Influence of Heat Treatment on the Volatile Compounds of Milk

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In this work dynamic headspace capillary gas chromatography coupled with multivariate statistical techniques was applied to distinguish milks subjected to different heat treatments (pasteurization, direct ultrahigh-temperature method, and "in-bottle" sterilization). Results were obtained by taking into account a reduced, though significant, number of volatile compounds (acetone, 2-butanone, 2-pentanone, 2-heptanone, 3-methylbutanal, pentanal, hexanal, heptanal, dimethyl disulfide, toluene, and limonene). Moreover, the concentration of these volatile compounds, especially aldehydes and ketones, provided discriminant information about storage time and storage temperature in both whole and partially skimmed UHT milk. The analytical method was quite simple, rapid, and reliable and permitted the analysis of a large number of milks to be carried out by using a small volume of sample. This work represents an attempt to apply a method, generally used only for research purposes, to the quality assessment of heat-treated milk.

INTRODUCTION

Cows' metabolism and feed, chemical, microbial, and enzymic reactions, and technological treatments can influence the composition of milk volatile compounds (Calvo and de la Hoz, 1992). Heat treatments and storage conditions have been studied by several authors (Scanlan et al., 1968; Jeon et al., 1978; Shibamoto et al., 1980; Badings et al., 1981; Early and Hansen, 1982; Rerkrai et al., 1987; Adhikari and Singal, 1992; Van Eijk, 1992; Moio et al., 1994; Andersson and Oste, 1995) because they could be responsible, especially in ultra-high-temperature (UHT) milk, for the development of off-flavors, due to a change in concentration of particular compounds, such as aldehydes and methyl ketones. The origin of the substances that occur in the profile of volatile compounds of milk has been studied by Duin (1965), Bassette et al. (1966), Gordon and Morgan (1972), and Grosch (1982). Most of the above-mentioned investigations applied methods (i.e. vacuum distillation followed by solved extraction) to extract a large number of compounds. These methods, however, are time-consuming and require a large volume of sample and, consequently, cannot be applied to a large number of samples and be used for routine monitoring.

On the contrary, dynamic headspace technique has been shown to be an easier, more rapid, and reliable method for the determination of the composition of volatile compounds by using a small volume of sample (Badings et al., 1985; Kim and Morr, 1996). By analyzing a larger number of samples, statistical multivariate techniques can be applied to build useful models for milk quality assessment, as demonstrated by Vallejo-Cordoba and Nakai (1994a,b) and Vallejo-Cordoba et al. (1995) on pasteurized milk.

The aim of our work was to study the possibility of using the profile of volatile compounds as a marker of both the type of milk heat treatment, on samples taken immediately after production, and the influence of storage time and temperature on UHT milk at the end of shelf life. The research was carried out by using dynamic headspace (purge-and-trap off-line method), previously tested by Contarini and Leardi (1994) and Miliazza and Stani (1995), which allowed us to extract the most volatile compounds at room temperature without using solvent. Data were evaluated by a multivariate statistical approach.

MATERIALS AND METHODS

Sampling. Eighty-eight commercial samples of milk subjected to different heat treatments were collected at a factory. Milk samples were produced by different heat treatments (Table 1) and packed aseptically in both 1000 mL Tetra Brik cartons (pasteurized and UHT milks) and 1000 mL bottles (sterilized milks).

The characteristics of milk samples and the time of sampling are reported in Table 2.

In addition, a very inexpensive UHT milk, purchased in a discount shop, was analyzed. Consequently, no information could be given about the technological treatment of this sample.

Analytical Method. Milk (20 g) at room temperature was supplemented with 1 mL of an aqueous solution of methyl butanoate (Aldrich Chemical Co., Milwaukee, WI) (0.05 mg/100 mL of distilled water). Volatile compounds were stripped by nitrogen (flow rate = 25 mL/min) for 1 h and adsorbed on Tenax trap (TA 60−80 mesh, catalog no. 16252, Chrompack, Middelbourg, The Netherlands). The Tenax trap was fitted in the thermal desorption cold trap injector (TCT, Chrompack), connected directly to the GC system. Compounds were desorbed from Tenax by heating at 240 °C for 10 min and transferred by a carrier gas stream (H₂, flow rate = 10 mL/min) to a fused-silica trap, cooled at −120 °C by liquid nitrogen. Injection into the capillary system was performed by flash-heating (260 °C for 1 min) of the cold trap on which the volatile compounds had been reconcentrated. After each run, TCT was automatically cleaned by a back-flush cycle. Blank runs were included between each analysis to confirm that the compounds had been removed completely.

