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Elizabeth Guichard, Marie Repoux, E.M. Qannari, H  l  ne Labour  , Gilles Feron. Model cheese aroma perception is explained not only by in vivo aroma release but also by salivary composition and oral processing parameters. *Food and Function*, 2017, 8 (2), pp.615-628. 10.1039/C6FO01472K . hal-01564771

HAL Id: hal-01564771

<https://u-bourgogne.hal.science/hal-01564771v1>

Submitted on 22 Feb 2018

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1 **Model cheese aroma perception is not only explained by *in vivo* aroma**
2 **release but also by salivary composition and oral processing parameters**

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8
9 **Abstract**

10 The aim of the present paper was to determine, on four model cheeses differing in fat content and firmness consumed by
11 fourteen well characterised subjects, the respective impacts of *in vivo* aroma release, bolus rheology, chewing activity,
12 mouth coating and saliva composition on dynamic aroma perception. The originality of the approach was to consider all the
13 parameters together and to be able to evaluate their relative contribution using multi-block partial least square (MB-PLS)
14 regression. Fruity aroma perception of the more hydrophilic compound (ethyl propanoate) was related to its dynamic
15 release parameters before swallowing whereas blue cheese aroma perception of the more hydrophobic compound (nonan-
16 2-one) was related to its dynamic release parameters after swallowing and was highly impacted by mouth coating.
17 Moreover MB-PLS approach made it possible to evidence the combined effects of saliva composition and cross-modal
18 interactions to understand why in some cases dynamic aroma perception could not be explained by dynamic *in vivo* aroma
19 release data. Subjects with low sodium content in saliva perceived fruity aroma which is not congruent with saltiness as less
20 intense and salt- congruent (blue cheese) aroma as more intense, which was explained by their higher sensitivity to salt.
21 Subjects with a high lipolysis activity perceived fruity aroma which is not congruent to fat as less intense and fat-congruent
22 (blue cheese) aroma as more intense, which should be explained by the link between lipolysis activity and fat sensitivity.
23 These results could be considered for the reformulation of foods towards specific populations taking into account
24 nutritional recommendations.

25 **Key words:** aroma perception, aroma release, cheese, bolus rheology, saliva composition, chewing behaviour

26 Introduction

27 Aroma perception is an important aspect of food acceptability by consumers. However it is often very difficult to
28 directly explain aroma perception during food consumption by the amount of aroma compounds in the food. One
29 explanation is that aroma compounds have first to be released from the food matrix to the vapour phase in the
30 buccal cavity then transferred into the nasal cavity via the velum to reach the olfactory receptors¹. The partition of
31 aroma compounds between the food matrix and the vapour phase depends on the affinity of aroma compounds
32 for the macromolecules present in the food² which can be determined by measuring the vapour/matrix partition
33 coefficients of aroma compounds. These coefficients depend both on the physico-chemical properties of the
34 aroma but also on the food matrix composition. As an example hydrophobic aroma compounds are more soluble
35 in fat than in water and thus are better released in the vapour phase from products with a reduced fat content³.
36 Proteins are able to form reversible or irreversible binding with aroma compounds which will impact aroma
37 release in the vapour phase^{4,5}. In case of irreversible binding such as the formation of Schiff base between amino
38 groups of proteins and aldehydes⁶, the aroma compounds cannot be released in the vapour phase and thus cannot
39 be perceived. In case of reversible binding the aroma perception is lowered⁷. The major effect of hydrocolloids is
40 to increase food viscosity and thus decrease the transfer of aroma compounds from matrix to vapour^{8,9}, however
41 inclusion complexes are formed between amylose and specific aroma compounds which also impacts aroma
42 perception¹⁰. During consumption, food is mixed with saliva and broken down during the masticatory step to form
43 a swallowable bolus. During this step, aroma compounds are transferred from food to saliva before being released
44 in the nasal cavity. This transfer of aroma compounds is highly dependent on food composition and texture but
45 also on subject's physiology¹¹. Thanks to the development of on line aroma release technologies, such as
46 atmospheric pressure chemical ionisation (APCI-MS)¹² or proton transfer reaction mass spectrometry (PTR-MS)¹³,
47 it was possible to better understand the impact on dynamic *in vivo* aroma release of food matrix composition and
48 structure^{14,15} and of physiological parameters such as chewing, swallowing, saliva flow^{16,17}. Concerning the impact
49 of food matrix, increasing fat content results in a decrease in maximum intensity and overall *in vivo* aroma release
50 together with an increase in aroma remanence as was observed in gelatin gels¹⁸, whey protein gels¹⁵ or model
51 cheeses¹⁹, which was explained by a lower solubility of hydrophobic aroma compounds in fat. Even if food
52 rheology could explain chewing activity during consumption²⁰, bolus properties and in-mouth bolus rheology
53 better explain *in vivo* aroma release²¹. The amount of cheese remaining in the mouth after swallowing has also
54 been found important to explain *in vivo* aroma release. A higher amount of fat product remaining in the mouth
55 leads to a lower release of the more hydrophobic compounds before swallowing due to their high affinity for the
56 food bolus and to a longer aroma persistence in the breath^{16,22}. The less hydrophobic compounds are less

57 impacted by mouth coating but are more quickly released from firmer cheeses, due to a higher food breakdown¹⁶,
58 ¹⁷. When consuming solid foods, differences in chewing activities between subjects are responsible for differences
59 in aroma release, an intense chewing work leads to a higher amount of aroma release¹⁷.

60 However despite the development of *in vivo* measurements, results in the literature in the field of dairy products
61 showed that no clear relationship exists between *in vivo* aroma release and aroma perception due to the co-
62 existence of physicochemical and cognitive mechanisms²³. Cognitive mechanisms are due to cross-modal
63 interactions which vary according to the type of texture²⁴. In the case of semi-liquid foods such as yoghurts, an
64 increase in viscosity induced a decrease in both aroma release and aroma perception²⁵, whereas for solid cheeses
65 aroma perception decreased with firmness without any noticeable difference in aroma release²⁶. This finding was
66 explained by the attention paid by the subjects to the texture perception which likely led to a decrease in the
67 aroma perception. By delivering simultaneously different kinds of texture and different odorants, Bult et al.²⁷
68 showed that the perceived aroma intensity was reduced with increasing viscosity when the odour was presented
69 either ortho or retronasally. Moreover the perceived thickness was increased only when the odour was presented
70 retronasally simultaneously with swallowing time. An adaptation phenomenon was also evidenced in gel candies
71 as being responsible for the influence of taste and texture on aroma perception²⁸, by comparing the *in vivo* aroma
72 release curves and sensory time intensity curves which were recorded simultaneously. This study showed that
73 time at maximum chewing activity came first followed by time at maximum in nose concentration then time at
74 maximum perceived intensity. The time delays between release and perception increased when the product
75 remained longer in the mouth. The difficulties to relate results from analytic and sensory approaches was also
76 pointed out in brandies²⁹ and explained by sensory interactions between fruity and woody aroma³⁰. In coffee, the
77 burnt sensory notes could be associated with pyrazines detected by *in vivo* PTR-MS analysis but the effect of
78 adding sugar produced a change in aroma perception from burnt to caramel which could not be associated to any
79 change in aroma composition³¹, confirming the existence of sweet-aroma interactions²³. Odour-taste interaction
80 basically depends on the capacity of the two stimuli to form an appropriate combination in a food product context
81 (congruency)³². Thus, odour-taste integrated perception highly depends on learned associations, the context in
82 which the food is consumed and the consumer's previous experience³³. In experimental situations, odour-taste
83 interactions were found to be affected by the so-called dumping and halo effects³⁴, which resulted from the
84 tendency for sensory panellists to dump their sensations on several available scales (taste and/or odour).
85 Moreover, in more complex cheese flavour mixtures, NaCl, lactic acid, and aroma were found to be able to
86 enhance cheese flavour intensity and to compensate each other towards cheese flavour intensity³⁵, thus
87 suggesting complex taste-taste and taste-aroma interactions involved in the overall cheese flavour. So far, no
88 study has investigated cross-modal interactions using parameters related to oral physiology.

89 During the last decade, saliva has gained more and more interest in the field of sensory science. Indeed, due to its
90 role in oral clearance, bolus moisturizing and hydrolytic properties, saliva promotes the disintegration of the
91 matrix and thus the release of palatable active substances during food consumption. For example the salivary flow
92 rate influences aroma release³⁶ by a dilution effect³⁷. Salivary flow has also been positively associated with liking
93 for fat³⁸. Moreover, amylase activity can modify the bolus rheological properties³⁹ and thus modulates the
94 perception of salt in starch-based matrices⁴⁰. Even subject to controversy, lipase might be responsible for
95 hydrolysis of food triglycerides and thus fat perception^{41, 42}. Finally the catalytic activity of saliva against some
96 aroma compounds (e.g. esters, aldehydes) has been demonstrated *in vivo* and *in vitro*⁴³⁻⁴⁶ with consequences on
97 aroma release and thus on perception. Saliva is also composed of small and large molecules that contribute to
98 maintain a "salivary homeostasis" whose background level can regulate the dynamics of molecule release and thus
99 their sensory impact (sensory or nutritional)⁴⁷. For instance, it has been shown that a human subject with a high
100 salivary sodium concentration is less sensitive to saltiness⁴⁸.

