# AGRICULTURAL AND FOOD CHEMISTRY

# The Role of Fat in Flavor Perception: Effect of Partition and Viscosity in Model Emulsions

SARA BAYARRI, ANDREW J. TAYLOR,\* AND JOANNE HORT

Samworth Flavour Laboratory, Division of Food Sciences, School of Biosciences, University of Nottingham, Sutton Bonington Campus, Loughborough LE125RD, United Kingdom

Decreasing the fat content of a food, while maintaining the same aroma content, changes both aroma release (due to partition effects) and the viscosity of the food. To understand the relative contribution of these two factors on flavor perception, a series of flavored emulsions were prepared to control aroma release and viscosity using different aroma, oil, and hydroxypropyl methyl cellulose (HPMC) contents. Samples were formulated to deliver the same aroma-release in vitro and in vivo, and their viscosity was measured using the Kokini oral shear stress parameter. Despite the in vivo aroma release being constant, there were perceptual differences between the samples, and the flavor intensity decreased as in-mouth viscosity increased. For these iso release samples, the Kokini oral shear stress parameter correlated well with the decrease in perception, suggesting that there may be a viscosity stimulus or that the viscosity affects release of tastant and hinders aroma–taste interactions.

KEYWORDS: APCI-MS; aroma release; flavor perception; viscosity; fat; HPMC

## INTRODUCTION

Although fat is an essential part of the human diet, overconsumption of fat can lead to undesirable medical conditions (1), and there is pressure to reformulate food products with reduced fat contents. However, removing significant amounts of fat from foods usually results in poor flavor and texture. Consequently manufacturers are obliged to adjust these properties to produce an acceptable product. Since it is now established that flavor perception occurs through a cross-modal system (2) (i.e., the senses of aroma, taste, and texture interact to form the perception), it is obvious that changing one modality, such as viscosity, can affect the overall perceived flavor. Therefore studies of the effect of fat should, ideally, measure the changes in each modality involved so that appropriate corrective strategies can be applied.

Fat has a significant effect on the partition of volatile compounds between the food and the air phases with lipophilic aroma compounds being the most affected. If fat content is reduced, the amount of lipophilic aromas in the flavor formulation also needs to be reduced to maintain the same profile of aroma release from the product (3-9). Partition can be measured under static equilibrium headspace conditions but does not always relate well to the release profile that is observed in vivo during consumption of a product (4). Using in vivo release measurements in a large group of people (about 90) and rebalancing the aroma content to produce the same maximum intensity of release in simple regular and low-fat milk systems resulted in no significant difference in flavor intensity perception between the two systems (10).

The effect of fat on viscosity and rheological behavior can be measured in the emulsion sample before consumption but this does not necessarily relate to rheological behavior in vivo. Some authors have suggested measuring viscosity at a shear rate of 50 s<sup>-1</sup>, as this represents the shear rate in mouth (11). Richardson et al. (12) reported a good correlation of sensory viscosity with the dynamic viscosity ( $\eta^*$ ) determined at 50 rad  $s^{-1}$ , while the Kokini oral shear stress parameter has also been used as a measure of in vivo conditions (13, 14). Besides these rheological factors, the role of the tastant in the perception of flavor should not be ignored. It is well-established that mixtures of some tastants and aromas produce synergistic and antagonistic sensory effects (15), and therefore delivery of the tastant to the taste buds on the tongue also needs to be considered. There is some evidence that the decline in perceived flavor as sample viscosity increases is due to poor mass transfer of the tastant to the tongue (16), as no significant difference in aroma release can be seen (14, 17, 18). The published work has been carried out in predominantly monophasic systems, but biphasic systems like emulsions mean that the concentration of a tastant in the aqueous phase will change with the oil fraction of the emulsion unless corrections are made, and this factor also needs to be taken into consideration.

The hypotheses to explain the effect of decreasing fat in a food on flavor perception are therefore (1) aroma release changes and affects flavor perception; (2) viscosity changes which may act as a stimulus in its own right or modify the release of tastant and affect the interaction between sugar and aroma, thus reducing the perceived flavor. To rule out hypothesis 1, the aroma content of each sample was adjusted to produce the same release in vivo (iso-release), by formulating a series of flavored emulsions with selected compositions, so that the effect of fat

<sup>\*</sup> Correspondence to: A.J. Taylor; Phone: +44 115 951 6144; Fax: +44 115 951 6154; e-mail: andy.taylor@nottingham.ac.uk.

#### The Role of Fat in Flavor Perception

content and viscosity on flavor perception could be analyzed as independent variables. To take into account the possibility of taste—aroma interactions, emulsions contained both fruity aromas and sugar and used odor- and taste-free emulsifier and oils. The aroma release, in-mouth viscosity, and initial flavor perception of each sample were measured and the data analyzed to identify trends and correlations.

