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Effect of Fat Content on Odor Intensity of Three Aroma Compounds in Model Emulsions: δ -Decalactone, Diacetyl, and Butyric Acid

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The purpose of this investigation is to try to explain the odor intensity of three aroma compounds in model emulsions using physicochemical measurements (liquid/liquid partition coefficients and vapor/liquid partition coefficients) and to evaluate how odor intensity is affected by the oil content and the pH of the emulsion. The aroma compounds chosen for this study were diacetyl, δ -decalactone and butyric acid. The physicochemical analyses agree with the sensory data. Hydrophilic molecules (diacetyl and butyric acid) are all the more odorant as the oil content is high. In contrast, odor of hydrophobic molecules (δ -decalactone) is more pronounced in aqueous media. The odor and the vapor/liquid partition coefficient of butyric acid are influenced by pH. This study also provides the means to predict the vapor/liquid partition coefficient of a compound in an oil/water emulsion on the basis of a few physical measurements.

Keywords: Partition coefficient; odor intensity; δ -decalactone; diacetyl; butyric acid

INTRODUCTION

Flavors consist of a combination of numerous compounds, each with peculiar individual physical properties. These properties determine several types of interactions with the other molecules, which may lead to different flavoring behaviors. The lack of quantitative measurements of these interactions and their relationships with odor or flavor properties allows the flavorist's work to be quite empirical. Quantitative data may help the flavorist to adapt concentrations of the aroma compounds with the type of food or beverage to flavor.

The study of interactions between flavor compounds in food is hindered by the complexity of food. It is especially difficult to separate the effects of fat from the effects of proteins. For this reason, many authors used model systems to study interactions between flavors and components of food systems (D'Dios Vega and Brewer, 1994; Desamparados et al., 1994; Druaux et al., 1995). However, a few foods are mainly two-phase systems, being oil-in-water or water-in-oil emulsions with a low concentration of nonfat molecules, as is the case of butter or margarine. The interactions between the volatile with the non-volatile compounds depend on their hydrophobicity, but also on the water content (Le Thanh et al., 1992). This dependency underscores the importance of the matrix and that it is essential to evaluate the importance of aroma compounds in model systems similar to that of the original product (Guadagni et al., 1972; Urbach et al., 1972; Buttery et al., 1973; Le Thanh et al., 1993).

From a sensorial point of view, the lipophilic aroma compounds tend to be more odorous in aqueous than in lipid media. They demonstrate higher odor thresholds in vegetable oil than in water solutions (Guadagni et al., 1972). From an analytical point of view, most studies reported in the literature deal with vapor/liquid partition phenomena, in particular with the effect of medium composition on the equilibrium headspace concentrations. Thus, lipophilic compounds are influenced by the presence of lipids in the medium and at the equilibrium their headspace concentrations are higher in aqueous than in lipid solutions (Land, 1979; Van Boekel and Lindsay, 1992). Unfortunately, in the experiments with volatile compounds, equilibrium headspace concentrations have not been related to sensory scores. Thus, up to now, no parallel has been drawn in the same medium between odor intensity and vapor/liquid partition coefficients.

In the case of foods in which volatile fatty acids and other ionizable compounds are important aroma compounds, the pH of the aqueous phase can markedly influence perception by governing the state of dissociation of these acids (Bills et al., 1969; Baldwin et al., 1973; Hartwig and McDaniel, 1995). Generally, the more volatile flavorful fatty acids have pK values between pH 4 and 5 and are most potent below this range (Baldwin et al., 1973). In contrast to the numerous studies dealing with the effect of pH on flavor, the effect of pH on odor and vapor/liquid partition coefficients has received very little attention.

The aroma compounds chosen for this study are diacetyl, presenting a butter flavor; δ -decalactone, with a coconut flavor; and butyric acid, with a fruity or cheesy flavor depending on the concentration. These aroma compounds are common in numerous dairy products (Kinsella, 1975; Badings and Neeter, 1980; Schieberle et al., 1993) and were chosen because of their very different physical properties. The purpose of this investigation is to perform, in parallel and on model

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emulsions, physicochemical measurements (liquid/liquid and vapor/liquid partition coefficients) and sensory evaluations (odor intensity) with each of these aroma compounds. Two parameters, the oil content and the pH, were studied. Then comparisons are done between the vapor/liquid partition coefficient measured by head-space analysis and the one calculated by the Buttery model equations (Buttery et al., 1973).

MATERIALS AND METHODS

Chemicals. Isoamyl acetate (used as a reference scale for the intensity evaluation), diacetyl, δ -decalactone, and butyric acid were purchased from Aldrich (Saint-Quentin Fallavier, France) and were respectively, 99, 97, 99, and 99% pure. The hydrophobicities of diacetyl, butyric acid, and δ -decalactone were established with the log (P) values (calculated from Rekker, 1977) and are, respectively, -2.0 , 0.8 , and 3.4 . In this work, the oily phase in the model medium is paraffin oil (Primol 352), which was purchased from GHM SIGG (Marseille, France). This oil is completely free of background odors. Moreover, it is composed of saturated alkanes (average molar weight, $490 \text{ g}\cdot\text{mol}^{-1}$) and is inert towards chemical compounds. Its density at 20°C is $0.86\text{--}0.87$. The aqueous phase was ultrapure water (Milli-Q Reagent Water System, Millipore Corporation, Bedford, MA). The emulsifier (SP50 - HLB = 11; Sisterna, Roosendaal, The Netherlands) used was a sucrose stearate-palmitate ester with a purity of $>90\%$, a water content of $<2\%$, and a bulk density of $0.4\text{--}0.5$.

