

Application of headspace analysis in dairying

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Summary. The paper presents a brief survey of the problems concerning headspace gas chromatographic (HSGC) analysis of volatile substances as well as its application possibilities in dairying. On the basis of numerous data from literature and the author's practical experience the basic methods of direct HSGC analysis of volatile substances as well as the methods based on the preliminary concentration of volatile substances with subsequent gas chromatographic analysis of the obtained concentrates have been described.

The paper includes schemes of the individual methods and results obtained (volatile substances in milk, milk products, packing materials, etc.).

The paper is of informative character. It should be used as the basic information, especially for the staff of dairy research and inspection.

Composition and quantity of volatile substances are determined quite frequently using analysis of the vapour phase or its condensate present over the sample. This method is generally known as a headspace analysis [19].

From the physical aspect the headspace analysis can be characterized as a method for getting information on the content of volatile substances in condensed material, indirectly, using the analysis of co-existing gaseous phase [20].

Headspace analysis means a remarkable progress and development particularly in connection with gas chromatography (HSGC). In case of headspace analysis high sensitivity, GC resolving power and selectivity of chromatographic detectors are emphasized.

HSGC is employed where determination of trace quantities of volatile substances is required, which cannot be analysed by any other method without changing their original structure and proportionality. It has found extensive application possibilities in biology, chemistry, judicial chemistry, agriculture,

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foodstuff industry, environment protection, etc. HSGC has significantly contributed to the study of aroma. Therefore, it has been given a symbolical term the "gas chromatographic nose" [35].

In analytical practice there are known numerous versions of headspace analysis. They can be classified into the following two basic groups [20, 24]:

— methods employing the state of a thermodynamic equilibrium between the sample and the vapour phase, so-called direct HSGC,

— and methods where the above-mentioned equilibrium is not reached, so-called open system.

Drozď and Novák [20] specify the HSGC methods in more detail and classify them into groups as follows:

1. Direct analysis of the vapour phase over the sample (headspace).

2. Methods based on the concentration of the headspace substances with subsequent gas chromatographic analysis of the concentrate obtained. The concentrate can be collected as follows: a) in empty freezing columns; b) on different sorbents.

3. Release (extraction) of volatile substances from the sample by a stream of gas. In this case the following modifications can be applied: a) direct analysis of the obtained gaseous extract of volatile substances; b) concentration of obtained volatile substances and their subsequent analysis; c) release and collection of volatile substances from the sample is carried out in a closed system.

There is a number of summary works dealing with the problems of HSGC from different aspects [18, 20, 24, 35, 38, 55, 63, 77].

Because of its universality, simplicity, exactness and other advantages HSGC has found application possibilities in dairying, particularly in the study of characteristic and defect flavour of milk, milk products, starter cultures, in the inspection of the quality of fermented milk products in particular, in the study of the effects of packing materials on the quality of packed milk products, etc.

Attention is also paid to a survey of some applications of HSGC in dairying with particular focus on many year' experience gained with the method in question in our department.

Application of direct HSGC in dairying

Direct HSGC analysis is widely used in dairying. The most frequently used apparatus employed by us for the direct HSGC analysis of volatile substances is shown in Fig. 1. The vessel — in our case a 25 ml volumetric flask with a cut

neck at the measuring mark — is hermetically closed with a rubber stopper. In the flask there is the sample to be analysed (5 g of liquid milk product + 3 g Na_2SO_4). After tempering the flask for 10 min at 60°C , 1 ml portion is taken from the headspace and directly injected into the gas chromatograph.

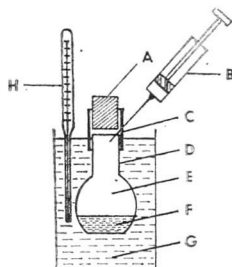


Fig. 1. An apparatus for HSGC analysis of volatile substances [63]. A — glass closure, B — syringe, C — rubber tube, D — volumetric flask (25 ml), E — headspace, F — sample + Na_2SO_4 (anhydrous), G — water bath, H — thermometer.

The volatile substances of the analysed portion are determined by gas chromatography.

Quantitative interpretation of the method described is as follows [35]:

$$\begin{aligned} F_i &= P_i c_i \\ P_i &= P_i^0 x_i \gamma_i \\ F_i &= c_i P_i^0 \gamma_i x_i \end{aligned}$$

where F_i is peak area for the volatile substance i in the chromatogram, c_i — detector characteristic for the i constituent, P_i^0 — vapour pressure of the pure constituent i , P_i — partial pressure for the i constituent, γ_i — activity coefficient for the i constituent, x_i — molar fraction of the i constituent in the sample.

A study dealing with operational conditions of the above-mentioned method with respect to the analysis of easily volatile substances in milk products has been published by Palo and Kátra [63].

Bassette et al. [9, 11] applied as soon as in 1962 and 1963 the direct HSGC technique designed originally for the analysis of volatile components in diluted aqueous solutions [15] for the volatile substances in mil. The sample of milk to be analysed was placed in a serum vessel (5 ml of milk + 2.5 g Na_2SO_4), and after tempering 1 ml of headspace volatile substances was injected into the gas chromatograph. In milk of different quality the presence of acetaldehyde, propionaldehyde, acetone and 2-butanone was determined in this way: it was possible to identify them even at a concentration of 0.1 ppm. In the development of the method they also used experience of other authors with HSGC analysis of volatile substances of aroma of raspberries [82], cabbage [4] and other foods [43, 78].

Using the above-mentioned method Bassette and Claydon [5] analysed successfully also volatile substances present in milk cultures with the aim of their quick differentiation, or in individual species of *Escherichia coli* and *Aerobacter*

cultivated in milk [6, 13]. In this way they studied also the content of methylsulphide with regard to the flavour of milk [22, 76, 79]. They also employed the above-mentioned method in testing the volatile substances (acetone, methylsulphide, ethanol and 2-butanone) in blood, milk and stale of dairy cows fed different fodder [12, 21, 41]. It was successfully employed also for the study of changes in chemical and organoleptic properties of concentrated milk during storing [40]. Reliability of the above-mentioned method for the analysis of volatile substances in biological solutions was verified by Bassette and Ward [7]. They proved that reliable results can be achieved when keeping the principles of the method.

