Available online at www.sciencedirect.com
ScienceDirect
Food Chemistry 103 (2007) 960–967
Food Chemistry
www.elsevier.com/locate/foodchem

Seasonal variation in the odour characteristics of whole milk powder

Andrea Biolatto [a,*], Gabriela Grigioni [a,b], Martín Irurueta [a], Ana M. Sancho [a], Miguel Taverna [c], Norma Pensel [a]

[a] Instituto Tecnología de Alimentos, Centro de Investigación de Agroindustria, Instituto Nacional de Tecnología Agropecuaria INTA, CC 77 (B1708WAB) Morón, Buenos Aires, Argentina
[b] Consejo Nacional de Investigaciones Científicas y Técnicas CONICET, Av. Rivadavia 1917 (C1053AAY) Buenos Aires, Argentina
[c] EEA Rafaela, Instituto Nacional de Tecnología Agropecuaria INTA, Ruta Nacional 34-Km. 27, CC 22 (2300), Rafaela, Santa Fe, Argentina

Received 10 February 2006; received in revised form 15 September 2006; accepted 28 September 2006

Abstract

Seasonal changes in whole milk powder odour characteristics were monitored using SPME-GC and E-nose methodologies. ANOVA showed a significant effect of the season on dimethyl sulphide, *n*-pentanal, *n*-hexanal, and butyric acid. Whole milk powder manufactured in summer season had significantly ($p < 0.05$) higher levels of *n*-hexanal, *n*-pentanal and dimethyl sulphide as compared to autumn and winter seasons. On the other hand, butyric acid showed significant differences ($p < 0.05$) among autumn and spring. Applying linear discriminant analysis, seasonal variations in the odour profiles assessed by electronic nose were observed. A better classification outcome was obtained when volatile compounds and electronic nose data were analysed together, obtaining a success rate of 89.5% and 70.7% of the original cases and after cross-validation respectively. As in other application reported in the literature, E-nose approach represents an alternative technique to traditional methods of odour measurement.

Keywords: Whole milk powder; Solid-phase microextraction-gas chromatography; Electronic nose; Multivariate analysis

1. Introduction

Sensory quality is defined by a complex set of features including appearance, flavour and texture. As stated by Piggot (1994), each sensory character can be viewed as being a single and important part in the entire set of quality features. Flavour is the result of a combined sensation of taste and odour (Furia & Bellanca, 1975) and it is mainly perceived by the interactions of the volatiles with the olfactory epithelium in the nasal cavity (Piggot, 2005). The changes that occur in the food matrix play a key role in flavour perception (Mariaca & Bosset, 1997).

Dairy products provide a great sense of eating pleasure due to their flavour and smooth taste characteristics. In fresh milk, flavour and off-flavour compounds could likely be created through cow's metabolism and be transferred from feed systems to milk *via* the rumen (Badings, 1991). In addition, off-flavours occur in milk through a variety of mechanisms such as action of native milk enzymes or bacterial enzymes and chemical changes catalysed by light or heavy metals (Badings, 1991; Vallejo-Cordoba & Nakai, 1993).

Sensory features of raw milk depend on milk composition, which are related to dairy cattle management: genetic, herd health, nutrition and feeding programmes. Significant changes in fat, an important source of flavours, can be achieved *via* the level of lipid supplementation and nutritional modifications. Fat-soluble vitamins, protein and lactose contents are subject to seasonal modifications due to changes in the feeding availability. In the case of mineral contents, there are different published results about the influence of feed (Badings, 1991; Christian et al., 1999;

* Corresponding author. Tel.: +54 11 4621 0446/0457; fax: +54 11 4621 2012.
E-mail address: abiolatto@cnia.inta.gov.ar (A. Biolatto).

0308-8146/$ - see front matter © 2006 Elsevier Ltd. All rights reserved.
doi:10.1016/j.foodchem.2006.09.050

Grummer, 1991; Lindmark-Månsson, Fondén, & Petterson, 2003; O'Brien, Mhra, Connolly, & Harrington, 1999; Smit, Schönfeldt, deBeer, & Smith, 2000).

Milk flavour profiles are caused by a complex mixture of volatiles at different levels that could be affected by the manufacture process. Several of these compounds can be more or less manifested in different types of milk depending on the presence of pro-oxidants, antioxidants, storage conditions and so on.

Hall and Andersson (1983) determined by multiple regression analysis that the straight-chain aldehydes (*n*-pentanal, *n*-hexanal, *n*-heptanal, *n*-octanal, *n*-nonanal) contributed significantly to describe the flavour acceptance in whole milk powder. Others authors (Fenaille, Visani, Fumeaux, Milo, & Guy, 2003; Marsili, 1999; Ulberth & Roubicek, 1995) have also identified these compounds in milk products. These fat oxidation products are generally associated to green odours and flavours.

Dimethyl sulphide, a sulfur compound, is related to feed flavours that may be caused by transfer of components from the green forages, silage, etc. and it is associated to forage or silage descriptors. In addition, the growth of psychrotrophic bacteria in milk at temperatures above 4 °C causes an increase in dimethyl sulphide content (Badings, 1991).