A Chrompack 9001 gas chromatograph equipped with TCT, flame ionization detector, and CP Sil 5 CB (Chrompack) capillary column (50 m length, 0.32 mm i.d., 5 μm film thickness) was used. The oven temperature was held at 50 °C for 10 min, programmed to 230 °C at a rate of 8 °C/min,

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Table 1. Technological Conditions

	type of milk	heat treatment		flash cooling (°C)
		°C	*t*	
pasteurization	whole	78	15.0 s	
UHT (direct)	whole	148	3.5 s	83
UHT (direct)	whole	144	3.5 s	88
UHT (direct)	whole	150	3.5 s	88
UHT (direct)	whole	144	3.5 s	95
UHT (direct)	partially skimmed	144	3.5 s	88
in-bottle sterilization	whole	120	30 min	

Table 2. Characteristics of the Studied Milk Samples

type of sample	no. of milk samples	code	time of sampling	storage temp
pasteurized whole milk	8	A	immediately after production	
UHT whole milk	30	B	immediately after production	
	19	C	at the end of shelf life[a]	room temp
	11	D	at the end of shelf life[a]	4 °C
UHT partially skimmed milk	6	E	immediately after production	
	5	F	at the end of shelf life[a]	room temp
	4	G	at the end of shelf life[a]	4 °C
in-bottle sterilized whole milk	5	H	immediately after production	
commercialized UHT whole milk	1	K	1 month after production	room temp

[a] 90 days.

and held at 230 °C for 20 min. The detector temperature was 260 °C; the flow rate of hydrogen carrier gas was 1 mL/min. Each sample was analyzed in duplicate.

Quantitative results were calculated by comparison with the amount of methyl butanoate added to the sample and were expressed as parts per billion. At this stage of the research, no response factors of the volatile compounds with respect to the internal standard were calculated. As a result, the quantitative values, like arbitrary units, were not representative of the real content of volatile compounds, but they were useful to compare different samples.

For peak identification, some samples were analyzed by using a mass spectrometry detector (5989A Hewlett-Packard, Palo Alto, CA) coupled with an HP 5890 gas chromatograph, adopting the gas chromatographic operating conditions mentioned above.

Statistical Analysis. Data for volatile compounds were subjected to multivariate statistical analysis by using the PARVUS package (Forina et al., 1988). Eighty-eight milk samples (objects) and 11 volatile compounds (variables) were considered (Table 3).

Four data sets (Table 2) were obtained according to the following criteria:

Forty three whole milks (A+B+H) were sampled immediately after production and categorized according to the kind of heat treatment (pasteurization, UHT, "in-bottle" sterilization). In this data set the results of the UHT milk sample purchased in the discount shop were added as a test sample (K).

Thirty UHT whole milks (B) were sampled immediately after production and categorized according to the temperatures of sterilization/flash cooling (degassing) (144/88, 144/95, 148/83, 150/88 °C).

Sixty UHT whole milks (B+C+D) were categorized according to both the time of sampling and the temperature of storage (immediately after production; at the end of shelf life at room temperature and at 4 °C).

Fifteen partially skimmed milks (E+F+G) were categorized according to the time of sampling (immediately after production; at the end of shelf life at room temperature and at 4 °C).

These data sets were subjected to the following statistical analyses.

Data Standardization by Autoscaling (Massart et al., 1988). Autoscaling was applied to all data to consider all variables independently of their different numerical values. Hence, all variables had the same weight since they had a mean = 0 and unitary variance.

Principal Component Analysis (PCA) (Massart et al., 1988). The PCA is an exploratory data analysis, which, through the calculation of linear combinations of original variables, allows

the number of dimensions to be considerably reduced while maintaining most of the information of the data set, expressed as percent variance.

Rotation of Principal Components by Varimax (Massart et al., 1988). This technique performs orthogonal rotation of the principal components and, when applied to the matrix of loadings, it aims at maximizing the grouping of variables along the axes. This makes the interpretation of the new axes easier.

