Combinatorial Approach to Flavor Analysis. 2. Olfactory Investigation of a Library of *S*-Methyl Thioesters and Sensory Evaluation of Selected Components

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The odor characteristics of individual components present in a library comprised of *S*-methyl thioesters were determined independently by two laboratories using similar but not identical techniques. The odor potency was assessed by values of *b*est *e*stimate−GC−*lo*wer *a*mount *d*etected by *s*niffing (BE−GC−LOADS). For small and medium chain *S*-methyl thioesters, these values were found to increase from 6 ng for *S*-methyl thiobutanoate to 90 ng for *S*-methyl thiostearate. All assessors detected a "green", "floral", or "pineapple" odor for *S*-methyl thiohexanoate and described thioesters containing a 2−6 carbon chain length as "cheesy". The results of this preliminary analysis were confirmed by a more extensive study of selected compounds, namely *S*-methyl thioacetate, *S*-methyl thiopropionate, *S*-methyl thiobutanoate, and *S*-methyl thiohexanoate, using a trained panel of 18 subjects. The subjects confirmed the presence of the "green" and "fruity" notes in the odor of *S*-methyl thiohexanoate. The analysis also revealed a significant difference in the odor of *S*-methyl thiopropionate relative to that of *S*-methyl thioacetate and *S*-methyl thiobutanoate. When "cheesy" characteristics were mentioned, the majority of panelists clearly associated the flavor of *S*-methyl thiopropionate with Camembert with almost 20% of all the descriptors given referring specifically to this cheese variety as compared to about 2 and 5% in the case of *S*-methyl thioacetate and thiobutanoate, respectively. Prompted by this observation, two samples of Camembert prepared from unpasteurized and pasteurized milk were analyzed and relatively large amounts of *S*-methyl thiopropionate were found in the former but not in the latter cheese. The results obtained in the course of this work suggest that the sensory analysis of combinatorial libraries is a useful new approach in the search for new commercial flavors and/or identification of characteristic flavors in foods.

Keywords: *Combinatorial synthesis; flavor libraries; thioester; olfactory analysis; flavor potency; sensory evaluation*

INTRODUCTION

Sulfur-containing compounds are flavor constituents in various foods (Mussinan et al., 1994) and are known to be prevalent in a range of dairy products (Grill et al., 1966; Cuer et al., 1979). In particular, short chain *S*-methyl thioesters have been identified in numerous cheeses, e.g., Domiati (Collin et al., 1993), Fontina and Beaufort (Bosset et al., 1993), Langres (Dumont et al., 1974), Limburger (Parliment et al., 1982), Pont l'Evêque (Dumont et al., 1974), Saint-Nectaire (Verdier et al., 1995), and Vacherin (Dumont et al., 1974). However, despite the significance of this class of flavors, relatively few attempts have been made to characterize these compounds in detail (see, for example, Cuer et al., 1979).

In the accompanying paper (Khan et al., 1999), we demonstrated the feasibility of a "one pot" synthesis of a *S*-methyl thioester library and quantified the individual components present. The flavor library was also used to determine physicochemical properties of the *S*-methyl thioesters, e.g., lipophilicity coefficients (log k_w), to illustrate that these parameters can be obtained for a relatively large number of compounds in a single experiment. However, olfactory analysis is probably the most demanding and labor intensive area in flavor chemistry research. Consequently various rapid and cost-effective techniques e.g. GC−sniffing methods such as CHARM (Acree et al., 1984, Marin et al., 1988), AEDA (Grosch, 1994), and OSME (Miranda-Lopez et al., 1992) have been developed and applied successfully to the analysis of flavor extracts from foods. In this paper we demonstrate the feasibility of using GC−sniffing to assess the odor potency and individual flavor characteristics of compounds present in a library of *S*-methyl thioesters. In addition, we report on the results of a more detailed sensory evaluation of some *S*-methyl thioesters, which were selected on the basis of this preliminary GC−olfactory assessment. It was shown that GC−sniffing of a flavor library can provide rather accurate information about the "quality" of individual

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flavors, and this was confirmed subsequently by traditional sensory analysis.