Strecker aldehydes, such 3-methyl butanal, are generally described as having fruity odours and flavours when are present in milk at low concentrations (Furia & Bellanca, 1975). Milk contaminated with Streptococcus lactis var. maltigenes during production or stored for some period of time under insufficient cooling (around 10 °C), may develop a malty flavour, mainly as a result of 3-methyl butanal, 2-methyl butanal and methyl propanal formation (Badings, 1991; Fernández-García, Gaya, Medina, & Nuñez, 2004).

Short chain fatty acids, such as butyric acid (C4:0) give sharp and tangy flavours. These compounds, derived from the hydrolysis of milk triglycerides, contribute to both desirable and undesirable flavours to dairy products (Chen, Daniel, & Coolbear, 2003).

Milk powder is widely employed as raw material in food formulation. Therefore flavours and off-flavours could appear in final products (Shiratsuchi, Shimoda, Imayoshi, Noda, & Osajima, 1994). Consequently, flavour monitoring is a part of the research, development, production and quality control of food.

Flavour analysis has been commonly carried out by human assessment. Trained panels can determine descriptors to better assess a certain product quality and detect modification due to taints, off-odours or off-flavours. Nevertheless sensory panels are time-consuming and have their limitations related to human susceptibility and variability, both within and among subjects (Pearce & Gardner, 1998). Alternative method for rapidly measuring odour quantitatively, reproducibly and objectively will therefore be most convenient to food industry and its associated research facilities. Instrumental techniques like gas chromatography–mass spectrometry (GC–MS) give information concerning individual compounds. Different methodologies had been proposed including several sample preparations to isolate and concentrate the odour-active compounds (Chialva, Gabri, Liddle, & Ulian, 1982; Grimm, Lloyds, Miller, & Spanier, 1997; Marsili, Miller, Kilmer, & Simmons, 1994; Marsili, 1999; Nickerson & Likens, 1996; Werkhoff & Bretschneider, 1987; Weurman, 1969). Among them, solid-phase microextraction (SPME) is a solventless extraction technique that has been shown to be a simple and effective tool for detecting low levels of flavour compounds (Arthur & Pawliszyn, 1990; Harmon, 1997; Yang & Peppard, 1994). These extraction methods have the drawback that volatile compounds are not embedded in the original matrix. Due to this reason there is a great interest in using the electronic nose (E-nose) approach in food odour analysis, which allows investigating volatile compounds directly in their original matrix. Basically, this device is a sensor-based instrument designed to respond to the volatile compounds present in the headspace of a sample, combined with a suitable pattern recognition routine. E-nose has been successfully used to classify samples with similar odours characteristics of agricultural product (Persaud, Khaffaf, Hobbs, & Sneath, 1996), grains (Börjesson, Eklöv, Johnsson, Sundgren, & Schnürer, 1996), sheepmeat (Braggins & Frost, 1997), ground beef (Spanier & Braggins, 1999), processed beef (Grigioni, Margaría, Pensel, Sánchez, & Vaudagna, 2000), milk (Brudzewski, Osowszy, & Markiewicz, 2004), etc. However, the number of studies dedicated to dairy products is still limited, probably due to the complexity of their matrixes (Ampuero & Bosset, 2003).

In view of the remarkable progress and the advantages and disadvantages of each instrumental technique, there is no universal method for the simultaneous analysis of all volatile (flavour) compounds (Mariaca & Bosset, 1997). Taking this into account, the objective of this work was to monitor odour seasonal changes in whole milk powder complementing SPME-GC analysis and E-nose approach.

2. Materials and methods

2.1. Milk samples

The commercially processed spray-dried whole milk was obtained from a factory of the Central Dairy Area of Argentina. Samples were collected over a period of one year corresponding to three batches in autumn (in southern hemisphere this season comprise the months of March, April and May), six in winter (June, July and August), five in spring (September, October and November) and five in summer (December, January and February). Special care was taken during powder manufacture to keep to minimum variations throughout the industrial process. The high-heat powders (WPNI 0.72 mg/g) were manufactured from standardised milk, with a fat to solids-not-fat ratio of 0.375,

being the heat treatment used of 90-93 °C during 3 min. Samples of 800 g spray-dried whole milk were packed in polyethylene bags and cardboard boxes and they were transported from the factory to our laboratory. After that, each sample was transferred into caramel glass bottles and they were kept at -20 ± 1 °C until analysis.

3. SPME-GC

3.1. Materials

Volatile compounds standards, SPME fibers, vials and seal with Black Viton septa were purchased from Sigma-Aldrich of Argentina S.A.

3.2. Sample preparation

Whole milk powder was reconstituted to 13% total solids with distillate water at 40 °C and it was stirred during 4 min. Then, 10 ml of the reconstituted milk with 10 μl of internal standard solution (50 μg/g 4-methyl-2-pentanone) and a microstirring bar were placed in a 27 ml glass GC vial (60 mm high and 30 mm in diameter) and capped.

3.3. Extraction of volatile compounds

A set of eight volatile compounds were selected due to their contribution to the flavour characteristics of milk powder, i.e.: dimethyl sulphide, 3-methyl butanal, *n*-pentanal, *n*-hexanal, *n*-heptanal, *n*-octanal, *n*-nonanal and butyric acid (Badings, 1991).