#### MATERIALS AND METHODS

**Preparation and Composition of Oil/Water (o/w) Emulsions.** Emulsions were prepared from rapeseed oil (RO) (AarhusKarlshamn AB, Karlshamn, Sweden), hydroxypropyl methyl cellulose (HPMC) (Methocel K4M, Enorica GMBH, Norderstedt, Germany), sucrose stearate (E-473) (Sisterna SP70, S. Black Ltd., Herts, UK), sucrose (as a tastant), and a mixture of aroma compounds. Rapeseed oil, hydroxypropyl methyl cellulose (HPMC), and sucrose stearate (used as fat stimulus, thickening agent, and emulsifier, respectively) were chosen because they were the most odorless and tasteless materials available at food grade (7, 19).

Oil-in-water emulsions were prepared at three rapeseed oil concentrations (0.5, 3, and 30 g/100 g w/w), each with three HPMC contents (0, 0.6 and 1.2 g/100 g). Sucrose and emulsifier contents were kept constant in all the samples, i.e., 5 g sucrose/100 g emulsion and 1 g emulsifier/100 g emulsion. These concentrations were chosen to represent the typical range of fat and viscosity in liquid emulsions such as milkshake.

Sucrose stearate was dispersed in distilled water for 15 min. Rapeseed oil was added and the mixture agitated using a high-shear blender (Silverson Machines Ltd., Chesham, UK) for 30 min. This premix was passed through a two-stage valve homogenizer (Panda 2k, Niro Soavi S.P.A., Sheffield, UK) at pressures of 500 and 50 bar for the first and second stages, respectively. HPMC and sucrose solutions were prepared in distilled water. The emulsions and solutions were stored at  $4 \pm 1$ °C overnight. The next day, HPMC and sucrose solutions were added to the o/w emulsion and gently mixed for 60 min. Emulsions were flavored with a mixture of four volatiles (Sigma-Aldrich Ltd., Gillingham, UK), which represented a fruity flavor and varied in hydrophobicity and concentration as explained in Results: ethyl hexanoate (LogP = 2.83), isoamyl acetate (LogP = 2.26), ethyl butyrate (LogP = 1.85) and *cis*-3-hexen-1-ol (LogP = 1.61) (LogP values fromEPI Suite, U.S. Environmental Protection Agency). The volatiles were predissolved in propylene glycol and added to the emulsion. Flavored emulsions were mixed overnight on a roller bed (SRT-2, Stuart Scientific, Redhill, UK) at 4 °C  $\pm$  1 °C and stabilized at room temperature for 2 h ( $22 \pm 1$  °C) before measurement. The final emulsion had a fairly symmetrical particle size distribution: 90% of the particles were  $<0.70 \,\mu\text{m}$  with an average particle size of  $0.26 \pm 0.05 \,\mu\text{m}$  (D<sub>3.2</sub>) (measured using a Malvern Mastersizer, Malvern Instruments, UK).

**Rheological Measurements.** The flow characteristics of each o/w emulsion were measured at 22 °C using a CVO rheometer (Bohlin Instruments, Cirencester, UK) at increasing shear rates from 1 to 100 s<sup>-1</sup>. Double-gap (40/50) geometry was used for the less-viscous emulsions, and cone and plate geometry (4°/40 mm) for the remainder. Three batches of each emulsion composition were prepared. One measurement was performed on each new batch. The power law region of each flow curve was fitted to eq 1 in order to estimate the power law parameters of the samples:

$$\eta = K\dot{\gamma}^n - 1 \tag{1}$$

where  $\eta$  is the apparent viscosity,  $\gamma$  is the shear rate, *K* is the consistency index, and *n* is the flow behavior index. The power law parameters (*K* and *n*) were used to calculate the Kokini oral shear stress ( $\tau$ ) according to eq 2 (*13*):

$$\tau = KV^{n} \left[ \frac{1}{h_{0}^{(n+1)/n}} + \left( \frac{F}{R^{n+3}} \times \frac{n+3}{2\pi K} \right)^{1/n} \times \frac{(n+1)t}{2n+1} \right]^{n2/(n+1)}$$
(2)

where  $\tau$  is the Kokini oral shear stress, V is the velocity of tongue (2



**Figure 1.** Viscosity curves of the nine o/w emulsions at different RO concentrations  $\bigcirc = 0.5$ ;  $\square = 3$ ;  $\triangle = 30$  g/100 g) and at different HPMC concentrations (without line = 0; solid line = 0.6; dotted line = 1.2 g/100 g).

cm s<sup>-1</sup>), *F* is the normal force (1N), *R* is the radius of plug (2.5 cm), *t* is time (1 s),  $h_0$  is the initial plug height (0.2 cm), *K* is the consistency index, and *n* is the flow behavior index.

