	130	160	160	165	170	435	181	160
19 Caraway IV	75	120	160	165	175	180	465	117	150
20 Caraway V	90	140	190	200	250	410	540	92	175
21 Coriander I	85	135	160	165	170	180	480	197	160
22 Coriander II	85	140	165	170	175	180	480	190	170
23 Coriander III	85	140	165	170	175	180	480	180	170
24 Coriander IV	70	130	170	230	370	470	550	118	135
25 Angelica root I	60	110	135	145	180	265	525	141	135
26 Angelica root II	60	115	145	150	155	175	515	173	150
27 Angelica root III	60	110	135	145	180	265	525	157	135
28 Carrot seed I	80	140	200	220	300	465	550	75	135
29 Carrot seed II	60	115	170	180	195	205	500	82	192
30 Sweet marjoram I	70	125	155	160	170	205	515	139	150
31 Sweet marjoram II	80	130	160	165	170	205	500	167	160
32 Pine needle I	70	115	140	145	155	180	530	185	140
33 Pine needle II	50	110	145	150	155	160	520	167	155
34 Pine needle III	50	100	130	135	140	150	470	175	135
35 Pine needle IV	50	100	130	135	140	150	460	185	140
36 Silver fir	50	110	140	145	150	160	465	163	145
37 Valerian I	110	170	220	240	310	480	560	90	210
38 Valerian II	90	150	200	210	220	230	480	96	210

Testing procedure

The DTA, TG and DTG curves in thermal decomposition of essential oils were recorded using the OD-103 Derivatograph (MOM, Budapest, Hungary).

All measurements were carried out under the same conditions. A weighed sample of 200 mg of oil in a platinum crucible was heated under the atmosphere of the furnace at a temperature increase rate of 5 °C min^{-1} up to the final temperature of 700 °C. α-Al_2O_3 was used as reference material. Each curve was recorded at least three times. The temperatures of the onset and the end (T_{100}) of thermal decomposition were read from the TG and DTG curves, whereas the temperatures for 5, 30, 70, 80, 90 and 95% losses in mass (T_5, T_{30}, T_{70}, T_{80}, T_{90} and T_{95}) were read exclusively from the TG curves. DTG peak height (h) was read as the distance between the baseline of the DTG curve and the peak tip. T_m represents the temperature of maximum height of the DTG peak.

The density of the essential oils was determined using a pyknometer of 5 ml volume at a temperature of exactly 20 °C. For the determination of the refractive indices, an Abbé refractometer (Carl Zeiss, Jena, G.D.R.) was used. The measurements were made at a temperature of exactly 20 °C using a sodium discharge lamp of wavelength 589 nm. The specific optical rotation was determined using a polarimeter (Carl Zeiss, Jena, G.D.R.) equipped with a sodium discharge lamp of wavelength 589 nm.

Calculations

A data matrix **X**, consisting of $K = 1, 2 \ldots, k$ variables and $N = 1, 2 \ldots, n$ objects, was the starting point for further chemometric investigations. The variables used were the temperatures which represent respective mass losses (T_5, T_{30}, T_{70}, T_{80}, T_{90}, T_{95} and T_{100}), the DTG peak height (h) and the temperature of maximum height of the DTG peak (T_m). From the data matrix **X** its standardized version **Z** and correlation matrix **R** were calculated. The correlation matrix **R** was used as a starting matrix in principal component analysis. Principal components (PC) were determined by considering eigenvalues and associated eigenvectors. For plotting purposes only, three or two first principal component score vectors (t_1, t_2 and t_3) were used. These account for over 80% of variability in each case. In this way, nine variables were reduced to three or two principal component scores.

For evaluation of the data, software developed in our laboratory for IBM-PC compatible microcomputers was used.