Sensory Evaluation. *Olfactive Purity of Diacetyl, δ -Decalactone, and Butyric Acid.* Sniffing analysis was used to verify the olfactive purity of diacetyl, δ -decalactone, and butyric acid solutions (10, 10, and 20 ppm, respectively in dichloromethane). Analyses were made with a modified Hewlett-Packard 5890 Series II gas chromatograph equipped with a $30 \text{ m} \times 0.32 \text{ mm}$ cross-linked capillary free fatty acid phase (FFAP) column (Chrompack, France). At the end of the capillary column, the effluent was split 1:1 into a FID and a sniffing port. The sniffing port consisted of a glass funnel as described by Abbott et al. (1993). The column effluent directed to the sensory port was mixed with humidified air ($200 \text{ mL}/\text{min}$) to avoid dehydration of nasal mucus of the sniffer. Splitless/split injections were made, and operating conditions were as follows: H_2 carrier gas, $1.5 \text{ mL}/\text{min}$; temperature at injection port, 240°C ; detector temperature, 250°C ; oven temperature, $40\text{--}240^\circ\text{C}$ at $5^\circ\text{C}/\text{min}$.

Sensory Test Samples. The emulsifier was used at a concentration of 1% (w/w). The emulsions with diacetyl or δ -decalactone were evaluated at 10 ppm, and the emulsions with butyric acid were evaluated at 20 ppm. These concentrations were chosen on the basis of preliminary sensory evaluations to obtain a reasonable intensity perceived by all the panel. Moreover, the concentrations were chosen to obtain iso-intensive aqueous solutions, as far as possible. For each aroma compound, the water content of the emulsions ranged from 0 to 99% (w/w); that is, 0, 16, 50, 84, and 99% (w/w). The water content 100% (medium without emulsifier) was also investigated in order to evaluate the effect of the emulsifier on the odor intensity and on the partition coefficients. The pH values of 4.5 and 5.2 were investigated because they are on both sides of butyric acid pK. Thirty odorous emulsions (six with diacetyl at pH 5.2; six with δ -decalactone at pH 4.5 and 5.2, and six with butyric acid at pH 4.5 and 5.2) were evaluated. The pH of the aqueous phases containing the aroma compound was adjusted to pH 4.5 or 5.2 with a 0.01 N NaOH solution or a 0.01 N HCl solution. The pH was measured by a pH electrode (model A90333; Bioblock Scientific, France) with a microprocessor pH/mV meter (Model A93313; Bioblock Scientific, France). The emulsions were realized with an IKA-ULTRA-TURAX T25 (Janke & Kunkel, Staufen, Germany; 30 s for 100 mL). The preparation of emulsions was made at 4°C to minimize the aroma compound loss.

Panelists. Twenty subjects who had previous sensory experience participated this study. Because they had participated in other sensory studies, they were trained and selected

for their capacity to discriminate and memorize tastes and other sensations, to recognize and memorize odors and to describe their perceptions when tasting food products.

Sensory Test Procedure. Samples were presented at $21 \pm 1^\circ\text{C}$ in coded 60 mL brown capped flasks. The booths were lighted with red light. Each flask contained 20 mL of sample. Sample presentation was established with the latin square design to take into account serving order and carry over effects of samples (MacFie and Bratchell, 1989; Schlich, 1994). A 10-point category scale, adapted from the technique of Punter et al. (1984), was used as a reference scale for the intensity evaluation. Points 1, 3, 5, 7, and 9 were symbolized by five isoamyl acetate solutions ($0.04\text{--}10 \text{ ppm}$ with a 4 geometrical ratio); points 2, 4, 6, and 8 were intermediate points between the isoamyl acetate solutions; and point 10 corresponded to an intensity higher than the 10 ppm isoamyl acetate solution. Panelists were instructed to smell each sample, rate its intensity on the reference scale, and describe the odor that they have perceived. All testing sessions were limited to a single pH level for each aroma compound studied. In the course of four sessions, two replicates were completed for each odorous compound. With a paired t test, no session effect was observed (paired \pm test), indicating that the subjects and the samples preparation were replicated. With this technique, all the samples are evaluated with regard to the same odor category scale. However, the odor quality of the category scale is different from one of the samples, and some panelists have had difficulty evaluating only odor intensity of aromatized model emulsions.

Liquid/Liquid Partition Coefficients. The equilibrium concentrations of the aroma between the hydrophobic phase and the aqueous phase at a given temperature were measured. Aqueous solution (2 mL) of containing the aroma compound (diacetyl, butyric acid, and δ -decalactone concentrations of 100, 200, and 200 ppm , respectively) was in contact with 2 mL of paraffin oil. The two phases were gently stirred, to avoid formation of an emulsion, for at least 48 h until equilibrium was reached at $25 \pm 1^\circ\text{C}$. The equilibrium concentrations were determined with a calibration scale, and the liquid/liquid partition coefficient was finally expressed as

$$P^\circ = C_o/C_w \quad (1)$$

where C_o and C_w are, respectively, the concentrations of aroma compound in oil and in water ($\mu\text{L}/\text{L}$). For each sample, three replications were completed.