The volatile compounds in the headspace of whole milk powder were obtained using SPME. The SPME fiber used was 75 μm Carboxen-PDMS. For thermal desorption, the SPME fiber remained in the injector for 5 min. The setting on the SPME holder assembly scale was adjusted to 1.0 scale unit to ensure that the fiber was positioned in the headspace above the sample in exactly the same way from run to run. With the sample exposed, the sample vial was placed in a 45 °C water bath, and the sample was stirred to high speed. After 45 min, the fiber was retracted into the needle assembly and removed from the vial. The setting on the SPME holder assembly was changed to 3.4 scale units prior to injection into the GC injector port, which was fitted with a special insert for SPME analysis.

3.4. GC analysis

A Shimadzu series 14B gas–liquid chromatograph, equipped with a flame ionisation detector was used. The injector was operated in the splitless mode at a temperature of 280 °C and the detector at a temperature of 280 °C. Nitrogen was used as carrier gas at 1 ml/min. A 30 m × 0.25 mm i.d. × 0.25 μm FFAP capillary column was used (J&W Scientific, Folson, CA). The following column temperature-programming sequence was used: an initial temperature of 40 °C was maintained for 2 min before being increased to 180 °C at 4 °C/min, then it was raised at 40 °C/min to 235 °C and held for 10 min. Peak identifications were based on comparison of retention time of unknowns with authentic compounds. The relative concentrations of individual compounds were determined by comparing the peak area of the compound in each chromatogram with that of the added 4-methyl-2-pentanone internal standard (10 μl of 50 μg/g 4-methyl-2-pentanone solution) and considering the relative response factors of each compound studied. The internal standard quantification method was based on that described by Vichi, Castellote, Pizzale, and Conte (2003) with the following modifications: standard mixtures with concentration in the range of 10–50 μg/kg and 500–2500 μg/kg for dimethyl sulphide, 3-methyl butanal, *n*-pentanal, *n*-hexanal, *n*-heptanal, *n*-octanal, *n*-nonanal and butyric acid, respectively, were analysed under the conditions described above; the absolute response factor for each compound were then obtained from the slope of the linear regression of the total peak area as a function of the concentration, and the relative response factors were calculated as the ratio between the absolute response factors of each compound to that of the internal standard (obtained at the concentration in whole milk powder samples). The relative concentrations are the average of three separate SPME extractions collected from each sample.

4. E-nose

4.1. Materials

Standard compounds for sensor diagnostic, vials and septa were purchased from Sigma–Aldrich of Argentina S.A.

4.2. Sample preparation

An aliquot of 3 g of whole milk powder was placed in a 22 ml glass vial (70 mm high and 24 mm in diameter) and capped. Analyses were carried out in triplicate.

4.3. E-nose analysis

An electronic nose comprised a detector array of 28 conducting polymer sensors with an automatic multisampler for 50 vials was used (OSMETECH PLC, England). The Acquisition method was set with an initial reference period of 27 s (time *T*), a sample period of 60 s, a wash time of 10 s (2% butanol-water solution as cleansing agent) and a reference interval of 120 s. Nitrogen (oxygen free) was used as carrier gas with a reference humidity of 50% (relative humidity at 30 °C). The multisampler method was the Stopped Flow technique. The loop-fill time used was 0.25 s with an equilibration time of 0.20 s. Before data were obtained (Acquisition process), samples were held at 45 ± 1 °C for 10 min with an internal vial pressure of 0.21 atm in order to stabilise headspace conditions.

4.4. Statistical analysis

The differences in volatile compounds among seasons were examined applying a fixed effects model. The model was $Y_{ij} = \mu + \alpha_i + \xi_{ij}$, where Y_{ij} is the dependent variable; μ is an overall mean response, α_i is the effect of the ith level of season; ξ_{ij} is the residual error with zero mean and variance σ^2 (Weber & Skillings, 2000). Least square means (LSMs) were used in order to compare the value of volatile compounds among seasons. The statistical analysis was performed by means of the General Linear Model (PROC GLM) procedure of SAS®.

GC-SPME and E-nose data were analysed applying Linear Discriminant Analysis (LDA) with Wilks' lambda stepwise method for variable selection. The criterion used was the significance of F with a maximum of 0.05 to enter and a minimum of 0.10 to exit. First, all the variables x_i were transformed into standardised z_i values.

$$z_i = \frac{x_i - \bar{x}}{\sigma^2} \quad (1)$$

This transformation allowed an equal weighing of all the variables. LDA was applied as a classification procedure to obtain an equation by which milk samples could be classified. This method maximises the variance between and within categories. SPSS®Advanced Statistics 12 software (SPSS Inc., Chicago, IL) was used.

5. Results and discussion

Table 1 shows the concentration, standard deviation and ANOVA results of the volatile compounds detected in all samples analyzed in each season. Besides, Table 1 shows the concentration and standard deviation of *n*-heptanal, *n*-octanal, *n*-nonanal, and 3-methyl butanal in those samples where they have been quantified, but they were not considered in the ANOVA. According to the ANOVA results, season had a significant effect for *n*-pentanal ($p < 0.01$) and *n*-hexanal, dimethyl sulphide and butyric acid ($p < 0.0001$). It can be observed that whole milk powder manufactured in summer had significantly higher levels of *n*-pentanal and *n*-hexanal as compared to autumn and winter. It is important to remark that in this assay milk powder samples were evaluated without storage, the observed behaviour of the volatile compounds could be associated to the raw material characteristics. Although, *n*-heptanal, *n*-octanal, and *n*-nonanal were not detected in all samples, it is important to mention that they showed, in general, a higher concentration in samples collected in spring and summer compared to those collected in autumn and winter. Different authors (García, Pensel, Margaría, Olga Rosso, & Machado, 1999; Larick & Turner, 1989; Melton, Black, Davis, & Backus, 1982; Yang, Lanari, Brewster, & Tume, 2002) showed that pasture feeding programs led to increased values of highly unsaturated fatty acids. A previous study carried out with milk from the Central Dairy Area of Argentina showed that the concentrations of linoleic ($n-6$) and linolenic ($n-3$) acids were higher in milk from animals fed with pasture-based diet compare to those fed with grain based diet (Páez et al., 2002). These fatty acids are precursors of aldehydes compounds such *n*-pentanal and *n*-hexanal. It is important to remark that *n*-pentanal and *n*-hexanal levels found in the present study were below of their odour threshold in milk of 130 mg/kg and 50 mg/kg, respectively (Fenaille et al., 2003).