In our conditions direct HSGC analysis of volatile substances in milk products was used for the first time in 1965. The original method [9, 11] was modified for the analysis of cheese [60]: In a 20 ml vessel (penicillin vial) 5 ml of 40% aqueous solution (homogenate) of cheese was added to 3 g Na_2SO_4 . Fur-

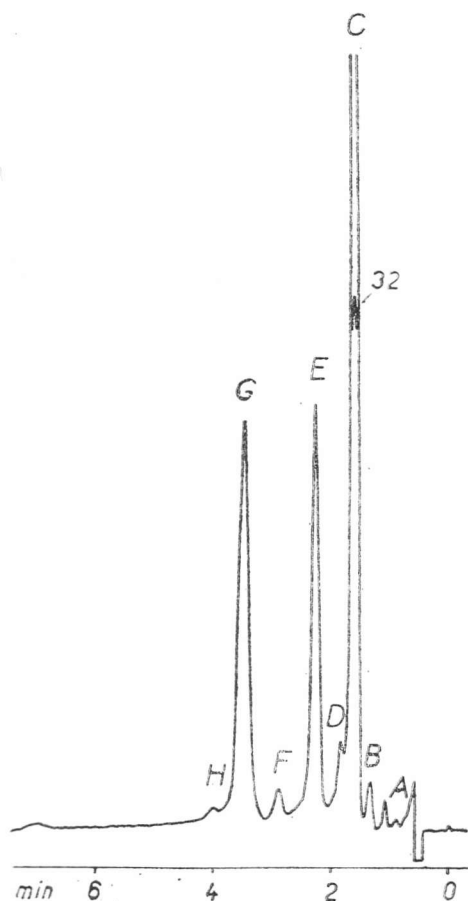


Fig. 2. GC spectrum of volatile substances of typical Slovak sheep cheese (bryndza) in consumers packing, stored for four days at 20°C [60]. Analysis conditions: Apparatus: Fractovap Model C (Carlo Erba, Milan) with FID; Column: glass, 16 m \times 4 mm packed with 13% of Di-(2)-ethylhexylsebacate and 2% of polyethylene glycol 400 on Celit 545 (0.125—0.150); Temperature: -88°C ; Carrier gas: nitrogen (3×10^{-2} MPa); Dosing: 1 ml (headspace). A — acetaldehyde, B — acetone, C — diacetyl, ethanol, D — isopropanol, isobutyraldehyde, E — 2-butanone, butyraldehyde, F — propanol, G — sec. butanol, H — iso-valeraldehyde.

ther procedure remained unchanged (see the description of the technique in the Introduction). A GC spectrum of volatile substances of sheep cheese (bryndza) obtained in this way is shown in Fig. 2.

Later, the vessel (penicillin vial) was replaced by a modified 25 ml volumetric flask (see Fig. 1). This resulted also in a higher precision of the method. Reproducibility of the method is documented in Table 1.

Table 1. Reproducibility of direct HSGC method — GC spectrum of volatile substances of cream culture [63]

No. of sample	Peaks [mm]			
	Acetone	Ethanol	2-Butanone	Diacetyl
1	65	71	12	5,5
2	74,5	68,5	12,5	5
3	69	61	12,5	5
4	75	82	13	6
5	74	81	13	6
6	73	83	13	6
7	71	73	12	5
8	68	69	12,5	5
9	72	79	14	5,5
10	74	82	13	5,5
\bar{x}	72,3	75,8	12,75	5,45
s_x	1,78	3,04	0.726	0.685
$V[\%]$	2.46	4.0	5.7	12.55

By this method volatile substances were analysed in connection with the study of the flavour of sheep cheese [67], aroma profile of stored (barrel) sheep cheese [54, 68], "bryndza" [57] and Olomouc cake-cheese [65]. It was used also in the extensive study of the character of volatile substances present in the individual types of Czechoslovak [59] and Italian cheeses [16, 53].

Direct HSGC technique was used for the analysis of volatile substances — for checking the quality of cream culture [37, 57, 66], in the study of yoghurt ageing [25, 26, 34] and kefir ageing [27].

It was found useful also in the investigations of the origin of defect flavour of milk [55]. In Fig. 3 there is a GC profile of volatile substances of milk without odour and with shed, cow odour.

Greig and Manning [28] used the direct HSGC technique in the analysis of changes in the content of acetaldehyde in pasteurized milk during storing.

The method in question was employed also for identification of volatile substances in milk products analysed by gas chromatography. The individual

volatile substances of certain functional group present, for instance in kefir, were masked by addition of a special reagent into the analysed sample. They were studied using the direct HSGC technique [51, 52].

A special method of direct HSGC analysis of volatile substances in hard

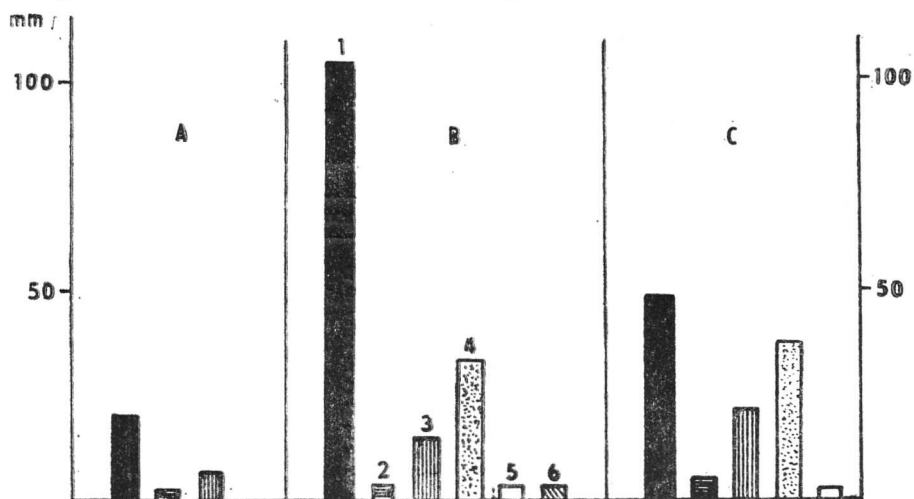


Fig. 3. Representation of volatile substances of milk without aroma and with different intensities of cow aroma expressed in terms of peak heights in chromatogram [55]. A — without aroma, B — intensive aroma, C — slight aroma. 1 — acetone, 2 — ethanol, 3 — 2-butanone, 4 — isopropanol, 5 — acetaldehyde, 6 — non-identified.