**Aroma Release Measurements.** Aroma release from each of the flavored o/w emulsions was analyzed by an Atmospheric Pressure Chemical Ionization-Mass Spectrometer (APCI-MS; Micromass, Manchester, UK) (20). Samples were allowed to equilibrate at room temperature ( $22 \pm 1$  °C) for 2 h before measurement. Volatile molecules were ionized by a 4 kV corona discharge using different cone voltages (CV) as follows: m/z 145 (ethyl hexanoate, CV = 20), m/z 131 (isoamyl acetate, CV = 18), m/z 117 (ethyl butyrate, CV = 18) and m/z 83 (*cis*-3-hexen-1-ol, CV = 24).

Static Headspace Measurements. Two replicate aliquots (40 g) of each thickened-flavored emulsion were weighed into individual 100 mL Schott bottles (Fisher Scientific, Loughborough, UK) fitted with stoppered lids. The resultant headspaces were sampled in turn into the APCI-MS at 5 mL/min. Intensities of each compound were recorded as peak height ion counts, as data analysis was comparative and absolute concentrations of the aroma compounds were not required.

In Vivo Release Measurements. Nine panelists were asked to breathe in, sip 5 mL of sample emulsion from a spoon, close their mouths, swallow the sample, and then exhale and continue to breathe normally while resting their nose on the APCI-MS nasal sampling tube. Air from the nose was sampled into the APCI-MS source at 40 mL/min. Each assessor consumed all nine samples in a single session, with a break of 5 min between each sample. Plain crackers and water were used as palate cleansers. Exhalations were studied for 1 min after sample consumption, so that the temporal changes in breath aroma compounds could be followed. The breath by breath data were processed as described previously, and the data were then analyzed to extract two parameters, the maximum aroma intensity ( $I_{max}$ ) and the cumulative area under the 1 min release profile ( $A_{cum}$ ) (21).

Sensory Evaluation. A panel of 22 assessors (18 females and 4 males, aged 40–65), experienced in sensory techniques, evaluated the fruity flavor intensity of the o/w emulsions using a nine sample multiple paired comparison test (22) where all possible pairs of samples were evaluated by all panelists. Samples were presented in pairs at room temperature ( $22 \pm 1$  °C) in plastic cups, fitted with a lid, and labeled with a random three-digit code. In order to familiarize themselves with the flavor, all subjects were given a sample of the fruity-flavored emulsion prior to assessment. Panelists tasted the same volume of each sample from a spoon (5 mL) and were asked to judge (forced choice mode) which emulsion gave the most intense, initial, fruity flavor. All 36 possible pairings were evaluated by each panelist over four separate sessions with nine pairs of emulsions presented in each session. A break of 15 min was given between each set of two pairs to prevent fatigue. Plain crackers, lime juice, and still mineral water were supplied to assist

**Table 1.** Rheological Properties of the o/w Emulsions. Average Values and Standard Deviation (n = 3) of Flow Behavior Index, Consistency Index, and Kokini Oral Shear Stress

HPMC (g/100 g)	RO (g/100 g)	flow behavior index n <sup>a,b</sup>	consistency index K <sup>a,b</sup> (Pa s <sup>n</sup> )	Kokini oral shear stress $ au^b$ (Pa)
0	0.5	0.98 ± 0.01 a	$(1.6 \pm 0.2) \times 10^{-3}$ a	0.79 ± 0.06 a
	3	1.00 ± 0.01 a	$(1.7 \pm 0.1) \times 10^{-3}$ a	0.87 ± 0.03 a
	30	$0.89\pm0.06$ b	$(15.2 \pm 6.8) \times 10^{-3}$ a	1.88 ± 0.16 a
0.6	0.5	$0.88 \pm 0.01 \text{ b}$	0.13 ± 0.02 a	$5.91 \pm 0.38$ b
	3	$0.88 \pm 0.01 \text{ b}$	0.13 ± 0.02 a	$5.98\pm0.48$ b
	30	$0.79 \pm 0.01 \text{ c}$	$0.67 \pm 0.12 \text{ b}$	12.63 ± 1.14 c
1.2	0.5	$0.77 \pm 0.01 \text{ c}$	$1.43 \pm 0.12 \text{ c}$	18.64 ± 0.74 d
	3	$0.78 \pm 0.01 \text{ c}$	$1.45 \pm 0.18 \text{ c}$	19.32 ± 1.05 d
	30	$0.61\pm0.01~\text{d}$	$9.81\pm0.21~\text{d}$	$50.23\pm2.03~\text{e}$

<sup>a</sup> n and K calculated by fitting experimental data to the Power Law model ( $\eta = K \dot{\gamma}^{n-1}$ ) 0.996 < r < 1. <sup>b</sup> Different letters denote significant differences between samples ( $\alpha = 0.05$ ).

in cleansing the palate between samples. Presentation order of samples was randomized and balanced across the panel. Evaluations were conducted in isolated booths under controlled lighting conditions.

