# TeRiFiQ

**Project no. 289397**

Combining Technologies to achieve significant binary Reductions in Sodium, Fat and Sugar content in everyday foods whilst optimizing their nutritional Quality

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# Deliverable D1.5

# Report on cheese with high level of unsaturated fatty acids

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**Contributor: ACTIA (ACTALIA)**

| Dissemination level | |
|---|---|
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| **CO** Confidential, only for members of the consortium (including the Commission Services) | [ ] |

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# 1. Summary

The objective of this work was to study the influence of the reduction of the salt content (-30% in comparison with standard cheese) and of the reduction of cheese saturated fatty acids SFA (-10% reduction in SFA = enrichment in unsaturated fatty acids UFA) on the composition and sensorial characteristics of white mould ripened Brie-type cheese.

These trials were achieved in March 2014 using milk from a local herd affiliated to the *Bleu Blanc Coeur* association, promoting the use of extruded linseed and alternative protein concentrates.

These low-SFA milks are commercial milks, which is a specificity of this study, compared with other works on experimental milks.

This milk was skimmed and standardized in fat with its own cream (rich in Unsaturated Fatty Acids) or with cream coming from conventional milk from an industrial cheese plant (standard milk). We obtain milk with -7/8% of saturated fatty acids SFA as forecast, even if the difference is not far from 60% vs 70% SFA instead of 70 vs 80% which was forecast. This difference is realistic and corresponds to real UFA enriched milks.

Cheeses with standard or reduced salt content were made with both types of milk.

These reductions of SFA and NaCl had a little influence on the sensory profile. Similar conclusions can be drawn from the technological results. We conclude that it is possible to make good Brie-type cheese in these conditions.

# 2. Material and methods

## 2.1 Milk treatment

Raw milk was purchased from a local dairy herd (EARL Gibet, F-35 Chevaigné) and from the reception of a cheese factory (Entremont-Sodiaal, F-35 Montauban de Bretagne). The farm milk (enriched in UFA) was warmed at 55°C in a plate-exchanger and skimmed at 500 L/h in a pilot centrifuge (Elecrem, Vanves, France), heat treated at 90°C for 60 sec (Actijoule, Evian, France) and cooled at 50°C.

At the same time, a casein concentrate was prepared by dispersing microfiltration retentate powder (PROMILK 852B, IDI, Arras, France) and milk permeate powder (IDI, Arras, France) in warm water (10/5/85) using a Tri-Blender. After 90 min rehydration at 50°C, the casein concentrate was added to skim milk and microfiltered on 1.4 µm ceramic membrane with a gradient of porosity (Membralox GP, Pall-Exekia, Tarbes, France).

Running conditions were 50-55°C, VCF 20, TMP 1,2b, permeation rate 500 $L.h^{-1}.m^2$. The permeate was cooled at <4°C in a plate heat-exchanger and stored in two separated double jacked stainless steel cheesemaking vats (with circulated iced water).

Conventional milk (rich in SFA) was skimmed using the procedure described above. Both UFA-rich cream and SFA-reach cream were analyzed by IR (Lactoscope, Delta Instruments) and their fat content was adjusted to 400 g/kg by adding skim milk. Standardized cream was then heat treated at 120°C for 1 min. in order to inactivate thermoresistant bacteria. Finally, each cream type was added to the microfiltered casein enriched skim milk to standardize cheesemilk with either SFA-rich fat or UFA-rich fat. Both batches of milk were stored at 2-4°C until use (up to 60 hours). The target composition of the cheese milk was:

- Casein 36 g/Kg
- Denatured whey proteins: 2 g/kg
- Fat 75 g/L
- Lactose 45 g/kg

## 2.2 Cheesemaking

Cheese manufacture was performed on the dairy technology platform of the UMR STLO (INRA Rennes) using the soft cheese pilot line (Technal, F-79024 Niort, France). The temperature and hygrometry of the room is controlled during the manufacture: 33°C – 97% relative humidity. The flow chart of the cheesemaking process is given in table I in Annex.

The milk ripening was done with 50 L of each milk type in double jacked stainless-steel vats in order to make two twin-vats (interval 5 min), one being dedicated to standard salt content, the other to reduced salt-content. When the target value of renneting was reached, the milk was transferred into two 50L semi-cylindrical stainless-steel vats (25kg of standardized milk per vat).

The coagulum was cut up with vertical knives and a knife grid into 1.5 x 1.5 x 1.7 cm. The stirring step was automatically performed with a single paddle. After having taken a part of the whey off, the moulding was carried out manually by pouring the curd into mould (20 cheeses per vat).

The experimental design is given in Table II (below). The four experimental conditions were randomly made during three successive days.

The salt concentration of the static brine was adjusted after the brining of each batch of cheeses: around 120 g of NaCl were added and dispersed in the brine.