MATERIALS AND METHODS

Chemicals. Calcium hydride, citric acid, dichloromethane, hexanoyl chloride, propionyl chloride, anhydrous sodium sulfate, sodium hydrogen carbonate, and sodium thiomethoxide were obtained from Aldrich Chemical Co. (Dorset, U.K.). *S*-Methyl thioacetate and *S*-methyl thiobutanoate were supplied by Lancaster Synthesis Ltd. (Lancashire, U.K.). All solvents were of the highest purity available and distilled over calcium hydride prior to use.

Analysis of Individual Thioesters. This was performed using an HP 5890 GC (Split) System with a BPX-5 capillary column (12 m × 0.3 mm; film thickness = 0.33 μm) connected to a FID and using helium as a carrier gas at a flow rate of 0.7 mL min^{-1}. Samples were injected using a split/splitless injector, split ratio 1/100. During analysis, the injection temperature was set to 40 °C and the detector was at 370 °C. For the analysis of individual thioesters (**C2**, **C3**, **C4**, and **C6**) the oven temperature was programmed from 40 to 145 °C at 10 °C min^{-1}.

Synthesis of *S*-Methyl Thioesters. A flavor library of 11 *S*-methyl thioesters was prepared as described in the accompanying paper (Khan et al., 1999). Individual compounds were synthesized as follows.

*S-Methyl Thiohexanoate (**C6**).* Hexanoyl chloride (10.00 g, 74.29 mmol) was dissolved in dry dichloromethane (180 mL) at 0 °C, and sodium thiomethoxide (6.50 g, 92.86 mmol) was added in small portions (approximately 0.5 g) with vigorous stirring over 30 min under dry nitrogen (*Caution!* exothermic). Stirring was continued for 3 h at 0 °C followed by 16 h at room temperature. The reaction mixture was filtered, and the organic phase was washed with ice-cold 5% aqueous citric acid (100 mL) and 5% $NaHCO_3$ (100 mL). The organic layer was then dried over Na_2SO_4 and filtered and the solvent removed in vacuo (10 mmHg) at 0 °C to obtain a colorless oil which was purified further by three successive distillations under vacuum (bp 46−48 °C, 10 mmHg; lit. bp 62 °C, 15 mmHg (Vinokurov et al., 1991)) to yield *S*-methyl thiohexanoate (**C6**) as a colorless liquid (6.93 g, 64%); GC retention time = 5.42 min.

*S-Methyl Thiopropionate (**C3**).* This was prepared using the same procedure as described above to afford the thioester (**C3**) as a colorless liquid (bp 18−20 °C, 10 mmHg; lit. bp 120−121 °C, 760 mmHg (Mayer et al., 1966)); GC retention time = 2.42 min.

GC−Olfactometry. GC−olfactometry was performed independently in two different laboratories at Université Catholique de Louvain, Louvain-la-Neuve, Belgium (group 1), and INRA, Grignon, France (group 2), using the experimental protocols described below.

Group 1. This was performed using a Chrompack CP9001 gas chromatograph which was equipped with a splitless injector maintained at 250 °C and opened after 0.5 min. Compounds were analyzed using a 50 m × 0.32 mm, wall-coated open tubular (WCOT) apolar CP-SIL 5 CB capillary column (film thickness, 1.2 μm) connected to a FID detector using a standard temperature gradient as described previously (Khan et al., 1999). To assess the olfactory potential of the compounds, a T-junction was used at the end of the capillary column. Then, 50% of the eluent was sent to a FID detector maintained at 250 °C and connected to a Shimadzu CR3A integrator, while the other part was directed to a GC−odor port at 250 °C. In the latter case, the eluent was diluted with a large volume of air (20 mL min^{-1}) previously humidified using an aqueous copper(II) sulfate solution. Then, 2 μL of the *S*-methyl thioester library in dichloromethane (5 g L^{-1}) was injected to determine the flavor quality of the library components.