**Statistical Analysis.** *Rheological and Aroma Release Data Analysis.* Two-way ANOVA with interactions was applied to rheological and aroma release data. Experimental designs included two three-level factors: HPMC concentration (0; 0.6; 1.2 g/100 g), and RO concentration (0.5; 3; 30 g/100 g). Significant differences between individual samples were determined by the Tukey's test ( $\alpha = 0.05$ ). Analyses of variance were carried out using Statgraphics Plus 4.1.

Sensory Data Analysis. Panel judgements on each pair were subjected to a Friedman analysis (22). Rank sums were calculated for each sample, by assigning a score of 2 to the row total ("is more fruity flavor than") and 1 to the column total ("is less fruity flavor than"). Significant differences in flavor between samples were identified by calculating the Tukey's HSD value for comparing two rank sums ( $\alpha = 0.05$ ).

#### **RESULTS AND DISCUSSION**

Influence of Rapeseed Oil and HPMC Concentration on Rheological Properties. The effect of emulsion formulation on rheological behavior was studied at 22 °C. This temperature was chosen as the samples were consumed by a sip and swallow procedure, and therefore the samples were unlikely to experience significant changes from the serving temperature of the emulsions. The viscosity curves in Figure 1 demonstrate that emulsions without HPMC showed Newtonian behavior but emulsions with HPMC showed shear thinning behavior at the highest HPMC levels. The flow behavior indices in Table 1 reinforced this (and show statistical significances) as values around 1 indicate Newtonian behavior while values less than 1 indicate shear thinning. The fat content of the emulsion only had an effect on viscosity when 30 g/100 g fat was present; there was little if any difference between the viscosities of the 0.5 and 3 g/100 g emulsions (Figure 1). Analysis of variance showed that both rapeseed oil concentration and HPMC concentration effects were significant (P < 0.001) on all three variables considered, flow behavior index, consistency index, and oral shear stress.

The oral shear stress has been proposed as a more relevant indicator of in-mouth shear conditions and showed a range from around 0.8 to 50 Pa. This parameter increased significantly ( $\alpha = 0.05$ ) in the emulsions with HPMC when 30 g/100 g of oil was added. There was evidence of a HPMC-oil interaction (P < 0.05). The increase in both oral shear stress and consistency index due to the addition of 30 g/100 g of rapeseed oil was significantly higher ( $\alpha = 0.05$ ) in the emulsions with the higher HPMC concentration. The results are in agreement with previous studies showing that the amount of thickener and fat content interact to modify rheological characteristics particularly viscosity and flow behavior index (23).

Table 2.	Aroma	Conc	entratio	on (mg/	kg) Ad	Ided to	the	o/w	Emulsions	in
Order to	Produce	e the	Same	Aroma	Relea	se uno	der S	tatic	Headspac	е
Conditior	is and ir	n Vivo	)							

RO concentration (g/100 g)	ethyl hexanoate	isoamyl acetate	ethyl butyrate	<i>cis</i> -3-hexen-1-ol
	Static Hea	Idspace		
0.5	5	10	20	250
3	25	25	40	250
30	200	100	200	400
	In Viv	0		
0.5	5	10	20	250
3	5	10	20	250
30	30	30	30	250

Influence of Rapeseed Oil and HPMC Concentration on Aroma Release. Static Headspace Results. Initially, all the emulsions were flavored with the same mixture of volatile compounds (ethyl hexanoate, 5 mg/kg; isoamyl acetate, 10 mg/ kg; ethyl butyrate, 20 mg/kg; cis-3-hexen-1-ol, 250 mg/kg) and the headspace concentrations measured at static equilibrium to study the effect of oil and HPMC on partition and binding of the volatile compounds. ANOVA of the headspace data showed that rapeseed oil concentration had a significant effect (P <0.001) on partition; however, HPMC concentration did not show any significant effect on the volatile headspace release (P >0.05), indicating that binding between these volatiles and the thickener was not significant. It is known that some compounds, such as allyl disulfide, do bind to HPMC (18). The decrease in headspace concentration when fat content was increased was higher for the more lipophilic compounds. The headspace concentration was reduced by factors of 37, 17, 12, and 2 for ethyl hexanoate, isoamyl acetate, ethyl butyrate, and cis-3-hexen-1-ol, respectively, as fat content increased from 0.5 to 30 g/100 g and the changes were consistent with the hydrophobicity of the compounds.