Butyric Acid. Analyses were carried out on a Hewlett-Packard HP 5890 Series II gas chromatograph equipped with a flame ionization detector and a $30 \text{ m} \times 0.32 \text{ mm}$ crosslinked capillary FFAP column (Chrompack, France). Operating conditions were as follows: N_2 carrier gas, $2 \text{ mL}/\text{min}$; temperature at injection port, 123°C ; column temperature, 120°C ; and detector temperature (200°C). A Shimadzu CR3A recorder-integrator was used.

Diacetyl and δ -Decalactone. Analyses were carried out on a Chrompack CP9000 gas chromatograph equipped with a flame ionization detector and a $3.0 \text{ m} \times 2.0 \text{ mm}$ stainless steel Carbowax 20M column (Chrompack, France) packed with Chromosorb W-AW ($100\text{--}120 \text{ mesh}$). Operating conditions were as follows: N_2 carrier gas, $16 \text{ mL}/\text{min}$; temperature at injection port, (190°C), column temperature, 100°C for diacetyl and 180°C for δ -decalactone; and detector temperature 200°C .

Vapor/Liquid Partition Coefficients. The equilibrium concentrations of the aroma compounds between the liquid phase and the vapor phase at a given temperature were measured. This temperature was 25°C for diacetyl and butyric acid and 80°C for δ -decalactone. An inert gas (nitrogen) passed through the liquid phase at a constant flow rate (20 , 60 , and $100 \text{ mL}/\text{min}$, respectively, for diacetyl, butyric acid, and δ -decalactone) and carried the volatile compounds into the headspace. A sample of the vapor phase was automatically injected into the gas chromatograph at regular intervals. The equilibrium vapor/liquid was considered to be reached when the concentration of aroma compounds in the

Table 1. Odor Intensity, Odor Description, and Vapor/Liquid Partition Coefficient of δ -Decalactone in Model Emulsions Containing 1% Emulsifier^a

fat content (% w/w)	odor intensity (21 °C)		coconut quotations 1 st /2 nd rep.		K^{∞} ($\times 10^4$) dimensionless (80 °C)	
	pH 4.5	pH 5.2	pH 4.5	pH 5.2	pH 4.5	pH 5.2
0	5.47 (1.83) ^a	6.29 (2.22) ^a	6/3	6/5	0.87 (0.02)	1.03 (0.10)
15	4.03 (2.11) ^{bc}	4.59 (2.22) ^b	7/4	7/5	ND	ND
49	3.41 (2.06) ^{cd}	4.09 (2.11) ^b	5/5	7/6	ND	ND
83	3.12 (2.17) ^d	3.56 (1.92) ^b	6/3	6/5	ND	ND
99	4.29 (2.26) ^b		2/3		ND	

^a Values in the same column with different letters are significantly different ($p < 0.05$); ND (nondetected); standard deviation is in parentheses after each value.

Table 2. Effect of Emulsifier and pH on Odor Intensity, Odor Description, and Vapor/Liquid Partition Coefficients (K^{∞}) of δ -Decalactone (Performed at 80 °C), Diacetyl (Performed at 25 °C), and Butyric Acid (Performed at 25 °C)^a

product	emulsifier	odor intensity (21 °C)		descriptor quotation 1 st /2 nd rep.		K^{∞} ($\times 10^4$) dimensionless	
		pH 4.5	pH 5.2	pH 4.5	pH 5.2	pH 4.5	pH 5.2
δ -Decalactone	with	5.47(1.83) ^{a,b}	6.29(2.22) ^{a,a}	6/3	6/5	0.87(0.02) ^{a,a}	1.03 (0.10) ^{b,a}
	without	4.85(2.37) ^{a,a}	5.35(1.84) ^{b,a}	4/4	5/2	1.05(0.10) ^{a,b}	1.81(0.10) ^{a,a}
diacetyl					butter		
	with	—	4.18(2.15) ^a	—	caramel	—	4.76(0.06) ^a
					1/1		
	without	—	3.79(2.04) ^a	—	3/2	—	5.63(0.06) ^a
butyric acid					0/2		
	with	5.38(2.37) ^{a,a}	3.56(1.86) ^{a,b}	4/5	rancid	5/5	1.31(0.03) ^{b,a}
	without	5.76(1.78) ^{a,a}	4.35(2.12) ^{a,b}	10/10	5/4	3.09(0.07) ^{a,a}	0.52(0.08) ^{b,b*}

^a For the odor intensity or for K^{∞} of each aroma compound, values with different letters are significantly different ($p < 0.05$); standard deviation is in parentheses after each value. First letter corresponds to emulsifier effect and second to pH effect. *Measured from a butyric acid solution at 1000 ppm (infinite dilution domain).

gas phase remained constant. The vapor/liquid partition coefficient was expressed as

$$K^{\infty} = (C_g/C_l) \quad (2)$$

where C_g and C_l are, respectively, the concentrations of aroma compound in the gaseous phase and in the liquid phase determined with a calibration scale (expressed in $\mu\text{L/L}$). For each sample, three replications were completed.

Samples. For the measurement of partition coefficients, the emulsions were prepared as for the sensory evaluation, but the concentrations were 100 ppm for diacetyl, 200 ppm for δ -decalactone, and 500 ppm for butyric acid to have a significant response of the apparatus. The measurements were realized in infinite dilution domain, where the aroma concentration does not affect the partition coefficient, so these results can be compared with the sensory data.