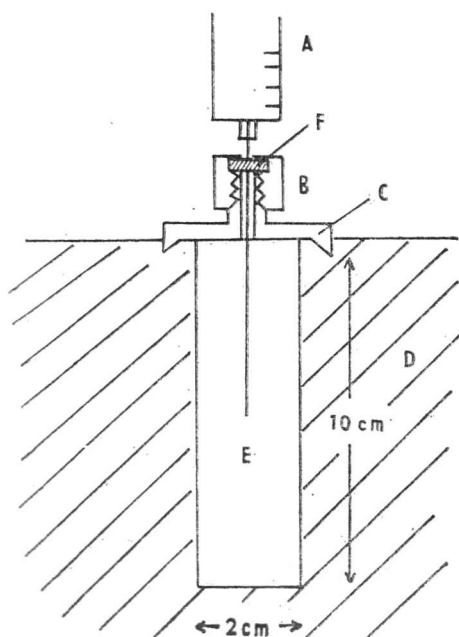


Fig. 4. Schematic illustration of the isolation of volatile substances from a drilled hole in cheese according to Manning and Moor [44]. A — syringe, B — a nut for fixing the rubber septum F, C — enclosure of the drilled hole in cheese (from stainless steel), D — cheese, E — hole in cheese.

cheese has been described by Manning and Moore [44]. To the cylindrical hole in the cheese a septum was applied hermetically (see Fig. 4). After reaching the equilibrium between the cheese and volatile substances released in the hole, 1 ml of headspace vapours was taken through the septum. This sample was directly injected into the gas chromatograph. In comparison with the already-mentioned method — analysis of volatile substances from a cheese solution [60] — a poorer GC spectrum of volatile substances was obtained by the Manning method (see Fig. 5).

Direct HSGC was employed also in the study of permeability of packing materials for volatile substances present in packed milk products [57, 69].

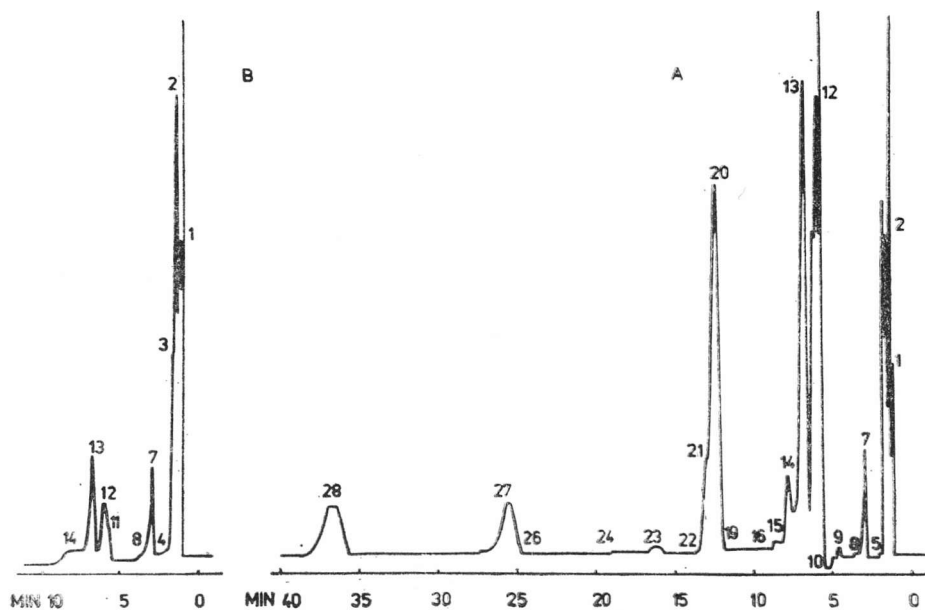


Fig. 5. GC spectrum of volatile substances of Roquefort-type cheese called Niva. A — headspace method using cheese solution [60], B — headspace method using hole in the cheese [44].

Permeability was determined using a simple glass apparatus made of ground-in glass. It was used for releasing and transfer of volatile substances from the product through the packing material. The apparatus consisted of a two-section vessel; in the lower one the cheese sample was placed. The cheese was in direct contact with the packing material tested. The bottom section of the vessel was hermetically closed by the upper section having a volume of 10 ml. The tested packing material separated both sections of the vessel. The volatile substances of cheese, passing through the packing material into the upper section of the vessel were then introduced into the gas chromatograph.