Dimethyl sulphide showed the same seasonal behaviour observed for *n*-pentanal and *n*-hexanal (Table 1). Urbach (1990) reported that when cows consume feeds, such as alfalfa, the dimethyl sulphide content of the milk increases compared to the level obtained when cows are fed on a reduced protein regimen. Lane et al. (2002) established that

Table 1
Volatile compounds detected in whole milk powder in different seasons

	Autumn	Winter	Spring	Summer	Significance level of ANOVA[D]
					Season
Aldehydes[A]					
n-Pentanal	9.50 ± 2.66b	11.62 ± 3.31b	13.24 ± 2.89b	16.24 ± 2.34a	*
n-Hexanal	2.32 ± 0.35b	2.45 ± 1.08b	3.13 ± 0.53ab	5.19 ± 1.32a	**
n-Heptanal[C]	nd	4.00 ± 0.44	7.83 ± 1.74	5.90 ± 1.12	–
n-Octanal[C]	4.76 ± 1.14	nd	4.58 ± 0.98	8.62 ± 5.13	–
n-Nonanal[C]	7.47 ± 1.77	7.60 ± 0.19	9.81 ± 3.34	14.63 ± 3.59	–
3 Methyl butanal[C]	0.93 ± 0.06	0.95 ± 0.08	1.17 ± 0.16	1.31 ± 0.44	–
Sulphur compounds[A]					
Dimethyl sulphide	2.32 ± 0.69b	2.84 ± 1.11b	4.79 ± 1.26ab	5.13 ± 1.58 a	**
Free fatty acid[B]					
Butyric acid	7.39 ± 2.38 a	5.42 ± 1.44ab	3.90 ± 0.46b	4.95 ± 1.69ab	**

nd: no detected.
Different letters in the same row indicate significant differences ($p < 0.05$) by least square means.
[A] Concentrations expressed in μg/kg.
[B] Concentrations expressed in mg/kg.
[C] Concentration values corresponding to few samples in each season. These compounds were not included in ANOVA analysis.
[D] Significance at $^{*}p < 0.001$; $^{**}p < 0.0001$.

the concentration of dimethyl sulphide was significantly lower in milk from the "total mixed ration" (TMR)-fed cows than in milk from pasture-fed cows. These authors proposed that the reduced degradation of the parent amino acids might account for the lower concentrations of dimethyl sulphide in milk from TMR-fed cows. Bendall (2001), using gas chromatography – olfactometry method, associated the observed differences between dimethyl sulphide content in milk from TMR and pasture fed cows to variations in rumen degradation of methionine. It is important to remark that concentrations, like the ones measured for dimethyl sulphide in all the seasons (Table 1), had been related to desirable flavour notes in milk powder (McSweeny, Nursten, & Urbach, 1997).

In accordance to Bugaud, Buchin, Coulon, Hauwuy, and Dupont (2001) and Ramaswamy et al. (2001) results, the behaviour found for the oxidations products studied and dimethyl sulphide could be explained by the variations in feed regimens employed in the Central Dairy Area of Argentina. In autumn and winter feeding systems are based on silage, grains and alfalfa (*Medicago sativa* L.) hay. In spring and summer, alfalfa pasture is introduced as *ad- libitum* feeding, while silage, grain and hay are sudden reduced from diet. Furthermore, it is important to note that *n*-heptanal, *n*-octanal, *n*-nonanal and 3-methyl butanal were not included in the ANOVA because they were not detected in most samples. However, in Table 1 are shown the concentration data in those samples where they have been quantified. As can be seen, in general, spring and summer showed higher concentration of *n*-heptanal, *n*-octanal, *n*-nonanal compared to autumn and winter. In the same way that *n*-pentanal and *n*-hexanal, the differences in the *n*-heptanal, *n*-octanal, *n*-nonanal level found between seasons can be produced by the variations in feed regimens. On the other hand, the 3-methyl butanal level, a Strecker aldehyde, could be resulted of the conditions employed during manufacture of whole milk powder.

Butyric acid showed significant differences ($p < 0.05$) among autumn and spring season. This result could be associated to the development of lipolysis in raw milk. This parameter showed the same pattern throughout the season (data not published).