Table II: Experimental design

| Fat composition<br>Salt content | UFA enriched fat | Standard fat |
|---|---|---|
| Standard | X | X |
| Reduced | X | X |

## 2.3 Analysis

The milks and creams fat and proteins, from the various steps of the cheesemilk standardization, were analysed using infrared spectrometer (Lactoscope, Delta Instruments).

The concentration of denatured whey proteins was calculated as the sum of the denatured proteins present in the cream and in the casein enriched skim milk.

Denatured whey proteins from the cream =

$$Q_{cream} \times (NCN_{\text{cream before heat treatment}} - NCN_{\text{cream after heat treatment}}) + Q\,\text{cream} \times (NCN_{\text{whole milk before heat treatment}} - NCN_{\text{whole milk after heat treatment}}) \times (1-Fat_{cream})/(1- Fat_{\text{whole milk}})$$

where Q is : quantity, NCN: non casein nitrogen and Fat: milk fat content

Denatured whey proteins from the casein-enriched milk =

$$Q_{\text{skim milk}} \times (NCN_{\text{whole milk before heat treatment}} - NCN)/(1- Fat_{\text{whole milk}})$$

Sensory analysis

The sensory analysis was realized with 15 panellists who have been trained to the sensory evaluation of cheeses according to ISO 8586-1. In order to validate the attributes, a training session was made before the determination of the sensory profile of the cheeses (3 sessions: 3 products per session).

The cheeses were tempered at 15°C for two hours before the sensorial analysis.

Three sessions were made with the 4 cheese types of each day of cheesemaking. Anova was made using a block factor (day of cheese manufacture + day of sensory analysis) and three factors: salt content (standard or reduced), fat composition (standard or UFA enriched) and panellists.

For Principal Component Analysis, the sole descriptors showing a *p value* < 0.15 for the factors salt, fat or interactions between fat and salt were selected.

The descriptors are listed below:

| | Lower limit (0) | higher limit (10) |
|---|---|---|
| **Aspect of the rind** | | |
| Rind Colour | white | ▫ yellow |
| Rind thickness | thin | ▫ thick |
| **Aspect of the body** | | |
| Interior colour | white | ▫ yellow |
| Runny aspect | not runny | ▫ very runny |
| **Odour** | | |
| Global odour intensity | low | ▫ strong |
| Mushroom odour | low | ▫ strong |

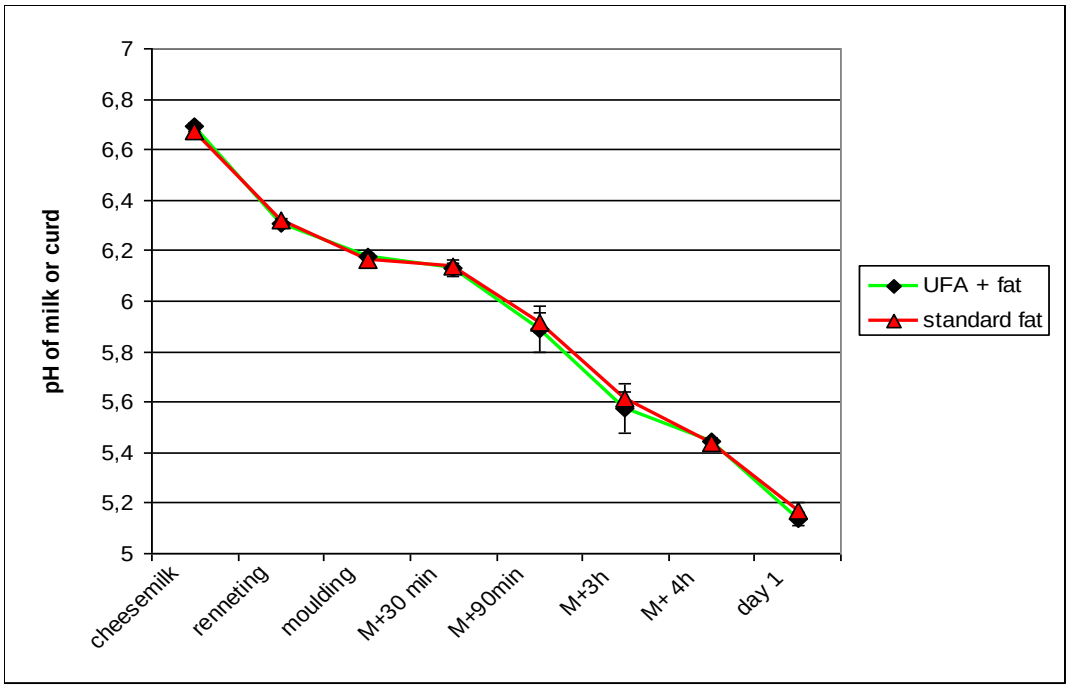
| | | | |
|---|---|---|---|
| Ammonia odour | low | ▫ | strong |
| Texture | | | |
| Flexible | not flexible | ▫ | flexible |
| Firmness | soft | ▫ | firm |
| Granular | low | ▫ | strong |
| Creamy | low | ▫ | strong |
| Rind Perception | low | ▫ | strong |
| **Flavour** | | | |
| Global taste Intensity | low | ▫ | strong |
| Salty taste | not salty | ▫ | very salty |
| Acid taste | not acid | ▫ | very acid |
| Mushroom aroma | low | ▫ | strong |
| Soap aroma | weak | ▫ | strong |
| Aromatic richness | low | ▫ | strong |
| Taste | | | |
| Bitter taste | not bitter | ▫ | very bitter |
| Metallic taste | absent | ▫ | strong |
| Pungent taste | not pungent | ▫ | very pungent |
| Rancid taste | not rancid | ▫ | very rancid |