Linear Discriminant Analysis (LDA) (Massart et al., 1988). The LDA is a technique used to classify each sample according to the distance (Mahalanobis distance) from the category centroid. The method was validated by the "repeated evaluation set" technique, in which a predefined percentage of objects (20%) is removed from the data set to verify whether they are correctly classified when the "delimiter" between the two categories is calculated in the absence of these objects. This is repeated for a very large number of times (1000) and the final result, expressed as prediction ability, defines the capability of the method to correctly predict the category of an unknown object.

Stepwise Linear Discriminant Analysis (Jenrich, 1960). This technique was applied to select the most discriminant variables. It considers the ratio (Wilks's lambda) between the generalized within-category dispersion and total dispersion. The selected variable is the one that produces the maximum decrease of Wilks' lambda, tested by a suitable *F* test for the input of a new variable or for the deletion of a previously selected one.

RESULTS AND DISCUSSION

Figures 1 and 2 show the profile of volatile compounds obtained by analyzing a sample of UHT milk by FID and mass spectrometry detector, respectively. The identification was performed by comparison of mass spectra with both the Wiley library (McLafferty and Stauffer, 1988) and authentic standards analyzed under the same conditions. Other substances were found, but they were not taken into account because their identification was not complete.

Before the sampling was begun, the precision of the analytical method was tested by evaluating the results of six replicates of one sample of UHT milk. The values obtained are reported in Table 3. The standard deviation was not comparable with that normally obtained for other gas chromatographic analyses, but it is worth noting that the high volatility of these compounds, even though extracted under standardized conditions, strongly
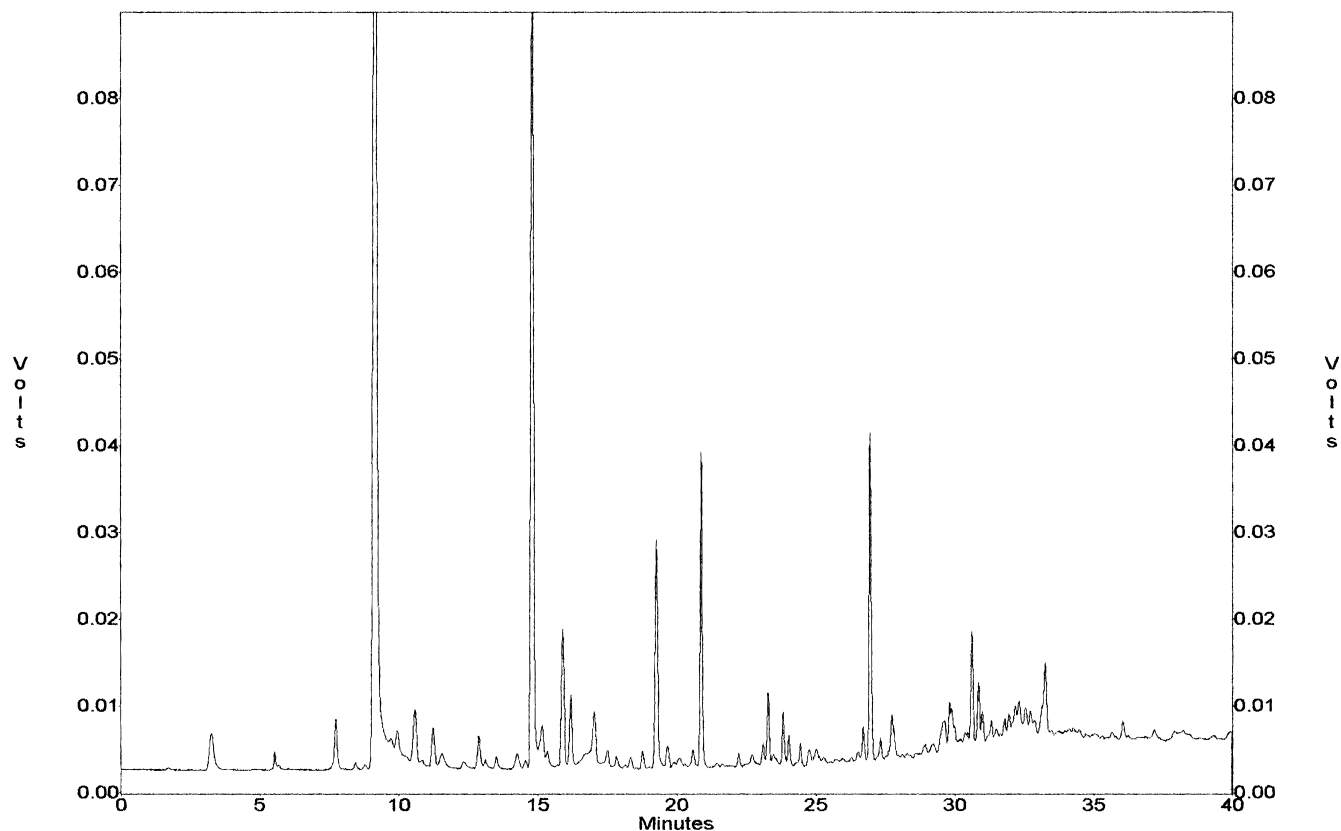
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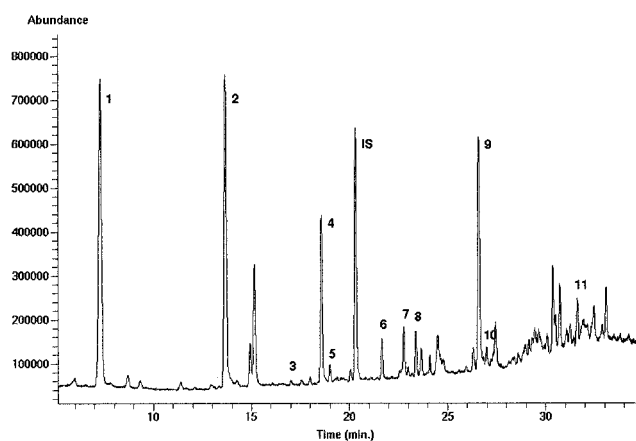**Figure 1.** GC/FID chromatogram of UHT milk.