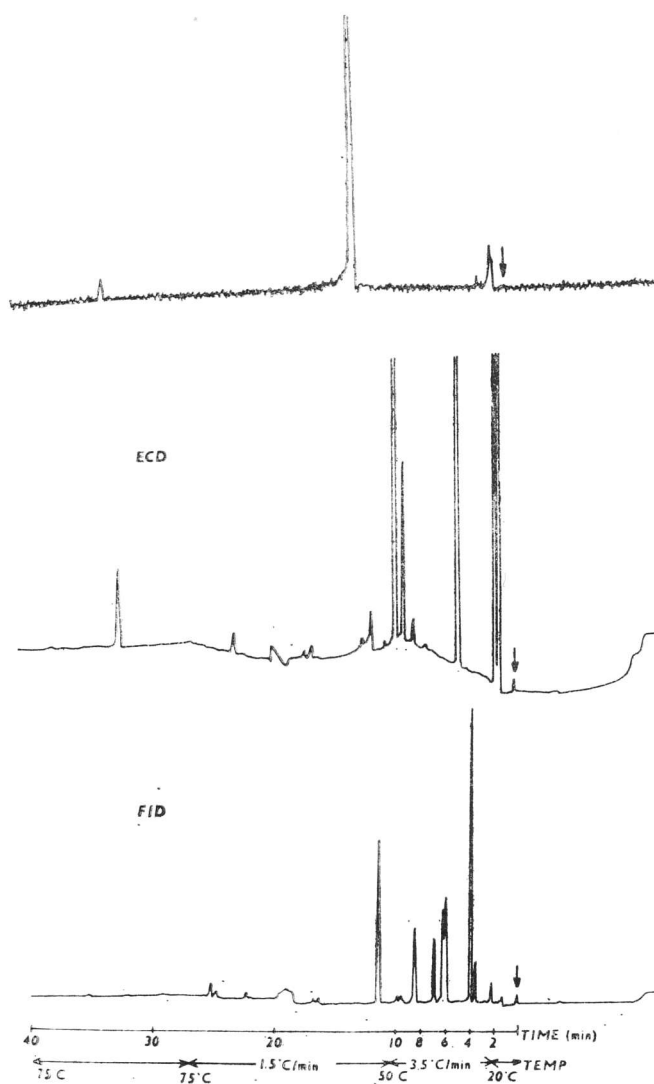


Fig. 6. Chromatogram of volatile substances in Olomouc cake-cheese obtained by direct HSGC using an apparatus with automatic dosing and multidetection equipment [56]. Apparatus: Fractovap 2 900 ser. with HS sampler model 250 equipped with FID, ECD and FPD (Carlo Erba, Milan); Column: gas capillary WWCOT, 40 m, diameter 0.6 mm, impregnated with Carbowax 400; Carrier gas: hydrogen (4.5 ml min^{-1}); Temperatures: column PT, detectors 200°C , injector 150°C , sample 60°C (40% aqueous solution of cheese), syringe 85°C ; Dosing: 2 ml (splitter 1 : 4); Sample heating: 10 min; Auxiliary gas: nitrogen.

The direct HSGC has become the subject of interest of several manufacturers of gas chromatographs. The principle of this technique was used for the development of several types of automatic analysers of volatile substances. For instance, Perkin — Elmer has been manufacturing such devices for 10 years. It consists of an electro-pneumatic dosing device which withdraws automatically, by means of a syringe, the released volatile substances from the thermostat sample reservoir (e.g. for 50 samples in penicillin vials), and introduces them into the gas chromatograph. More detailed information on the above-mentioned system is available in the corresponding prospectuses, which — together with the description of application possibilities — presents, e.g. Kolb [38].

The Italian company Carlo Erba Strumentazione, Milan, also manufactures an automatic system for direct HSGC analysis of volatile substances [83]. This is equipped with an automatic dosing device which enables to analyse as many as 40 samples (in penicillin vials) tempered to temperatures of 30—120°C. It is equipped with an electro-pneumatic system for injecting the sample by means of a gas-tight temperable syringe with controllable volume of 0.1 to 2.5 ml. The Company offers the equipment together with a gas chromatograph (capillary analytical columns) equipped with a multidetection system (e.g.

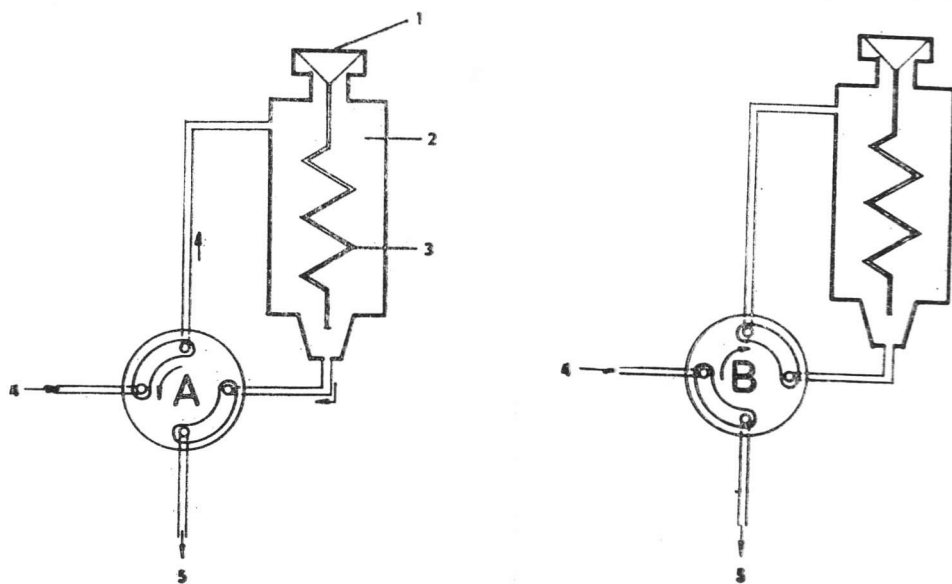


Fig. 7. Scheme of an apparatus for HSGC analysis of volatile substances in packing materials [23]. A — carrier gas flow rate through the chamber (when applying volatile substances on the anal. column), B — tempering of packing material in the chamber (release of volatile substances from packing material). 1 — septum, 2 — chamber, 3 — metallic clamps (spiral) for the packing material sample, 4 — carrier gas inlet, 5 — carrier gas outlet (application to analytical column).

FID, ECD and FPF). The mentioned system is automatic and fully reliable. An analysis of volatile substances in Olomouc cake-cheese, using the above-mentioned apparatus, is demonstrated in Fig. 6 [56].

To the direct HSGC there also belongs the method employing an apparatus developed by Carlo Erba Strumentazione, Milan [23]. It is designed for the determination of volatile substances in packing materials. It consists of a tempered „extraction“ chamber connected to the injector of a gas chromatograph. A rolled-up piece of packing material is placed in the chamber which is then tempered. Volatile substances released from the packing material are then transported in a carrier gas stream directly to the analytical column of the chromatograph (see Fig. 7). In Fig. 8 there is a chromatogram of the residue of solvents present in varnished cellophane. By this method also the volatile substances present in the packing materials used in the Czechoslovak dairy industry [70, 72] were determined, and the technique was used in the study of the effect of PE on the quality of consumable types of milk and cream [73—75].

The above-mentioned method was tested also with respect to the possibility of determining the volatile substances directly in the milk product [71]. In this case a sample of the milk product in a small glass tube was placed in the chamber instead of the packing material roll. Reliability of the method expressed in terms of yields is shown in Table 2.

Methods based on the concentration of headspace substances with subsequent gas chromatographic analysis

Among such methods also the HSGC analysis of the distillate of volatile substances of some milk products can be included. Analysis of the distillate is

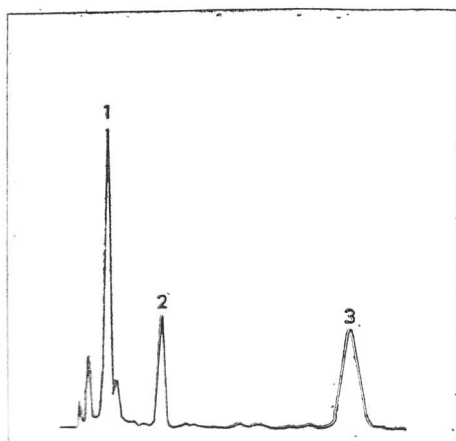


Fig. 8. Chromatogram of solvent residues in a sample of varnished cellophane [83]. Sample: varnished cellophane 0.25 dm²; Extraction conditions: 100°C/20 min; Total solvent residue: 9.9 mg m⁻². 1 — ethyl acetate + tetrahydrofurane, 2 — toluene, 3 — cellosolve.