101 A previous paper²² highlighted the role of bolus rheology and composition of stimulated saliva on aroma release
102 from cheeses varying in fat content and firmness. The aim of the present paper is to better understand the
103 physicochemical and physiological parameters which drive dynamic aroma perception. For that purpose, a group
104 of fourteen subjects were selected as representative of those participating to our previous study in order to
105 evaluate aroma perception as a function of time and to determine, on these well characterised subjects, the
106 respective impacts of *in vivo* aroma release, bolus rheology, chewing activity, mouth coating and saliva
107 composition on dynamic aroma perception.

108 **Experimental**

109 **Cheese products**

110 Four processed model cheeses were used¹⁶. They were composed of a mixture of cheddar, soft cheese, butter, melting
111 salts, protein powder (casein), salt and water. Two levels of texture (S=soft, F=firm) were obtained by varying the water
112 content and two ratio of fat to dry matter, a low level at 25% for low fat cheeses (lFf and lFfS) and a high level at 50% for high
113 fat cheeses (hFf and hFfS). The pH ranged from 5.27 to 5.55. The rheological properties of the cheeses were measured in a
114 large deformation at a rotation of 0.01 rad.s⁻¹ for 240 s using a Haake Viscotester (VT550 - Thermo electron GmbH,
115 Karlsruhe, Germany), as previously described¹⁶. The breakdown stress corresponds to the maximum strength necessary to
116 cause cheese breakdown, with the lowest values for the softest cheese (8129 ± 469 Pa for lFf and 8022 ± 1309 Pa for hFfS)
117 and the highest values for the firmest cheese (15253 ± 1231 Pa for lFf and 15556 ± 2307 Pa for hFf). The critical strain at
118 breakdown corresponds to the maximum rotation angle required to cause breakdown, with the lowest values for cheeses

119 with the highest fat content (0.273 ± 0.022 rad for hfS and 0.348 ± 0.061 rad for hfF), and the highest values for cheeses
120 with the lowest fat content (0.804 ± 0.056 rad for lfs and 0.836 ± 0.036 rad for lff). Two aroma compounds were added
121 during cheese production, a hydrophobic ketone, nonan-2-one (NO: $\log P = 2.9$, $6 \text{ mg}\cdot\text{kg}^{-1}$) and a hydrophilic ester, ethyl
122 propanoate (EP: $\log P = 1.4$, $25 \text{ mg}\cdot\text{kg}^{-1}$).

123

124 **Subjects**

125 In a first step, 48 subjects (23 female and 25 males) were selected from a group of 100 volunteers based on their good
126 dental and oral status, and on the repeatability of physiological parameters (salivary flow rate under resting and stimulated
127 conditions, respiratory flux, salivary composition)^{16, 20}. They were characterised for their oral volume, saliva composition
128 and flow, *in vivo* release measurement, chewing activity, bolus saliva content, bolus rheology and mouth coating. From this
129 group of 48 subjects, a subgroup of fourteen subjects (6 females and 8 males, average age: 40 years \pm 9) was selected for
130 the sensory analyses, on their ability to detect and recognise the two aroma notes. The subjects were not allowed to
131 smoke, eat or drink starting at least one hour before the different test sessions. All the subjects were informed of the
132 observational nature of this study. They gave their signed consent and received a financial compensation for their
133 participation. The study protocol was submitted to an Ethics Committee and was approved on 17 April 2008 by the Comité
134 de Protection des Personnes Est-1 (N°2008/15) and on 8 August 2008 by the Direction Générale de la Santé - France (N°
135 DGS2008-0196).

136 Subjects were characterised for specific physiological parameters described in the following sections.

137

138 **Oral volume**

139 Oral volume was measured using an Eccovision® acoustic pharyngometer (Hood Laboratories, USA), as described
140 previously⁴⁹. This device consists of two microphones and a horn driver mounted on a wave tube and connected to a PC-
141 compatible computer with signal conversion capabilities. The signal was converted into the surface change (cm^2) as a
142 function of the length of the oral cavity (cm). The subjects held the mouthpiece in their mouth with their teeth against the
143 flange and their tongue in a low position. To prevent air leaks, which could cause measurement errors, the subjects placed
144 their lips over the flange, sealing the mouthpiece. The subjects were instructed to breath with their nose during the
145 experiment. Values are expressed in cm^3 and correspond to the average of 10 measures.

146

147 **Saliva samples and flow**

148 Since our previous study²² highlighted that resting saliva poorly contributed to explaining aroma release whereas
149 stimulated saliva significantly impacted *in vivo* aroma release from soft cheeses, only results obtained with stimulated saliva

150 are taken into account in the present paper. Stimulated saliva was collected as previously described⁴⁹. The subjects chewed
151 a piece of Parafilm™ (0.5 g ± 0.2 g) for a period of 5 min and spit out the saliva every 30 seconds into a pre weighed cup
152 over a period of 5 minutes. The cups were weighted and the salivary flow rates were expressed in mL.min⁻¹. Immediately
153 after collection, the saliva samples were standardized by a first step of centrifugation for 30 min at 15000-x g to remove
154 bacteria and cellular debris. Thereafter the supernatants were stored at -80°C to stop metabolism until subjected to
155 biochemical analyses.

156

157 **Biochemical analyses of saliva samples**

158 **Protein concentrations.** Protein concentrations (Prot) were measured with a standard Quick Start Bradford protein assay
159 (Bio-Rad, France) with bovine serum albumin as the calibration standard.

160 **Enzyme activities.** All enzyme activities were expressed in International Enzyme Activity Units (U) per ml of saliva. One U is
161 defined as the amount of enzyme that catalyses the conversion of 1 micromole of substrate per minute. The lipolytic
162 (Lipolysis), proteolytic (Proteolysis), lysozymal (Lysozyme) and amylolytic (Amylase) activities were determined as
163 previously described^{42, 50, 51}.

164 **Sodium analysis.** The saliva samples were diluted to 1/20 (50 µL saliva in 950 µL filtered 18 mΩ Milli-Q-water (Millipore,
165 Bedford, MA, USA)) and filtered through a membrane (pore size = 0.45 µm, C.I.L., Sainte-Foy-La-Grande, France). The
166 amounts of sodium (Na) in saliva were determined by HPLC ionic chromatography using a Dionex ICS2500 ion
167 chromatographic system (Dionex, Voisins le Bretonneux, France) as previously described⁵². Quantifications were performed
168 using calibration curves realised with sodium standard solutions ranging from 0.1 to 10 mM in 22 mM sulfuric acid
169 (R²=0.999).

170

171 **Sensory analyses**

172 All sessions took place in an air-conditioned (21 ± 2°C) sensory testing room of the ChemoSens platform (Centre des
173 Sciences du Goût et de l'Alimentation, INRA, Dijon) using standardized booths equipped with computers. Subjects were
174 instructed to place each piece of cheese (6 g) in the mouth, and freely consume it while keeping the lips closed. The
175 products were presented in a random order at 17° C. All measurements were done in triplicate. Bread, apple and water
176 were used as mouth cleansers between two tests.

177 The subjects were firstly trained during eight sessions to recognise the odour of the two aroma compounds, blue cheese for
178 nonan-2-one (NO) and fruity for ethyl propanoate (EP), using triangular tests and recognition tests. Other training sessions
179 were conducted to familiarize the subjects with the discontinuous time-intensity methodology used. The last training

180 sessions were dedicated to the use of a continuous scale (0 to 10) to score the intensity of the two aroma compounds. Two
181 test sessions were then done using Fizz[®] software to score the intensity of the two aroma notes during the mastication, to
182 indicate the time of the first swallowing and then to score the intensity of the two aroma notes after each swallowing
183 event, during a total time of 3 min. During mastication and at each swallowing time, the intensity of each aroma note was
184 scored on the continuous scale (0-10). Each of the four cheeses was presented three times, in a random order, at 17°C.
185 From the aroma intensity perception at each swallowing event and the time of each swallowing event, time-intensity curves
186 were reconstituted for each aroma note and the following parameters were extracted: I_{max_S} for maximum intensity,
187 T_{max_S} for time to reach maximum intensity, I_{deg_S} for intensity at the first swallowing and T_{end_S} for time to reach the
188 end of perception (Fig. 1). The rate of perception ($V_{max_S} = I_{max_S}/T_{max_S}$) was then calculated as a fifth variable.

189 Insert Figure 1

190 ***In vivo* aroma release measurement**

191 The same protocol was applied for cheese consumption than that described for sensory analysis. The release of the two
192 aroma compounds was followed simultaneously in the nasal cavity as previously described¹⁶ using Atmospheric Pressure
193 Chemical Ionisation-Mass Spectrometry (APCI-MS) with an ion trap Esquire-LC mass spectrometer (Bruker Daltonique,
194 Wissembourg, France) according to their protonated molecular ion (MH⁺), which is the main ion: ethyl propanoate
195 (m/z=103) and nonan-2-one (m/z=143). Air was sampled from the nose at an average flow rate of 37 mL.min⁻¹ via a fused
196 silica capillary tubing (i.d. = 0.53 mm) heated at 150 °C and to which a 5 kV positive ion corona pin discharge was applied.
197 Each subject was asked to position the plastic tube in one nostril (the same for all the experiments) and to breathe
198 normally. This period (breath-blank phase) was used to record the potential residual signal of the previous sample until
199 return to the baseline and to control the regularity of breathing.

200 The curves were smoothed using a wavelet decomposition method to eliminate signal fluctuations due to the subjects'
201 breathing patterns. Two release phases were identified, the chewing phase (1) extended from placing the cheese in the
202 mouth to the first swallowing, and the post-swallowing phase (2) extended from the first swallowing to the time at which
203 the signal returned to its baseline level. For both release phases and for each aroma compound, four main parameters were
204 extracted from each individual release curve: the area under the curve (A_{1_P} and A_{2_P} (a.u.: arbitrary unit)) representing
205 the quantity of aroma released, the maximum intensity (I_{Max1_P} and I_{Max2_P} (a.u.)), the time to reach maximum intensity
206 (t_{Max1_P} and t_{Max2_P} (min)) and the release rate ($V_{max_P} = I_{Max1_P} / t_{Max1_P}$ (a.u./min)). These data, also not
207 quantitative, allow a direct comparison of the different release curves.