Butyric Acid. Analyses were carried out on the same gas chromatograph apparatus equipped with a 0.5 m \times 2.0 mm stainless steel HayeSep Q (Chrompack, France) column (80–100 mesh). Operating conditions were as follows: N_2 carrier gas, 31 mL/min; injection port temperatures, 190 °C; oven temperature, 180 °C; and detector temperature, 200 °C. A Chroma (Biosystems, Couteron, France) recorder-integrator was used for quantification.

Diacetyl and δ -Decalactone. Analyses were carried out on the same apparatus as used for the determination of liquid/liquid partition coefficients. Quantification was done in the same way as for butyric acid.

Statistical Analyses. All the statistical analyses were made with the Statistical Analysis System (SAS Institute, Inc., Cary, NC). An analysis of variance (ANOVA) was used on the sensory and physical results to determine significant differences among samples. A Student–Newman–Keuls test was used to perform a multiple comparison of means.

RESULTS

δ -Decalactone. *Sensory Analysis and Vapor/Liquid Partition Coefficients.* The average intensities over the

two replicates and the average vapor/liquid partition coefficients over the three replicates are reported in Table 1. For the media containing 1% emulsifier, at pH 4.5 and pH 5.2, the media without oil present had the highest odor intensity ($p < 0.05$). For the two pH values the odor intensity decreases when the fat content increases, except for the medium without water at pH 4.5. The odor intensity is then equivalent to that of the 15% oil content medium. Very few panelists quoted the 'coconut' term to describe the odor of this emulsion. Contrarily, whatever the pH and the oil content between 0 and 83%, panelists quoted more frequently the 'coconut' term (Table 1).

At 25 °C, the aroma quantity present in the gaseous phase was too weak to be detected, so the measurements of partition coefficients were performed at 80 °C. Nevertheless, partition coefficients for water contents of 99 and 100% were the only ones measurable (Table 2).

A pH effect on odor intensity was observed only in the emulsified medium without oil. However, a pH effect on vapor/liquid partition coefficient was observed in the nonemulsified medium without oil. At pH 4.5, the vapor/liquid partition coefficients, like the odor intensity, were not significantly affected by the emulsifier (Table 2). A significant effect of the emulsifier is only observed at pH 5.2. In presence of emulsifier, the odor intensity was significantly more intense ($p < 0.05$) and the term 'coconut' was more frequently quoted. However, the vapor/liquid partition coefficient decreased significantly in the presence of emulsifier. This result is in disagreement with the sensory data (Table 2).

Liquid/Liquid Partition Coefficients. The liquid/liquid partition coefficients show the great affinity of δ -decalactone for the oil (Table 3). The molecule is mainly dissolved in organic phase. These results are

Table 3. Effect of Emulsifier and pH on Liquid/Liquid Partition Coefficients of δ -Decalactone, Diacetyl, and Butyric Acid^a

product	emulsifier	pH 4.5	pH 5.2
δ -Decalactone	without	9.79 (0.79) ^{a,a}	10.03 (1.78) ^{a,a}
	with	2.88 (0.13) ^{b,a}	3.77 (0.20) ^{b,a}
diacetyl	without	—	5.48×10^{-2} (0.42×10^{-2}) ^a
	with	—	3.20×10^{-2} (0.43×10^{-2}) ^a
butyric acid	without	12.7×10^{-2} (0.58×10^{-2}) ^{a,b}	15.4×10^{-2} (0.59×10^{-2}) ^{a,a}
	with	2.77×10^{-2} (0.29×10^{-2}) ^{b,a}	3.44×10^{-2} (0.32×10^{-2}) ^{b,a}

^a For each aroma compound, values with different letters are significantly different ($p < 0.05$); standard deviation is in parentheses after each value. First letter corresponds to emulsifier effect and second to pH effect.

Table 4. Odor Intensity, Odor Description, and Vapor/Liquid Partition Coefficient of Diacetyl in Model Emulsions Containing 1% Emulsifier^a

fat content (% w/w)	odor intensity (21 °C)	quotations		K^{∞} ($\times 10^4$) dimensionless (25 °C)
		butter 1 st /2 nd rep	caramel	
0	4.18 (2.15) ^b	7/9	1/1	4.76 (0.06) ^c
15	3.91 (2.06) ^b	9/10	5/2	5.33 (0.23) ^c
49	3.82 (1.96) ^b	8/11	3/4	10.2 (0.8) ^c
83	4.38 (2.27) ^b	8/11	5/3	21.3 (1.1) ^b
99	5.35 (2.32) ^a	5/7	3/1	54.0 (7.0) ^a

^a Values in the same column with different letters are significantly different ($p < 0.05$); standard deviation is in parentheses after each value.

confirmed by the hydrophobic behavior of the molecule ($\log P = 3.4$; Rekker, 1977). Although P^{∞} is independent of the pH, it depends on the presence of the emulsifier. At pH 4.5 or 5.2, P^{∞} is significantly higher without emulsifier than with emulsifier (Table 3).

Diacetyl. *Sensory Analysis and Vapor/Liquid Partition Coefficient.* The diacetyl structure (2,3-butanedi-one) does not change with the pH. For this reason all the measurements were realized at pH 5.2. Odorous notes are mean values of two replicates, and vapor/liquid partition coefficients are mean values of three replicates (Table 4). In media containing 1% emulsifier, the higher the oil content, the more intense is the odor. The medium without water is significantly different ($p < 0.05$) from the other media. The evolution of vapor/liquid partition coefficients with the oil content agrees with sensory data, but three groups instead two are distinguished by the variance analysis of results (Table 4). If K^{∞} increases, the diacetyl concentration in gaseous phase also increases. This increase leads to a higher odor intensity. The terms "butter" or "caramel" are less frequently quoted for the aqueous media than for oily media. The description of odor intensity also changes with the oil content (Table 4).