RESULTS AND DISCUSSION

Thermal decomposition of essential oils

Since essential oils comprise a multicomponent mixture of organic compounds of approximate physico-chemical properties, the DTA, TG and DTG curves of their thermal decomposition are plots of the physico-chem-

ical phenomena which occur in the sample when it is heated. Thermal effects on the DTA curve result from the superposition of endo- and exo-thermic effects due to transitions of particular components. This creates great difficulties for the identification of the reaction responsible for the appearance of a definite thermal effect. On the other hand, the loss in weight on the TG (DTG) curves is the total weight loss associated with the thermal decomposition of components contained in the product examined. Thus it is not feasible to identify the weight loss associated with the decomposition of a definite component of an essential oil.

Figure 1 shows the changes in the shape of DTA, TG and DTA curves of thermal decomposition for two sage oils (6, sage I and 7, sage II) as the effect of differences in their chemical composition. It can be stated in general that the thermal decomposition of essential oils proceeds in two stages. The first stage is the distillation of the volatile oil fraction. The distillation process, which begins at ca. 30 °C, is common to all essential oils. Moreover, for some essential oils only the first stage is observed, as is evident for 6, sage oil I (Fig. 1A). This type of thermal decomposition is observed in the cases of oils 1, peppermint I, 3, peppermint III, 5, spearmint II, 6, sage I, 10, thyme I, 11, thyme II, 12, dill, 14, fennel II, 17–19, caraway II–IV, 21–23, coriander I–III, 29, carrot seed II, 36, silver fir, and 33–35, pine needle II–IV. For all the remaining essential oils two stages of thermal decomposition are observed. In the second stage the components of the

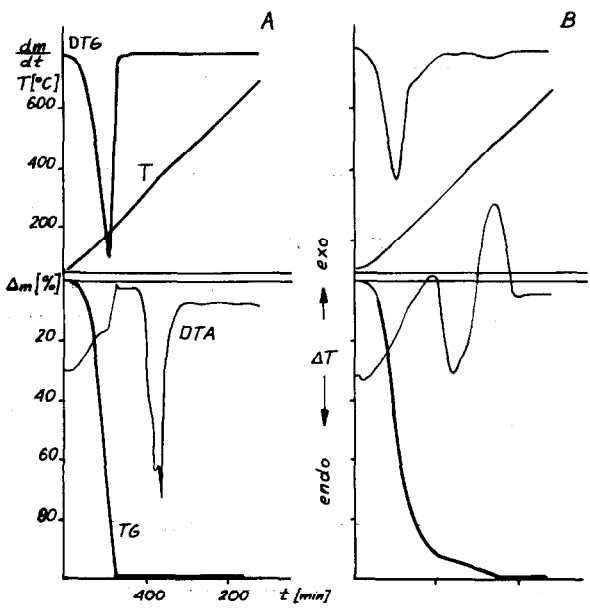

Fig. 1. DTA, TG and DTG curves of the thermal decomposition of sage oils: (A) 6, sage I and (B) 7, sage II.

essential oils which do not distil are decomposed and burnt. This is shown on the example of 7, sage II oil (see Fig. 1B). The group of essential oils characterized by two stage decomposition can be divided into two subgroups. In the first subgroup, which includes oils 4, spearmint I, 7, sage II, 8, clary sage I, 9, clary sage II, 13, fennel I, 15, fennel III, 20, caraway V, 24, coriander IV, 25, 27, angelica root I and III, 28, carrot seed I, and 37, valerian I, the second stage of thermal decomposition is distinctly marked. The second subgroup of essential oils consists of 2, peppermint II, 16, caraway I, 26, angelica root II, 30, sweet marjoram I, 31, sweet marjoram II, 32, pine needle I, and 38, valerian II. For these oils the residue burnt in the second stage is very small.

