Sensory Investigation of Yogurt Flavor Perception: Mutual Influence of Volatiles and Acidity

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The sensory properties of traditional acidic and mild, less acidic yogurts were characterized by a trained panel using a descriptive approach. Many of the descriptive attributes varied almost linearly with pH, showing either a positive or negative correlation with increasing acidity. The panel was very sensitive to acidity differences, as demonstrated by the linear relationship between acidity perception and pH. Important flavor differences were found between the two classes of yogurt. They were mainly due to differences in acidity and not to different concentrations of the three impact aroma compounds, acetaldehyde, 2,3-butanedione, and 2,3-pentanedione. This emphasizes the importance of acidity in yogurt flavor. Deodorization and impact aroma compound addition had much less influence on yogurt flavor than pH variations.

Keywords: *Sensory evaluation; fermentation; milk; yogurt; flavor; acidity; aroma; volatiles; taste*

INTRODUCTION

Consumer preference for food is driven by many criteria and especially by its flavor. Plain yogurt has a weak but distinctive and fragile flavor that is influenced by different factors, for example, viscosity, nonvolatile components, and aroma. We recently showed that yogurt aroma is a superposition of both volatiles initially present in milk and compounds produced during fermentation (Ott et al., 1997). Correct ratios among the different key compounds are essential for a balanced aroma.

To our knowledge only a limited number of sensory studies on yogurt flavor are available in the literature. Muir and Hunter (1992) developed a vocabulary for fermented milks and found a loose relationship between pH and a number of sensory factors such as flavor intensity, acidity/sourness, bitterness, lemon flavor, rancidity, and bitter aftertaste. These trials were, however, on commercial products. Harper et al. (1991) showed that perceived sourness (trained panel) was correlated with acceptance (consumer panel) of the samples, with a general preference for the less acidic ones. Rohm et al. (1994) found a positive correlation between flavor and acidity and attributed this effect to a coupled acid and acetaldehyde production. Viscosity and ropiness, on the other hand, correlated negatively with acidity and flavor. Kneifel et al. (1992) found acetaldehyde to most prominently participate in the typical yogurt aroma; products with acetaldehyde levels <10 ppm were generally rated as “low” in flavor intensity. These samples also showed rather low acidity values, with one exception. They concluded from this finding that acetaldehyde is an important aroma compound but that acidity and degradation of proteins also influence the flavor.

In recent years, consumers have shown a preference for milder, less acidic yogurts (Hunger, 1985; Eberhard et al., 1995). Such products were, however, rated less flavorful (Kneifel, 1992; Kneifel et al., 1992). In mild, less acidic yogurts produced with β-galactosidase negative strains (lac$^-$) of *Lactobacillus bulgaricus*, we recently detected important differences in the concentrations of three impact aroma compounds: acetaldehyde concentration was decreased and 2,3-butanedione and 2,3-pentanedione concentrations were increased compared to a traditional acidic yogurt (Ott et al., 1999). These mild yogurts also contained less lactic acid. As the mild yogurts were judged to have less typical yogurt flavor, we focused our interest on the influence of (1) pH and (2) the three impact aroma compounds mentioned above, as well as a combination of both criteria, on the perception of the yogurt flavor. Our previous quantitative results (Ott et al., 1999) showed that acetaldehyde content and growth potential of lactic acid were correlated. As growth generates acidity, it was unclear whether acetaldehyde and acidity were simultaneously required to impart an intense flavor to the yogurt. To answer this question, the present study uses a descriptive sensory approach and was conducted with the same microorganisms as in the previous investigation so that analytical results can be related to sensory data. Variations of pH as well as deodorization and volatile addition were investigated to separately determine the contribution of acidity and volatiles to yogurt flavor.

EXPERIMENTAL PROCEDURES

Chemicals. Acetaldehyde and 2,3-pentanedione were from Merck (Merck AG, Les Acacias, Switzerland) and 2,3-butanedione, L-threonine, glycine, and DL-lactic acid (~90%) from Fluka (Fluka AG, Buchs, Switzerland). They were of analytical grade.

Preparation of Fermented Milk Samples. *Microorganisms and Strains.* The following microorganisms and strains from the Nestlé culture collection were used (Table 1).

Fermentation. Fermented milk samples were prepared using the strains of *Lactobacillus delbrueckii* ssp. *bulgaricus* and *Streptococcus thermophilus* listed in Table 1 with fermentation conditions and milk preparation as previously described (Ott

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Table 1. Properties of the *S. thermophilus* and *L. delbrueckii* Subsp. *bulgaricus* Strains Used in This Study

bacterial strain		properties	abbrev
S. thermophilus	YS4+ YS7	Lac $^+$, nonropy, slow acidifier	slow St
	YS33	Lac $^+$, nonropy, fast acidifier	fast St
L. bulgaricus	YL30	Lac $^+$, slightly ropy, fast acidifier	Lac$^+$ Lb
	LB52	Lac $^-$ mutant of YL30	
	LFi5	Lac $^+$, ropy, fast acidifier	
	LFi31	Lac $^-$ mutant of LFi5	
mixed cultures[a]	YL30 + YS4 + YS7		mix lac$^+$YL30
	LFi5+ YS4 + YS7		mix lac$^+$LFi5
	LB52 + YS4 + YS7		mix lac$^-$LB52
	LFi31 + YS4 + YS7		mix lac$^-$LFi31

[a] Mixed cultures: 0.5% (v/v) starter of each *S. thermophilus* YS4 and YS7 and 1% (v/v) starter of *L. delbrueckii* ssp. *bulgaricus* (YL30, LFi5, LB52, or LFi31).

Table 2. Sensory Descriptors Used by the Panel for the Characterization of Fermented Milks

odor	appearance and texture on the spoon	flavor	mouthfeel	aftertaste
milky	yellowish	creamy	light	persistent
yogurt	bubbles	buttery	thick	milky
cottage cheese	heterogeneous	cottage cheese	floury	sour milk
sour milk	compact	acid	sandy	acid
pungent	lumpy	sweet	small lumps	lemon
onion	thick	cooked		astringent
		bitter		bitter
		astringent		
		sour milk		

et al., 1997, 1999). Fermentation was performed in 2 L closed glass bottles containing 1 L of milk each. Glycine (40 mg/L) or l-threonine (120 mg/L) was added to hot milk after pasteurization and before or after fermentation. All samples were stored for 2 weeks at 4 °C in the dark and homogenized by vigorous shaking prior to sensory tests. The pH of fermented milk samples was adjusted to the desired values either with DL-lactic acid (Fluka) or with NaOH (6 M).