On the basis of ASTM 679 (ASTM, 1975) recommendations for the determination of odor thresholds using standard methods of sensory analysis, best estimate−GC−lower amount detected by sniffing (BE−GC−LOADS) was defined as the geometric mean between the lowest mass of compound perceived at the outlet of the GC−odor port and the highest undetected amount injected onto the column (Meilgaard et al., 1993). Group BE−GC−LOADS were calculated as the geometric mean of the individual BE−GC−LOADS. Experiments were performed using three independent measurements of a dichotomized dilution of a given compound as follows: a standard solution (C_0) and 75%, 50%, 25%, etc. of this initial concentration were injected onto the GC until a dilution ($C_n = C_0/2^n$) was no longer perceived at the odor port. This was then followed by injection of a mid-dilution between C_{n-1} and C_n until the compound was no longer detected at two consecutive dilutions. Sensory analysis was performed by three subjects working independently, and a verbal description of the odor was obtained at the same time.

Group 2. This was performed using a Girdel Series 30 GC instrument. Samples were applied manually by splitless injection onto a nonpolar capillary column (HP-5MS, 30.0 m × 0.25 mm, 0.25 μm film thickness, 5% diphenyl and 95% dimethylpolysiloxane) using hydrogen as a carrier gas at a flow rate of 1.6 mL min^{-1}. Compounds were detected with FID using air and hydrogen as carrier gases at flow rates of 300 and 20 mL min^{-1} respectively. The injection and detector temperatures were set at 250 °C, and the oven temperature was held at 40 °C for 2 min, increased to 250 °C at 10 °C min^{-1}, and then maintained at 250 °C for 10 min. Data were collected (2 points/s) and processed on a NEC Powermat SX Plus computer equipped with a Maxima 820 chromatography workstation.

The eluent from the chromatograph was split in a 10:90 ratio at the column exit to allow transfer of 10% to FID and 90% to the sniffing port, which contained humidified air to minimize desiccation of the nasal membrane. Two trained subjects participated in the sensory study and recorded data independently on an acquisition computer (Maxima 820 chromatography workstation). Upon initial perception, a button was pressed and held until the odor was no longer detected. At this point, the button was released and a verbal description of the compound was given by each panelist.

Initially, the flavor library was diluted with dichloromethane (1:20 v/v) and a geometrical series of eight successive 3-fold dilutions were prepared. These samples were tested by the two subjects in a random order and for each compound, the individual and group BE−GC−LOADS were determined as described for group 1 above.

Sensory Evaluation of Individual Compounds in Solution. Four of the thioesters studied by GC−sniffing of the flavor library, i.e., *S*-methyl thioacetate (**C2**), *S*-methyl thiopropionate (**C3**), *S*-methyl thiobutanoate (**C4**), and *S*-methyl thiohexanoate (**C6**), were subjected to more detailed sensory analysis. Five solutions in water (c1−c5) were prepared for each compound by 3-fold dilution of the samples which were then stored in 60 mL coded brown flasks (25 mL per flask) at 4 °C overnight. Prior to sensory evaluation by the panelists, these cooled samples were equilibrated at 20 °C for 1 h. Eighteen subjects participated in the sensory sessions, nine of which had previous experience in tasting cheese and fresh dairy products. In their own vocabulary, panelists were asked to describe the nature of the odor perceived and could provide an unlimited number of terms to describe each compound. During each sensory session, 16 samples of a single aroma compound were presented to the panelists in a sequential manner starting with a “warm-up” medium dilution (c3) followed by triplicate samples of five dilutions (c1−c5). These solutions were evaluated by the subjects in separate booths in an air-conditioned room at 20 °C under white light. Analysis of the descriptive results for each compound was performed by calculating the global quotation frequency (GQF) of each term category (for the five concentrations) by dividing the number of quotations of each term category by the total number of quotations for the compound.

Quantification of Thioesters Present in Cheese. A sample of Pont l’Evêque, Munster, mass-produced Camembert prepared from pasteurized milk (A) and a sample of long ripened raw milk Camembert (B) were purchased at a local supermarket. Each cheese (30 g) was mixed with water (20 mL) using an Ultrathurax blender (2 × 30 s), and aliquots (5 mL) were withdrawn for analysis. The volatile compounds present were isolated by a dynamic headspace analyzer (Purge and Trap concentrator, HP7695A, Hewlett-Packard, Avondale, PA) linked to a gas chromatograph (HP6850 series) equipped with a nonpolar capillary column (HP-5, 30.0 m × 0.25 mm, 0.25 μm film thickness) and a quadrupole mass spectrometer (HP 5972A).