The characteristics of the thermal treatment applied during milk processing carry out to different stages in Maillard reaction (Singh & Newstead, 1998). One of the most important steps of thermal aroma formation *via* Maillard reaction is the reaction between dicarbonyls with amino acids to yield, among others, Strecker aldehydes. The 3-methyl butanal, a branched aldehyde, resulted from of Strecker degradation (Hall & Andersson, 1985). Besides, milk contaminated with *Streptococcus lactis* var. *maltigenes* during production or storage for some period of time under insufficient cooling can produce 3-methyl butanal (Badings, 1991; Fernández-García et al., 2004). Thus, the 3-methyl butanal level found in the present study could be resulted of the conditions employed during manufacture of whole milk powder.

E-nose data were analysed using Linear Discrimant Analysis with Wilks' lambda stepwise method to investigate the grouping of the milk odour profiles as a function of the season. Three discriminant functions (DF) were obtained that explained 50.7%, 34.6% and 14.7% of the total variance respectively (Fig. 1), with a success rate of correct classification of each sample in their respective group (i.e.: season) of 72.4% and 65.5% of the original cases and after cross validation. The discriminant analysis applied to E-nose data included a calculation of the average values of the three canonical variables (Table 2, case a) and their coefficients, which shown the position of each season in the systems of the three coordinates. The canonical variables form the following equations of the corresponding linear discriminant function (DF_i with $i = 1$–3), where S_j represent sensor number j:

$$DF_1 = -0.915 * S_2 + 0.812 * S_6 - 0.911 * S_{22} - 0.285 * S_{25}$$
$$DF_2 = +0.188 * S_2 - 0.129 * S_6 + 0.043 * S_{22} + 1.062 * S_{25}$$
$$DF_3 = -0.160 * S_2 - 0.370 * S_6 + 0.803 * S_{22} - 0.132 * S_{25}$$

In the same way, GC-SPME data were analysed using Linear Discrimant Analysis with Wilks' lambda stepwise method. Two discriminant functions (DF) were obtained that explained 85.1%, 14.4% of the total variance, respectively (Fig. 2), with a success rate of correct classification 65.5% and 62.1% of the original cases and after cross validation. The discriminant analysis applied to SPME-GC data included a calculation of the average values of the two canonical variables (Table 2 case b) and their coefficients. The canonical variables form the following equations of the corresponding linear discriminant function (DF_i with $i = 1$–2):

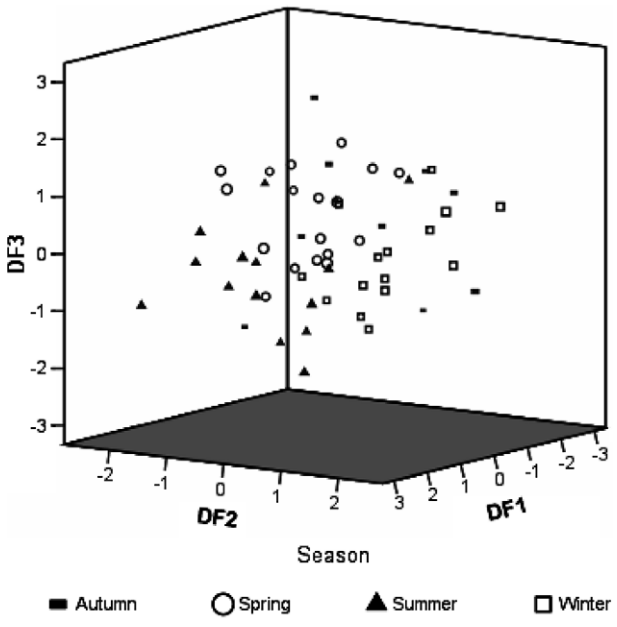

Fig. 1. Discriminant analysis of electronic nose data. The first, second and third discriminant functions explained 50.7%, 34.6% and 14.7% of the total variance, respectively.

Table 2
Results of the canonical analysis of applied to E-nose data (case a), and SPME-GC-FID (case b) and SPME-GC-FID plus E-nose data (case c)

Season	Means of the canonical variables								
	DF1			DF2			DF3		
	Case a	Case b	Case c	Case a	Case b	Case c	Case a	Case b	Case c
Autumn	−0.281	−2.101	−2.933	−1.593	0.699	0.783	0.113	ns	0.675
Winter	−0.220	−1.133	−1.007	0.429	−0.262	−0.125	0.601		−0.125
Spring	−0.933	0.787	0.819	0.325	−0.756	−1.562	−0.537		0.316
Summer	1.280	1.719	2.016	0.108	0.611	1.165	−0.237		0.063

ns: no significant ($p > 0.05$).

$$DF_1 = 0.742 * n\text{-hexanal} + 0.839 * \text{dimethyl sulphide} - 0.503 * \text{butyric acid}$$

$$DF_2 = 0.521 * n\text{-hexanal} + 0.044 * \text{dimethyl sulphide} + 0.811 * \text{butyric acid}.$$

Changes in the classification outcome were obtained by the incorporation of volatile compounds data. Three significant discriminant functions were obtained that explained 68.8%, 25.8% and 5.4% of the total variance respectively. A success rate of correct classification of 89.5% and 70.7% was obtained for the original cases and after cross-validation respectively (Fig. 3). Discriminant analysis revealed that the complement of both techniques resulted in a better discrimination between whole milk powders manufactured in autumn respect to those manufactured in summer. Similar results were observed in the ANOVA analysis of volatile compounds, i.e.: significant differences were found among autumn and summer for *n*-pentanal, *n*-hexanal and dimethyl sulphide.