PCA differentiation of whole sample set

There is no doubt that the shape of thermoanalytical curves is conditioned by the chemical composition of the essential oil being degraded. In chemotaxonomy [11] the individual chemicals which make up essential oils are classified as secondary metabolites. The quality and quantity of secondary metabolites in plant species differ greatly, not only from species to species but within the species as well. The causal agent of such variations is little known [12]. In such a case, besides genetic factors, both the macro- and the micro-environment could be responsible for the variations in chemical composition of essential oils. Hence it cannot be expected that the principal component analysis of the thermoanalytical data will provide a reflection of the botanic system. On the other hand, thinking about essential oils as commercial products, which should be standardized, some general specification of their chemical composition is needed. The PCA examination of the whole data set may answer the question whether thermoanalytical methods can support the chemical assessment of essential oils.

The data matrix for essential oil samples is listed in Table 1. It consists of 38 objects (oil samples) and nine variables. The following sequence of positive eigenvalues was calculated: 5.47, 1.54, 1.00, 0.50, 0.33, and four values less than 0.1. The number of PC scores was estimated from the sequence of the eigenvalues. Only eigenvalues greater than 1 were considered, and in this case over 88% of the total variance is explained by this rule. In this way, nine variables were reduced to three PC scores. Figure 2 shows the PC score plot for all 38 samples. Most of the samples cluster on the center of the plot, having t_1 values between -1.9 and 0.1. Nineteen samples are clearly distinguished from the others.

To the group of outliers with $\mathbf{t}_1 < -1.9$ belong all oils from the species of *Archangelica officinalis* (25–27, angelica root I–III), *Pinus sylvestris* (32–35, pine needles I–IV) and *Abies alba* (36, silver fir). Although these species have nothing in common from the point of view of botanic classification, the main chemical constituents of their essential oils are similar. All these oils

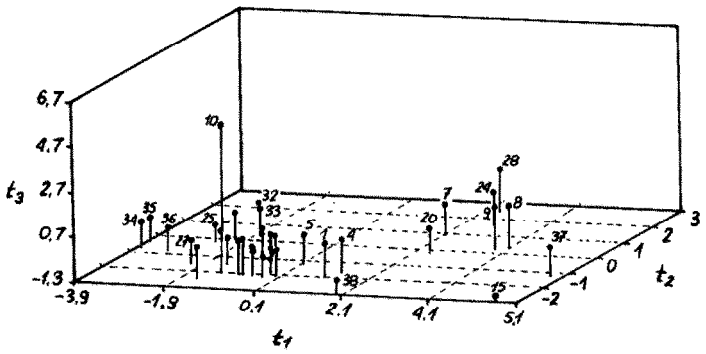

Fig. 2. Principal component scores plot derived from the thermoanalytical data for the whole sample set. Only outliers are numbered.

contain various amounts of terpenoids and sesquiterpenoids as the main fraction. Moreover, they have no single predominant compound. To the second group of outliers with $t_1 > 0.1$ belong all oils from the species of *Mentha spicata* (4 and 5, spearmint I and II), *Valeriana officinalis* (37 and 38, valerian I and II) and *Salvia sclarea* (8 and 9, clary sage I and II). Additionally, this group includes single representatives of essential oils from other species, such as 1, peppermint I, 7, sage II, 15, fennel III, 20, caraway V and 24, coriander IV. Hence, this group of outliers is not so uniform as the one with $t_1 < -1.9$. Nevertheless, this group encompasses all essential oils representative of three species, i.e. *Salvia sclarea*, *Valeriana officinalis* and *Mentha spicata*. Oils from these species contain mainly esters. In clary sage oil the predominant component is linalyl acetate (48–75%), whereas that in valerian oil is bornyl acetate. The oil obtained from *Mentha spicata* differs from other peppermint or spearmint oils because it contains, as the predominant components, linalyl acetate and linalool instead of menthol. Moreover, no other oil studied here contains esters as the main component. Hence it can be suggested that, using thermoanalytical methods, oils with high ester content can be distinguished from the others.