Dynamic Headspace Analysis. Samples were heated at 60 °C for 10 min in the pocket heater of the headspace analyzer and then purged with high purity helium at 30 mL min^{-1} for 15 min. Volatile compounds were adsorbed on a Tenax trap column (30 cm × 3.2 mm, Teckmar Inc., Cincinnati, OH). The column was then heated at 225 °C for 2 min to desorb compounds which were cryofocused on the head of a capillary column maintained at −150 °C with liquid nitrogen. To limit water transfer to the GC column, the analyzer was equipped with a moisture control system, (MCS). Following analysis, the trap and the MCS were heated for 10 min at 225 and 200 °C respectively to remove possible residual compounds and water.

GC−MS Analysis. The volatile compounds which condensed on the capillary interface between the headspace analyzer and the GC were applied automatically onto the column (splitless injection, helium flow rate of 1.6 mL min^{-1}) by heating the interface to 180 °C for 1 min. The oven temperature, initially held at 5 °C for 8 min, was programmed to raise from 5 to 20 °C at 3 °C min^{-1} and then from 20 to 150 °C at 10 °C min^{-1}.

The GC column was connected directly to a HP 5972A quadrupole mass spectrometer (ion source temperature, 280 °C; ionization voltage, 70 eV; mass scan range, *m*/*e* 29−300 amu; scan rate, 1.68 scans^{-1}; electron multiplier voltage, 2400 V with no solvent delay). Spectra were recorded automatically with a MSD analytical workstation (SHP G1701AA 6890). Chromatograms were reconstituted from the total ion current and integration of the thiomethyl fragment ion (*m*/*z* [SCH_3^+] = 47). For the quantification of thioesters present in the cheese samples, reference compounds were added to a cheese curd at 0, 0.05, 0.1, 0.25, and 0.5 ppm concentrations and these standards were used to construct a calibration curve using linear regression analysis.

RESULTS AND DISCUSSION

Characterization of the *S*-Methyl Thioester Library by GC−Olfactory Analysis. To assess the flavor potency of components in the thioester library, best estimate−GC−lower amount detected by sniffing (BE−GC−LOADS) was considered to be a more suitable approach than other established methods, e.g., CHARM (Acree et al., 1984; Marin et al., 1988), OSME (Miranda-Lopez et al., 1992), and AEDA and OAV (Grosch, 1994) as it could provide quantitative information on the odor potency of a mixture of compounds by injecting a known amount of the library at the GC inlet without the requirement to establish the concentrations of individual components at the GC−odor port. Other benefits of this approach include (i) the ability to analyze a library which does not need to contain all components at exactly the same concentration, (ii) the fact that it is not necessary to determine odor threshold values for individual compounds, and (iii) the ability to normalize the odor potency of the thioesters as the molar ratios of the library components are known (Khan et al., 1999).

The odor potency (BE−GC−LOADS) and individual flavor characteristics of the constituent compounds of a library of *S*-methyl thioesters were determined independently in two laboratories (groups 1 and 2) using