208

209 **Chewing activity**

210 Chewing activity was monitored during cheese consumption, simultaneously to aroma release. The muscle activity of the
211 superficial masseter and temporis muscles (left and right) during chewing was recorded by electromyography (EMG) using
212 gold surface electrodes (Grass technologies, West Warwick, RI, U.S.A), at 382 Hz, then the signal was amplified and
213 digitalized⁵³. The following parameters were extracted from EMG data: number of chewing cycles (Nber_Cycle), chewing
214 duration (Chew_time expressed in s), total muscle work (W_tot expressed in mV.s-1) and mean amplitude of contraction
215 (Ampl expressed in mV) which corresponds to a mean calculated from the amplitude values of each chewing cycle
216 registered in a whole chewing sequence²⁰.

217

218 **Bolus saliva content**

219 The percentage of dry matter and water content were determined using an infrared dryer for all cheeses and boluses
220 obtained just before swallowing. The percentage of moistening (Moist %) into the bolus was calculated from the bolus
221 water content (Bwc %), the bolus dry matter (Bdm %), the cheese dry matter (Cdm %) and the cheese water content (Cwc
222 %) as follows:

$$223 \text{ Moist}(\%) = \left(\frac{Bwc}{Bdm} \times Cdm \right) - Cwc$$

224 Three replicates per cheese and per subject were performed.

225

226 **Bolus rheology**

227 The subjects were instructed to chew the cheese samples until swallowing and to spit out the bolus into a truncated
228 syringe. Bolus rheological properties were measured using a compression test on an aliquot of 3 mL of bolus²⁰. The test was
229 performed using a mobile circle upper plate and a fixed circle lower plate as compression device, with a compression rate of
230 1 mm.s⁻¹. The bolus was subjected to a force F ranging between 0.01 N and 50 N. From the compression curve, particularly
231 two phases were highlighted. A "flow phase" during which the suspension begins to flow and the particles move
232 significantly in relation to one another at a height denoted as hflow (mm). Yield stress and viscous effects were described
233 respectively by the parameters sflow (Pa) and Kflow (Pa.s). A "particle phase" during which the mechanical response is
234 governed by the particle size which is represented by a height denoted hpart (mm) and the yield stress component denoted
235 spart (Pa). At the end of the compression, hend (mm) denotes the final height and Send (mm²) the area generated under
236 the maximal force. All measurements were done in triplicate.

237

238 **Mouth coating**

239 The amount of food that sticks to the oral surface after food ingestion (QRB) was quantified by the “mouth rinse” method⁵⁴.
240 Curcumin (Naturex, France) was added during cheese production (30 mg.kg⁻¹). Each subject was asked to place a piece of
241 cheese (6 g, at 17°C) in the mouth and to chew normally until swallowing. The subjects swallowed without cleaning
242 movement and then rinsed their mouth (with cleaning movements) with 4 mL of warm water at 50°C for 30 s, and spat it
243 into a vial. This rinsing procedure was applied two times consecutively and the spittle was cumulated in the same vials. The
244 fluorescence intensity of curcumin was quantified using a Perkin Elmer 1420 Multilabel Counter Victor 3V at an excitation
245 wavelength of 450 nm and an emission wavelength of 510 nm. All measurements were done in triplicate.

246

247 **Statistical analyses**

248 Analyses of variance (ANOVA) and Principal Component Analysis (PCA) were performed using XLSTAT[®] Software (Excel 97,
249 version 8.0, Paris, France). When a significant effect (p<0.05) was revealed by applying ANOVA, the Student-Newman-Keuls
250 test was used to compare the differences in least-squares (LS) means.

251 Statistical treatments for Partial Least Square (PLS) analysis were performed using the free software R 3.3.0 ([http://cran.r-](http://cran.r-project.org/)
252 [project.org/](http://cran.r-project.org/)), as already described for the treatment of *in vivo* release data obtained with the same cheeses and 48
253 subjects²². The main R package used for multivariate data analyses was «pls 2.5-0»⁵⁵. In a preliminary stage, the statistical
254 treatment consisted in a pre-processing step⁵⁶. More precisely, all the variables are mean centered; then the blocks of
255 variables are set to the same total variance. Finally, in order to explore the systematic variation patterns in the X blocks
256 which are likely to predict the systematic variation patterns in Y, Multiblock -PLSR (MB-PLSR) is applied.

257 The different variables used in the MB-PLS approach are presented in Table 1. They have been divided in six blocks. The Y
258 block corresponds to the variables to be explained that is, the sensory variables. The five other blocks correspond to the
259 explanatory variables (X1-X5). X1 is related to aroma release variables before (1) and after swallowing (2) as previously
260 described. The variables in the subsequent blocks were selected as highly impacting the *in vivo* aroma release²². X2 is
261 related to bolus rheological variables and bolus moistening. X3 is related to mouth coating and oral volume. X4 is related to
262 masticatory variables extracted from the EMG signals. X5 is related to the properties of stimulated saliva.

263 Insert Table1

264 **Results**

265

266 **Subjects' physiology**

267 The 14 subjects were selected from the 48 subjects participating to the *in vivo* release measurements¹⁶. Their physiological
268 characteristics (Table 2) cover the range of variability observed for the 48 subjects as presented in the PCA representation

269 (Fig.2). The principal plan represents 47% of the information. Axis 1 (26.76%) is explained by the salivary flow (Flux_S) and
270 the amount of sodium (Na_S) on its positive part, which are higher for subjects S027 and S094. Axis 2 (20.14%) is explained
271 by the amount of amylase (Amylase_S) , the amount of salivary proteins (Prot_S) and the oral volume (Oral_vol) on its
272 positive part, in relation with subjects S001, S052 and S086. At the opposite, S004 has a very low amount of amylase.

273

274 Insert Table 2.

275 Insert Figure 2

276 **Time intensity perception**

277 From the time intensity curves obtained for the four cheeses, 14 subjects and two odour notes, four variables
278 were extracted namely, maximum intensity (Imax_S), time to reach the maximum intensity (Tmax_S), intensity at
279 the first swallowing (Ideg_S) and time at the end of perception (Tend_S). The fifth variable, rate of perception
280 ($V_{max_S} = I_{max_S}/T_{max_S}$) was calculated. Due to the high inter-individual differences between subjects only two
281 variables show significant differences between the cheeses. Tmax_S is significantly higher ($p = 0.002$) for the blue
282 cheese aroma (NO) detected in low fat cheeses (IfS and lff) and Vmax_S is significantly higher ($p = 0.07$) for the
283 fruity aroma (EP) detected in high fat soft cheese (hfs). Means and standard deviations for the different cheeses
284 are reported in supplementary Table S1.

285 The high inter-individual variability was also observed on the *in vivo* release variables for the 48 subjects, which
286 was explained by differences in physiological parameters. Results of *in vivo* aroma release have already been
287 published¹⁶, However the data obtained for the 14 subjects selected for the sensory analysis are available in
288 supplementary Table S2. In order to better highlight the subject effect, a multiblock PLS analysis was performed on
289 these 14 subjects to explain sensory perception for each cheese and each aroma compound by the *in vivo* release
290 variables together with the physiological variables (Table 1).

291

292 **Relating sensory perception to *in vivo* aroma release and physiological parameters using MB-PLS**

293 MB-PLS is an extension of the PLS method, a class of regression models attempting to find relationships between
294 explanatory and response variables. In MB-PLS, the predictor variables are separated into subsets or blocks that
295 are standardized in order to balance for the size effect due to the measurement scale. It is a statistical approach
296 particularly relevant when different data sets reflecting different dimensions (physiology, physic, chemistry ..) and
297 with a different number of variables in each set are considered.

298 MB-PLS analyses were conducted on the different data sets to assess the extent to which the X blocks of variables
299 explain sensory perception during cheese matrix consumption (Fig. 3 and Fig. 5). The four cheeses and the two

300 aroma compounds nonan-2-one (NO) and ethyl propanoate (EP) were considered separately for the statistical
301 treatment.

302 Two different results are presented from MB-PLS analysis in the following sections, the importance of the block
303 and the projection of the variables in the correlation circle. At first, the importance of the blocks is calculated from
304 the sum of the beta-weight of the different variables constitutive of the block related to the corresponding
305 component and is expressed in %. It thus reflects the contribution of the block of variables to the determination of
306 the component. Higher is the percentage, higher is the contribution of the block for explaining sensory data.
307 Complementary to the importance of the blocks, the projections correspond to the depiction in a correlation circle
308 of the variables belonging to the different blocks. They thus represent the importance of the variables to the
309 components. Higher a variable is correlated to the components, higher it contributes to the model and thus to
310 explain perception.

311 The choice of the number of MB-PLS components to be retained for the importance of the block and the
312 projections was based on the total variance of block Y recovered by these components. For more details regarding
313 this aspect, we refer to a previous paper²². We restrict ourselves to the first two components because they explain
314 between 45.3% and 56% of the total variance of Y for the high fat cheeses and the two aroma compounds and
315 between 36.5 and 50.4% for the low fat cheeses and the two aroma compounds.