The emulsifier had no effect on odor intensity or on the vapor/liquid partition coefficient (Table 2).

Liquid/Liquid Partition Coefficient. Diacetyl shows an higher affinity for water than for oil (Table 3), in agreement with its hydrophilic character. This higher affinity is confirmed by the calculated $\log P$, which is -2 (Rekker, 1977). The emulsifier effect is significant at the 0.5% level.

Butyric Acid. *Sensory Analysis and Vapor/Liquid Partition Coefficient.* Two pH values, 4.5 and 5.2, were investigated. These values are on both sides of the pK of butyric acid, which is 4.8. The results of the measurements of odor intensity and of the vapor/liquid partition coefficients at pH 4.5 and 5.2 are reported in Table 5. In the emulsified medium without oil at pH 5.2, the gaseous phase concentration was too weak to be measured. Thus, the vapor/liquid partition coefficient was determined from a solution at 1000 ppm

(instead of 500 ppm; Table 5).

Whatever the pH, the odor intensity and the vapor/liquid partition coefficients increase when the percentage of paraffin oil increases (Table 5). This parallelism indicates that butyric acid has a higher affinity for aqueous phase than for oily phase. At pH 4.5, the medium without water presents an odor intensity and a vapor/liquid partition coefficient significantly higher ($p < 0.05$) than those of the other media. At pH 5.2, the analysis of variance indicated three groups (instead of two at pH 4.5) for the odor intensities and for the vapor/liquid partition coefficients. However, the panelists have had difficulties evaluating the odor intensity of the medium without oil. As a matter of fact, according to the variance analysis, the odor intensity of this medium is similar to the one of group b and to the one of group c. For each pH the note 'rancid' is more frequent when the fat content is high.

Except for the medium without water, where the pH effect does not exist, the odor intensities or the vapor/liquid partition coefficients at pH 4.5 are always above those obtained at pH 5.2. The difference between the two pH values is significant at the 5% level, with the exception of the medium with an oil content of 83%. The note 'rancid' is more frequently quoted at pH 4.5. The physical results fit with the sensory data.

The emulsifier influences the vapor/liquid partition coefficients, which are significantly higher in its absence (Table 2). In the case of sensory results, the difference is not significant, but shows that the emulsifier tends to reduce the odor intensity.

Liquid/Liquid Partition Coefficient. Butyric acid shows an higher affinity for water than for oil (Table 3). These results confirm the hydrophilicity of butyric acid, which has a calculated $\log P$ of 0.8 (Rekker, 1977).

A pH effect is observed in absence of emulsifier. In contrast, in the emulsified media, the values obtained at pH 4.5 are similar to those obtained at pH 5.2. Whatever the pH, the difference between the values with and without emulsifier is significant.

DISCUSSION

General Behavior of the Odorous Compounds.

Standard deviations of sensory analysis may appear large compared with physical results, but the tool used is also very different. Whatever the standard deviations, the sensory measurements showed statistically significant differences.

δ -Decalactone. In the media containing paraffin oil, δ -decalactone is highly retained by the oily phase and thus it was not possible to measure the vapor/liquid partition coefficients. Surprisingly, the odor intensity of the medium with 99% of paraffin oil is important compared with the other media (Table 1). Panelists may have perceived an odor that they could not clearly describe. If this odor was perceived as odd, they may have tended to give a high odor intensity note, as they

Table 5. Odor Intensity, Odor Description, and Vapor/Liquid Partition Coefficient of Butyric Acid in Model Emulsions Containing 1% Emulsifier^a

oil content (% w/w)	odor intensity (21 °C)		rancid quotations 1 st /2 nd rep.		$K^\infty (\times 10^4)$ dimensionless (25 °C)	
	pH 4.5	pH 5.2	pH 4.5	pH 5.2	pH 4.5	pH 5.2
0	5.38 (2.37) ^b	3.56 (1.86) ^{bc}	4/5	5/5	1.31 (0.03) ^b	0.52 (0.08) ^{c*}
15	4.97 (2.12) ^b	3.32 (1.68) ^c	8/10	8/6	1.17 (0.01) ^b	0.76 (0.02) ^c
49	5.15 (2.27) ^b	3.23 (2.04) ^c	9/11	7/9	1.48 (0.03) ^b	1.11 (0.06) ^c
83	4.76 (2.12) ^b	4.26 (2.27) ^b	9/11	5/7	1.95 (0.14) ^b	1.93 (0.02) ^b
99	7.12 (2.49) ^a		10/10		4.61 (0.72) ^a	

^a Values in the same column with different letters are significantly different ($p < 0.05$); standard deviation is in parentheses after each value. *Measured from a butyric acid solution at 1000 ppm (infinite dilution domain).

were instructed to evaluate the total odor intensity and not only the 'coconut' odor intensity.

In the pH range of this study, the structure of δ -decalactone probably remains the same because the open form (δ -hydroxy decanoic acid) is unstable and tend to spontaneously lactonize (Kinsella et al., 1967). However, for all the media, the odor intensity and the vapor/liquid partition coefficient are always higher at pH 5.2 than at pH 4.5, even if there is not always a significant pH effect (Table 2).