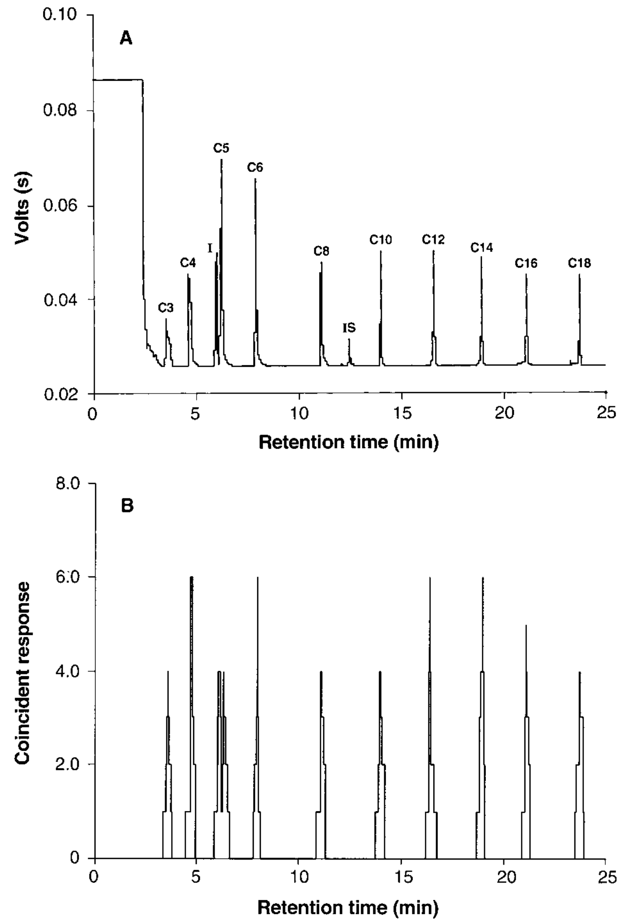

Figure 1. Comparison of FID−GC chromatogram (a) and aromagram (b) of the *S*-methyl thioester library: *S*-methyl thioacetate (**C2**), *S*-methyl thiopropionate (**C3**), *S*-methyl thiobutanoate (**C4**), *S*,*S*-dimethyldithiocarbonate (**I**), *S*-methyl thiopentanoate (**C5**), *S*-methyl thiohexanoate (**C6**), *S*-methyl thiooctanoate (**C8**), internal standard, tetradecane (**IS**), *S*-methyl thiodecanoate (**C10**), *S*-methyl thiododecanoate (**C12**), *S*-methyl thiotetradecanoate (**C14**), *S*-methyl thiohexadecanoate (**C16**) and *S*-methyl thiooctadecanoate (**C18**). The aromagram was obtained by height sensory experiments performed by group 2, and the peak height is proportional to the degree of dilution for individual flavor components.

similar but not identical techniques (see Materials and Methods). This detailed assessment by two independent groups was desirable to minimize differences in the description of flavor which are inevitably associated with these types of experiments. GC analysis of the flavor library (FID detection) was entirely consistent with previous results (Figure 1a) and showed the presence of a single impurity, *S*,*S*-dimethyl dithiocarbonate (Khan et al., 1999). Crucially, the olfactory purity of the thioester library was confirmed by comparison of the FID−GC chromatogram with aromagrams (Figure 1b) obtained in a 3^4−3^6 dilution range (Acree et al., 1984).

GC-sniffing was then performed to obtain BE−GC−LOADS values, and the results obtained by both groups are summarized in Figure 2. It should be noted that both groups found it difficult to assess *S*-methyl thioacetate (**C2**) and *S*-methyl thiopropionate (**C3**) because these compounds were eluted from the column too soon after the solvent peak. No attempt was made to adjust the analytical conditions as *S*-methyl thioacetate had already been well characterized in the literature (Cuer

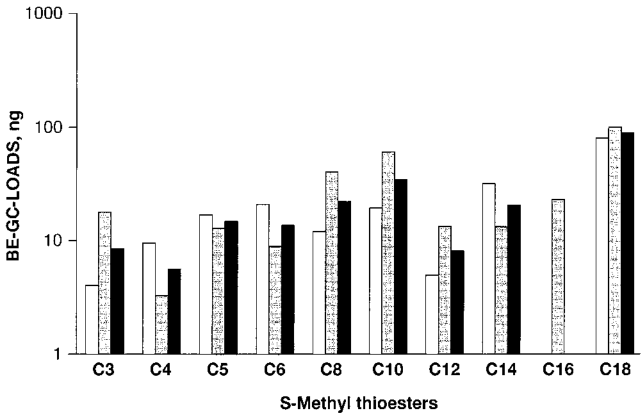

Figure 2. Variation in best estimate−GC−lower amount detected by sniffing (BE−GC−LOADS) as a function of thioester chain length during independent sensory assessment by groups 1 (white) and 2 (gray) (see Materials and Methods), and as the geometric mean of both studies (black).

et al., 1979; Wyllie et al., 1994), and we used the two compounds in a further sensory study (see below). Also, both groups observed an overall increase in BE−GC−LOADS values from *S*-methyl thiobutanoate (**C4**) to *S*-methyl thiodecanoate (**C10**), i.e., thioesters of intermediary carbon chain length. The apparent “break” in this trend for longer carbon chain length thioesters (**C12**−**C16**) could be attributed to their nauseating properties, which may have lead to a considerable underestimate in the BE−GC−LOADS values of these compounds.