Figure 2. GC/MS chromatogram of UHT milk (the numbering is the same as in Table 3).

Table 3. Results from Six Replicates of the Analysis of Volatile Compounds in a Milk Sample

no.	compound	retention time (min)	mean	SD	CV %
1	acetone	9.1	70.50	10.39	14.7
2	2-butanone	14.8	8.02	0.29	3.6
3	3-methylbutanal	17.8	0.87	0.12	13.8
4	2-pentanone	19.3	3.21	0.17	5.3
5	pentanal	19.7	3.58	0.87	24.3
6	dimethyl disulfide	22.2	1.24	0.05	4.0
7	toluene	23.3	4.63	0.41	8.8
8	hexanal	23.8	2.26	0.11	4.9
9	2-heptanone	26.9	10.39	1.04	10.0
10	heptanal	27.3	1.54	0.49	31.8
11	limonene	31.3	3.74	0.53	14.2

contributed to increase the variability of the results. For this reason the values obtained can be considered quite satisfactory.

The identified compounds belong to the following classes: ketones, aldehydes, terpenes, and sulfur and aromatic compounds.

Ketones represented the most abundant class. Those having a higher carbon number are responsible for heated milk flavor, as described by Badings et al. (1981). Ketones have different origins: acetone and 2-butanone mainly derive from cows' feed (Bassette et al., 1966; Gordon and Morgan, 1972); 2-pentanone and 2-heptanone have both a natural (Duin, 1965; Gordon and Morgan, 1972) and a technological origin (Shibamoto et al., 1980; Grosch, 1982; Badings, 1984). During the heat treatment the pathways of their formation are both β oxidation of saturated fatty acids, followed by decarboxylation, and decarboxylation of β ketoacids, naturally present in milk (Grosch, 1982).

The aldehydes detected in this research were 3-methylbutanal, pentanal, hexanal, and heptanal. 3-Methylbutanal can be found in raw milk as a consequence of microbial growth of *Streptococcus lactis* var. *maltigenes* (Morgan, 1970), but its presence in heated milk is due to nonenzymic browning reaction of leucine (Ramshaw and Dunstone, 1969). The other aldehydes are formed in raw milk by autoxidation of unsaturated fatty acids (Scanlan et al., 1968; Rerkrai et al., 1987; Moio et al., 1993), and heat treatment can promote this reaction as described by Tressl et al. (1978) and Forss (1979).

As far as sulfur compounds are concerned, only dimethyl disulfide was detected in our research due to the poor sensitivity of FID detector to sulfhydryl compounds. Dimethyl disulfide was found in raw milk as a consequence of cows' feed (Badings and Neeter, 1980), while its presence in heat-treated milk can be due to the oxidation of methanethiol (Dumont and Adda, 1978a; Calvo and de la Hoz, 1992).