Most of the oils, i.e. 2, peppermint II, 3, peppermint III, 6, sage I, 10, thyme I, 11, thyme II, 12, dill, 13, fennel I, 14, fennel II, 16, caraway I, 17, caraway II, 18, caraway III, 19, caraway IV, 21, coriander I, 22, coriander II, 23, coriander III, 28, carrot seed I, 29, carrot seed II, 30, sweet marjoram I and 31, sweet marjoram II, belong to the group with $-1.9 < t_1 > 0.1$. Because these oils cluster very closely they are not numbered in Fig. 2. Notably, this group of oils also has very similar t_2 and t_3 values, with the exception of 10, thyme I which possesses a very high t_3 value. All these oils have one or two predominant components (making up over 50%) of low molecular weight, as, for example, menthol in peppermint oil or limonene and carvone in caraway oil.

As was mentioned previously, separate examples of oils possessing predominant components (i.e. 1, peppermint I, 7, sage II, 15, fennel III, 20, caraway V and 24, coriander IV) belong also to the group with $t_1 > 0.1$. The density values of the aforementioned oils are in each case higher than required by the standard [13]. Moreover, direct inspection of the TG curve shows the presence of a non-distilling fraction. Therefore it can be suggested that these oils are contaminated by non-volatile impurities.

PCA differentiation in quality assessment of essential oils

In this work more than three examples of essential oils from each of three species (i.e. *Pinus sylvestris, Carum carvi* and *Coriandrum sativum*), were studied. Principal component analysis for these oils was carried out separately and the results were compared with the standardized assessment in accordance with the Polish Pharmacopeia [13].

The data matrix for caraway oil samples consists of five objects (oil samples) and nine variables. The following sequence of positive eigenvalues was calculated: 6.12, 1.86, 0.81, 0.20, and five values less than 10^{-12}. Considering only eigenvalues greater than 1, a two PC scores model was adopted which explains over 88% of the total variance. In this way, nine variables were reduced to two PC scores. The t_1 vs. t_2 plot is shown in Fig. 3. The standard requirements for caraway oils are as follows; d_{20}^{20} 0.902–0.916, $[\alpha]_D + 70–80°$. Three oils, i.e. 16, caraway I ($d_{20}^{20} = 0.9108$, $[\alpha]_D = +73°$), 17, caraway II ($d_{20}^{20} = 0.9134$, $[\alpha]_D = +70.6°$) and 19, caraway IV

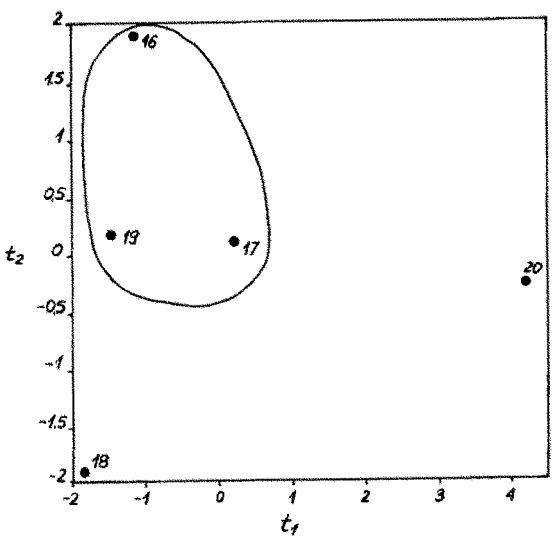

Fig. 3. Principal component scores plot derived from analysis of caraway oils. The oils which fulfil the standard are circled.