Table 2. Yields of the method [71]

Compound Milk product	Acetaldehyde			Acetone			Ethanol		
	Added [mg]	Deter- mined [mg]	Yield [%]	Added [mg]	Deter- mined [mg]	Yield [%]	Added [mg]	Deter- mined [mg]	Yield [%]
Milk	0.261	0.166	63.6	0.337	0.281	76.4	0.422	0.378	89.7
	0.348	0.226	65.2	0.450	0.378	84.2	0.663	0.620	97.9
	0.522	0.378	72.4	0.562	0.484	86.1	0.949	0.887	93.5
	0.969	0.514	73.9	0.787	0.681	86.6	1.266	1.225	96.8
Yoghourt	0.126	0.910	72.2	0.450	0.363	80.1	0.605	0.599	99.0
	0.348	0.244	70.1	0.563	0.469	83.3	0.949	0.917	96.7
	0.522	0.333	63.8	0.675	0.551	81.7	1.110	1.044	94.1
	0.696	0.476	68.5	0.788	0.665	84.5	1.433	1.437	99.6
Cheese	0.261	0.147	56.2	0.337	0.269	79.6	0.474	0.424	89.3
	0.435	0.251	57.8	0.563	0.424	75.3	0.633	0.583	92.2
	0.609	0.337	55.4	0.675	0.552	77.3	0.791	0.713	90.1
	0.783	0.463	59.1	0.788	0.616	78.2	0.949	0.869	91.6

performed similarly as that of the direct HSGC technique mentioned in case of milk [9, 11]. Such a simple procedure of the concentration of headspace substances with the use of steam distillation in a micro-Kjeldahl apparatus and subsequent gas chromatographic analysis of the distillate by direct HSGC technique was described by Bassette and Ward [8]. Thus, volatile substances were determined in milk at ppb concentrations.

Similarly, the concentration of volatile substances from the cheese fat was found successful, providing application of high-vacuum distillation. In such a case volatile substances were entrapped in the freezing column. From there the volatile substances, after the column heating, were introduced into the gas chromatograph [46, 61]. Volatile substances of fat of stored sheep cheese, determined in this way, are shown in Fig. 9.

As early as in 1962 Badings and Galesloot [3] concentrated the volatile substances of cream culture, yoghurt and butter by a 10 min lasting application of nitrogen flow through 100 ml of liquid sample at 20°C, or 125 g of melted butter (37°C). The volatile substances were trapped into an U-shaped tube cooled with liquid nitrogen (cold trap). This was then connected to the injector of a gas chromatograph (heated to 40°C) and the released volatile substances were introduced to the column in the carrier gas flow.

A similar method was described by Day and Morgan [17]: 5 to 10 ml of a sample of milk or liquid milk product is placed in a closed vessel. After heating the content to 60°C, the nitrogen-released volatile substances condense in an U-shaped pre-column, cooled with dry ice. The volatile substances are transported from the pre-column — after its heating — by carrier gas into the

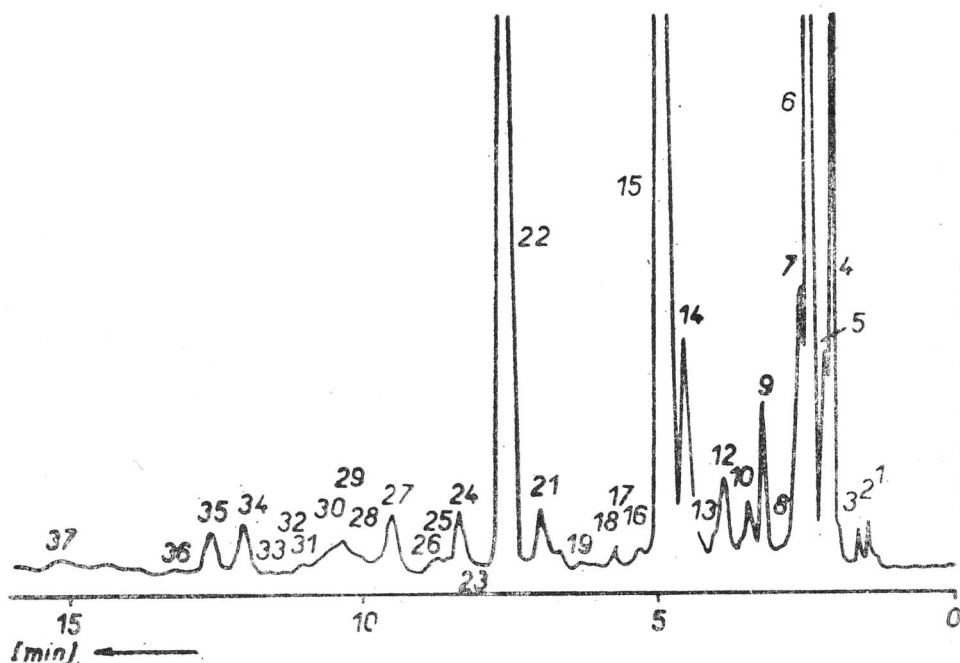


Fig. 9. GC spectrum of volatile substances separated from the fat of stored sheep cheese by high-vacuum distillation — HSGC distillate [61]. Operating conditions: distillation of volatile substances from fat of cheese at 40°C and at reduced pressure 0.4 Pa; Apparatus: Fractovap Model D (Carlo Erba, Milan) with flame-ionization detectors (FID) (dual column system); Column: glass (1.60 m × 4 mm) packed with Chromosorb W (acid washed) 60—80 mesh impregnated with 10% of Octoil S; Temperature: (8°C/min) 40 to 140°C; Carrier gas: nitrogen; Dosing: 1 ml by heating the released volatile substances (60°C — 10 min). 3 — acetaldehyde, 6 — ethyl formate, 7 — ethyl acetate, 8 — acetone, 9 — ethyl alcohol, butyraldehyde, 10 — i-propyl alcohol (?), 12 — diacetyl, 13 — n-propyl alcohol, 14 — 2-butanone, 15 — sec. butylalcohol, 18 — ethyl propionate, 20 — 2-pentanone, propyl acetate, 31 — 2-heptanone, 37 — i-butyric acid.

gas chromatograph. This method ensures good reproducibility of the results (see Table 3).