316 In order to avoid cumbersome graphical displays on the projections, only the variables with a correlation
317 coefficient with one of the two first components above 0.5 are depicted. The font size for each variable on the
318 projection (Fig. 4 and Fig. 6) reflects the importance of the correlation coefficient with the first two components;
319 with large font size indicating large correlations.

320 The results from the MB-PLS are presented and discussed successively for high fat cheeses and for low fat cheeses.

321

322 ***Relative Importance of the blocks in the projection for high fat cheeses***

323 The importance of the blocks of variables for the two first MB-PLS components is shown in figure 3 for the two
324 high fat cheeses and the two aroma compounds. The release block is important for the high fat firm cheese (hfF)
325 and both molecules and this on the two components (31.7% for NO on component 1 and 37% for EP on
326 component 2) and less important for high fat soft cheeses (with a maximum of contribution of 18.5%). The bolus
327 rheology block is mainly reflected by the second component for EP (25.9% for hfF-EP and 31.5% for hfS-EP) and
328 less reflected for NO. The importance of coating and oral volume is higher for EP than for NO, on the first
329 component. The masticatory variables take an important part in the explanation for both cheeses and both aroma
330 compounds. Notice also the high contribution of stimulated saliva, mainly for hfS-NO (48.9% on component 1 and

331 24.8% on component 2) for which the masticatory variables are less important (12.5% on component 1 and 21.3%
332 on component 2).

333 Insert Figure 3

334 Insert Figure 4

335 ***Projection of the main variables for high fat cheeses***

336 Figure 4 shows the projection of the variables from each block for the two high fat cheeses (hFF and hFS) and the two aroma
337 compounds (EP and NO). The sensory variables from the Y block (variables to be explained) are negatively correlated with
338 component 1 and for the high fat soft cheese, the rate of perception is negatively correlated with component 2. The other
339 variables (explanatory variables) are projected differently according to both the cheese and the aroma compound. For the
340 high fat firm cheese and ethyl propanoate (hFF-EP), the masticatory parameters are correlated with component 1, with the
341 chewing time (Chew_time) and number of chewing cycles (Nber_Cycle) on the positive part and the amplitude (Ampl) on
342 the negative part, the release parameters are correlated with component 2, with a higher correlation coefficient for
343 Vmax_P and lmax1_P, the amount of product remaining in the mouth (QRB) is projected at the opposite of the maximum
344 intensity perceived (lmax_S). Among the salivary parameters, the amount of proteins (Prot_S) is positively correlated with
345 component 1, opposite to the amount of sodium (Na_S) and of amylase (Amylase_S). The amount of lipolysis (Lipolysis_S) is
346 projected close to the QRB. For the high fat soft cheese and ethyl propanoate (hFF-EP), the most correlated explanatory
347 variables are the bolus rheology properties, with Send and Moist negatively correlated with component 1 together with the
348 sensory variables, opposite to hend and hpart. The masticatory parameters are also correlated with component 1, with
349 Chew_time and Nbe_Cycle on the positive part and Ampl on the negative part. Among the release parameters, only
350 lmax1_P and A1_P are positively correlated with component 1 and Vmax_P with a lower correlation. QRB is also projected
351 opposite to lmax_S. Concerning the high fat firm cheese and nonan-2-one (hFF-NO), the masticatory parameters
352 (Chew_time and Nbe_Cycle) are also positively correlated with component 1 and negatively with component 2, the release
353 parameters before swallowing (lmax1_P and A1_P) are positive correlated with component 1 whereas the release
354 parameters after swallowing (lmax2_P, A2_P) are negatively correlated with component 2. Concerning the high fat soft
355 cheese and nonan-2-one (hFS-NO), the salivary parameters are well represented on the projection, mainly Flux_S and Na_S,
356 positively correlated with component 1 and component 2 and opposite to the maximum intensity perceived. The release
357 parameters have only low correlations with these two components as represented by their small font size, but the time to
358 reach maximum intensities both before and after swallowing (tmax1_P and tmax2_P) are positively correlated with
359 component 2, opposite to the rate of perception. The masticatory parameters and QRB are negatively correlated with
360 component 1.

361

362 ***Relative Importance of the blocks in the projection for low fat cheeses***

363 The importance of the blocks of variables for the two first MB-PLS components is shown in figure 5 for the two low fat
364 cheeses and the two aroma compounds. The relative importance of the release block is always below 28%, it is higher for
365 nonan-2-one and the firm cheese (lff-NO, 27.5% on component 1 and 14.9% on component 2) and for ethyl propanoate
366 and the soft cheese (lfs-EP, 20.7% on component 1 and 19.8% on component 2). The bolus rheology block is reflected on
367 the two components with a contribution of 19.2% on component 1 and 6.8% on component 2 for lff-NO, 17.4% on
368 component 1 and 29% on component 2 for lfs-NO and intermediates for EP. Coating and oral volume are highly important
369 for lff-NO (25.2% on component 1 and 29.8% on component 2) and lfs-EP (48.7% on component 2). The masticatory
370 properties have a high impact on fruity aroma (EP) in the low fat firm cheese (46.5% on component 1 and 47% on
371 component 2), and in the low fat soft cheese (41.3% on component 1) but they seem to impact less blue cheese aroma
372 (NO). For nonan-2-one the salivary parameters are more relevant to explain sensory perception with 22.2% of contribution
373 on component 1 and 28.5% on component 2 for low fat firm cheese and 14.8% on component 1 and 46.5% on component 2
374 for low fat soft cheese.

375 Insert Figure 5

376 Insert Figure 6

377 ***Projection of the main variables for low fat cheeses***

378 Figure 6 shows the projection of the variables from each block for the two low fat cheeses and the two aroma
379 compounds on the two first MB-PLS components.

380 For the low fat firm cheeses (lff), the sensory variables related to aroma persistence (Tmax_S and Tend_S) are
381 negatively correlated with component 1, whereas the rate of perception (Vmax_S) and the maximum intensity of
382 perception (Imax_S) are negatively correlated with component 2 for ethyl propanoate and positively for nonan-2-
383 one. For the low fat soft cheeses, the rate of perception (Vmax_S) is positively correlated with component 1, the
384 maximum intensity (Imax_S) negatively with component 2, whereas the times (Tmax_S and Tend_S) are negative
385 correlated with component 1 for ethyl propanoate and negatively correlated with component 2 for nonan-2-one.
386 Concerning the explanatory variables, the masticatory parameters (Chew_time and Nber_Cycle) are negatively
387 correlated with component 1 for lfs cheese and the 2 aroma compounds and for lff cheese and ethyl propanoate
388 but negatively correlated with component 2 for lff cheese and nonan-2-one, whereas the amplitude is negatively
389 correlated with component 2 only for lff cheese and ethyl propanoate. Concerning the bolus rheology parameters
390 Hend, hpart and Kflow are always positive correlated with component 1 and component 2 whereas Moist and
391 Send are negatively correlated with component 1 and component 2. Concerning the release parameters, tmax2_P
392 is always negative correlated with component 1, the other parameters have lower correlations on this plan, except
393 Vmax_P on the positive part of component 1 for lff cheese and ethyl propanoate, lmax1_P on the positive part of
394 component 1 and negative part of component 2 for lff cheese and nonan-2-one, A2_P and A_P for both cheese

395 and nonan-2-one. The salivary parameters have only small correlations on this plane for the low fat soft cheese
396 but for the low fat firm cheese, the salivary flow (Flux_S) and the amount of sodium (Na_S) are negatively
397 correlated with component 2.

398

399 **Discussion**

400 Considering the results of MB-PLS on high fat cheeses (Fig. 4), different trends are observed for firm and soft cheeses. For
401 the firm cheeses, the perception of fruity aroma is more related to the release of ethyl propanoate (EP) before swallowing
402 whereas the perception of blue cheese aroma is more related to the release of nonan-2-one (NO) after swallowing. Ethyl
403 propanoate which is less hydrophobic is released faster from the fat cheese matrix¹⁶, and thus perceived at the beginning of
404 the oral processing for the firm cheese whereas nonan-2-one which is hydrophobic is released later and mainly after
405 swallowing¹⁹, which explains the longer time to reach maximum intensity and time to reach the end of perception. For the
406 soft cheeses the same correlations are found for nonan-2-one whereas no such correlation is found between sensory
407 parameters and release parameters for ethyl propanoate. Regarding the masticatory variables, it appears that the mean
408 amplitude of contraction (Ampl) is well correlated with the rate of fruity perception (EP) on component 1 for firm cheese
409 and component 2 for soft cheeses. This variable was already highlighted in our previous paper²² as responsible for a higher
410 rate of EP release. Thus, we confirm that subjects with high amplitude per burst will release more rapidly the hydrophilic
411 compound (EP) and we demonstrate that this higher release rate induce a more rapid perception. The other masticatory
412 variables better explain the blue cheese perception (NO) and more specifically aroma persistence. Subjects with a longer
413 chewing time (Chew_time) and larger number of bursts (Nber_Cycle) have a longer duration of blue cheese aroma (Tmax_S
414 and Tend_S) for both cheeses. This could also be explained by a longer time to reach the maximum NO release intensity
415 after swallowing (tMax2_P). The bolus rheology properties impact fruity perception (EP) more, with a positive relationship
416 between bolus moistening (Moist) and time to reach the end of perception (Tend-S), which could be explained by an
417 important bolus spreadability (Send). Bolus moistening (Moist) impacts EP release intensity (Imax1_P), a high bolus
418 moistening decreases the amount of release before swallowing. This could be explained by dilution with saliva which
419 decreases the rate of release of hydrophilic aroma compounds as already proposed using *in vitro*⁵⁷ and *in silico* models⁵⁸
420 and thus will delay the perception by increasing the persistence. The amount of product remaining in the mouth (QRB)
421 explains the perception of both aroma notes. QRB corresponds to the fat coating at the surface of the tongue and oral
422 mucosa after swallowing⁵⁴. For blue cheese perception (NO), it could be thus due to the retention of NO, hydrophobic
423 aroma compounds, in the fat, which delays its release and then enhances its perception. Conversely, the fruity perception
424 (EP) is projected opposite to fat coating in the mouth, i.e. higher is the coating lower is the perception. Many salivary
425 parameters impact aroma perception. Sodium concentration in saliva (Na_S) positively influences fruity perception (EP) and
426 negatively blue cheese perception (NO), whereas it was not found to have a strong impact on *in vivo* aroma release²².