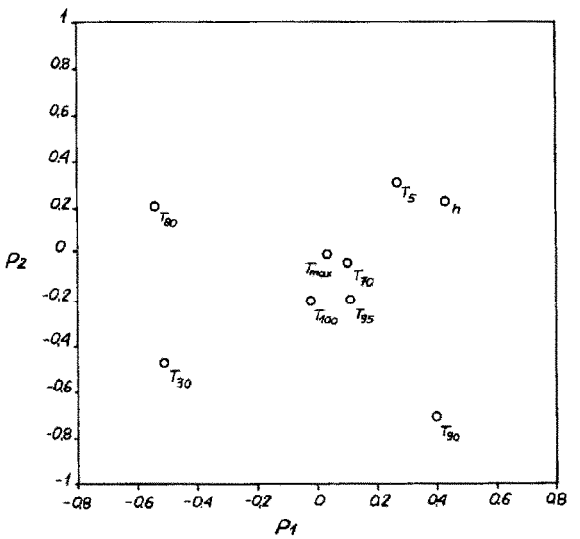

Fig. 4. Loading plot corresponding to Fig. 3.

($d_{20}^{20} = 0.9124$, $[\alpha]_D = +74.0°$) cluster in the upper left corner of the plot. As can be seen these oils are in accordance with the standard. The two remaining oils, i.e. 18, caraway III ($d_{20}^{20} = 0.8531$, $[\alpha]_D = +96.2°$) and 20, caraway V ($d_{20}^{20} = 0.9760$, $[\alpha]_D = +48.8°$), differ from the standard in both density and optical rotation values. Hence our results suggest that the quality control problem for caraway oils can be fully resolved on the basis of thermoanalytical data. To illustrate which variables are responsible for the observed behavior the loading vectors $\mathbf{p}_1$ and $\mathbf{p}_2$ were plotted. The loading plot is shown on Fig. 4. From this figure it can be seen that the variables T_{30}, T_{80}, h and T_{90} were largely responsible for the separation into groups.

The data matrix for coriander oil samples consists of four objects and nine variables. The following sequence of positive eigenvalues was calculated: 8.30, 0.69, 0.01, and six values less than 10^{-14}. The one PC score model explains 92% whereas the two PC model explains over 99% of the total variance. Thus either model can be adopted. For the sake of comparison with the other plots the two PC scores model was chosen. The $\mathbf{t}_1$ vs. $\mathbf{t}_2$ plot is shown in Fig. 5. The standard requirements for coriander oils are as follows; d_{20}^{20} 0.864–0.877, n_D^{20} 1.462–1.472, $[\alpha]_D$ + 8–13°. Three oils which are in accordance with the standard, i.e. 21, coriander I ($d_{20}^{20} = 0.8717$, $n_D^{20} = 1.4643$, $[\alpha]_D = +8.6°$), 22, coriander II ($d_{20}^{20} = 0.8715$, $n_D^{20} = 1.4639$, $[\alpha]_D = +9.2°$) and 23, coriander III ($d_{20}^{20} = 0.8708$, $n_D^{20} = 1.4644$, $[\alpha]_D = +11.4°$) are clearly distinguished on the right-hand side of the plot from 24, coriander IV ($d_{20}^{20} = 0.9766$, $n_D^{20} = 1.4731$, $[\alpha]_D = +4.8°$), which differs from the standard. Hence our results suggest that the quality control problem for coriander oils can be fully resolved on the basis of thermoanalytical data. To

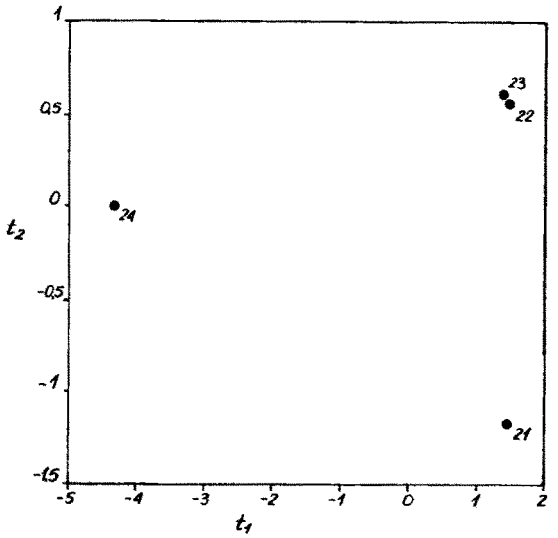

Fig. 5. Principal component scores plot derived from analysis of coriander oils.

illustrate which variables are responsible for this behavior, the loading vectors $\mathbf{p}_1$ and $\mathbf{p}_2$ were plotted. The respective loading plot is shown on Fig. 6, which suggest that the T_{30}, T_{70}, T_{80}, h and T_{95} variables were mostly responsible for the differentiation.