Diacetyl. In the medium without emulsifier, we found an air/water partition coefficient for diacetyl at 25 °C of $(0.56 \pm 0.01) \times 10^{-3}$. This value is in good agreement with the air/water partition coefficient measured by Dumont and Land (1986) of 0.53×10^{-3} and that determined by Desamparados et al. (1994) of 0.45×10^{-3} . However, Overbosch et al. (1991) reported a slightly higher value of 0.70×10^{-3} . All these coefficients were measured at 25 °C.

Butyric Acid. With a pK of 4.8, 67% of butyric acid is present in protonated forms (which are odorous) at pH 4.5, whereas only 29% of the butyric acid at pH 5.2, is in the protonated forms. For this reason the odorous forms concentration increases in the gaseous phase when pH decreases below the pK , leading to a higher odor intensity and to a higher vapor/liquid partition coefficient at pH 4.5 (Table 5). When pH decreases, not only the odor but also the flavor, is enhanced. Thus, Baldwin et al. (1973) stated that the flavor thresholds of butyric acid was reduced from 6.1 to 0.4 when pH was reduced from 6.0 to 3.2. Hartwig and McDaniel (1995) observed that flavors of citric acid, malic acid, lactic acid and acetic acid were very intense at pH 3.5 and very weak at pH 6.5.

Emulsifier Effect. In the case of liquid/liquid partition coefficients, the emulsifier effect could favor the solubility of the aroma compound in water or form a barrier between oil and water, preventing the aroma compound from diffusing in the oily phase. For this reason, P^∞ decreases. Marion and Doublier (1992) have proposed that the emulsifier could make crystalline structures around the aroma compounds in aqueous phase.

In the case of vapor/liquid partition coefficients and odor intensity, the emulsifier could interact with molecular forms of the aroma compound leading to lower K^∞ values and odor intensities. The effect of emulsifier depends on the molecular form concentration and, consequently, on the pH. Thus, with butyric acid at pH 5.2, the emulsifier retains the majority of protonated forms, at such a level that the quantity in the vapor phase is not sufficient to be detected at 500 ppm. At pH 4.5, the effect is the same, but the quantity of protonated forms remains sufficient to be measured (Table 2). From a sensorial point of view, the difference

in odor intensity seems not enough marked to be perceived by the panelists.

With δ -decalactone, the emulsifier seems to interact with the aroma compound, leading to a decrease of K^∞ values (Table 2). However, these results are in contrast to the sensory results and could be the result of the temperature at which the sensory and the physicochemical measurements were made (respectively, 21 and 80 °C). On one hand, it is probable that the sensory differences between the medium with emulsifier and the medium without emulsifier were not enough marked at 21 °C to be perceived by the panelists. On the other hand, the emulsifier effect on vapor/liquid partition coefficient could have been enhanced because of the temperature (80 °C).

Comparison of Headspace Equilibrium Concentrations with Those Calculated with the Buttery Model. The effect of concentrations of aroma compounds on odor perception depends on their partial pressure (P_i) over the food matrix. This vapor pressure is determined at a fixed temperature (T), pressure (P), and chemical environment by the chemical potential (μ_i) of the compounds

$$\mu_i = \mu_i^\circ + RT \ln a_i \quad (3)$$

where μ_i° is the chemical potential in standard conditions, R the perfect gas constant, and a_i is the activity of compound i (Perez and Romulus, 1993).

In the present case, the equilibrium between three phases is considered, that is, the water phase, the oil phase and the gas phase. At equilibrium, there is equality between the chemical potentials in the different phases and thus

$$a_{iw} = a_{io} \quad (4)$$

$$\gamma_{iw}x_{iw} = \gamma_{io}x_{io} \quad (5)$$

where a_{iw} and a_{io} are, respectively, the activity of the compound i in water and oil phase; γ_{iw} and γ_{io} are, respectively, the activity coefficients in the water and oil phase; P is the total pressure; and x_{iw} and x_{io} are the molar fractions in water and in oil phase, respectively. Therefore, at a constant concentration of flavor in the emulsion, x_{iw} and x_{io} will change when the proportion of aqueous phase in the emulsion are modified.

If P^∞ is the partition coefficient between the oil phase and the water phase (eq 1), it can be related to K_w and K_o which are, respectively, the air/water and the air/oil partition coefficients:

$$P^\infty = C_o/C_w = K_w/K_o \quad (6)$$

Buttery et al. (1973) developed equations for determining the vapor/liquid partition coefficient (K) in a three-phase system. Because K is the ratio between the solute concentration in the air and the solute concentration in the mixture, with the air/water partition coefficient (K_w), the air/oil partition coefficient (K_o), the volumic fraction of water in mixture (F_w), and the volumic fraction of oil in mixture (F_o), one can calculate the air/mixture partition coefficient as follows:

$$K = 1/(F_w/K_w + F_o/K_o) \quad (7)$$

With eq 6, this expression can be written as

$$K = K_w/(F_w + F_o P^\circ) \quad (8)$$

In the case of the δ -decalactone, we could not determine experimentally the air/oil partition coefficient (K_o) because the majority of molecules was retained by the oily phase and the gaseous concentration was too weak to be measured. With eq 8, we can evaluate the partition coefficient in media containing oil.