427 Salivary lipolysis (Lipolysis_S) was depicted in the same direction of QRB in three projections (hF-EP, hF-NO and hS-NO)
428 witnessing a different impact on sensory perception depending on the fruity (EP) or blue cheese (NO) note. Salivary flow
429 (Flux_S), protein content (Prot_S), amylase activity (Amylase_S) also impact sensory perception but differently according to
430 the cheese and the aroma notes.

431

432 As for high fat cheeses, different trends are observed between firm and soft low fat cheeses (Fig. 6). For the low
433 fat firm cheese (lF) aroma persistence (Tmax_S and Tend_S) can be explained by the time to reach maximum
434 aroma release after swallowing (tMax2_P), the amount of product remaining in the oral cavity (QRB) and the oral
435 volume (Oral_Vol) for the two aroma compounds whereas the chewing time (Chew_time) and the number of
436 chewing cycles (Nber_Cycle) mainly explain fruity perception (EP). These correlations are logical because a higher
437 amount of product remaining in the mouth increases the amount of aroma released after swallowing²² which
438 should increase aroma persistence. Moreover a longer chewing time increases the total amount of aroma
439 released¹⁷ and thus aroma perception. The mean amplitude of contraction (Ampl) is depicted very similarly to the
440 rate of perception (Vmax_S) of the fruity note as it was the case for high fat cheeses. Similar to high fat cheeses,
441 stimulated saliva composition and in particular the sodium content (Na_S) impact aroma perception. High sodium
442 content is related to a high fruity aroma (EP) and low blue cheese aroma (NO) intensity.

443 For the low fat soft cheeses (lS), similar trends are observed for the fruity aroma (EP) but not for blue cheese
444 aroma (NO). The blue cheese aroma perception (NO) cannot be logically explained by NO release but is related to
445 saliva composition and more precisely to lipolysis (Lypolysis_S). Bolus moistening and bolus rheology highly
446 contribute to the perception of the two aroma notes. A higher aroma perception is related to a higher moistening
447 (Moist) and higher bolus spreadability which is explained by the area at the end of compression (Send). These
448 results are also in line with the higher salivary flow (Flux_S).

449

450 Thanks to MB-PLS statistical approach, some general trends can be highlighted concerning the explanation of
451 sensory perception by release and physiological properties. When consuming solid foods containing fat such as
452 cheeses, masticatory behaviour highly impacts the dynamic of aroma release and thus aroma perception. A large
453 mean amplitude of contraction increases rate of release and rate of perception. A longer chewing time increases
454 the time to reach maximum intensity after swallowing and the time to reach maximum perceived intensity.
455 Concerning the variables from the other blocks, the relationships are not always so simple to interpret, which may
456 be due to interactions between these variables. A high salivary flow leads to a high bolus moistening, a large bolus
457 area at the end of compression indicating a high bolus spreadability, which results in a lower rate of aroma
458 release²² but the direct consequence on aroma perception is not clearly evidenced.

459 However some variables were found to be related to sensory perception and not to aroma release. This is the case
460 for specific variables of saliva composition. The most striking finding is that lipolysis which is depicted in the same
461 direction to fat coating for NO and EP, is also depicted in the NO projection in the same direction to sensory
462 variables, while the opposite is observed for EP. This was particularly emphasized for high-fat cheeses. Salivary
463 lipolysis has been described as a marker of fat sensitivity and liking in human⁴¹. For example, a positive correlation
464 was found between the level of lipolysis and perceived fat intensity while the opposite was observed for liking in
465 subjects⁵¹ with a normal weight. On obese subjects, *in vivo* inhibition of lingual lipase led to a significant
466 enhancement of their sensory threshold for triolein⁵⁹. Moreover, compared to normal-weight subjects, obese
467 individuals exhibited a significant low level of salivary lipolysis thus reflecting the higher liking for fat as reported in
468 this population^{48, 60}. Concerning coating, fatty perception of oil in water emulsions has been previously related to
469 the level of fat retention at the surface of the tongue after consumption^{61, 62}. Altogether, these findings tend to
470 substantiate a positive correlation between the level of salivary lipolysis, fat coating and fat perception. These
471 findings support our hypothesis of cross-modal interactions between aroma perception and fattiness that differs
472 depending on the aroma. For NO (blue cheese aroma), the similar depiction of lipolysis, fat coating and sensory
473 variables should indicate a congruency between aroma perception and fat perception. Contrariwise, for EP (fruity
474 aroma), the depiction between lipolysis, fat coating and sensory variables should indicate a negative interaction
475 because of the non-congruency between this aroma and fat perception.

476 A similar hypothesis can be proposed for the effect of sodium content in stimulated saliva on aroma perception.
477 This property is depicted in the same direction as sensory variables for EP whereas it is in the opposite direction
478 for NO. The amount of sodium in saliva has already been described as an important factor which contributes in the
479 sensitivity for salt, i.e. the higher the concentration, the higher the detection threshold⁶³⁻⁶⁶. As for lipolysis, it is
480 likely that subjects exhibiting a high sodium concentration had a lower saltiness perception. Following our
481 hypothesis regarding the existence of salty-aroma interactions and considering that blue cheese aroma, unlike
482 fruity aroma, could be congruent to saltiness, subjects with a low salivary sodium content may perceive the
483 cheeses as being saltier. Since the subjects were instructed to rate the aroma intensity only and not the salty taste
484 in the cheese products during the sessions, they should have reported their salty perception on the blue cheese
485 aroma which is congruent with salty perception, due to a dumping effect³⁴. Contrariwise, the perception of a fruity
486 note may be increased for the subjects with high sodium content who perceived the cheeses as less salty, because
487 this aroma is not congruent with salty.

488 Conclusions

489 As a conclusion, this study allowed us to highlight the respective impacts of *in vivo* aroma release and physiological
490 properties on sensory perception. The originality of the approach is to consider all the variables together and
491 evaluate their relative contribution. Our study confirms the important role of masticatory variables for firm
492 cheeses and that of salivary properties for soft cheeses. The perception of the fruity aroma of the more
493 hydrophilic compound (ethyl propanoate) is related to its dynamic release parameters before swallowing whereas
494 the blue cheese aroma of the more hydrophobic compound (nonan-2-one) is related to its dynamic release
495 parameters after swallowing and is highly impacted by mouth coating. Moreover, it was evidenced that dynamic *in*
496 *vivo* aroma release does not always explain sensory perception. MB-PLS approach made it possible to evidence the
497 combined effects of saliva composition and cross-modal interactions to understand why in some cases dynamic
498 aroma perception could not be explained by dynamic *in vivo* aroma release data. Our study confirms the key role
499 of salivary sodium content and lipolysis activity in sensory perception. The main finding is that subjects with a high
500 sodium content in saliva perceive aromas which are not congruent with saltiness as more intense and salt-
501 congruent aromas as less intense and that subjects with a high lipolysis activity perceive aromas which are not
502 congruent to fat as less intense and fat-congruent aromas as more intense. These findings could help to better
503 understand the inter-individual differences in aroma perception and could be considered for the reformulation of
504 foods targeted for specific populations taking into account nutritional recommendations.

505 **Acknowledgements**

506 The acknowledgements come at the end of an article after the conclusions and before the notes and references.
507 This work received financial support from the French National Research Agency (ANR-07-PNRA-014), the Regional
508 Council for Burgundy and FEDER (European Union). The authors thank Etienne Sémon from ChemoSens Platform
509 (CSGA) for APCI experiments, Vincent Gigot from CSGA for APCI curve smoothing, Claude Yven for
510 electromyography, Chantal Septier for subject's physiology and sensory analysis, Elsa Ropiteau for sensory
511 analysis, Laboratoire de Rhéologie (Université Joseph Fourier Grenoble, France) for bolus rheology, Fromageries
512 Bel SA, Soredab (groupe Soparind Bongrain) and the panellists.