The above-mentioned method has found wide application possibilities in dairying. For instance, it was used for analysis of volatile substances as components of aroma of dairy cultures *S. lactis* var. *multigenes* [47]; conversion of acetaldehyde to ethanol by dairy streptococci and leuconostocs [14], etc. was studied.

The method using a syringe instead of the freezing column to trap the released volatile substances of the sample (see Fig. 10) can be considered a certain modification of the above-mentioned method. After the syringe has been heated, the released volatile substances are introduced into the gas chromatograph [42, 80].

Table 3. Concentration [ppm] of compounds identified in 10 samples of Cheddar cheese [15]

Peak in chromatogram No.	Compound	1	2	3	4	5	6	7	8	9	10
2	Acetaldehyde	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.2	0.1	0.1
4	Acetone	0.3	0.1	0.2	0.4	0.5	0.7	0.1	0.1	0.5	0.3
5	Ethyl acetate	0.4	0.6	0.3	0.2	—	—	—	—	—	—
7	Ethanol	450.0	140.0	5.5	35.5	62.0	27.0	23.0	620.0	11.0	35.0
8	2-Butanone	0.4	0.1	0.1	0.1	0.1	11.0	2.7	—	0.1	19.0
9	2-Butanol	0.9	34.0	0.1	0.2	—	1.1	36.0	1.1	0.3	30.0
10	n-Propanol	—	11.0	—	1.9	—	1.0	11.0	0.5	—	7.6
11	Ethyl butyrate	—	—	—	—	1.2	—	—	0.2	—	—

In the study of the origin of fruit aroma of milk, the automated headspace concentration of volatile substances (ethyl esters of lower fatty acids) was used directly connected to the capillary analytical column of the gas chromatograph [81].

Hrivňák et al. [36] have described a technique for concentrating the volatile substances from cheese into a capillary pre-column (see Fig. 11). For instance, cheese fat mixed with Chromosorb W is treated with nitrogen and the released components are trapped — frozen in the capillary pre-column. The pre-column is then connected to the capillary analytical column, the analytical capillary column is permanently tempered. Connection of the pre-column requires only to stop the carrier gas supply. Dosing of the volatile substances from the pre-

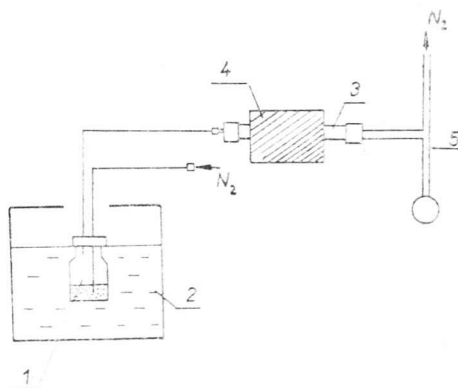


Fig. 10. Scheme of an apparatus for separation and concentration of volatile substances by freezing in a syringe [80]. 1 — vessel with the sample, 2 — water bath (60°C), 3 — syringe packed in dry ice 4, 5 — flow meter.

column is made possible such that the carrier is connected to the pre-column and this is tempered to 60°C. The released volatile substances are analysed by gas chromatography. A chromatogram of separation of volatile substances from 3 µl of fat of the Niva cheese, using the above-mentioned method and a capillary with PEG 1500, involved as many as 150 symmetric peaks. A similar method was also used by Grob and Grob [30], which differed from the former

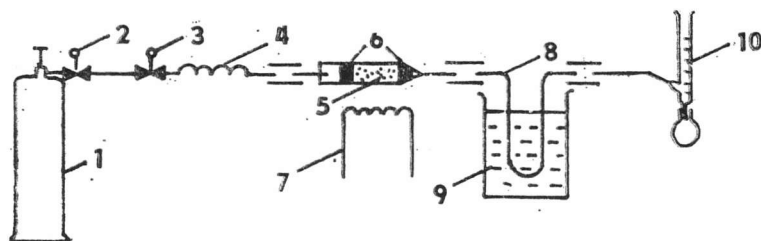


Fig. 11. Scheme of an apparatus for separation and concentration of volatile substances from fat of cheese into the capillary pre-column [36]. 1 — reserve flask with nitrogen, 2, 3 — pressure reducing valves, 4 — resisting capillary column, 5 — column packed with fat of cheese mixed with Chromosorb W, 6 — sealing (glass wool), 7 — heating coil, 8 — capillary pre-column, 9 — vessel with freezing mixture, 10 — flow meter.

one in that the employed pre-columns were longer and were placed in the chromatograph thermostat. However, in this case the apparatus had to be stopped when connecting the pre-column to the analytical column.

In a very similar way as described by Hrivňák et al. [36] also Arnold [1] and Arnold and Barnhard [2] analysed volatile substances from the fat of milk products. However, he froze the volatile substances directly to the analytical capillary column. By its intensive heating the released components were directly analysed by GC.

An advantageous modification of the above-mentioned methods [30, 36] is the substitution of the freezing U-shaped pre-column by a short capillary. This is then specially connected to the capillary analytical column. The temperature of the injection section of the apparatus and carrier gas release the volatile substances which are then analysed using GC [62]. It is not necessary to stop the apparatus or to stop the carrier gas flow, which simplifies the whole analytical operation and improves GC separation of volatile substances. The system for separation and concentration of volatile substances and their introduction from the freezing capillary to the analytical column is illustrated in Figs. 12 and 13.