The data matrix for pine needle oils consists of four objects and nine variables. The following sequence of positive eigenvalues was calculated: 6.27, 2.46, 0.26, and six values less than 10^{-12}. Considering only eigenvalues

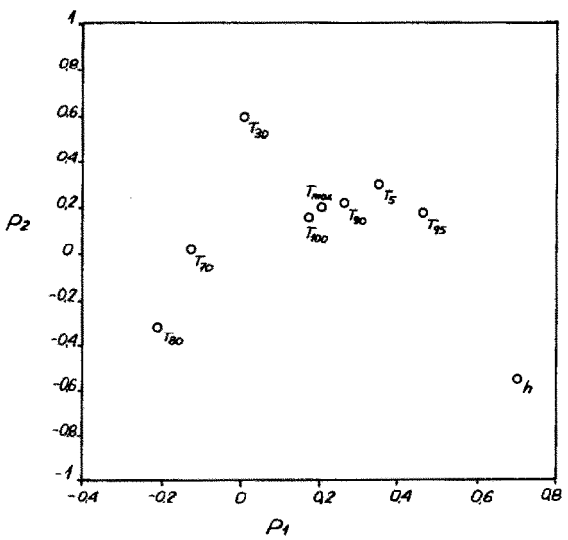

Fig. 6. Loading plot corresponding to Fig. 5.

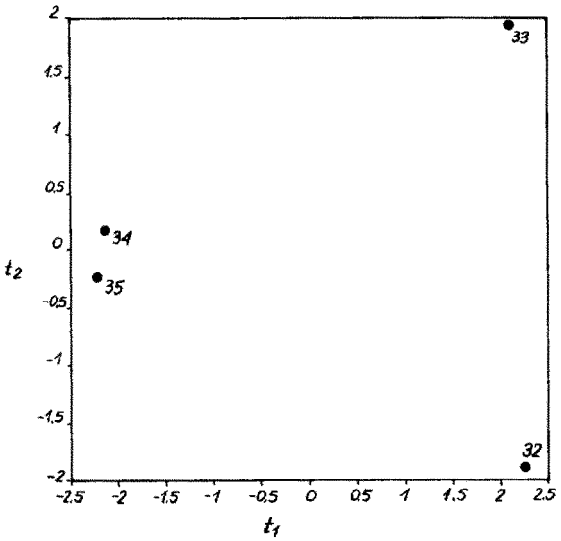

Fig. 7. Principal component scores plot derived from analysis of pine needle oils.

greater than 1, the two PC scores model was adopted, which explains over 98% of the total variance. In this way, nine variables were reduced to two PC scores. The t_1 vs. t_2 plot is shown in Fig. 7. The standard requirements for pine needle oils are as follows; d_{20}^{20} 0.861–0.881, $[\alpha]_D$ −20° to +13°. All oils investigated, i.e. 32, pine needle I ($d_{20}^{20} = 0.8674$, $[\alpha]_D = +9.2°$), 33, pine needle II ($d_{20}^{20} = 0.8670$, $[\alpha]_D = +9.6°$), 34, pine needle III ($d_{20}^{20} = 0.8691$, $[\alpha]_D = +9.6°$) and 35, pine needle IV ($d_{20}^{20} = 0.8663$, $[\alpha]_D = +8.2°$), are in