Inevitably, there was some variation in BE−GC−LOADS values calculated from the data obtained with different subjects which is common in sensory analysis (Meilgaard et al., 1993). However these variations may also be explained by slight differences in experimental procedures used by the laboratories. For example, a blind random sampling technique was employed by group 2, while in group 1 subjects performing the analysis were offered dichotomized dilutions of samples (see Materials and Methods). In general, given the qualitative nature of the tests there was good overall correlation between the results obtained in the two laboratories in different countries.

During the analysis, the subjects were also asked to provide a qualitative description of the flavor perceived (Table 1). All three assessors in group 1 described the **C2**−**C6** thioesters as “cheesy” and this was the descriptor most frequently used. However, particular differences were also noted including the pleasant “floral” and “pineapple” notes of *S*-methyl thiohexanoate (**C6**). It is worth noting that *S*-methyl thiopropionate (**C3**), *S*-methyl thiobutanoate (**C4**), *S*-methyl thiopentanoate (**C5**), and *S*-methyl thiohexanoate (**C6**) were all described as “cheesy” and occasionally as “Camembert”, while the descriptor most frequently associated with the longer chain thioesters was “rancid”. Overall there was very good agreement between the three subjects in group 1 and most encouragingly, a very similar flavor description was obtained independently by group 2 (Table 1). The two assessors in group 2 also registered the “floral” and “green” notes of *S*-methyl thiohexanoate (**C6**) and used descriptors “cheese” for the **C3**−**C5** thioesters whereas the longer chain thioesters were attributed with “vegetable” notes (“cauliflower” and “cabbage”), although the term “rancid” was used too. Surprisingly, among the higher molecular weight

Table 1. Qualitative Description of *S*-Methyl Thioesters by GC Sniffing of the Flavor Library

thioester	flavor descriptor: group 1	flavor descriptor: group 2
C2	cheese (Herve), rotten cabbage	
C3	cheese, hydrogen sulfide	ripened cheese, Camembert
C4	Herve, Gruyère	ripened cheese, Munster
C5	cheese (Camembert)	cheese rind, cheese
C6	cabbage, well matured Camembert, rancid, floral, pineapple	floral, green (grass, matured pineapple)
C8	rancid, foot	cooked cabbage, cheese
C10	rancid	cabbage, dust
C12	nauseating, irritating	cauliflower
C14	rancid, cheese	cabbage, rancid, cheese
C18	paint, solvent	burnt smell

thioesters, both groups identified *S*-methyl thiotetradecanoate (**C14**) in particular as being “cheesy.”

Characterization of Selected *S*-Methyl Thioesters by Traditional Sensory Analysis. Having established the feasibility of library assessment by GC−sniffing, we turned our attention to those compounds which were noted by the subjects as interesting or peculiar (“hits”). In particular, we selected *S*-methyl thiopropionate (**C3**) and *S*-methyl thiohexanoate (**C6**) as targets for a more detailed investigation and compared their properties with two other thioesters, *S*-methyl thioacetate (**C2**) and *S*-methyl thiobutanoate (**C4**), which are commercially available and relatively well characterized (Cuer et al., 1979; Wyllie et al., 1994). Approximately 120 different descriptive terms were generated by the panel to describe the samples, and as requested, no hedonic term was proposed. These terms varied significantly both in nature and respective numbers, and to facilitate interpretation of these results, they were regrouped into seven categories which included those most frequently mentioned (Table 2).