With the assurance that HPMC concentration did not affect the static equilibrium headspace behavior of the volatile compounds used in this experiment, the amount of each compound was adjusted to achieve the same static headspace concentrations as found in the 0.5 g/100 g oil emulsions (**Table 2**). For the most lipophilic compound (ethyl hexanoate)  $40 \times$ more was added to the 30 g/100 g emulsion while, for the least lipophilic compound, the factor was 1.8. Intermediate levels were added to the 3 g/100 g oil emulsion.

The headspace concentrations of these samples were measured (**Figure 2**), and ANOVA analysis showed no significant effect of HPMC or rapeseed oil concentration on the headspace concentration of ethyl hexanoate ( $P_{\text{HPMC}} = 0.12$ ;  $P_{\text{RO}} = 0.79$ ), isoamyl acetate ( $P_{\text{HPMC}} = 0.088$ ;  $P_{\text{RO}} = 0.099$ ), ethyl butyrate



Figure 2. Aroma intensity of ethyl hexanoate (a), isoamyl acetate (b), ethyl butyrate (c), and cis-3-hexen-1-ol (d) in static headspace at different HPMC concentrations (black bars = 0; light gray bars = 0.6; dark gray bars = 1.2 g/100 g).



Figure 3. Maximum intensity of *cis*-3-hexen-1-ol (a) and ethyl hexanoate (b) in vivo at different HPMC concentrations (black bars = 0; light gray bars = 0.6; dark gray bars = 1.2 g/100 g). Different letters denote significant differences between samples ( $\alpha = 0.05$ ). The same behavior was observed for the  $A_{cum}$  data of these compounds (results not shown).

 $(P_{\rm HPMC} = 0.80; P_{\rm RO} = 0.46)$  and *cis*-3-hexen-1-ol ( $P_{\rm HPMC} = 0.62; P_{\rm RO} = 0.14$ ). Thus the reformulation of the volatile compounds to produce the same headspace concentrations had been successful, and these samples exhibited iso-release in terms of static equilibrium headspace.

In Vivo Release Results. The emulsion samples adjusted for iso-static headspace release (as described above) were fed to nine panelists, and the maximum concentrations and total amounts delivered to the nose were measured (in vivo measurements). Analysis of variance showed that delivery to the nose ( $I_{max}$  and  $A_{cum}$ ) was not significantly affected by HPMC concentration (P > 0.58) for any of the volatile compounds used in this study (**Figure 3**). Oil content did not affect the delivery ( $I_{max}$  and  $A_{cum}$ ) of the least lipophilic compound (*cis*-3-hexen-1-oi; P > 0.05; **Figure 3a**). However, the delivery of the more lipophilic compounds (ethyl hexanoate, isoamyl acetate, and ethyl butyrate) was affected by the oil content (P < 0.01).  $I_{\text{max}}$  values of ethyl hexanoate were significantly higher ( $\alpha = 0.05$ ) when panelists consumed the emulsion with 30 g/100 g of oil compared to the emulsions with lower oil content (**Figure 3b**). The same behavior was observed for isoamyl acetate and ethyl butyrate and for the  $A_{\text{cum}}$  data of these compounds (results not shown).

Delivery to the nose was higher than expected from static headspace measurements when consuming 30 g/100 g fat emulsions, and this is in agreement with previous reports that the retention effect of oil on aroma release is smaller under in vivo conditions than under static headspace conditions (4–6).

Following these experiments, the amount of the aroma added to the samples was adjusted (**Table 2**) to give equal aroma

**Table 3.** Effects of HPMC and Oil Content of Emulsions on in Vivo Aroma Release Obtained from Two-Way ANOVA Analysis. Probability (*P*) Values<sup>a</sup>

	ethyl hexanoate		isoamyl acetate		ethyl butyrate		cis-3-hexen-1-ol	
	I <sub>max</sub>	A <sub>cum</sub>						
main effects	Р	Р	Р	Р	Р	Р	Р	Р
A: HPMC concentration	0.60	0.19	0.78	0.31	0.97	0.27	0.29	0.08
B: rapeseed oil concentration	0.21	0.00	0.10	0.02	0.07	0.62	0.21	0.64
interactions: $A \times B$	0.23	0.22	0.34	0.41	0.71	0.67	0.80	0.55

<sup>a</sup> If P value is less than 0.05, this factor has a significant effect on in vivo aroma release at the 95.0% confidence level.



Figure 4. APCI-MS breath results showing the effect of rapeseed oil concentration on the temporal release profile of ethyl hexanoate released from samples during consumption. Samples were formulated to deliver the same maximum intensity of aroma in vivo. The samples used to illustrated the effect in this figure contained no HPMC.

release in-nose from the emulsions (iso-release in vivo). It was only necessary to adjust levels in the 30 g/100 g oil emulsion, as the 0.5 g/100 g and 3 g/100 g emulsion (adjusted for isorelease headspace) showed very similar volatile delivery in vivo. The amounts of *cis*-3-hexen-1-ol (relatively hydrophilic) were the same for the three different oil contents but ethyl hexanoate, isoamyl acetate and ethyl butyrate required increases of  $6\times$ ,  $3\times$ , and  $1.5\times$ , respectively, to achieve the same in vivo release from the 30 g/100 g emulsion as the 0.5 g/100 g emulsion.