No references were found about the influence of heat treatment on the concentration of toluene and limonene;

Table 4. Average and Standard Deviation of Variables Detected

no.	variables	pasteurized milk (8)[a]		UHT milk (30)		"in-bottle" sterilized milk (5)	
		mean	SD	mean	SD	mean	SD
1	acetone	166.85	80.34	35.87	13.88	224.58	35.94
2	2-butanone	81.89	38.05	8.16	2.69	84.37	19.50
3	3-methylbutanal	0.00	0.00	0.54	0.40	0.85	0.52
4	2-pentanone	3.12	1.70	3.88	1.48	48.08	4.29
5	pentanal	1.79	0.33	2.31	1.87	4.82	3.33
6	dimethyl disulfide	0.00	0.00	1.50	1.20	1.45	1.11
7	toluene	2.90	1.09	3.40	2.46	8.64	4.26
8	hexanal	3.77	0.78	2.64	0.58	3.58	0.65
9	2-heptanone	2.63	1.08	9.07	1.17	64.47	9.14
10	heptanal	2.43	1.55	2.08	1.03	2.48	1.10
11	limonene	4.08	3.73	6.63	4.61	3.26	3.67

[a] Numbers in parentheses indicate the number of samples of each group.
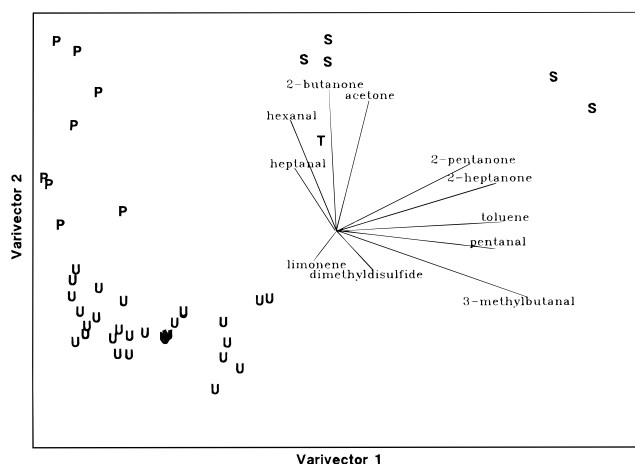

Figure 3. Plot of the loadings of the 11 variables and of the scores of the 44 samples: (U) UHT milks; (P) pasteurized milks; (S) in-bottle sterilized milks; (T) commercial UHT milk.

toluene, in raw milk, is a product of degradation of β-carotene, as reported by Moio et al. (1993); limonene, which has been detected in raw milk and cheese by several authors (Dumont and Adda, 1978b; Bosset et al., 1994; Moio et al., 1994), is probably transferred from forages.

Comparison between Pasteurized, UHT, and In-Bottle Sterilized Milk. Table 4 reports the results [average and standard deviation (SD)] obtained from the samples categorized according to the heat treatment and analyzed immediately after production.

Data were subjected to multivariate statistical analysis, and the results from PCA and Varimax rotation are shown in Figure 3, which represents the plot of the loadings and the scores on the first and second varivector, which explained 55.8% of the total variance. This type of representation shows the mutual relationships between objects and variables.

The compounds detected by the TCT method seemed to be able to distinguished between the three types of milk. The first component allowed pasteurized (P) and in-bottle sterilized (S) milk to be distinguished. The major contribution to this component was given by 2-pentanone, 2-heptanone, toluene, pentanal, and 3-methylbutanal. The second component allowed pasteurized and UHT milk (U) to be distinguished. The major contribution to this component was given by acetone, 2-butanone, and hexanal.

The concentration of acetone and 2-butanone, which are the most volatile ketones, was lower in UHT milk, probably due to the degassing procedure that resulted in the stripping of these compounds. In-bottle sterilized milk was characterized by high values for all the

variables, which highly contributed to both the components. As a result, the samples of this type of milk fall into the top right-hand corner of the plot, and all the above-mentioned variables contribute to the separation of in-bottle sterilized milk from the other two types of milk.

Figure 3 also reports the behavior of the sample of UHT whole milk purchased in a discount shop (T), which showed an unfavorable overall flavor impression characterized by a very pronounced "caramelized flavor". This object was assigned to "test set", which means that its values did not contribute to the calculation of the model. Its projection on the model, which permits comparison with the "known" objects, shows that this UHT milk sample is more similar to the in-bottle sterilized milks than to the UHT milks. On the basis of this behavior, it was assumed that this product may derive from milk of low microbiological quality subsequently subjected to severe and/or repeated heat treatments. This may also explain the low cost of this sample.