The discriminant analysis applied to both GC-SPME and E-nose data included a calculation of the average values of the three canonical variables (Table 2 case c) and their coefficients, which shown the position of each season in the systems of the three coordinates. The canonical variables form the following equations of the corresponding linear discriminant function (DF_i with $i = 1$–3), where S_j represent sensor number j:

$$DF_1 = -0.358 * S_2 - 0.558 * S_7 + 0.131 * S_{22} + 0.777 * n\text{-hexanal} + 0.878 * \text{dimethyl sulphide} - 0.584 * \text{butyric acid}$$

$$DF_2 = -0.694 * S_2 + 0.725 * S_7 + 0.859 * S_{22} + 0.490 * n\text{-hexanal} - 0.021 * \text{dimethyl sulphide} + 0.668 * \text{butyric acid}$$

$$DF_3 = 0.401 * S_2 + 0.340 * S_7 - 0.736 * S_{22} + 0.102 * n\text{-hexanal} + 0.232 * \text{dimethyl sulphide} + 0.306 * \text{butyric acid}$$

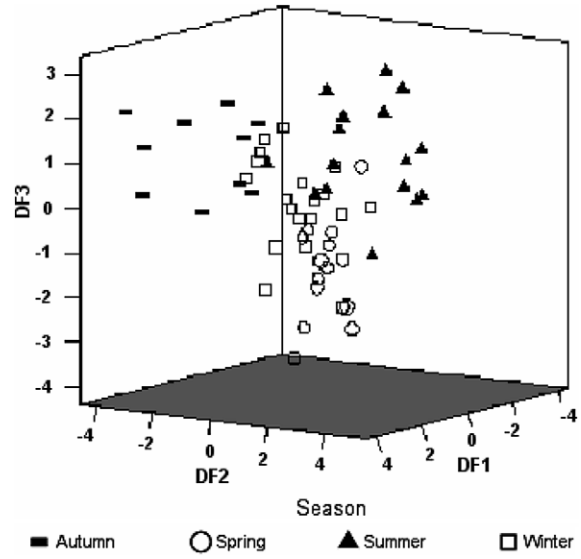

Fig. 3. Discriminant analysis of SPME-GC-FID and electronic nose data. The first, second and third discriminant functions explained 68.8%, 25.8% and 5.4% of the total variance, respectively.

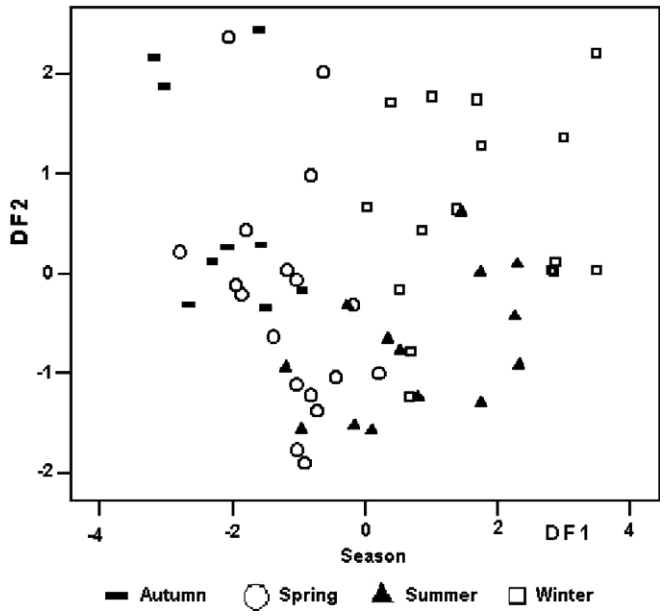

Fig. 2. Discriminant analysis of SPME-GC-FID data. The first and second discriminant functions explained 85.1% and 14.4% of the total variance, respectively.

6. Conclusion

The results presented in this study showed that the odour seasonal changes in whole milk powder can be monitored

using SPME-GC analysis or E-nose approach. As in other application reported in the literature, E-nose approach represents an alternative technique to traditional methods of odour measurement. Moreover, for a complete understanding of the classification scheme resulted from E-nose results it is adequate to relate the data to that obtained by traditional methodologies. This stage is relevant when this kind of approach is intended to be applied to food quality control. In our study, the complement of SPME-GC and E-nose has shown a better classification of the whole milk powders manufactured in the different seasons.

Acknowledgements

The study described herein is a part of a major project financed by Instituto Nacional de Tecnología Agropecuaria INTA and Agencia Nacional de Promoción de Actividades Científicas y Tecnológicas, Contract number BID 802/OC-AR-PICT 09-0494.

References

Ampuero, S., & Bosset, J. O. (2003). The electronic nose applied to the dairy products: a review. *Sensor and Actuators B, 94*(1), 1–12.

Arthur, C. L., & Pawliszyn, J. (1990). Solid-phase microextraction with thermal desorption using silica optical fibers. *Analytical Chemistry, 62*(19), 2145–2148.

Badings, H. T. (1991). Milk. In H. Maarse (Ed.), *Volatile compounds in food and beverages* (pp. 91–106). New York: Marcel Dekker Inc.

Bendall, J. G. (2001). Aroma compounds of fresh milk from New Zealand cows fed different diets. *Journal of Agricultural and Food Chemistry, 49*(10), 4825–4832.

Börjesson, T., Eklöv, T., Johnsson, A., Sundgren, E., & Schnürer, J. (1996). Electronic nose for odour classification of grains. *Cereal Chemistry, 73*(4), 457–461.