The term yogurt is used throughout this paper when both *L. bulgaricus* and *S. thermophilus* were used for fermentation.

Deodorization of Yogurt Samples. Traditional acidic yogurt was deodorized in a rotary evaporator apparatus with an automated vacuum control (both Büchi AG, Flawil, Switzerland) using the following parameters: temperature of water bath, 50 °C; vacuum conditions, 15 min at 150 mbar, then 15 min at 100 mbar, and finally 30 min at 50 mbar. Rotation was at maximal speed to minimize foaming. The amount of lost water was replaced by the same quantity of distilled water (Fontavapor, Büchi).

Nondeodorized samples were subjected to similar stirring in a rotary evaporator, under the same temperature, but the sample was previously hermetically sealed and no vacuum was applied. The pH of fermented milk samples was adjusted to pH 4.5 with NaOH (6 M).

Analysis of the Samples. Fermented milk samples were analyzed for volatiles according to previously reported methods: static headspace gas chromatography (S-HS-GC) for acetaldehyde [procedure 2 of Ott et al. (1999)]; static-and-trapped headspace gas chromatography (S&T-HS-GC) for all other volatiles [procedure 1c of Ott et al. (1997)].

Addition of Volatiles. Acetaldehyde, 2,3-butanedione, and 2,3-pentanedione were added as aqueous solutions to the deodorized samples. The volume of introduced water was corrected accordingly for all samples. Final concentrations in the deodorized samples were 15, 1, and 0.1 ppm, respectively.

Sensory Evaluation. In this paper, odor, taste, and flavor terms will be used with ISO meaning (Technical Committee ISO, 1992).

Odor refers to the organoleptic attribute perceptible by the olfactory organ on sniffing certain volatile substances.

Taste refers to sensations perceived by the taste organ when stimulated by certain soluble substances

Flavor refers to a complex combination of the olfactory, gustatory, and trigeminal sensations perceived during tasting.

The term "*aroma*" is used herafter, like the *odor* term, without any hedonic aspect.

Panel Selection and Training. Twelve panelists were selected among the staff of the Nestlé Research Center (NRC), on the basis of their ability to recognize basic tastes in solution and their availability and interest for taking part in this study. Training consisted of five sessions. During the first session, after a brief explanation of training and testing procedures, panelists were presented with four samples of yogurt prepared with some of the strains mentioned above and were requested to list the terms appropriate to describe the appearance, texture with the spoon, smell, flavor, mouthfeel, and aftertaste of the samples. After each sample, each panelist read out and explained the terms he/she had found. After the fourth sample, a total of 55 terms had been collected. The following four sessions were used to (a) expose panelists to more yogurt samples, and possibly identify new terms; (b) present other dairy products (e.g., cottage cheese, kefir) to help panelists characterize some specific descriptive terms; (c) reduce the total number of terms by eliminating redundant ones or those for which the panel could not reach a consensus; (d) agree on precise definitions of the terms and on the tasting protocol; and (e) practice the use of the rating scale and make sure that panelists rated samples coherently.

After the fifth session, the panel had agreed on a list of 33 clearly defined terms (Table 2) and on how to perform the evaluation.

Sample Preparation and Tasting Protocol. Fermented milk samples were delivered to the sensory laboratory in the glass bottles used for fermentation and kept refrigerated at 4 °C until needed. To prepare the samples, the bottles were vigorously shaken until the yogurt was homogeneous, and then it was poured into small glass pots, which were closed to retain volatiles. Samples were left for 20 min to reach room temperature before serving.

In each session, four samples were presented monadically to panelists with random three-digit codes and in balanced presentation order. Panelists were requested to open the lid and to evaluate first the smell attributes and then the appearance and texture with the spoon. After putting some sample in the mouth, they rated flavor and texture (in-mouth) attributes. Finally, 10 s after having swallowed the sample, they evaluated the aftertaste attributes. Between each sample,

Table 3. Tested Samples of Fermented Milks, Their pH, and Concentration of Three Impact Aroma Compounds

			final concentration (mg/L)		
trial	sample[a]	pH	acetaldehyde	2,3-butanedione	2,3-pentanedione
1	slow St	4.9	3.8	0.8	0.08
2	fast St	4.5	13.7	1.1	0.10
3	Lac$^+$ Lb	4.1	12.6	0.5	0.21
4	mix lac$^+$LFi5	4.2	7.4	1.5	0.16
5	mix lac$^+$YL30	4.1	16.6	1.4	0.11
6	mix lac$^+$YL30 + 3.5 mg/L 2,3-butanedione[b]	4.2	16.6	5.0	0.12
7	mix lac$^+$YL30 + 8.5 mg/L 2,3-butanedione[b]	4.2	16.6	10.0	0.08
8	mix lac$^+$YL30 + 40 mg/L glycine[c] + 1.5 mg/L 2,3-butanedione[b] + 0.1 mg/L 2,3-pentanedione[b]	4.2	5.0	3.0	0.24
9	mix lac$^+$YL30 + 40 mg/L glycine[c]	4.2	5.0	1.4	0.08
10	mix lac$^+$YL30 + 40 mg/L glycine[b]	4.3	16.6	1.4	0.13
11	mix lac$^-$LFi31	4.4	3.8	1.9	0.23
12	mix lac$^-$LB52	4.4	2.4	2.9	0.20
13	mix lac$^-$LB52 + 120 mg/L L-threonine[c]	4.6	15.0	2.9	0.21
14	mix lac$^-$LB52 + 120 mg/L L-threonine[b]	4.5	2.4	2.9	0.18

[a] Abbreviations according to Table 1. [b] Addition after fermentation. [c] Addition before fermentation.

panelists were instructed to cleanse their palates with distilled water and unsalted crackers.

Data Handling and Analysis. The computerized data acquisition system FIZZ (Biosystems, Couternon, France) was used to collect intensity ratings on-screen. Each attribute was associated with a 12 cm unstructured linear intensity scale with two anchors at 3 mm from each extremity.

Rating marks on the scale were converted to numerical values (left anchor = 0; right anchor = 100) and given in percent. Data were processed either with statistical routines of the FIZZ program or with commercial statistical software NCSS 6.0 (NCSS, Kaysville, UT). The smooth, nonparametric curves were computed using the local regression method “Loess” in S-PLUS 2000 (MathSoft, Seattle, WA).