Reformulation was successful as the measured in vivo  $I_{max}$ values for all nine emulsions showed no statistical difference for any of the four compounds, but there were differences in  $A_{\rm cum}$ , especially for the more hydrophobic compounds (Table 3). Figure 4 illustrates the situation for ethyl hexanoate when flavor delivery data from nine panelists consuming nine emulsions was averaged. The higher oil contents prolonged the delivery of the lipophilic compounds such as ethyl hexanoate, an observation that has been reported previously (3, 24). The same behavior was observed for isoamyl acetate. Release of the least lipophilic flavor probes such as ethyl butyrate and cis-3-hexen-1-ol did not show much change, and their release profiles were unaffected by the oil concentration. Ideally, release should be balanced for both  $I_{\text{max}}$  and for  $A_{\text{cum}}$ . Practically this is difficult, and the significance of the change in  $A_{\rm cum}$  on flavor perception is not well documented. A previous experiment with flavored low and regular fat milk samples showed that balancing aroma delivery based on  $I_{\text{max}}$  release in vivo (10) produced samples whose flavor was indistinguishable by a panel of 90 people. In some food systems,  $A_{cum}$  may have a significant effect on perception, but, for these emulsion samples, sensory tests were conducted using samples with the same  $I_{\text{max}}$  but with different Acum values.

Less fr	uity fla	ivour
---------	----------	-------

More fruity flavour

218 220	245	269 270 277 281	295 301	
Ц	I	11 11		
Aª Dª	$B^{ab}$	$C^{cb}E^{cb}\ G^{cd}\ F^{cd}$	H <sup>cd</sup> I <sup>d</sup>	

*Key:* A=1.2% HPMC-30% RO

 A=1.2%
 HPMC-30%
 D
 D=0.6%
 HPMC-30%
 RO

 B=1.2%
 HPMC-3%
 RO
 E=0.6%
 HPMC-3%
 RO

 C=1.2%
 HPMC-0.5%
 RO
 F=0.6%
 HPMC-0.5%
 RO

G=0% HPMC-30% RO H=0% HPMC-3% RO I =0% HPMC-0.5% RO

**Figure 5.** Line diagram representations of the rank sum scores for fruity flavor in the multiple paired comparison test emulsions. Values above the line are the actual rank sums, different superscript letters denote significant differences between samples ( $\alpha = 0.05$ ).



**Figure 6.** Effect of in-mouth viscosity (measured by Oral Shear Stress) on flavor perception from a series of emulsions. All samples delivered the same aroma release in vivo but contained different amounts of oil and HPMC. For sample key, see Figure 5.

3. Influence of Rapeseed Oil and HPMC Concentration on Flavor Perception of Emulsions Adjusted To Deliver Isorelease in Vivo. The nine, iso-release emulsions were presented to the 22 panelists in a multiple-paired comparison test (9 emulsion samples and 36 pairs of samples), and the panelists were asked to choose (forced choice test) which sample had the most initial fruity flavor. The sensory results were processed to produce rank sums which indicate differences between the 9 emulsion samples as measured by the panelists. The Tukey's HSD value for this experiment was 30, which means that the rank sums have to differ by this value to be significantly different (P = 0.05) (22). Figure 5 shows the rank sums for each of the 9 emulsions. Sample A had the lowest initial fruity flavor intensity and sample I the most initial fruity flavor intensity, and these corresponded to the samples with the highest and lowest viscosities, respectively.

The samples could be divided into four groups based on their significant differences (P < 0.05). The first group was formed

by samples A (high HPMC-high RO), D (medium HPMChigh RO), and B (high HPMC-medium RO). This group was judged as the lowest fruity flavor. The second group was formed by the samples B (high HPMC-medium RO), C (high HPMClow RO), and E (medium HPMC-medium RO). The third group was formed by samples C (high HPMC-low RO), E (medium HPMC-medium RO), F (medium HPMC-low RO), G (low HPMC-high RO), and H (low HPMC-medium RO). Finally, the fourth group (F (medium HPMC-low RO), G (low HPMChigh RO), H (low HPMC-medium RO), and I (low HPMClow RO)) was judged as the strongest fruity flavor.