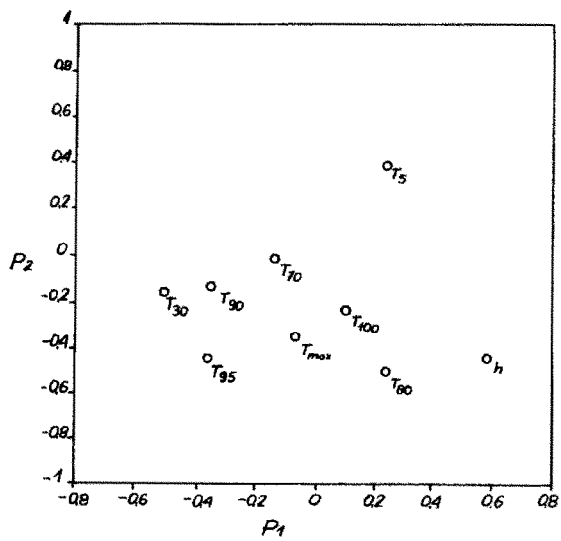

Fig. 8. Loading plot corresponding to Fig. 7.

accordance with the standard. Nevertheless, on Fig. 7 oil pairs 34, 35 and 32, 33 are well separated. Moreover the separation is not correlated with the small differences observed in d_{20}^{20} and $[\alpha]_D$ values. Thus it can be suggested that, besides differentiation comparable with that of classical methods, which is observed in the cases of caraway and coriander oils, thermoanalytical methods also reflect some other differences in quality of the oils, although in this initial work it would be premature to make a definite statement about the nature of those differences. To illustrate which variables are responsible for this the loading vectors $\mathbf{p}_1$ and $\mathbf{p}_2$ were plotted. The loading plot is shown in Fig. 8, which suggests that the variables T_{30}, T_{90}, T_{95}, h, T_{80} and T_5 impart the most important information.

CONCLUSIONS

Our preliminary results indicate that thermal analysis might be a useful supplement in the analytical determination of the identity and purity of essential oil samples. In particular, it allows the detection of non-volatile impurities which are difficult to determine using gas chromatography.

Using principal component analysis, the essential oils can be divided into three groups: oils containing terpenoids and sesquiterpenoids without a predominant compound, oils with a single predominant compound of low molecular weight, and oils with esters as the main fraction.

In quality assessment, our results demonstrate the comparable classification ability of classical and thermoanalytical methods. However, in the case of thermal analysis, some other, at present unknown, properties of essential oils can be also detected.

REFERENCES

1 K. Bauer and D. Garbe, Common Fragrances and Flavor Materials, Preparation, Properties and Uses, VCH, Weinheim, 1985, p. 112.
2 T. Adzet, R. Vila, C. Ibañez and S. Cañigural, Planta Med., 54 (1987) 369.
3 A. Cotroneo, S. Dugo, L. Favretto and L.G. Favretto, J. Chromatogr., 449 (1988) 183.
4 R. Ivanić and K. Savin, Planta Med., 30 (1976) 25.
5 J. Hannewijk and A.J. Haighton, J. Am. Oil Chem. Soc., 35 (1958) 457.
6 R.L. Hassel, J. Am. Oil Chem. Soc., 53 (1976) 179.
7 H.J. Nieschlag, J.W. Hagemann, J.A. Rothfus and D.L. Smith, Anal. Chem., 46 (1974) 2215.
8 M. Wesołowski, Fat Sci. Technol., 89 (1987) 111.
9 S. Wold, Pattern Recognition, 8 (1976) 127.
10 H. Lamparczyk and M. Wesołowski, Thermochim. Acta, 155 (1989) 327.
11 R. Hegnauer, G. Bot. Ital., 120 (1986) 15.
12 D. Vokou and N.S. Margaris, Int. J. Biometeorol., 30 (1986) 147.
13 Farmakopea Polska III (IIIrd Polish Pharmacopeia), PZWL, Warsaw, 1954.