References

1. A. Buettner, S. Otto, A. Beer, M. Mestres, P. Schieberle and T. Hummel, *Food Chemistry*, 2008, **108**, 1234-1246.
2. M. Kopjar, I. Andriot, A. Saint-Eve, I. Souchon and E. Guichard, *Journal of the Science of Food and Agriculture*, 2010, **90**, 1285-1292.
3. L. Boisard, C. Tournier, E. Semon, E. Noiro, E. Guichard and C. Salles, *Flavour and Fragrance Journal*, 2014, **29**, 95-106.
4. S. Lubbers, P. Landy and A. Voilley, *Food Technology*, 1998, **52**, 68-214.
5. A. Tromelin, I. Andriot and E. Guichard, in *Flavour in food*, eds. A. Voilley and P. Etiévant, Woodhead Publishing Limited, Cambridge, CB1 6AH, UK, CRC Press edn., 2006, vol. Part 2 Flavour retention and release from the food matrix, pp. 172-207.
6. A. Meynier, V. Rampon, M. M. Dalgalarondo and C. Genot, *International Dairy Journal*, 2004, **14**, 681-690.
7. I. Andriot, M. Harrison, N. Fournier and E. Guichard, *Journal of Agricultural and Food Chemistry*, 2000, **48**, 4246-4251.
8. J. Delarue and P. Giampaoli, in *Flavour in food*, eds. A. Voilley and P. Etiévant, Woodhead Publishing Limited and CRC Press LLC, Cambridge, CB1 6AH (GBR), CRC Press edn., 2006, pp. 208-228.
9. S. Lubbers and E. Guichard, *Food Chemistry*, 2003, **81**, 269-273.
10. C. Heinemann, M. Zinsli, A. Renggli, F. Escher and B. Conde-Petit, *LWT - Food Science and Technology*, 2005, **38**, 885-894.
11. C. Salles, M. C. Chagnon, G. Feron, E. Guichard, H. Laboure, M. Morzel, E. Semon, A. Tarrega and C. Yven, *Critical Reviews in Food Science and Nutrition*, 2011, **51**, 67-90.
12. A. J. Taylor, R. S. T. Linforth, B. A. Harvey and A. Blake, *Food Chemistry*, 2000, **71**, 327-338.
13. W. Lindinger, A. Hansel and A. Jordan, *International Journal of Mass Spectrometry and Ion Processes*, 1998, **173**, 191-241.
14. E. Aprea, F. Biasioli, F. Gasperi, T. D. Märk and S. van Ruth, *Flavour and Fragrance Journal*, 2006, **21**, 53-58.
15. M. Mestres, N. Moran, A. Jordan and A. Buettner, *J. Agric. Food Chem.*, 2005, **53**, 403-409.
16. M. Repoux, H. Laboure, P. Courcoux, I. Andriot, E. Semon, C. Yven, G. Feron and E. Guichard, *Flavour and Fragrance Journal*, 2012, **27**, 414-423.
17. A. Tarrega, C. Yven, E. Sémon and C. Salles, *International Dairy Journal*, 2008, **18**, 849-857.
18. A. B. Boland, C. M. Delahunty and S. M. van Ruth, *Food Chemistry*, 2006, **96**, 452-460.
19. L. Boisard, I. Andriot, C. Martin, C. Septier, V. Boissard, C. Salles and E. Guichard, *Food Chemistry*, 2014, **145**, 437-444.
20. C. Yven, J. Patarin, A. Magnin, H. Labouré, M. Repoux, E. Guichard and G. Feron, *Journal of Texture Studies*, 2012, **43**, 309-318.
21. S. Prakash, D. D. Y. Tan and J. S. Chen, *Food Research International*, 2013, **54**, 1627-1635.
22. G. Feron, C. Ayed, E. M. Qannari, P. Courcoux, H. Labouré and E. Guichard, *PLoS One*, 2014, **9**, 1-15.
23. C. Tournier, C. Sulmont-Rosse and E. Guichard, in *Food*, ed. G. S. Books, Global Science Books Ltd., Royaume-Uni (GBR), 2007, vol. 1, pp. 246-257.
24. I. Gierczynski, E. Guichard and H. Laboure, *Flavour and Fragrance Journal*, 2011, **26**, 141-152.
25. A. Saint-Eve, N. Martin, H. Guillemain, E. Sémon, E. Guichard and I. Souchon, *Journal of Agricultural and Food Chemistry*, 2006, **54**, 7794-7803.
26. I. Gierczynski, H. Laboure and E. Guichard, *Journal of Agricultural and Food Chemistry*, 2008, **56**, 1697-1703.

27. J. H. F. Bult, R. A. de Wijk and T. Hummel, *Neurosci. Lett.*, 2007, **411**, 6-10.
28. S. Leclercq and G. Blancher, *Chem. Senses*, 2012, **37**, 689-700.
29. G. Fiches, A. Saint Eve, S. Jourden, I. Deleris, P. Bruneriea and I. Souchon, *Flavour and Fragrance Journal*, 2016, **31**, 31-40.
30. B. Atanasova, T. Thomas-Danguin, D. Langlois, C. Chabanet, S. Nicklaus and P. Etievant, *Chem. Senses*, 2005, **30**, A28.
31. M. Charles, A. Romano, S. Yener, M. Barnaba, L. Navarini, T. D. Mark, F. Biasoli and F. Gasperi, *Food Research International*, 2015, **69**, 9-20.
32. H. N. J. Schifferstein and P. W. J. Verlegh, *Acta Psychologica*, 1996, **94**, 87-105.
33. R. J. Stevenson, R. A. Boakes and J. Prescott, *Learning and Motivation*, 1998, **29**, 113-132.
34. C. C. Clark and H. T. Lawless, *Chem. Senses*, 1994, **19**, 583-594.
35. J. Niimi, A. I. Eddy, A. R. Overington, P. Silcock, P. J. Bremer and C. M. Delahunty, *International Dairy Journal*, 2014, **39**, 222-228.
36. A. Tarrega, C. Yven, E. Semon and C. Salles, *International Dairy Journal*, 2011, **21**, 358-364.
37. M. Doyennette, C. de Loubens, I. Deleris, I. Souchon and I. C. Trelea, *Food Chemistry*, 2011, **128**, 380-390.
38. C. Méjean, M. Morzel, E. Neyraud, S. Issanchou, C. Martin, S. Bozonnet, C. Urbano, P. Schlich, S. Hercberg, S. Péneau and G. Feron, *PLoS One*, 2015, **10**, e0137473.
39. L. Engelen, P. A. M. van den Keybus, R. A. de Wijk, E. C. I. Veerman, A. V. N. Amerongen, F. Bosman, J. F. Prinz and A. van der Bilt, *Archives of Oral Biology*, 2007, **52**, 518-525.
40. A. L. S. Ferry, J. R. Mitchell, J. Hort, S. E. Hill, A. J. Taylor, S. Lagarrigue and B. Valles-Pamies, *Journal of Agricultural and Food Chemistry*, 2006, **54**, 8869-8873.
41. G. Feron and J. Poette, *Oilseeds and fats, Crops and Lipids*, 2013, **20**, 102-107.
42. J. Poette, J. Mekoué, E. Neyraud, O. Berdeaux, A. Renault, E. Guichard, C. Genot and G. Feron, *Flavour and Fragrance Journal*, 2014, **29**, 39-49.
43. A. Buettner, *Journal of agricultural and food chemistry*, 2002, **50**, 3283-3289.
44. A. Buettner, *Journal of Agricultural and Food Chemistry*, 2002, **50**, 7105-7110.
45. S. Pagès-Hélary, I. Andriot, E. Guichard and F. Canon, *Food Research International*, 2014, **64**, 424-431.
46. P. Piombino, A. Genovese, S. Esposito, L. Moio, P. P. Cutolo, A. Chambery, V. Severino, E. Moneta, D. P. Smith, S. M. Owens, J. A. Gilbert and D. Ercolini, *PLoS One*, 2014, **9**, e85611.
47. R. Matsuo, *Crit. Rev. Oral Biol. Med.*, 2000, **11**, 216-229.
48. L. M. Bartoshuk, V. B. Duffy, J. E. Hayes, H. R. Moskowitz and D. J. Snyder, *Phil. Trans. R. Soc. B*, 2006, **361**, 1137-1148.
49. A. Mishellany-Dutour, A. Woda, H. Laboure, P. Bourdiol, P. Lachaze, E. Guichard and G. Feron, *PLoS One*, 2012, **7**, e41276.
50. S. R. Drago, M. Panouille, A. Saint-Eve, E. Neyraud, G. Feron and I. Souchon, *Food Hydrocolloids*, 2011, **25**, 659-667.
51. E. Neyraud, O. Palicki, C. Schwartz, S. Nicklaus and G. Feron, *Archives of Oral Biology*, 2012, **57**, 556-566.
52. M. Emorine, C. Septier, T. Thomas-Danguin and C. Salles, *Food Research International*, 2013, **51**, 641-647.
53. L. Mioche, P. Bourdiol and S. Monier, *Archives of Oral Biology*, 2003, **48**, 193-200.
54. M. Repoux, C. Septier, O. Palicki, E. Guichard, G. Feron and H. Labouré, *Archives of Oral Biology*, 2012, **57**, 81-86.
55. B. H. Mevik and R. Wehrens, *J. Stat. Softw.*, 2007, **18**, 1-24.
56. S. Hassani, H. Martens, E. L. Qannari, M. Hanafi and A. Kohler, *Chemometrics and intelligent laboratory systems*, 2012, **117**, 42-53.
57. S. Odake, J. P. Roozen and J. J. Burger, *Nahrung*, 1998, **42**, 385-391.
58. M. Doyennette, I. Délérís, G. Féron, E. Guichard, I. Souchon and I. C. Trelea, *Journal of Theoretical Biology*, 2014, **340**, 209-221.

59. M. Y. Pepino, L. Love-Gregory, S. Klein and N. A. Abumrad, *Journal of Lipid Research*, 2012, **53**, 561-566.
60. C. Vors, J. Draï, L. Gabert, G. Pineau, M. Laville, H. Vidal, E. Guichard, M.-C. Michalski and G. Feron, *International Journal of Obesity*, 2015, **39**, 1425-1428.
61. S. Camacho, K. Liu, A. Linden, M. Stieger and F. Velde, *Journal of Texture Studies*, 2015, **46**, 399-410.
62. S. Camacho, V. van Riel, C. de Graaf, F. van de Velde and M. Stieger, *Journal of Agricultural and Food Chemistry*, 2014, **62**, 5789-5795.
63. L. M. Bartoshuk, *The American Journal of Clinical Nutrition*, 1978, **31**, 1068-1077.
64. R. J. Contreras and F. A. Catalanotto, *Behavioral and neural biology*, 1980, **29**, 303-314.
65. J. Delwiche and M. O'Mahony, *Physiology & Behavior*, 1996, **59**, 605-611.
66. T. Morino and H. G. Langford, *Physiology & Behavior*, 1978, **21**, 45-48.