RESULTS AND DISCUSSION

In a previous paper (Ott et al., 1999) we reported important differences in impact aroma compounds for milks fermented with different strains of *L. bulgaricus* and *S. thermophilus*. Yogurts produced with the lac$^-$ mutants of *L. bulgaricus*, in which the β-galactosidase gene is inactivated, showed important differences in the impact aroma compounds acetaldehyde, 2,3-butanedione, and 2,3-pentanedione compared to their parent strains (lac$^+$) containing an intact β-galactosidase gene (Table 3, trials 4, 5 and 11, 12). Our interest was therefore focused on sensory description of these different samples with the aim of correlating the analytical results with sensory description. The trials were structured in three main parts as shown in Figure 1.

Influence of Microorganisms, Their Properties, and Volatiles on Yogurt Flavor. Fermented milk samples produced with the same lot of milk but different microorganisms and strains were described using the 33 attributes created by the panel (Table 2). These samples had a wide pH range from 4.1 to 4.9 covering most of the fermented milks and yogurts found on the market as well as very different contents of volatiles (Table 3). On the basis of literature results, acetaldehyde production by the microorganisms was increased by threonine and decreased by glycine supplementation (Table 3, trials 8, 9, and 13) (Lees and Jago, 1976; Marranzini et al., 1989; Wilkins et al., 1986). 2,3-Butanedione and 2,3-pentanedione levels in the different samples, either different naturally or due to direct addition, should reveal the importance of these compounds. The influence of lac$^+$ and lac$^-$ strains of *L. bulgaricus*, containing a functional and nonfunctional β-galactosidase gene, respectively, is shown in trials 4 and 5 and 11 and 12. Trials with milk fermented with either *S. thermophilus* or *L. bulgaricus* alone compared to mixed cultures should give information about the respective contribution of both microorganisms (trials 1, 3, 5, and 12).

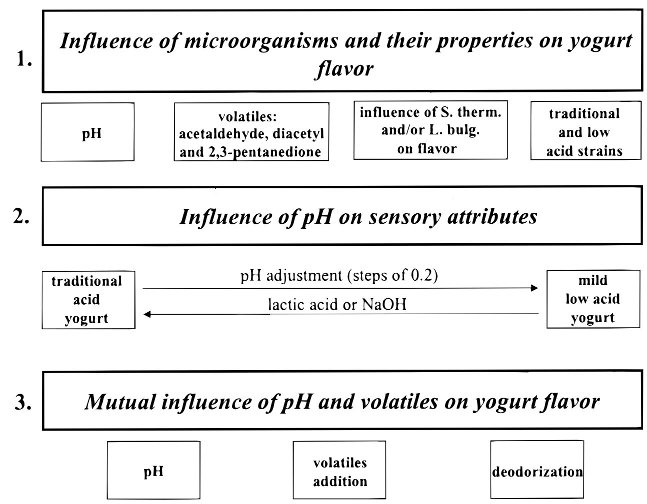

Figure 1. Overview of trials.

Influence of pH on Flavor. The intensity of the attributes buttery, sweet, cooked, creamy, cottage cheese, and milky aftertaste showed a positive correlation with increasing pH (Figure 2). The flavor descriptors astringent, bitter, and acid as well as the aftertastes persistent, astringent, and lemon showed a positive correlation with decreasing pH (Figure 2). Smooth trend lines in Figure 2 are added to improve the visual perceptions of the associations. They do not represent a model of the relationship (Cleveland, 1993).