Examination of the rank sums and oil content of the emulsions showed that there was no simple relationship between oil content and rank sum nor between HPMC content and rank sum. The relationship between in-mouth viscosity (as expressed by the Kokini Oral Shear Stress) (13) and rank sum is shown in **Figure 6**. The flavor perception of emulsions adjusted to deliver isorelease in vivo decreased due to an increase of in-mouth viscosity. A polynomial quadratic ( $y = y_0 + ax + bx^2$ ) regression between rank sums scores for fruity flavor and Log (oral shear stress) was calculated and the best fit ( $R^2 = 0.73$ ) was eq 3. Therefore a large proportion of the variation of flavor ranking can be expressed solely by the oral shear stress parameters, which are determined by the oil/HPMC composition of the emulsions.

relative flavor ranking = 
$$294.19 - 28.41x - 8.35x^2$$
 (3)

where *x* is the Log (oral shear stress).

There are various explanations for the sensory results. First of all, since aroma release was the same for all samples, this factor can be ignored. Second, the viscosity of the samples may trigger some trigeminal sensation, and this signal could then modify the signals from the taste (sweetness) and aroma receptors during neural processing, leading to a decrease in perception. Third, the different viscosities of the emulsions may affect the release and/or transport of sucrose to the taste receptors and result in a decreased sweetness signal. Since overall fruity flavor is a combination of the aroma and sweetness signals (15, 17), a decrease in sweetness would result in a decrease in fruity flavor. In this experiment, the emulsions with high fat contents also contained slightly higher concentrations of sucrose in the aqueous phase compared to the low fat emulsions (4.7 g and 6.6 g sucrose per 100 g water), but this slight change was probably masked by the greater changes in the sample viscosity.

The hypothesis that tastant release is affected by emulsion viscosity while aroma release is not can be explained by considering the different release mechanisms in mouth. In the case of the aroma compounds, the key release mechanism is partition at the air—liquid interface in mouth, where the liquid is in the form of a thin film. For the taste compounds, the mechanism is mass transfer across the saliva layer which depends on the ability of the sample and saliva layers to mix. Ferry et al. (*16*) have shown that the release of a dye from various hydrocolloid samples when mixed with water is very different, albeit under low shear conditions, and with water, rather than saliva. No definite conclusion can yet be drawn until further experiments have investigated this phenomenon.

This study indicates that, even when aroma release is the same, fat reduction can affect flavor perception due to a change in viscosity and/or tastant release. It is clear that further understanding of the complex contribution of fat to flavor perception is still required to meet the demand for acceptable low fat food products. Some researchers (25) have suggested

that fat has a taste as well as a viscosity stimulus which could also influence perceived flavor. Fat may also impart other qualities (e.g., creaminess) to the emulsions affecting flavor perception, and these will be investigated in future studies.

# ABBREVIATIONS USED

HPMC, hydroxypropylmethyl cellulose; RO, rapeseed oil;  $I_{\text{max}}$ , maximum aroma intensity;  $A_{\text{cum}}$ , cumulative area.

# ACKNOWLEDGMENT

The assistance of Dr. T. Hollowood in sensory evaluation and the results discussion, and Dr. R.S.T. Linforth in APCI-MS techniques, is acknowledged.