Table 1: presentation of the different blocks of variables used in the –MB-PLS Analyses. Y: variables to be explained, X: explanatory variables. a.u.: arbitrary units

Block	Abbreviation	Definition of the variable
Y: sensory parameters	Imax_S	Maximum intensity perceived (a.u.)
	Tmax_S	Time to reach the maximum intensity (s)
	Vmax_S	Rate of perception (a.u./s)
	Ideg_S	Maximum intensity at swallowing (a.u.)
	Tend_S	Time to reach the end of perception (s)
X1: aroma release parameters	A1_P	Area under the curve before 1 st swallowing (a.u.)
	A2_P	Area under the curve after 1 st swallowing (a.u.)
	A_P	Total area under the curve (A = A1_P + A2_P)
	IMax1_P	Maximum intensity before 1 st swallowing (a.u.)
	tMax1_P	Time to reach maximum intensity before 1 st swallowing (s)
	IMax2_P	Maximum intensity after 1 st swallowing (a.u.)
	tMax2_P	Time to reach maximum intensity after 1 st swallowing (s)
	Vmax_P	Rate of release (a.u./s)
X2: Bolus rheology	Moist	Moistening of the bolus just before the swallowing (%)
	sflow	Yield stress at flow phase of compression curve (Pa)
	spart	Yield stress at particle phase of compression curve (Pa)
	hpart	Bolus height at the beginning of the particle phase of compression curve (mm)
	Kflow	Consistency at the flow phase, which reflects bolus consistency (Pa.s)
	hflow	Bolus height at the beginning of the flow phase of compression curve (mm)
	hend	Bolus height at the end of compression (mm)
	Send	Area at the end of compression (mm ²)
X3:	QRB	Amount of product remaining in the oral cavity after swallowing (%)
Coating- oral vol	Oral_Vol	Volume of the oral cavity (cm ³)
X4: Masticatory parameters	Nber_Cycle	Number of chewing cycle
	Chew_time	Chewing duration (s)
	Ampl	Mean amplitude of contraction (mV)
	W_tot	Energy expended in chewing (mV/s)
X5: Properties of stimulated saliva	Flux_S	Salivary flow stimulated saliva (ml/min)
	Prot_S	Amount of salivary proteins stimulated saliva (mg/ml)
	Lipolysis_S	Amount of Lipolysis in stimulated saliva (mU/ml)
	Amylase_S	Amount of Amylase in stimulated saliva (U/ml)
	Proteolysis_S	Amount of Proteolysis in stimulated saliva (U/ml)
	Na_S	Amount of sodium in stimulated saliva (mM)

Table 2: oral physiological characteristics of the 14 subjects included in the study, mean, standard deviation, minimum and maximum values.

		minimum	maximum	mean	Standard deviation
Oral volume (cm ³)	Oral_Vol	26.85	62.16	39.03	9.73
Stimulated salivary flow (ml/min)	Flux_S	0.73	4.32	2.60	1.40
Protein (mg/ml)	Prot_S	0.57	1.31	1.04	0.31
Lipolysis (mU/ml)	Lipolysis_S	<0.01	1.32	0.90	0.74
Amylase (U/ml)	Amylase_S	1.27	30.6	20.86	11.00
Proteolysis (U/ml)	Proteolysis_S	0.04	0.51	0.13	0.12
sodium content (mM)	Na_S	2.09	37.82	15.82	11.03

Figure captions:

Figure 1. Time intensity curves obtained for one subject for blue cheese aroma (NO: nonan-2-one) for the four cheeses (hfS: high fat soft, hfF: high fat firm, lfS: low fat soft, lfF: low fat firm) showing the extracted parameters (Imax_S: maximum intensity, Tmax_S: time to reach maximum intensity, ldeg_S: intensity at swallowing and Tend_S: time to reach the end of perception).

Figure 2. Biplot representation from the PCA performed on the 14 subjects and their physiological parameters.

Figure 3. Bar charts representing the relative importance (%) of the different blocks of variables (X1-X5) for the different components obtained by means of MB-PLS analysis performed on fruity aroma (EP) and blue cheese aroma (NO) sensory data set and for the high fat (hfF & hfS) cheese products. Blue chart: X1-Aroma release, Green chart: X2-rheology, Orange chart: X3-coating and oral volume, Red chart: X4-EM data, Violet chart: X5-stimulated saliva composition.

Figure 4. MB-PLS results on dim1/dim2 for high fat (hf) cheeses: relationships between the X-blocks of explanatory variables (Blue arrows: X1-aroma release, Green arrows: X2-rheology, Orange arrows: X3-coating and oral volume, Red arrows: X4-EMG data, Violet arrows: X5-stimulated saliva composition) and the Y block of variables to be explained (Black arrows: sensory data for fruity aroma (EP) and blue cheese aroma (NO)). Top: firm high fat cheeses (hfF), bottom: soft high fat cheeses (hfS). The font size for each variable reflects the importance of the correlation coefficient with the two components.

Figure 5. Bar charts representing the relative importance (%) of the different blocks of variables (X1-X5) for the different components obtained by means of MB-PLS analysis performed on fruity aroma (EP) and blue cheese aroma (NO) sensory data set and for the low fat (lfF & lfS) cheese products. Blue chart: X1-Aroma release, Green chart: X2-rheology, Orange chart: X3-coating and oral volume, Red chart: X4-EM data, Violet chart: X5-stimulated saliva composition.

Figure 6. MB-PLS results on dim1/dim2 for low fat (lf) cheeses: relationships between the X-blocks of explanatory variables (Blue arrows: X1-aroma release, Green arrows: X2-rheology, Orange arrows: X3-coating and oral volume, Red arrows: X4-EMG data, Violet arrows: X5-stimulated saliva composition) and the Y block of variables to be explained (Black arrows: sensory data for fruity aroma (EP) and blue cheese aroma (NO)). Top: firm low fat cheeses (lfF), bottom: soft low fat cheeses (lfS). The font size for each variable reflects the importance of the correlation coefficient with the two components.

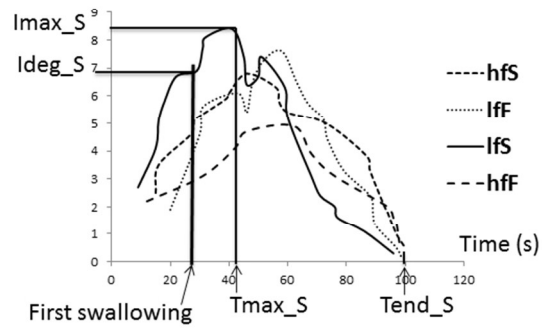


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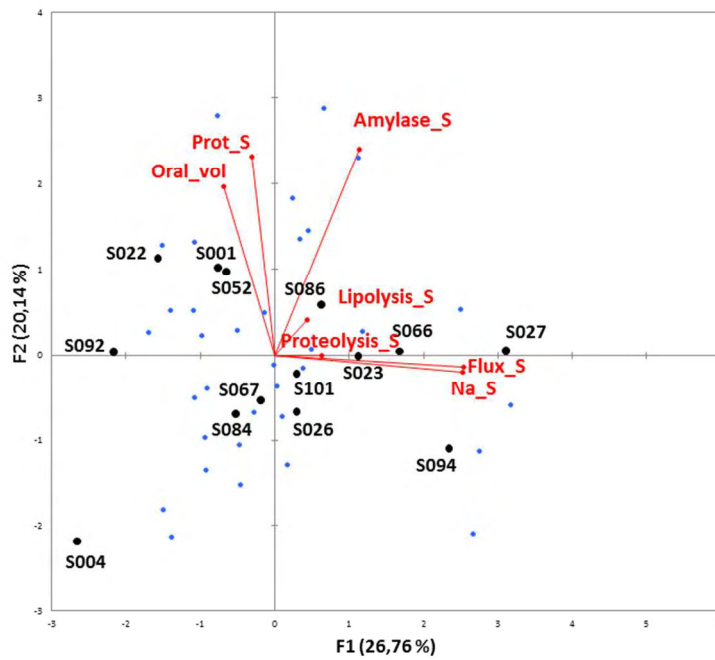


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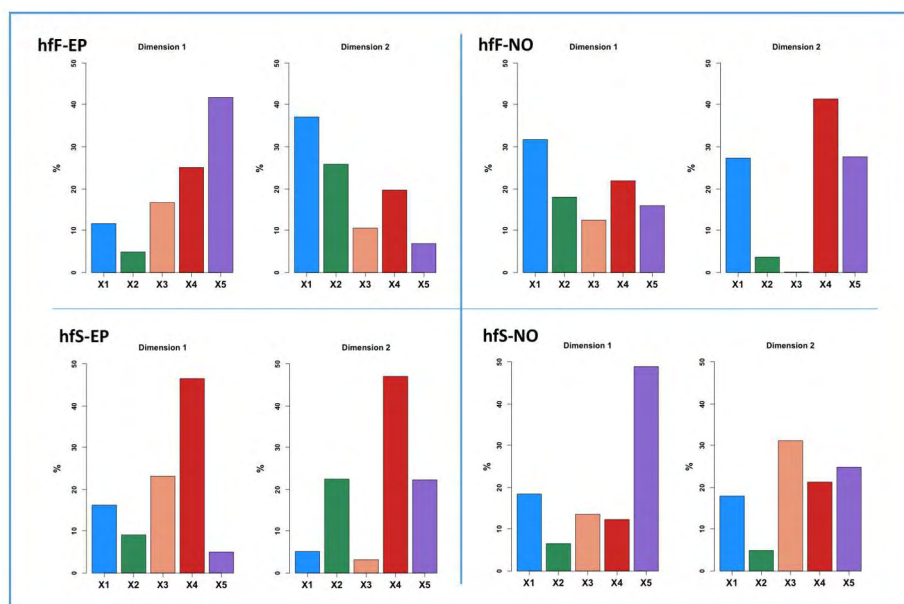


Figure 3. Bar charts representing the relative importance (%) of the different blocks of variables (X1-X5) for the different components obtained by means of MB-PLS analysis performed on fruity aroma (EP) and blue cheese aroma (NO) sensory data set and for the high fat (hff & hfs) cheese products. Blue chart: X1-Aroma release, Green chart: X2-rheology, Orange chart: X3-coating and oral volume, Red chart: X4-EM data, Violet chart: X5-stimulated saliva composition.