δ -Decalactone. We used eq 8 to estimate vapor/liquid partition coefficients of the media containing oil. The calculated data are probably underestimated because of the high P° value that characterizes the affinity of δ -decalactone for oil. The calculated vapor/liquid partition coefficients versus the oil content are represented in Figure 1A. When the medium does not contain oil, δ -decalactone concentrates in the gaseous phase and the vapor/liquid partition coefficient increases. The representation of the calculated vapor/liquid partition coefficients versus the odor intensity (Figure 2A) shows that the oil in the medium leads to a solubilization of the molecule in the organic phase and to a decrease of the gaseous phase concentration (which is no more measurable) and of the odor intensity. Though the sensitivity of our equipment did not permit checking of these values, the results obtained are in agreement with those of Kinsella (1975) who showed that lactones have higher thresholds in the fatty phase than in the aqueous phase. Moreover, Kinsella reported that in the case of butter, 96% of the lactones are in the lipid phase.

Diacetyl. We used eq 7 and the values obtained from only two measurements (vapor/liquid partition coefficient with 99% oil and with 99% water) for calculating theoretical vapor/liquid partition coefficients. Experimental and calculated vapor/liquid partition coefficients versus the oil content are reported in Figure 1B. The experimental data are in good agreement with the theoretical values, and the correlation between the measurements and the calculated values is excellent. The equation of the regression line established with 13 degrees of freedom is

$$(\text{experimental } K) = 0.960(0.039) \times (\text{theoretical } K) \quad (9)$$

with a correlation coefficient equal to 0.990. The distribution of the residues of the model is randomly organized around the straight line of the model. Thus, the estimation of the vapor/liquid partition coefficient is all the more accurate because the calculations were realized with the air/oil partition coefficient and the air/water partition coefficient. Figure 2B represents the effect of oil on vapor/liquid partition coefficients and on odor intensities. The oil addition in emulsion leads to

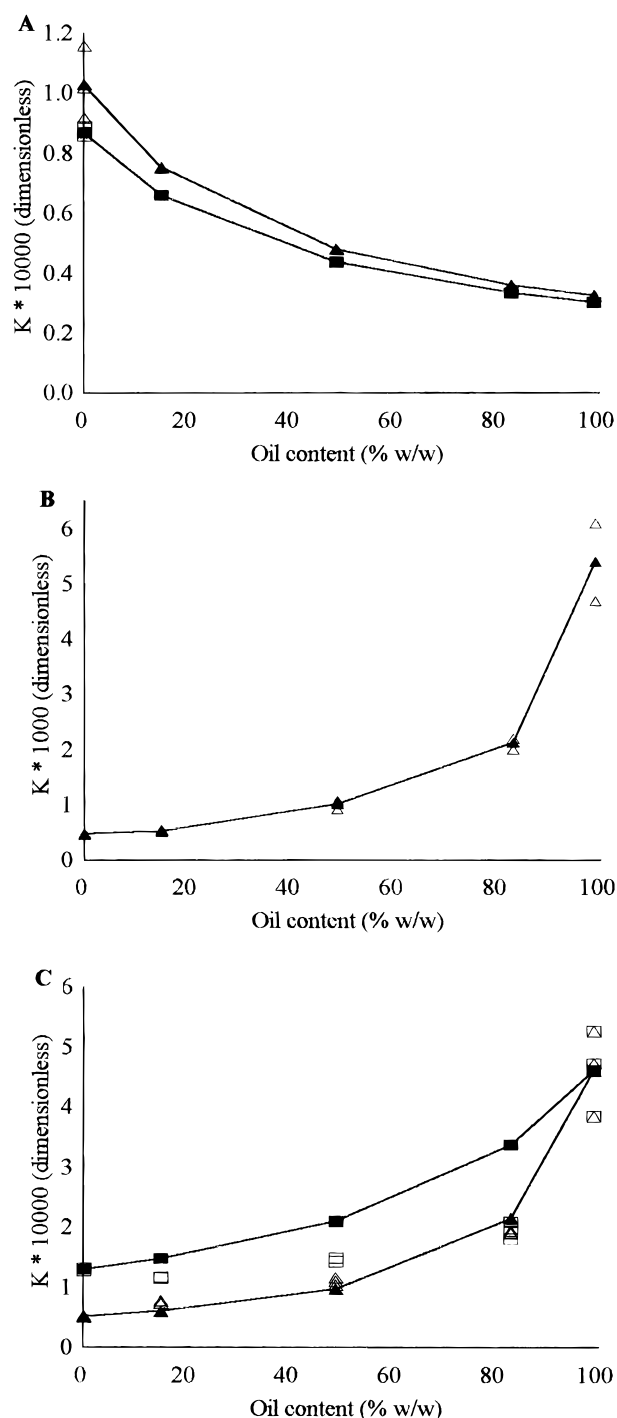


Figure 1. Correlation between vapor/liquid partition coefficient and oil content of (A) δ -decalactone, (B) diacetyl, and (C) butyric acid (ionized and molecular forms). (\square) pH 4.5 experimental; (\blacksquare) pH 4.5 theoretical; (\triangle) pH 5.2 experimental; (\blacktriangle) pH 5.2 theoretical.

an increase of the gaseous phase concentration, and thus of the odor intensity.