### LITERATURE CITED

- Kritchhevsky, D. Fats and oils in human health. In *Food Lipids. Chemistry, Nutrition, and Biotechnology*, 2nd ed; Akoh, C. C., Min, D. B., Eds.; Marcel Dekker: Inc.: New York, 2002; pp 543–558.
- (2) Small, D. N.; Prescott, J. Odor/taste integration and the perception of flavour. *Exp. Brain. Res.* 2005, 166, 345–357.
- (3) Miettinen, S.-M.; Hyvönen, L.; Linforth, R. S. T.; Taylor, A. J.; Tuorila, H. Temporal aroma delivery from milk systems containing 0–5.% added fat, observed by free choice profiling, time intensity, and atmospheric pressure chemical ionization-mass spectrometry techniques. *J. Agric. Food Chem.* 2004, *52*, 8111– 8118.
- (4) Doyen, K.; Carey, M.; Linforth, R. S. T.; Marin, M.; Taylor, A. J. Volatile Release from an Emulsion: Headspace and In-Mouth studies. *J. Agric. Food Chem.* 2001, *49*, 804–810.
- (5) Weel, K. G. C.; Boelrijk, A. E. M.; Burger, J. J.; Jacobs, M. A.; Gruppen, H.; Voragen, A. G. J.; Smit, G. Effect of emulsion properties on release of esters under static headspace, in vivo, and artificial throat conditions in relation to sensory intensity. *J. Agric. Food Chem.* **2004**, *52*, 6572–6577.
- (6) Roberts, D. D.; Pollien, P.; Antille, N.; Lindinger, C.; Yeretzian, C. Comparison of nosespace, headspace, and sensory intensity ratings for the evaluation of flavor absorption by fat. *J. Agric. Food Chem.* **2003**, *51*, 3636–3642.
- (7) Miettinen, S.-M.; Tuorila, H.; Piironen, V.; Vehkalahti, K.; Hyvönen, L. Effect of emulsion characteristics on the release of aroma as detected by sensory evaluation, static headspace gas chromatoraphy, and electronic nose. *J. Agric. Food Chem.* 2002, 50, 4232–4239.
- (8) van Ruth, S. M.; King, C.; Giannouli, P. Influence of lipid fraction, emulsifier fraction, and mean particle diameter of oilin-water emulsions on the release of 20 aroma compounds. J. Agric. Food Chem. 2002, 50, 2365–2371.
- (9) Guyot, C.; Bonnafont, C.; Lesschaeve, I.; Issanchou, S.; Voilley, A.; Spinnler, H. E. Effect of fat content on odor intensity of three aroma compounds in model emulsions: δ-decalactone, diacetyl and butyric acid. J. Agric. Food Chem. 1996, 44, 2341– 2348.
- (10) Shojaei, Z. A.; Linforth, R. S. T.; Hort, J.; Hollowood, T.; Taylor, A. J. Measurement and manipulation of aroma delivery allows control of perceived fruit flavour in low and regular fat milks. *Int. J. Food Sci. Technol.*, in press (published online at http:// www.blackwell-synergy.com/toc/ifs/0/0).
- (11) Wood, F. W. Psychophysical studies on the consistency of liquid foods. In *Rheology and texture of foodstuffs*; SCI Monograph No.27; Society of Chemical Industry: London, UK, 1968; pp 40–49.
- (12) Richardson, R. K.; Morris, E. R.; Ross-Murphy, S. B.; Taylor, L. J.; Dea, I. C. M. Characterization of the perceived texture of thickened systems by dynamic viscosity measurements. *Food Hydrocolloids* **1989**, *3*, 175–191.
- (13) Elejalde, C. C.; Kokini, J. L. The psychophysics of pouring, spreading and in-mouth viscosity. J. Texture Studies 1992, 23, 315–336.

- (14) Cook, D. J.; Hollowood, T. A.; Linforth, R. S. T.; Taylor, A. J. Oral shear stress predicts flavour perception in viscous solutions. *Chem. Senses* 2003, 28, 11–23.
- (15) Pfeiffer, J. C.; Hollowood, T. A.; Hort, J.; Taylor, A. J. Temporal synchrony and integration of sub-threshold taste and smell signals. *Chem. Senses* **2005**, *30*, 539–545.
- (16) Ferry, A.-L.; Hort, J.; Mitchell, J. R.; Cook, D. J.; Lagarrigue, S.; Valles Pamies, B. Viscosity and flavour perception: why is starch different from hydrocolloids? *Food Hydrocolloids*, in press.
- (17) Hollowood, T. A.; Linforth, R. S. T.; Taylor, A. J. The effect of viscosity on the perception of flavour. *Chem. Senses* 2002, 27, 583–591.
- (18) Cook, D. J.; Linforth, R. S. T.; Taylor, A. J. Effects of hydrocolloid thickeners on the perception of savory flavors. J. Agric. Food Chem. 2003, 51, 3067–3072.
- (19) Araujo, I; Rolls, E. Representation in the Human Brain of Food Texture and Oral Fat. J. Neurosci. 2004, 24, 3086–3093.
- (20) Taylor, A. J.; Linforth, R. S. T.; Harvey, B. A.; Blake, A. Atmospheric pressure chemical ionisation mass spectrometry for

in vivo analysis of volatile flavour release. *Food Chem.* **2000**, *71*, 327–338.

- (21) Linforth, R. S. T.; Taylor, A. J. Persistence of volatile compounds in the breath after their consumption in aqueous solutions. J. Agric. Food Chem. 2000, 48, 5419–5423.
- (22) Meilgaard, M.; Civille, G. V.; Carr, B. T. In *Sensory Evaluation Techniques*, 3rd ed.; CRC Press: New York, 1991.
- (23) Daget, N.; Joerg, M.; Bourne, M. Creamy perception I: In model dessert creams. J. Texture Studies 1987, 18, 367388.
- (24) Malone, M. E.; Appelqvist, I. A. M.; Norton, I. T. Oral behaviour of food hydrocolloids and emulsions. Part 2. Taste and aroma release. *Food Hydrocolloids* **2003**, *17*, 775–784.
- (25) Mattes, R. Fat: the sixth taste –implications for health. *Food Aust.* **2003**, *55*, 510–514.

Received for review June 1, 2006. Revised manuscript received September 5, 2006. Accepted September 17, 2006. S.B. is grateful to MECD (Spain) for financial support through a research grant.

JF061537K