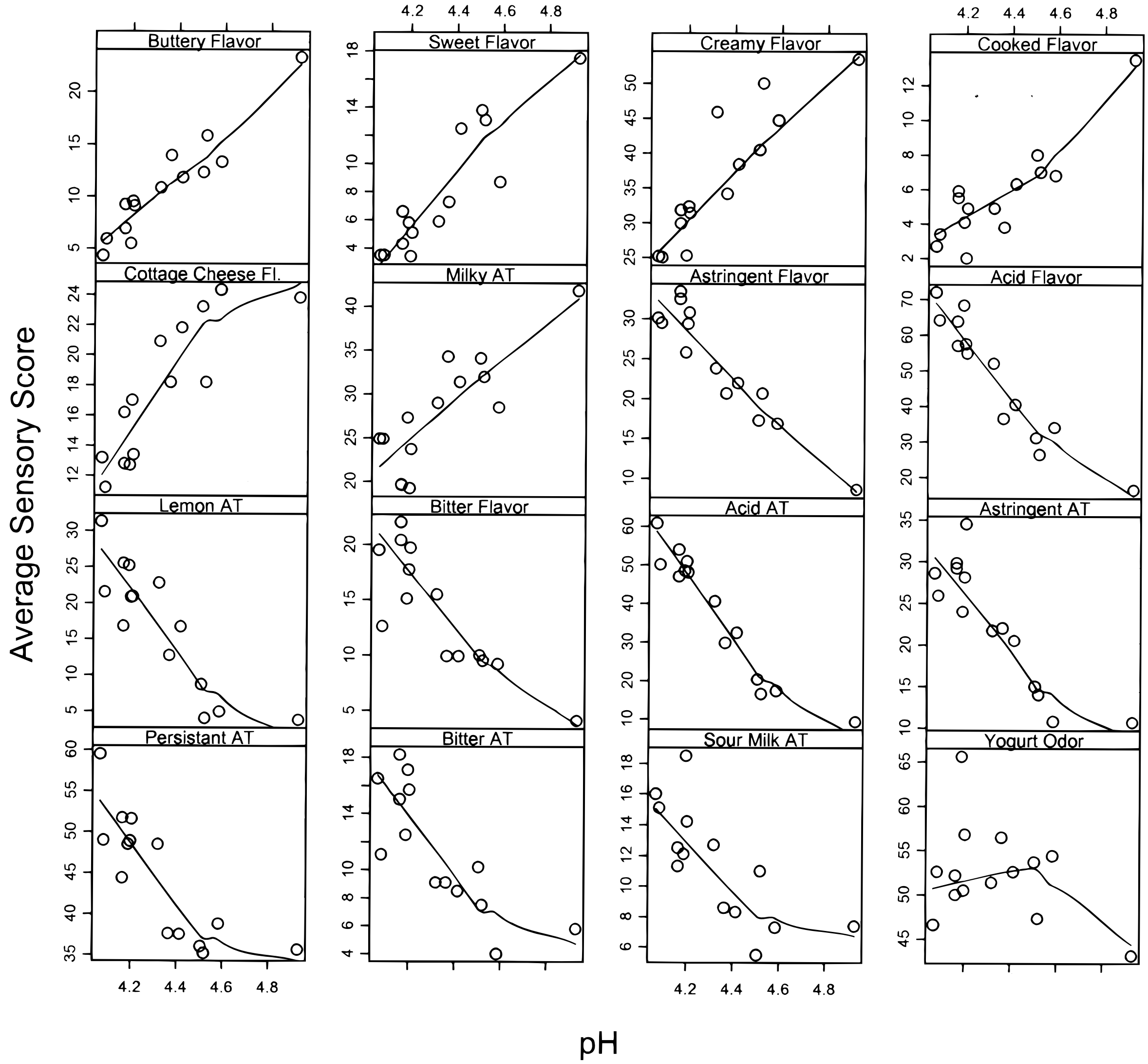

Figure 2. Correlation of pH with the sensory attributes of all the fermented milk samples listed in Table 3 in decreasing order of their correlation coefficient from left to right. AT, aftertaste; Fl, flavor.

The graphics highlight that the pH correlates very well with almost all attributes, except yogurt odor. Most of the relationships are clearly linear. The simple linear regression of acid flavor and pH, for example, cannot be improved significantly by a second-order polynomial model, but such models have to be interpreted carefully because of sample 1, which has a pH of 4.93, clearly higher than those of the other samples; such a point has a strong impact on regression models.

Figure 3 compares perceived sensory intensities for two traditional acid (Table 3, trials 4 and 5) and two mild, low-acid (trials 11 and 12) yogurts. The two samples belonging to the same group were very similar in most of the attributes. Only a slight difference in yogurt odor between the two acid samples and in sweetness between the two mild samples was observed. Intergroup differences were clearly distinguishable with higher ratings for buttery, creamy, cottage cheese, and sweet flavor attributes in the mild, less acidic group of yogurts. Acid and astringent tastes as well as persistent and acid aftertastes were, not surprisingly, rated higher in the acidic group. Differences between the two groups are in full agreement with the observation that acidity strongly influences many of the sensory attributes.

Influence of Volatiles on Flavor. Varying acetaldehyde, 2,3-butanedione, and 2,3-pentanedione concentrations of the different samples listed in Table 3 did not significantly influence aroma intensity (data not shown). It was clearly shown that the influence of pH on flavor perception was predominant over flavoring concentration, despite the wide concentration range of these three key compounds (Table 3). Increase or depletion of acetaldehyde formation by L-threonine and glycine addition, respectively, in the concentrations used, did not influence the sensory perception of the samples (data not shown).

To obtain a synthetic view of the overall sensory data, principal component analysis (PCA) of significantly different attributes (according to ANOVA) was carried out (on the correlation matrix). The resulting Biplot of the first two components (Figure 4) shows that the main variation perceived by the panel is in the attribute acid (77% of the total variation): samples on the right-hand side are high in the flavor attributes acid, bitter, and astringent and in the aftertastes of sour milk, bitter, persistent astringent, acid, and lemon. Those on the left-hand side are higher in the flavor scores sweet, creamy, buttery, cooked, and cottage cheese and have a milky

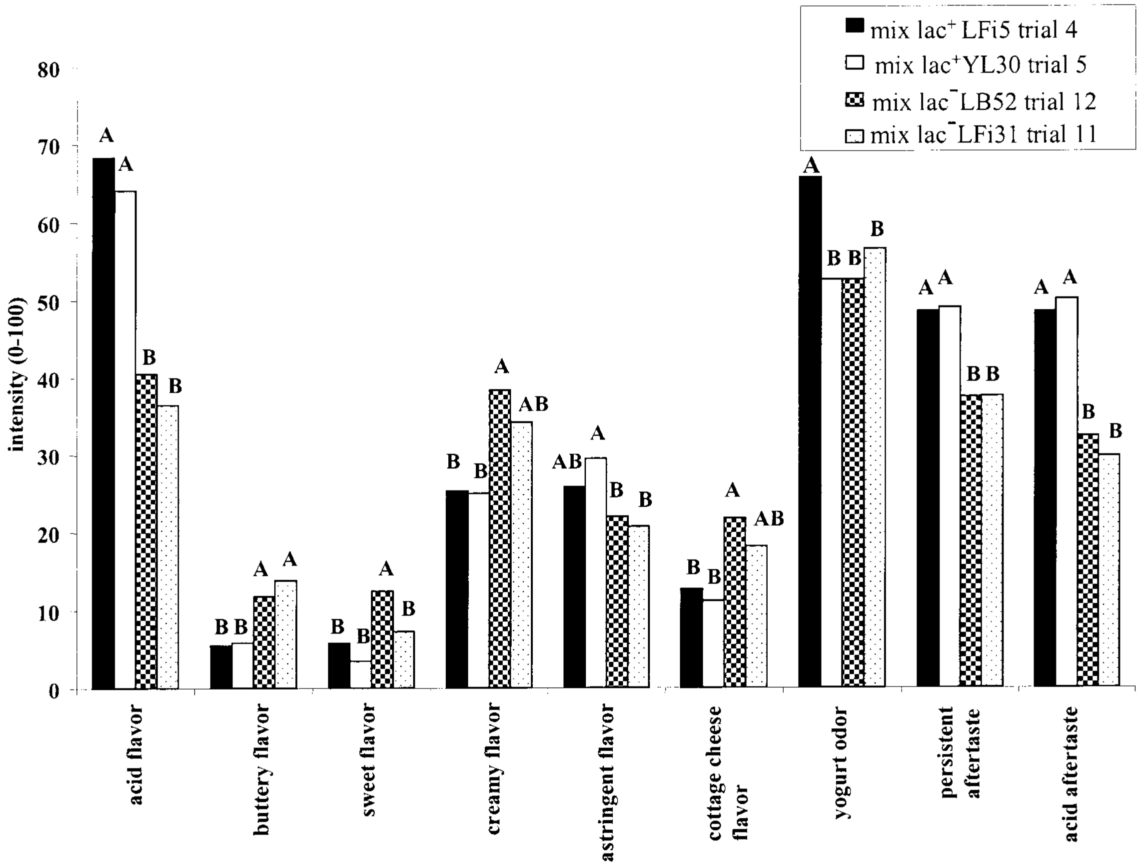

Figure 3. Attribute comparison of traditional acidic (mix lac$^+$) (Table 3; trials 4 and 5) and mild, less acidic (mix lac$^-$) yogurts (Table 3; trials 11 and 12). Only odor, flavor, or aftertaste attributes for which an ANOVA on these four products showed a significant difference ($P \leq 0.05$) are displayed. The difference between samples sharing the same letter is not significant (Duncan, $P = 0.05$).

aftertaste. Only trial 4 stands out, having a very high score in yogurt odor, compared to the other samples. However, as this sample does not contain higher concentrations (Table 3) of acetaldehyde, 2,3-butanedione, and 2,3-pentanedione, the increase of yogurt odor cannot be attributed to these compounds.