Due to the small number of objects of the categories pasteurized and in-bottle sterilized milk, no statistical classification procedures were applied.

Influence of Sterilization and Degassing Temperatures. To verify whether the analysis of volatile compounds was able to evaluate the effects of the severity of heat treatment applied during the UHT process (temperatures of sterilization and of degassing), the 30 samples of UHT milk, analyzed immediately after production (Table 2), were categorized according to the following sterilization/degassing temperatures: 144/88 °C (6 samples), 144/95 °C (6 samples), 148/83 °C (12 samples), and 150/88 °C (6 samples). The PCA applied to the samples divided into the above-mentioned four categories did not show any significant differences among the technological treatments.

Influence of Storage Conditions on UHT Milk. *Whole Milk.* To evaluate the influence of storage on the composition of volatile compounds, the samples of UHT whole milk were analyzed at the end of shelf life, keeping them both at room temperature and at 4 °C. The results were compared with those obtained immediately after production (Table 5).

The samples analyzed immediately after production (1) by the PCA (Figure 4), seemed to be mainly distinguished from those analyzed at the end of shelf life at room temperature (3) along the axis of the first eigenvector, where ketones (mainly 2-pentanone and 2-heptanone) and aldehydes (mainly hexanal and heptanal) have the most important loadings.

Milks stored at room temperature showed an increase particularly in 2-heptanone and 2-pentanone. The

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Table 5. Average and Standard Deviation of UHT Whole Milk Analyzed at Different Times

| | | immediately after production (30)[a] | | after 90 days | | | |
| | | | | at 4 °C (11) | | at room temp (19) | |
no.	variables	mean	SD	mean	SD	mean	SD
1	acetone[b]	35.87	13.88	46.97	3.52	46.29	16.01
2	2-butanone[b]	8.16	2.69	8.66	2.76	10.60	3.59
3	3-methylbutanal	0.54	0.39	0.51	0.63	0.60	0.39
4	2-pentanone[b]	3.88	1.48	5.65	0.90	9.80	2.32
5	pentanal[b]	2.31	1.87	1.14	0.77	1.70	1.45
6	dimethyl disulfide	1.50	1.20	1.23	0.47	1.47	0.71
7	toluene	3.40	2.46	4.01	3.06	4.04	2.05
8	hexanal[b]	2.64	0.58	1.80	0.28	2.02	0.97
9	2-heptanone[b]	9.07	1.17	12.47	2.67	19.08	4.40
10	heptanal[b]	2.08	1.03	1.18	0.41	1.38	0.73
11	limonene	6.63	4.61	6.46	2.83	4.90	3.61

[a] Numbers in parentheses indicate the number of samples for each group. [b] Variables selected by stepwise LDA.
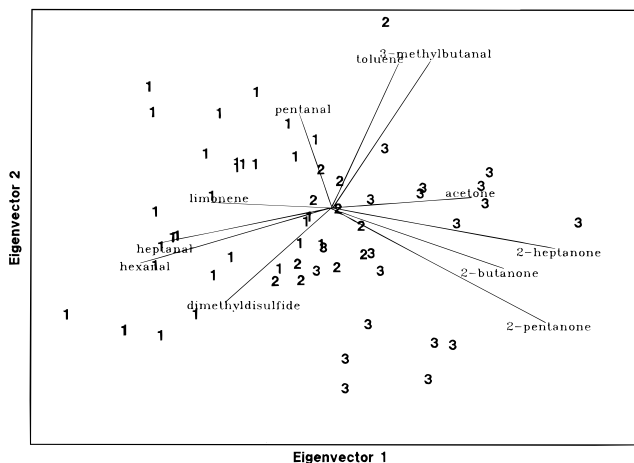

Figure 4. Plot of the loadings of the 11 variables and of the scores of the 60 UHT whole milk samples analyzed at different times/storage temperatures: (1) immediately after production; (2) after 90 days at 4 °C; (3) after 90 days at room temperature.