Besides the freezing pre-columns for trapping the released headspace volatile substances also short columns filled with a sorbent are employed successfully. These columns can be cooled in some cases as well. The trapped volatile substances are then thermally desorbed and applied to the gas chromatograph

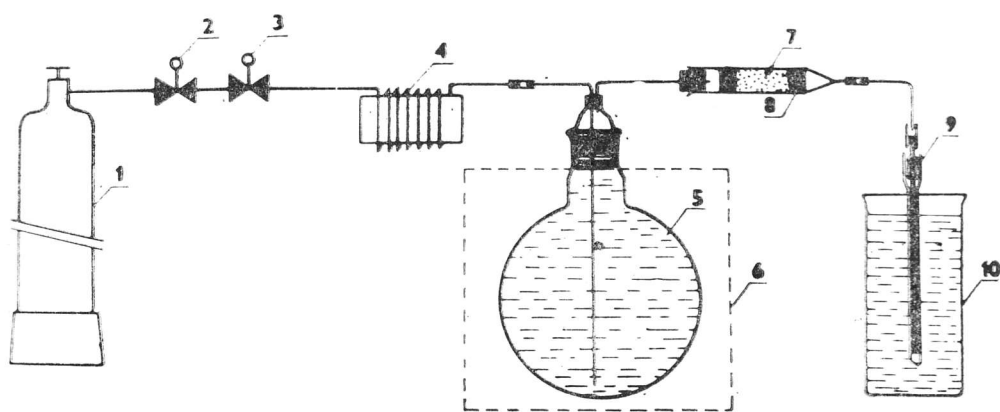


Fig. 12. Scheme of an apparatus for separation and concentration of volatile substances from milk [62]. 1 — reserve flask with nitrogen, 2, 3 — pressure reducing valves, 4 — resisting capillary column, 5 — vessel with sample, 6 — thermostat, 7 — tube with anhydrous magnesium perchlorate (30 mg), 8 — glass wool, 9 — capillary pre-column, 10 — freezing trap.

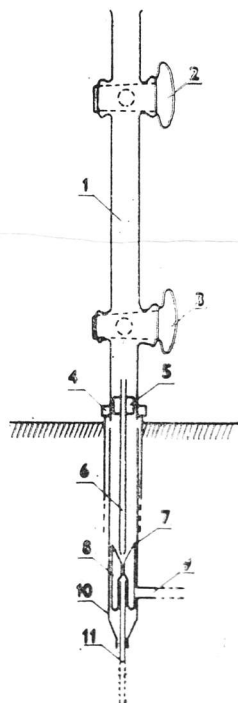


Fig. 13. An apparatus for transport of the capillary pre-column with frozen volatile substances into the injection space of the gas chromatograph [62]. 1 — glass tube with a pair of valves 2 and 3, 4 — silicone packing ring, 6 — capillary pre-column with a steel cylinder 5, 7 — injection section of the apparatus, 8 — brass connecting cylinder (connecting the capillary analytical column 11 with the capillary pre-column 6 into one unit), 11 — splitter with a needle valve of the splitter 9.

using a carrier gas. Extraction of volatile substances is frequently used instead of desorption. Adsorption material used in this case is usually activated charcoal, silica gel or polymer materials with low adsorption capacity of water vapours, e.g. Tenax, Amberlits, Norit, Porapak R and O, etc. [18, 24].

Manning and Price [45] described the above-mentioned method of concentration of volatile substances from milk. The volatile substances are adsorbed on the layer of activated charcoal.

Kroger and Patton [39] used for the determination of volatile substances in cheese a method of placing the cheese directly in a syringe. Using the piston of the syringe, the released volatile substances were dosed and applied directly to the chromatograph injector.

A special method of concentration of the headspace volatile substances consists in their continuous releasing and in trapping them in a closed system. This technique was described already in 1962 by Nawar and Fagerson [48] and later by Grob et al. [29, 31—33].

It was specially designed for the determination of organic pollutants in water.

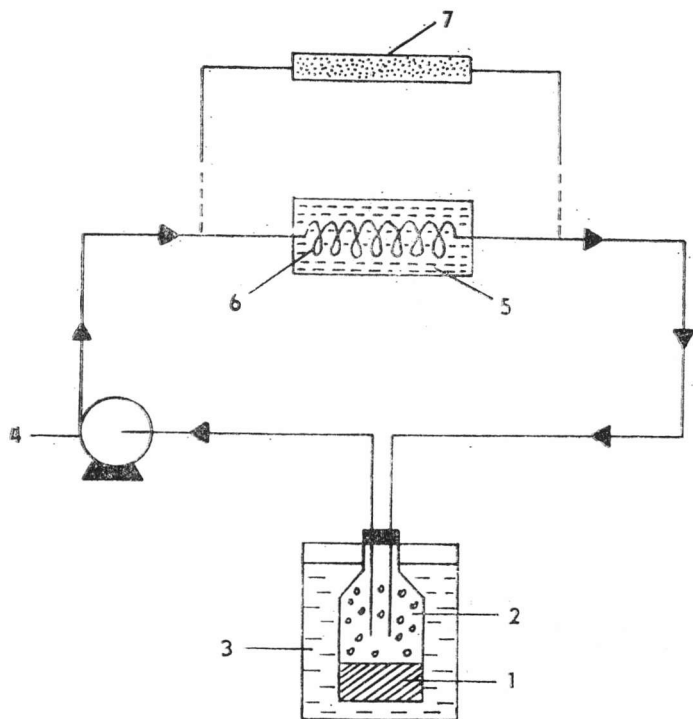


Fig. 14. Scheme of the circulation method of volatile substances concentration. 1 — sample, 2 — headspace, 3 — thermostat, 4 — pump, 5 — freezing trap, 6 — capillary pre-column, 7 — sorbent.