The total number of quotations for the four thioesters studied was 182 (**C2**), 195 (**C3**), 159 (**C4**), and 216 (**C6**). **C3** and **C6** presented the highest number of quotations, despite the absence of two subjects during this evaluation. The importance and ranking of the term categories according to their quotation frequency was quite different too (Figure 3). “Cabbage” and “cheesy” appeared to be the most frequent for **C2**, **C3**, and **C4**, and “cabbage” accounted for almost 45% of all the quotations for **C2**. “Cabbage” remained the most frequently used descriptor for **C3** and **C4** (~30%), but in these two cases, the difference between “cabbage” and “cheesy” was minor. Most interestingly however, the “cheesy” notes in the odor of *S*-methyl thiopropionate (**C3**) were to a large extent associated specifically with Camembert: 40 quotations as compared to just four and nine for **C2** and **C4**, respectively. The corresponding descriptions for these two latter compounds were much less precise (cheese, ripened cheese) and included other cheese varieties (Munster for **C2** and **C4** and Roquefort, Gruyère, and Parmiggiano for **C4**). The “rancid” category appeared in the third position according to the quotation frequency order for **C2** and **C4** and in the fourth for **C3**. In percentage terms, **C4** was perceived as more “rancid” than all the other thioesters. Garlic accounted for 24% of all the quotations referring to the odor of **C3**. Rather different results were obtained with

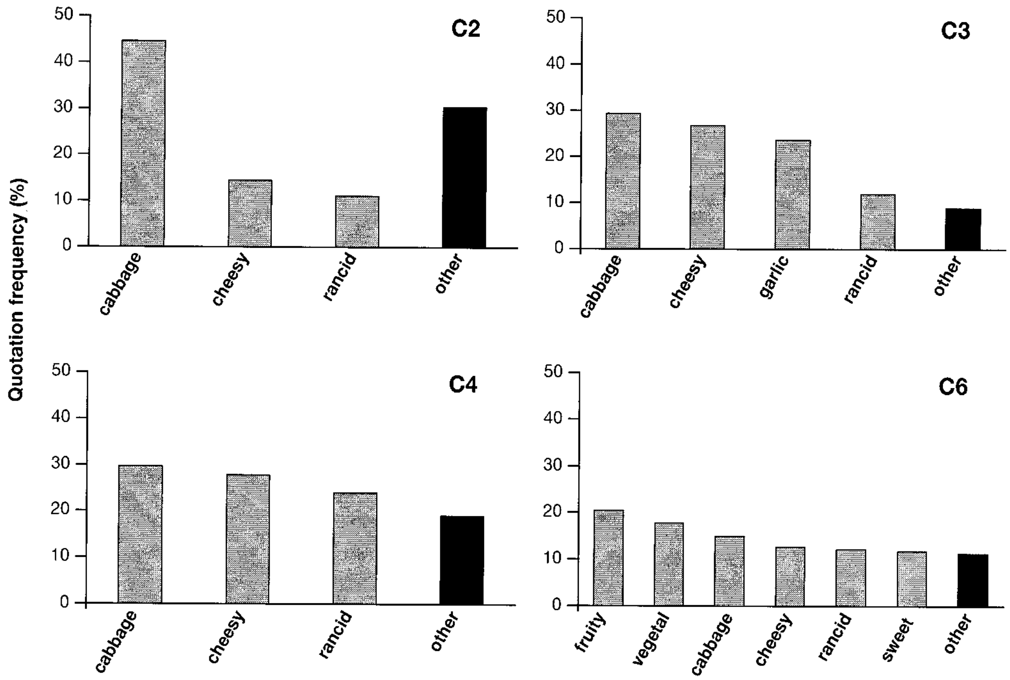

Figure 3. Quotation frequency of the most commonly mentioned descriptor terms used during sensory analysis of *S*-methyl thioacetate (**C2**), *S*-methyl thiopropionate (**C3**), *S*-methyl thiobutanoate (**C4**), and *S*-methyl thiohexanoate (**C6**) as individual compounds.