Braggins, T. J., & Frost, D. A. (1997). The effect of extended chilled storage in CO_2atmosphere on the odour and flavour of sheepmeat as assessed by sensory panel and an electronic nose. In: *Proceedings of the 43th international congress of meat science and technology* (pp. 198–199). Auckland: New Zealand.

Brudzewski, K., Osowszy, S., & Markiewicz, T. (2004). Classification of milk by means of an electronic nose and SVM neural network. *Sensors and Actuators B, 98*(2–3), 291–298.

Bugaud, C., Buchin, S., Coulon, J. B., Hauwuy, A., & Dupont, D. (2001). Influence of the nature of alpine pastures on plasmin activity, fatty acid, and volatile compound composition in milk. *Le Lait, 81*(3), 401–414.

Chen, L., Daniel, R. M., & Coolbear, T. (2003). Detection and impact of protease and lipase activities in milk and milk powders. *International Dairy Journal, 13*(4), 255–275.

Chialva, F., Gabri, G., Liddle, P. A. P., & Ulian, F. (1982). Quality evaluation of odourtic herbs by direct headspace GC analysis. Applications of the method and comparison with the traditional analysis of essential oils. *Journal of High Resolution Chromatography & Chromatography Communications, 5*, 182–188.

Christian, M. P., Grainger, C., Sutherland, B. J., Mayes, J. J., Hannah, M. C., & Kefford, B. (1999). Managing diet quality for Cheddar cheese manufacturing milk. Part 1. The influence of protein and energy supplements. *Journal of Dairy Research, 66*(3), 341–355.

Fenaille, F., Visani, P., Fumeaux, R., Milo, C., & Guy, P. A. (2003). Comparison of mass spectrometry-based electronic nose and solid phase microextraction gas chromatography-mass spectrometry technique to assess infant formula oxidation. *Journal of Agricultural and Food Chemistry, 51*(9), 2790–2796.

Fernández-García, E., Gaya, P., Medina, M., & Nuñez, M. (2004). Evolution of the volatile components of raw ewes' milk Castellano cheese: Seasonal variation. *International Dairy Journal, 14*(1), 39–46.

Furia, T. E., & Bellanca, N. (1975). In G. Burdock (Ed.), *Fenarolis handbook of flavor ingredients* (pp. 68–234). Cleveland, OH: CRC Press.

García, P. T., Pensel, N. A., Margaría, C. A., Olga Rosso, C. M., & Machado, C. (1999). Intramuscular fat, cholesterol, and 18:2 $n-6$/18:3 $n-3$ ratio in total lipids in two frame steers under different dietary regime. In: *Proceeding of the 45th international congress of meat science and technology* (pp. 76–77). Yokohama: Japan.

Grigioni, G., Margaría, C., Pensel, N. A., Sánchez, G., & Vaudagna, S. (2000). Warmed-over flavour analysis in low temperature-long time processed meat by an "electronic nose". *Meat Science, 56*(3), 221–228.

Grimm, C. C., Lloyds, S. W., Miller, J. A., & Spanier, A. M. (1997). The analysis of food volatiles using direct thermal desorption. In R. Marsili (Ed.), *Techniques for analyzing food aroma* (pp. 59–79). New York: Marcel Dekker Inc.

Grummer, R. (1991). Effect of feed on the composition of milk fat. *Journal of Dairy Science, 74*(9), 3244–3257.

Hall, G., & Andersson, J. (1983). Flavor changes in whole milk powder during storage. III Relationships between flavor properties and volatile compounds. *Journal of Food Quality, 7*, 237–253.

Hall, G., & Andersson, J. (1985). Flavor changes in whole milk powder during storage. III. Relationships between flavor properties and volatiles compounds. *Journal of Food Quality, 7*, 253–2327.

Harmon, A. D. (1997). Solid-phase microextraction for the analysis of flavors. In R. Marsili (Ed.), *Techniques of analyzing food aromas* (pp. 81–112). New York: Marcel Dekker Inc.

Lane, G.A., Frase, K., Kolver, E.S., Rowan, D.D., Allen, J.M., Mills, O.E., Abraham, A.S., Olney, S.D. (2002). Effect of a total mixed ration diet on the concentration of amino acid-derived volatiles in milk. In: *Proceedings of the New Zealand society of animal production*, Vol. 62, (pp. 242–245).

Larick, D. K., & Turner, B. E. (1989). Influence of finishing diet on the phospholipid composition and fatty acid profile of individual phospholipids in lean muscle of beef cattle. *Journal of Animal Science, 67*(9), 2282–2293.

Lindmark-Månsson, H., Fondén, R., & Petterson, H. E. (2003). Composition of Sweddish dairy milk. *International Dairy Journal, 13*(6), 409–425.

Mariaca, R., & Bosset, J. O. (1997). Instrumental analysis of volatiles (flavour) compounds in milk and dairy products. *Le Lait, 77*(1), 13–40.

Marsili, R. T., Miller, N., Kilmer, G. J., & Simmons, R. E. (1994). Identification and quantification of the primary chemicals responsible for the characteristic malodour of beet sugar by purge and trap GC–MS–OD techniques. *Journal of Chromatographic Science, 32*, 165–171.

Marsili, R. T. (1999). SPME–MS–MVA as an electronic nose for the study of off-flavour in milk. *Journal of Agricultural and Food Chemistry, 47*(2), 648–654.