Figure 5. Plot of the loadings of the 7 variables and of the scores of the 60 UHT whole milk samples analyzed at different times/storage temperatures: (1) immediately after production; (2) after 90 days at 4 °C; (3) after 90 days at room temperature.

formation of odd-carbon-numbered methyl ketones appeared to depend primarily upon storage temperature and to be independent of oxygen content (Jeon et al., 1978; Rerkrai et al., 1987). An important contribution to the distinction between milks of category 1 and milks of category 3 was also given by aldehydes, which showed a small decrease in milks stored at room temperature. Opinion differs as to the behavior of aldehydes during the storage time. Early and Hansen (1982) observed a decrease of the long chain (C > 10) *n*-alkanals during a storage of 24 weeks at 24 °C. The short-chain alkanals showed a slight increase after 8 weeks, followed by a decrease to traces. Jeon et al. (1978) and Rerkrai et al. (1987) reported a general increase of aldehydes, particularly hexanal, during storage and attributed this behavior to oxygen availability and storage temperature. Differences in heat treatment, type of packaging, oxygen content, quality of raw milk, and light exposure may account for conflicting results.

Poor contribution was given by dimethyl disulfide to the discrimination between the categories. Its concentration in samples immediately after production was affected by high variability, and mean values during storage time did not show significant changes, as observed by Dumont and Adda (1978a).

The samples of milk stored at 4 °C (2) (Figure 4) were grouped in an intermediate region between the other two categories. Milk storage at low temperature seemed to reduce the chemical modifications, which usually occur during the shelf life of UHT milk. Similar results have been obtained by other authors (Rerkrai et al.,
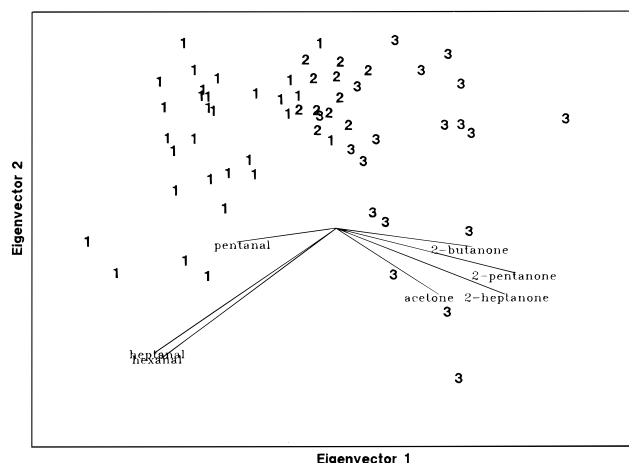
Table 6. Prediction Ability

time of sampling	cat.	% of correct prediction	prediction matrix		
immediately after production	1	93.97	5661	363	0
after 90 days at 4 °C	2	89.50	217	1893	5
after 90 days at room temp	3	93.24	0	260	3584
total		92.95			

1987; Adhikari and Singal, 1992; Andersson and Oste, 1995), who observed a minor increase in off-flavors such as "stale or oxidized" in milk stored under refrigerated conditions.

To improve the separation among the categories, the most discriminant variables (Table 5) were selected by using stepwise linear discriminant analysis, (LDA), and the principal components were recalculated on the reduced data set (60 objects and 7 variables).

Figure 5 shows the eigenvector projection of UHT milks, divided into the three categories. The first two eigenvectors explained 54.0% of variance and seemed to be able to better distinguish category 1 from category 3.

Table 6 reports the results of LDA used as a classification method on the reduced data set. The seven volatile compounds were able to discriminate the three categories, especially milk immediately after production, from the other milks. Moreover, for all the categories, the predictive ability was very satisfactory. From the prediction matrix it was noted that the samples analyzed immediately after production were never assigned

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Table 7. Average and Standard Deviation of UHT Partially Skimmed Milk Analyzed at Different Times

| | | immediately after production (6)[a] | | after 90 days | | | |
| | | | | at 4 °C (4) | | at room temp (5) | |
no.	variables	mean	SD	mean	SD	mean	SD
1	acetone	22.81	10.19	28.90	7.60	33.86	8.79
2	2-butanone	9.21	1.95	9.61	2.05	12.15	1.87
3	3-methylbutanal	0.52	0.46	0.19	0.07	0.40	0.17
4	2-pentanone	1.37	0.35	3.18	0.31	4.55	1.13
5	pentanal	0.63	0.35	1.05	0.27	1.56	0.22
6	dimethyl disulfide	0.57	0.31	1.33	0.37	1.74	0.91
7	toluene	2.43	0.53	1.81	0.14	5.33	3.84
8	hexanal	0.79	0.11	1.80	0.29	3.62	0.46
9	2-heptanone	8.93	1.87	7.41	0.49	9.85	1.51
10	heptanal	1.72	0.79	2.49	0.51	2.30	0.90
11	limonene	8.64	2.48	10.80	4.13	4.49	1.05