When oil is added to the emulsion, the odor looks less intense even if the difference is not significant between the medium without oil and the emulsion with an oil content of 15% (Figure 2B). The homogeneity of the medium seems to influence the diacetyl behavior. Land (1979) observed that the concentration of dimethylsulfide required to measure the same vapor pressure was greater in an emulsified medium than in an unemulsified but chemically identical system. Desamparados et al. (1994) concluded the same thing after stating that the rate of diacetyl release from an oil-in-water emulsion

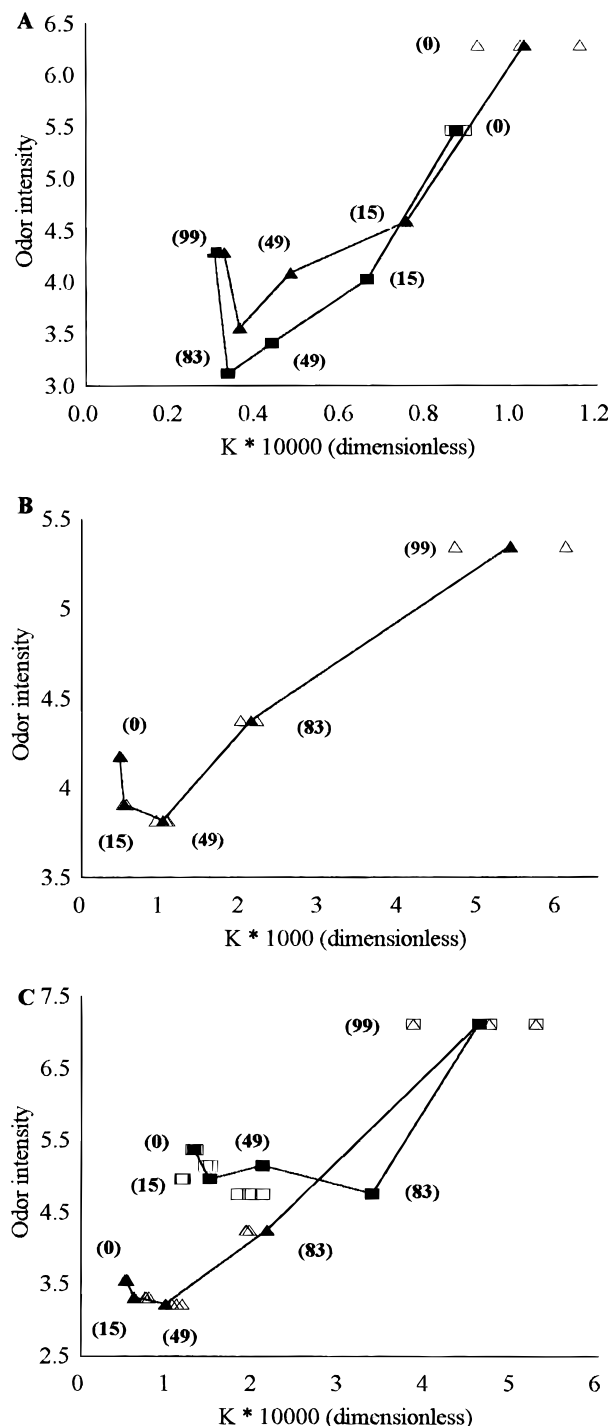


Figure 2. Correlation between odor intensity and vapor/liquid partition coefficient of (A) δ -decalactone, (B) diacetyl, and (C) butyric acid (ionized and molecular forms) in emulsions with different oil contents (..). (□) pH 4.5 experimental; (■) pH 4.5 theoretical; (△) pH 5.2 experimental; (▲) pH 5.2 theoretical.

was greater than from an water-in-oil emulsion. It is clear that the structure of the medium plays an important role with the composition and the use of a model may help the flavorist to adjust the composition of flavoring to the medium he wants.

Butyric Acid. As for diacetyl, we calculated theoretical vapor/liquid partition coefficient with eq 7 at pH 4.5 and 5.2. The calculated and the experimental vapor/liquid partition coefficients versus the oil content are shown in Figure 1C. At pH 5.2, the experimental data are in agreement with the calculated values. The correlation coefficient is equal to 0.981 and the equation

of the regression line established with 13 degrees of freedom is

$$(\text{experimental } K) = 0.970(0.053) \times (\text{theoretical } K) \quad (10)$$

However the residues are not really randomly distributed around the straight line of the model.

At pH 4.5, the calculated vapor/liquid partition coefficients are superior to experimental values (Figure 1C), leading to a correlation coefficient equal to 0.893 because of the nonrandom data distribution on both sides of the linear regression. The equation of the regression line established with 13 degrees of freedom is

$$(\text{experimental } K) = 0.960(0.146) \times (\text{theoretical } K) \quad (11)$$

Thus, in the case of butyric acid, it is more difficult to estimate vapor/liquid partition coefficients with the Buttery model. The odor intensity versus the experimental and calculated vapor/liquid coefficients is presented in Figure 2C. At a constant concentration, an increase in pH of the aqueous phase leads to a decrease of the protonated forms, which are the one volatile. As a consequence, gaseous phase concentrations are lower and lead to lower odor intensities.

As for diacetyl, the structure of the medium influences the butyric acid behavior, and the addition of oil in the medium leads to a decrease in odor intensity (Figure 2C).

Conclusions. Three aroma compounds with different physicochemical properties were shown to have various behaviors according to the medium composition. In most cases, physical results fit the sensory data. Hydrophobicity of the molecule is a key factor for the equilibrium between the emulsion and the vapor phase. Hydrophilic molecules (diacetyl and butyric acid) present a vapor/liquid partition coefficient, and consequently, an odor intensity, that is higher when the medium is hydrophobic. The reverse is evident for δ -decalactone. The effect of pH was also considered with success to explain vapor and odor levels. Whatever the oil content, in the case of diacetyl and (to some extent) butyric acid, the Buttery model allows good estimations of the vapor/liquid partition coefficients simply from the measurement of two partition coefficients.

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