Melton, S. L., Black, J. M., Davis, G. W., & Backus, W. R. (1982). Flavour and selected chemical characteristics of ground beef from steers background on pasture and fed corn up to 140 days. *Journal of Food Science, 47*(3), 699–704.

McSweeny, P. L., Nursten, N. E., & Urbach, G. (1997). Flavours and off-flavours in milk and dairy products. In P. F. Fox (Ed.), *Advanced dairy chemistry: Lactose, salts, and vitamin* (pp. 403–468). London: Chapman and Hall.

Nickerson, G. B., & Likens, S. T. (1996). Gas chromatographic evidence for the occurrence of hop oil components in beer. *Journal of Chromatography, 21*, 1–3.

O'Brien, B., Mhra, R., Connolly, J. F., & Harrington, D. (1999). Seasonal variation in the composition of Irish manufacturing and retails milks. Part 4. Mineral and trace elements. *Irish Journal of Agricultural and Food Research, 38*, 87–99.

Páez, R., García, P., Comerón, E., Aronna, M., Romero, L., Taverna, M., et al. (2002). Perfil de ácidos grasos en la leche de vaca Holando Argentino y Jersey sometidas a dos sistemas de alimentación. *Revista Argentina de Producción Animal, 22*, 43–44.

Pearce, T., & Gardner, J. (1998). Predicting organoleptic scores of sub-ppm flavour notes. Part 1. Theortical and experimental details. Part 2. Computational analysis and results. *Analyst, 123*, 2047–2066.

Persaud, K. C., Khaffaf, S. M., Hobbs, P. J., & Sneath, R. W. (1996). Assessment of conducting polymer odour sensors for agricultural malodour measurements. *Chemical Senses, 21*, 495–505.

Piggot, J. R. (1994). Understanding flavour quality: difficult or impossible? *Food Quality and Preference, 5*(1–2), 167–171.

Piggot, J. R. (2005). Quantifying aroma in berverages by chromatographic analysis and sensory methods. Correspondence. EnLibro de resúmenes. Jornadas de Análisis Sensorial. Tendencias Actuales y Aplicaciones. Buenos Aires, Argentina.

Ramaswamy, R., Baer, R. J., Schingoethe, D. J., Hippen, A. R., Kasperson, K. M., & Whitlock, L. A. (2001). Composition and flavor of milk and butter form cows fed fish oil, extruded soybeans, or their combination. *Journal of Dairy Science, 84*(10), 2144–2151.

Singh, H., & Newstead, F. (1998). Aspects of proteins in milk powder manufacture. In P. F. Fox (Ed.), *Advanced dairy chemistry, aspects of proteins in milk powder manufacture* (pp. 735–765). London: Elsevier Applied Science.

Shiratsuchi, H., Shimoda, M., Imayoshi, K., Noda, K., & Osajima, Y. (1994). Volatile flavor compounds in spray-dried skim milk powder. *Journal of Agricultural and Food Chemistry, 42*(6), 994–998.

Smit, L., Schönfeldt, H., deBeer, W., & Smith, M. (2000). The effect of locality and season on the composition of South African whole milk. *Journal of Food Composition and Analysis, 13*(4), 345–367.

Spanier, A. M., Braggins, T. J. (1999). Electronic nose: What are they and what are their potential uses in meat science. In: *Proceedings of the 52nd reciprocal meat conference of the American meat science association* (Vol. 52 pp. 103–112). Oklahoma, United States.

Ulberth, F., & Roubicek, D. (1995). Monitoring of oxidative deterioration of milk powder by headspace gas chromatography. *International Dairy Journal, 5*(6), 523–531.

Urbach, G. (1990). Effect of feed on flavor in dairy foods. *Journal of Dairy Science, 73*(12), 3639–3650.

Vallejo-Cordoba, B., & Nakai, S. (1993). Using a simultaneous factor optimization approach for the detection of volatiles in milk by dynamic headspace gas chromatographic analysis. *Journal of Agricultural and Food Chemistry, 41*(12), 2378–2384.

Vichi, S., Castellote, A. I., Pizzale, L., & Conte, L. S. (2003). Analysis of virgin olive oil volatile compounds by headspace solid-phase microextraction coupled to gas chromatography with mass spectrometric and flame ionization detection. *Journal of Chromatography A, 983*, 19–33.

Weber, D. C., & Skillings, J. H. (2000). *A first course in the design of the experiments: A linear models approach*. Boca Raton, USA: CRC Press.

Werkhoff, P., & Bretschneider, W. (1987). Dynamic headspace gas chromatography: concentration of volatile components after thermal desorption by intermediate cryofocusing in a cold trap. I. Principle and applications. *Journal of Chromatography A, 405*, 87–98.

Weurman, C. (1969). Isolation and concentration of volatiles in food odor research. *Journal of Agricultural and Food Chemistry, 17*(2), 370–384.

Yang, X., & Peppard, T. (1994). Solid-phase microextraction for flavor analysis. *Journal of Agricultural and Food Chemistry, 42*(9), 1925–1930.

Yang, A., Lanari, M. C., Brewster, M. J., & Tume, R. K. (2002). Lipid stability and meat colour of beef from pasture-and grain-fed cattle with or without vitamin E supplementation. *Meat Science, 60*(1), 41–50.