As traditional acidic and mild yogurts show important differences in acetaldehyde content and acidity, the following question had to be answered: Is the yogurt flavor influenced by the acidity, the acetaldehyde content, or both? Acetaldehyde concentration in the traditional acidic and mild, less acidic yogurts was therefore modified by addition of glycine (acetaldehyde ↓) or L-threonine (acetaldehyde ↑) to the samples before fermentation.

In this way from each of the traditional acidic and the mild, less acidic groups, yogurt samples containing low and high levels of acetaldehyde were produced (trials 5 and 9 for traditional acidic and trials 12 and 13 for mild, less acidic yogurts, containing 16.6, 5.0, 2.4, and 15.0 ppm of acetaldehyde, respectively). Sensory differences between the two groups remained, even when the acetaldehyde levels were similar. Descriptor intensities for a yogurt type were not significantly influenced by a change in acetaldehyde content (Figure 5). Most importantly, acetaldehyde concentration did not influence overall yogurt odor intensity. Acetaldehyde was judged to be a very important compound for yogurt flavor by many investigators during the past decades (Pette and Lolkema, 1950; Keenan and Bills, 1968; Bottazi and Dellaglio, 1967). Our results do not confirm this opinion. They rather suggest acidity (pH) contributes most importantly to yogurt flavor.

To obtain the desired concentration in the three aroma compounds, diketones were directly added to the traditional acidic yogurt (trial 5), and the acetaldehyde production was decreased by adding glycine (Marranzini et al., 1989). These modifications (trial 8) did not influence the final pH but led to levels of these three compounds almost identical to those in the untreated mild, low acidic yogurt (trial 12). Comparison of this sample (trial 8) with the untreated acidic yogurt (trial 5) showed only a minor difference in sweetness (data not shown). This result suggested that the differences found between traditional acidic and low-acid yogurts can be mainly attributed to the difference in pH and not to different amounts of the three key compounds, acetaldehyde, 2,3-butanedione, and 2,3-pentanedione.

Addition of different amounts of only 2,3-butanedione to traditional acidic yogurt yielding final concentrations of 5 and 10 ppm of this compound did not significantly influence the sensory attributes (trial 5 versus 6 and 7; data not shown). Considering the amount of 2,3-butanedione added, which cannot normally be reached in a traditional acidic yogurt by microbial action, it must be concluded that this compound has a rather small effect on the sensory attributes.

Influence of L. bulgaricus and/or S. thermophilus on Flavor. The cooperative growth of *S. thermophilus* and *L. bulgaricus* shortens the fermentation time in milk and could also influence the sensory properties of the final product. We were, therefore, interested to inves-

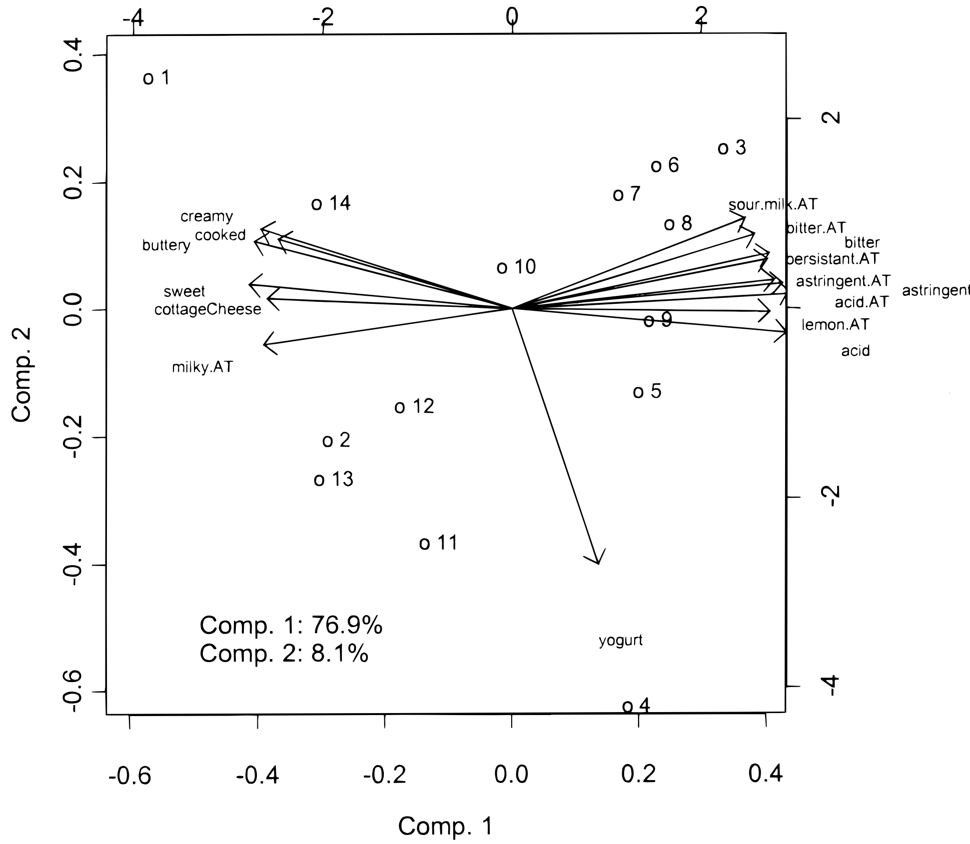

Figure 4. PCA of significant sensory attributes (ANOVA) of the fermented milk samples. Numbers refer to trials mentioned in Table 3.

tigate its influence on the sensory aspects. Fermentation of milk with *L. bulgaricus* alone (Table 3, trial 3) resulted in a product that was very similar to the yogurt produced in the presence of both *S. thermophilus* and *L. bulgaricus* (Table 3, trial 5). Only minor differences could be observed between the samples (Figure 6). This suggested that for the strains investigated, mainly *L. bulgaricus* gave its characteristics to the final product and *S. thermophilus* had little influence on generation of the yogurt flavor. Fermentation, however, was much more rapid in the presence of both microorganisms (Ott et al., 1999).