[a] Numbers in parentheses indicate the number of samples for each group.
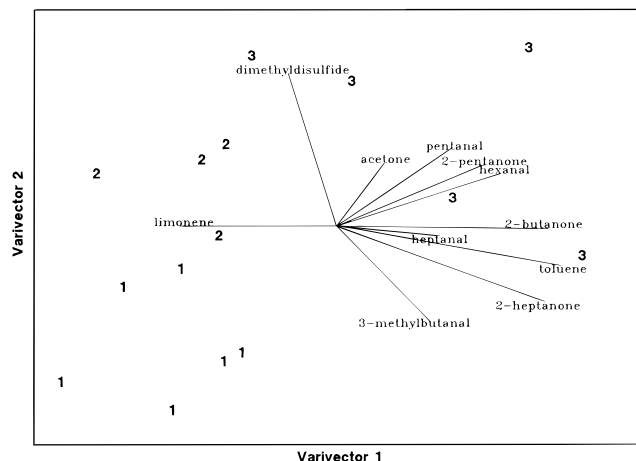

Figure 6. Plot of the loadings of the 11 variables and of the scores of the 15 UHT partially skimmed milk samples analyzed at different times/storage temperatures: (1) immediately after production; (2) after 90 days at 4 °C; (3) after 90 days at room temperature.

to the category of the milks sampled at the end of shelf life after storage at room temperature and vice-versa. The samples analyzed after a storage at 4 °C showed a less satisfactory percentage of correct predictions. It is worth noting that most of the objects uncorrectly classified were assigned to the category of milks sampled immediately after production.

Partially Skimmed Milk. The volatile compounds of 15 samples divided into 3 categories according to the storage conditions were evaluated (Table 7).

The total concentration of volatile compounds of partially skimmed milk (1.8% fat) was lower than that detected in whole milk. A comparison between the values of the compounds deriving from the fat in partially skimmed milk, immediately after production, and those obtained in whole milk showed that they were not directly proportional to the fat content. This result may be partially explained by the study of van Boekel and Lindsay (1992), who pointed out that the partition of volatiles over vapor/water is not only governed by the vapor pressure but also by the activity coefficient. As a result, more concentrated volatiles may be found in vapor over low-fat matrix than in vapor over full-fat matrix.

The concentration of pentanal, hexanal, and heptanal seemed to show a slight increase during storage. This result was in contrast with that obtained from whole milk samples, probably due to the concentration of phospholipids, rich in polyunsaturated fatty acids. This concentration is higher, with respect to the total fat

content, in partially skimmed milk than in whole milk (Forss, 1979).

The results of PCA analysis, followed by Varimax rotation, showed that storage conditions did influence the concentration of volatile compounds. Figure 6 reports the projection on the first and second varivectors (61.3% of total variance explained) of the scores and the loadings. Even though the samples appeared quite scattered, the milk stored at room temperature seemed to be well distinguished from the other two categories as observed above on UHT whole milk. The major contribution to the distinction between the categories was given by aldehydes, and the minor contribution was given by methyl ketones.

The classification procedure could not be applied due to the small number of samples.

Conclusions. The profile of volatile compounds obtained by TCT/GC method, under the conditions described above, allowed us to detect a significant though reduced number of substances that were correlated to the milk flavor. Heat treatment produced a general quali-quantitative increase of volatile compounds even though some differences were found to depend on the chemical–physical characteristics of each compound and the severity of heat treatment. The compounds detected seemed to be able to well distinguish between pasteurized, direct UHT, and in-bottle sterilized milk. The profile of volatile compounds also provided useful information to evaluate both the age of milk and the effects of the storage temperature. In this context, keeping UHT milk at 4 °C seemed to reduce the speed of processes that usually occur during 90 days of shelf life and, consequently, to decrease the development of flavor defects in milk.

It may be concluded that the TCT/GC method coupled with multivariate statistical techniques is a quite simple tool to monitor the quality of milk together with usual chemical–physical and microbiological parameters.

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