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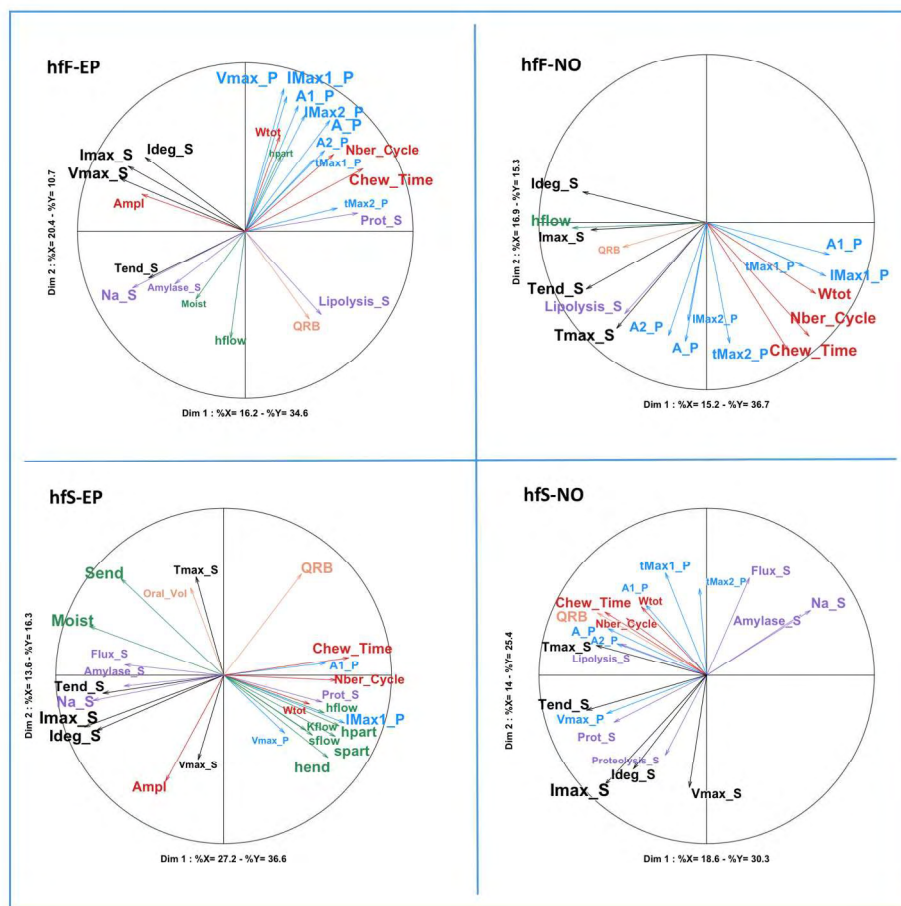


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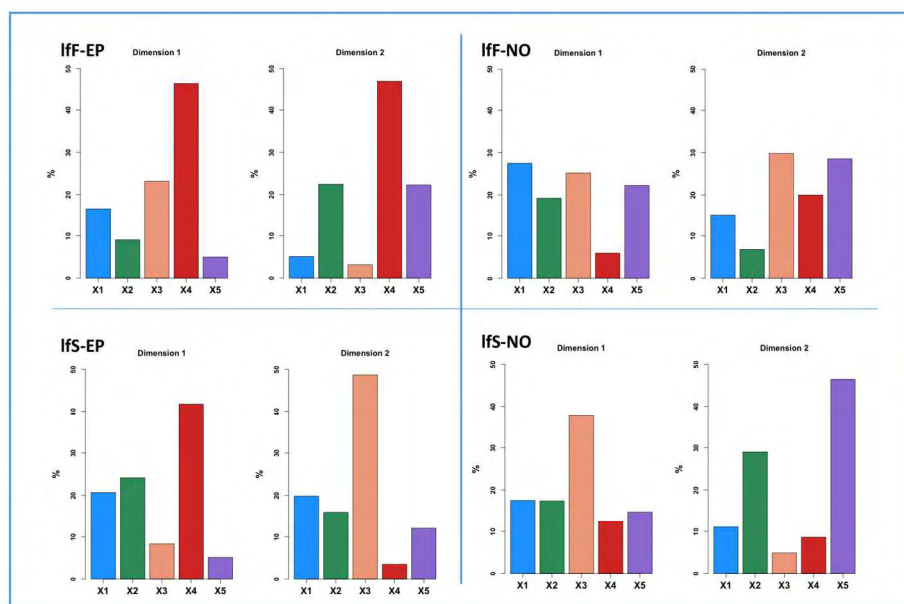


Figure 5. Bar charts representing the relative importance (%) of the different blocks of variables (X1-X5) for the different components obtained by means of MB-PLS analysis performed on fruity aroma (EP) and blue cheese aroma (NO) sensory data set and for the low fat (Iff & Ifs) cheese products. Blue chart: X1-Aroma release, Green chart: X2-rheology, Orange chart: X3-coating and oral volume, Red chart: X4-EM data, Violet chart: X5-stimulated saliva composition.

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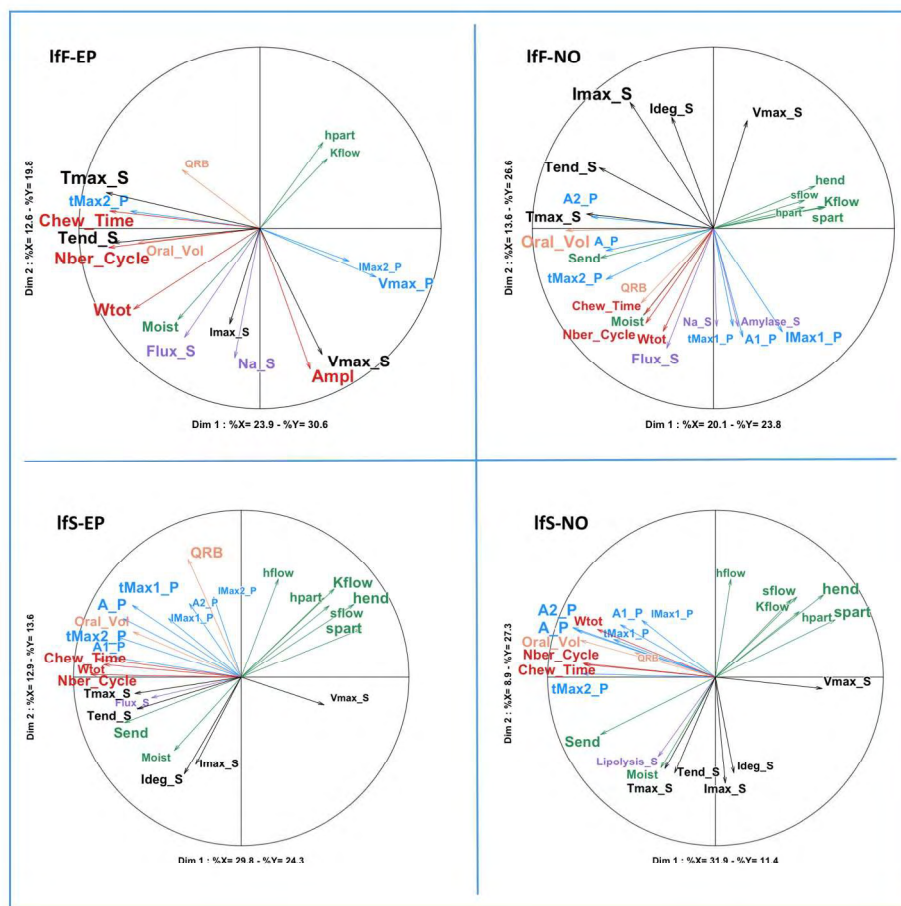


Figure 6. MB-PLS results on dim1/dim2 for low fat (If) cheeses: relationships between the X-blocks of explanatory variables (Blue arrows: X1-aroma release, Green arrows: X2-rheology, Orange arrows: X3-coating and oral volume, Red arrows: X4-EMG data, Violet arrows: X5-stimulated saliva composition) and the Y block of variables to be explained (Black arrows: sensory data for fruity aroma (EP) and blue cheese aroma (NO)). Top: firm low fat cheeses (Iff), bottom: soft low fat cheeses (Ifs). The font size for each variable reflects the importance of the correlation coefficient with the two components.

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