Fermentation of milk with *S. thermophilus* alone (Table 3, trial 1) gave a product that was considerably different from the product obtained in the presence of *L. bulgaricus* (Table 3, trial 5), having a higher rating for the attributes buttery, sweet, cooked, creamy, and cottage cheese (Figure 6). These differences were shown to be significant with the Duncan test at 5%. Despite these higher scores, levels of 2,3-butanedione and 2,3-pentanedione in trial 1 were lower than for the sample containing both microorganisms (Table 3, trial 5). On the other hand, the pH of the sample with *S. thermophilus* alone (Table 3, trial 1) was higher (4.9) than those of all other tested samples (range from 4.1 to 4.6). This reconfirmed that buttery and cottage cheese notes were strongly pH dependent. Differences in 2,3-butanedione and 2,3-pentanedione contents, as found between the different samples tested, did not significantly influence the intensity of these notes.

Influence of pH on Sensory Attributes. In all previous sections, a predominant sensory role of pH was found. However, the products compared also showed differences in volatile content and viscosity. It could, therefore, not be excluded that factors other than pH could also affect the sensory attributes. An investigation with pH as the only variable parameter was, therefore, conducted.

Two different types of yogurt, traditional acidic yogurt (Table 3, trial 5) and the corresponding mild, less acidic yogurt (Table 3, trial 12), were tested with regard to influence of the pH on perception of the different sensory attributes of yogurt. Each type of yogurt contained different concentrations of the three impact aroma compounds (Table 3, trials 5 and 12, respectively).

The pH of the two different yogurts was adjusted with either lactic acid or diluted NaOH (6 M) to 4.0, 4.2, 4.4, 4.6, 4.8, and 5.0 to create two series of varying pH. This range covered the acidity of many different kinds of yogurt, from very mild to very acidic.

Data were analyzed with a general linear model procedure (GLM-ANOVA), in which the type of yogurt and the pH are both fixed factors and the "taster" is a random factor. None of the six odor attributes was found to vary significantly with pH (figure not shown), and the only difference in odor attribute intensities between both yogurt types was the pungent odor. It was more intense but at the limit of significance ($p = 0.057$) in the traditional acidic yogurt at all pH values tested (Figure 7M). As acetaldehyde alone has been described as having a pungent fresh green aroma quality in GC-olfactometry (Ott et al., 1997) the difference in pungent odor could be attributed to the different concentrations of this compound in the two yogurt types. However, this difference did not appear in results of the previous experiment with various acetaldehyde concentrations. Presumably this is because the previous set of samples was more diverse in sensory properties.

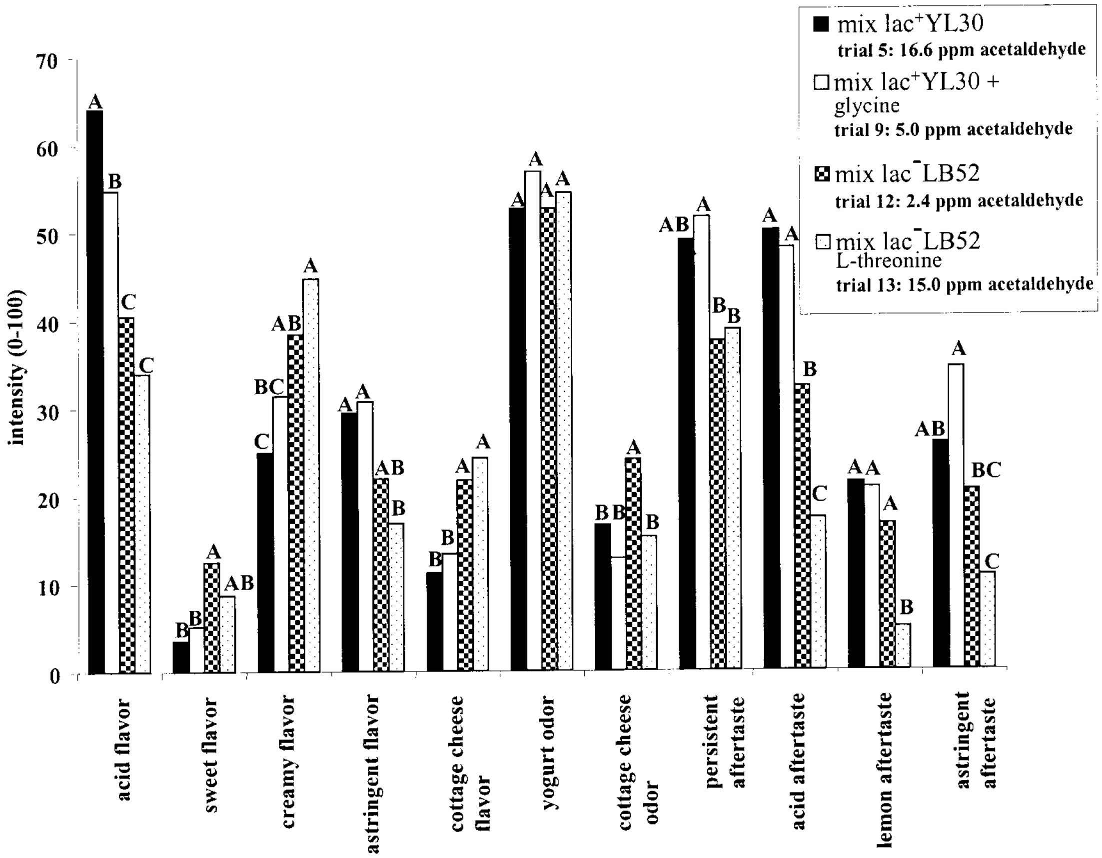

Figure 5. Influence of acetaldehyde content on sensory attributes of traditional acidic (mix lac$^+$) and mild, less acidic (mix lac$^-$) yogurt (Table 3; trials 5 and 9 and 12 and 13, respectively). Only odor, flavor, or aftertaste attributes for which an ANOVA on these four products showed a significant difference ($P \leq 0.05$) are displayed. The difference between samples sharing the same letter is not significant (Duncan, $P = 0.05$).