Table 2. Regrouping of the Descriptive Terms Obtained in Sensory Analysis for the Seven Categories Mentioned Most Frequently

category	descriptive term
garlic	*garlic*: cooked garlic
cheesy	*cheesy*: butter; milk; Camembert; Munster; Boursin; Parmigiano; Gruyère; ripened cheese; Camembert rind
cabbage	*cabbage*: cooked cabbage; uncooked cabbage; cauliflower; cooked cauliflower; fermented cabbage; sauerkraut; Brussels sprouts
fruity	*fruity*: strawberry; melon; blackberry; floral; violet; citrus fruit; acid; lemon; orange; grapefruit; pineapple
rancid	*rancid*: rancid butter; rotten; vomit; stale; urine; ammonia; hydrogen sulfide; garbage; rotten egg; egg; vinegar; Gruyère rind; sour-pickles; rotten; stable
vegetal	*green*: fresh; mint; menthol; grass; fennel; vetiver; camphor
sweet notes	anise; licorice; chocolate; vanilla

S-methyl thiohexanoate (**C6**). The "fruity" and "vegetal" categories were much more frequently used than the "cabbage," "cheesy", and "rancid" ones. Also, **C6** was the only compound to which "sweet" notes were attributed.

This study clearly indicated that the initial assessment of the thioester flavor library by GC−sniffing was largely correct. The "green" or "fruity" notes of *S*-methyl thiohexanoate (**C6**) detected by the subjects involved in the GC−sniffing experiments was unambiguously confirmed by the panel. The results of tests performed on *S*-methyl propionate (**C3**) were also very interesting. The "cheesy" note was unequivocally confirmed, but in addition, the majority of the panelists clearly associated the odor of this compound with Camembert, while the descriptors used for other thioesters were by far less precise (Munster, Gruyère, Parmiggiano, and Camembert were all mentioned). It was tempting to suggest that *S*-methyl thiopropionate (**C3**) is, at least partially, responsible for the characteristic flavor of mature Camembert, and if so, it could perhaps be used as a specific "chemical signature" for this cheese variety. To verify this conclusion, extracts from several cheeses were studied to determine their *S*-methyl thiopropionate (**C3**) content.

Analysis of Cheese Extracts. Two samples of Camembert were investigated, a mass produced cheese made from pasteurized milk (A) and another Camembert prepared by traditional methods (long ripened) from raw milk (B). In both cases, volatile compounds were extracted and analyzed using a dynamic headspace analyzer coupled to GC−MS (see Materials and Methods). Much to our delight, virtually no (<1 ppb) *S*-methyl thiopropionate (**C3**) was found in cheese A while in the traditional Camembert (B), the same compound was present at a concentration of 0.123 ppm. In a control experiment, we analyzed samples of Munster and Pont l'Evêque as these cheeses were also mentioned by the panelists and may have contained *S*-methyl thiopropionate (**C3**). Munster was shown to contain 0.032 ppm of *S*-methyl thioacetate and only 0.0061 ppm of *S*-methyl thiopropionate (**C3**), i.e., 20 times less than long matured Camembert (B) and only very small quantities of both thioesters were found in Pont l'Evêque. It is interesting to note that the amounts of *S*-methyl thiopropionate (**C3**) present (123 ppb in Camembert) and *S*-methyl thioacetate (**C2**) (32 ppb in Munster) are indeed significant in terms of their contribution to aroma as the thresholds of these compounds in dairy products are 100 and 5 ppb respectively (Cuer et al., 1979).

CONCLUSION

In conclusion, we have shown that GC−sniffing, using BE−GC−LOADS measurements, is a powerful tool for the characterization of both odor potency and flavor characteristics of a homologous series of compounds as evident from the assessment of the eleven component library of *S*-methyl thioesters. The "impression" of panelists which performed the initial assessment by GC−sniffing was confirmed unequivocably by subsequent investigation of selected compounds using a trained panel consisting of 18 subjects. Furthermore, following the analysis of flavor descriptors provided by the panel, *S*-methyl thiopropionate (**C3**) was identified in particular as a possible characteristic aroma of Camembert. This compound was indeed detected in relatively large quantities in long-ripened Camembert but not in the cheese which was mass produced from pasteurized milk. In general, the results obtained in the course of this work suggest that combinatorial libraries can be successfully used for the characterization of flavor compounds as well as for the identification of specific aromas in food products.

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