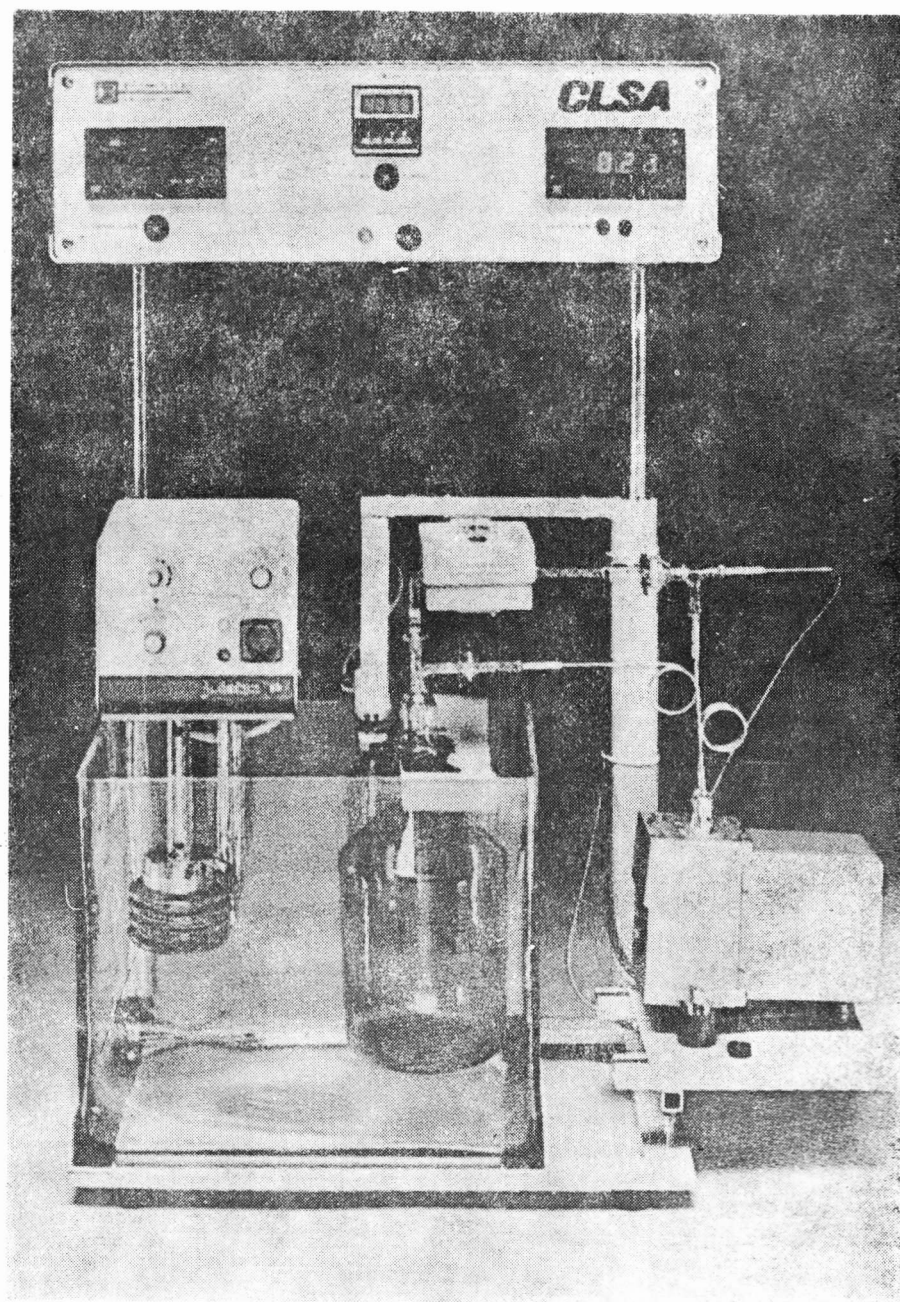


Fig. 15. Closed loop stripping apparatus — CSLA — for separation and concentration of volatile substances from water [84].

A scheme of this method is shown in Fig. 14. Headspace gas is transported by means of a pump into the freezing trap or on the adsorbent column, then, free from volatile substances, it is supplied into a vessel with the sample. Volatile substances are gradually released from the sample and collected in the freezing trap (or on the adsorbent column). The concentrate is then released and analysed.

Carlo Erba Strumentazione, Milan has recently introduced Grob's method into its production programme and offers the closed stripping loop apparatus (CSLA). Originally, it has been designed for the determination of organic compounds, present in water at a quantity of 1 part in 10^{13} parts of water, but it can be employed also for other liquid materials. The apparatus is shown in Fig. 15. The released volatile substances are trapped, in this case, on a small filter made of activated charcoal [84].

The above-mentioned method of concentration of volatile substances in a closed system has several advantages — if compared with other methods. These have been critically discussed in [20, 35, 49].

According to our experience, the above-mentioned method of concentration of volatile components in dairying was dealt with only scarcely [50].

Conclusion

Application of HSGC analysis of volatile substances in dairying contributed significantly to the study of the flavour of milk and milk products. Moreover, by its simplicity, ability to keep the original proportionality of volatile substances in the analysed product, as well as due to the possibility of determining even trace concentration of these substances the method ranks among the widely used ones in dairying. It has been successfully applied in research and inspection. Besides the above-mentioned cases, HSGC is employed in other fields as well. It is applied, for instance, in the study of characteristics of chemical reactions, in the study of physico-chemical properties of various substances, etc. However, this is beyond the framework of the dairying itself.

The paper does not provide a complete survey of studies published in this field. It is of informative character. Its aim was to point out to the variety and wide applicability of the HSGC in dairying in general, particularly under the conditions of the Czechoslovak dairy research and inspection.

In spite of over twenty years of its successful application in dairying HSGC still belongs to the frequent analytic methods and provides further possibilities for even more extensive applications.

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Использование headspace-анализа в молочной промышленности

Резюме

В работе обобщается проблематика headspace-газово-хроматографического (HSGC)-анализа летучих веществ и возможности его использования в молочной промышленности. На основе многочисленных данных из литературы и сведений, полученных автором в практическом опыте, описываются основные методы прямого HSGC-анализа летучих веществ, а также метода, основанного на предварительной концентрации летучих веществ с последующим анализом полученного концентрата при помощи газовой хроматографии.

Работа иллюстрируется схемами отдельных методов и результатами, полученными при их помощи (летучие вещества в молоке, в молочных продуктах и в упаковочном материале).

Работа носит информационный характер и призвана дать основную информацию главным образом работникам научно-исследовательских учреждений в молочной промышленности и работникам контроля.

Použitie headspace analýzy v mliekárstve

Súhrn

V práci je prehľadne zhrnutá problematika headspace-plynovochromatografickej (HSGC) analýzy prchavých látok a jej aplikačné možnosti v mliekárstve. Na základe mnohých údajov z literatúry a poznatkov získaných z autorových praktických skúseností sa opisujú základné metódy priamej HSGC analýzy prchavých látok, ako aj metódy založené na predbežnej koncentrácii prchavých látok s následnou plynovochromatografickou analýzou získaného koncentrátu.

V práci sú schémy jednotlivých metód a nimi získaných výsledkov (prchavé látky v mlieku, mliečnych výrobkoch, obalových materiáloch a pod.).

Práca má informatívny charakter. Má slúžiť ako základná informácia hlavne pre pracovníkov mliekárenského výskumu a kontroly.