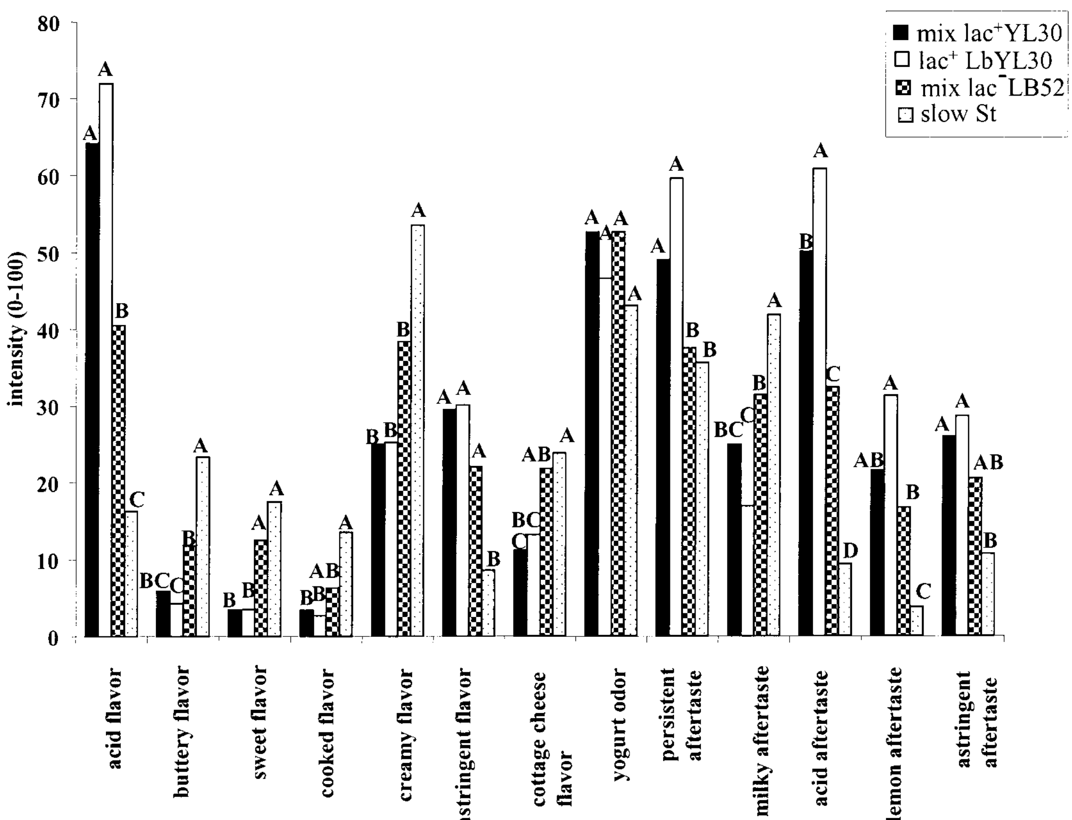

Figure 6. Influence of *S. thermophilus* and/or *L. bulgaricus* on yogurt flavor attributes. Apart from yogurt odor, only attributes for which an ANOVA on these four products showed a significant difference ($P \leq 0.05$) are displayed. The difference between samples sharing the same letter is not significant (Duncan, $P = 0.05$).

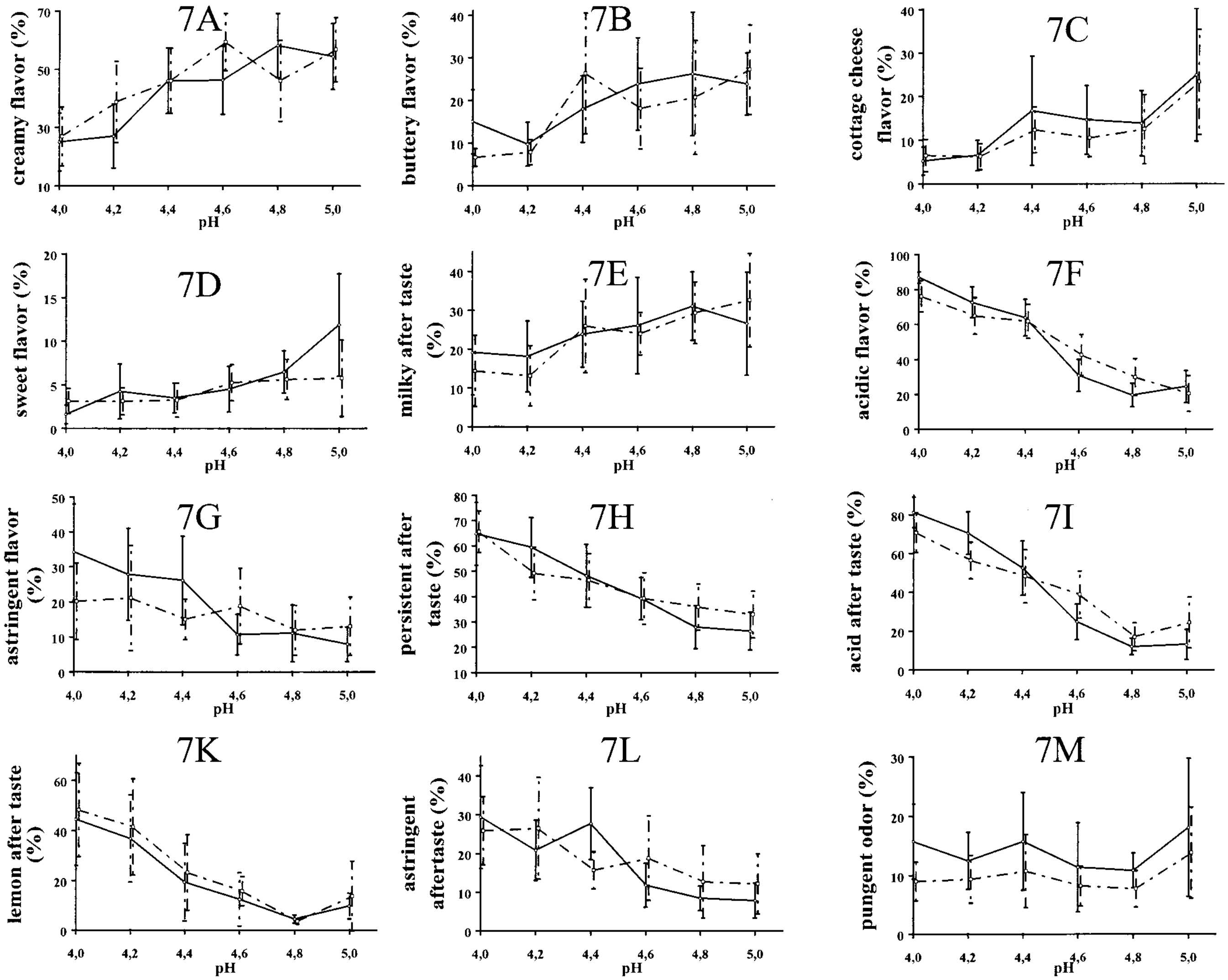

Figure 7. Influence of pH on different sensorial descriptors: (—) traditional acid; (···) mild, low-acid yogurt; (A) creamy flavor; (B) buttery flavor; (C) cottage cheese flavor; (D) sweet flavor; (E) milky aftertaste; (F) acidic flavor; (G) astringent flavor; (H) persistent aftertaste; (I) acid aftertaste; (K) lemon aftertaste; (L) astringent aftertaste; (M) pungent odor. Error bars indicate the standard errors of the means.

Perceived acidity of the samples was correlated with the pH of the yogurt (Figure 7F). The steep slope (from 80 to 90% intensity to ~20% over the pH range) indicated a strong sensitivity of the panel to changes in acidity of the products. The type of yogurt was not a significant factor for acidity perception. This suggests that the different contents of impact aroma compounds in each type of yogurt did not modulate the perception of acidity.

Many other flavor descriptors showed a variation with pH. Creamy, buttery, cottage cheese, and sweet as well as milky aftertaste intensities significantly increased with increasing pH (Figure 7A–E). The increase in sweet flavor with increasing pH was less obvious between pH 4 and 4.4 but was stronger from pH 4.6 to 5.0 and only in the acid yogurt (Figure 7D).

Astringent flavor as well as persistent, acid, lemon, and astringent aftertastes decreased with increasing pH (Figure 7G–L). Correlation of many of the sensory attributes with pH showed the importance of acidity in yogurt flavor perception and confirmed Muir and Hunter's (1992) observation of a relationship between pH and flavor intensity, for example, acid/sour flavor, bitter flavor, lemon flavor, rancid flavor, and bitter aftertaste. However, these authors compared very different products, that is, yogurt made with milks of different animal origin and fat contents as well as "fromage frais".

Significant differences in flavor attributes between the two types of yogurt were not found. Both yogurts showed very similar results over the whole pH range for all attributes. The descriptors buttery and cottage cheese, which were expected to be especially dependent on the concentration of the butter-like aroma compounds 2,3-butanedione and 2,3-pentanedione, did not reflect the differences in content of these two compounds between the two yogurt types (Table 3, trials 5 and 12, respectively).

Influence of pH, Deodorization, and Volatile Addition on Yogurt Flavor. Previous experiments suggested an important contribution of pH to yogurt flavor, and the contribution of volatiles seemed to be rather small. An experiment combining both factors should confirm these results.

Samples of a traditional acidic yogurt (Table 3, trial 5) fermented to pH 4.1 were adjusted to pH 4.5 by adding diluted NaOH (6 M). The volatiles present were removed under vacuum, and 15 ppm of acetaldehyde, 1 ppm of 2,3-butanedione, and 0.1 ppm of 2,3-pentanedi-

Table 4. Sample Treatment for pH Adjustment, Deodorization, and Volatile Addition (Acetaldehyde, 2,3-Butanedione, and 2,3-Pentanedione)[a]

	pH 4.1		pH 4.5	
treatment	no volatile addition	volatile addition	no volatile addition	volatile addition
untreated	16.1	32.2	16.1	32.2
deodorized	<2.0	16.1	<2.0	16.1

[a] Total amounts of the three volatiles are given as parts per million.

one were readded. These amounts corresponded to the concentrations of the three compounds found in a traditional acidic yogurt (Ott et al., 1999). This gave a total of eight samples (Table 4).

Deodorization removed 81% of total volatiles of the mild yogurt as calculated from the total surface areas of the FID chromatograms of normal and deodorized yogurt headspace volatiles.

An important influence of the pH on many sensory attributes was observed. A pH difference of 0.4 was sufficient to significantly influence creamy, buttery, cottage cheese, acid, and sweet flavor attributes and persistent, milky, acid, and lemon aftertaste (ANOVA; $\alpha = 0.05$). This confirmed the strong influence of pH on flavor described in the previous sections.

Deodorization influenced three attributes significantly (ANOVA; $\alpha = 0.05$): creamy flavor (↓), lemon (↑), and astringent (↑) after taste. Because creamy flavor was decreased by deodorization, this suggests a correlation between volatiles and this attribute. However, as readdition of the three compounds up to their original level did not significantly increase it, volatiles other than acetaldehyde, 2,3-butanedione, or 2,3-pentanedione are probably responsible. The influence of textural changes, however, cannot be totally excluded as deodorization changed the texture to some extent (data not shown).

Addition of the three volatiles acetaldehyde, 2,3-butanedione, and 2,3-pentanedione in concentrations found in traditional acidic yogurt had a significant effect only on yogurt odor (ANOVA; $\alpha = 0.05$). Yogurt odor intensity was systematically higher in products to which the three volatiles were added, but no significant change could be observed for any other flavor attributes. Contribution of the three compounds to the aroma of yogurt confirmed our previous results on the identification of yogurt impact aroma compounds (Ott et al., 1997).

CONCLUSIONS

From this work and our previous papers (Ott et al., 1997, 1999) it can be concluded that *the intensity of yogurt flavor perception is pH driven*.

The sensory panel demonstrated an extreme sensitivity to the pH, and the perception of acidity seems to condition the perception of the other attributes. A physicochemical interaction at the receptor level seems unlikely, because acidity is essentially perceived on the tongue and volatiles eliciting “yogurt” or “cottage cheese” recognition are perceived in the nose via the retronasal pathway. The question remains open whether the response is determined, more or less automatically, by the pattern of stimuli received simultaneously in the brain from various receptors or whether a cognitive mechanism takes place (“because it is acid, it cannot be creamy”). In any case, whereas physicochemical analyses are selective and, for instance, can measure independently pH and volatile concentrations, human perception is integrative by nature and cannot quantify one stimulus independently from the others. Although this may be considered as a weakness of sensory evaluation, it is truly representative of how consumers appreciate the sensory quality of products.

Consequently, *enhancing flavor formation by overexpressing lactic acid bacteria metabolism would not increase perceived intensity* and could unbalance the aroma due to an insufficient amount of milk-originating volatiles.

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