# 3. Results and Discussion

## 3.1 Cheesemaking and cheese composition

The Table IV (Annexes) shows the composition of the cheese milk used for the twelve batches of cheeses. Probably due to a dilution with water during microfiltration and continuous cooling, the casein content of both batches of cheesemilk was below the targeted value. Additionally, a dilution of cream with water during the heat treatment, UFA-enriched cheesemilk shows lower casein and fat content and a higher Fat/(Casein+ denatured whey proteins) ratio. However the Table V (annex) and the Fig. 1 (below) show that the cheese making parameters were similar except for cheese yield and small differences in day-1 cheeses (pH and Total Solids). The composition of fat of the two milk types was clearly different: 7% less saturated fatty acids, 1.5% more poly-unsaturated fatty acids and doubling the omega-3 content and trans fatty acids (especially vaccenic and rumenic acids) in UFA-enriched milk. Detailed results of the fatty acid profiles are given in (Annex A). This very significant difference is more realistic than a higher difference.

Fig1- Course of acidification




The composition of the cheeses at packaging (10 days) and at sensory evaluation (35 days) is shown in the Tables VI and VII respectively (Annexes).

The ANOVA highlights statistical differences ($P<0.05$) for the salt content of cheeses: 1.53 g/100g for control *versus* 1.22 g/100g for reduced salt cheeses i.e. a 20% reduction of NaCl content. As a consequence, the proteolysis of reduced salt cheese was significantly higher.

However a tendency ($P$ between 0.06 and 0.09) can be seen on total cheese solids TS (salt effect), on fat in dry matter FDM (fat type effect) and moisture in not fat cheese MNFS (salt effect). Moreover, a block effect is statistically significant on the proteolysis (SN/TN, water soluble nitrogen/total nitrogen %), the Brie cheeses made the first day being strongly proteolysis (data not shown).

The cheeses were free from Salmonella and *Listeria monocytogenes* (absence in 25 g). The coagulase positive staphylococci and *Escherichia coli* counts were below 10 CFU/g, allowing the sensory analysis.

The hygienic status was the same at D+35, for the degustation. The composition of the cheeses is given in Table VII. UFA-enriched cheeses showed significantly higher Fat, FDM, MNFS and pH than standard fat cheeses. The differences in fatty acid composition were

similar to those seen in the milk (see annexe B for detailed results).
Significant effect of brining duration is evident on the salt content (1.60 g/100g vs 1.38 g/100g) and on salt-in-moisture (3.54% vs 3.04%). The salt reduction increases dramatically the lipolysis in cheese: increase of 60% of the Free Fatty Acids (FFA). All fatty acids were affected except C4, C6 and C17:1 (cf annexe C). A tendency (*P=0.06 to 0.07)* can be seen for $C_{16:1}$, $C_{18:0}$, $C_{18:1}$, $C_{18:2}$ and $C_{18:3}$. This could be understood as butyric (C4) and caproic acids (C6) are known as a poor indicators of lipolysis. There is no significant effect of fat composition or interaction with salt reduction.
The fat composition influenced the colour of the paste measured by chromameter CR310 on a longitudinal slice of cheese (half cheese like a reblochon for tartiflette meal). UFA-enriched cheeses showed significant (*P*=0.01) higher b* values (yellow axis): 27.9 +/- 0.9 a.u. *Versus* 26.0 +/- 0.6 a.u. Luminance and a* values were similar for each type of Brie-cheese.

### 3.2- Sensory analysis

The results of the ANOVA of the sensory profile of cheeses are given in table VIII. Salt content effect and fat composition effect can be seen on various descriptors but interactions between these two factors occurred only for bitterness.

Statistical **effects of fat composition** were shown on various items, UFA-fat cheeses being characterized by:

- a slightly yellower colour of the rind and of the body
- a more pronounced runny aspect. However a strong block effect can be seen on this item (4.35, 3.46 and 2.99 for the days 1, 2 and 3 respectively)
- a less intense mushroom odour
- a softer texture in mouth
- a more salty taste, which corresponds to the numerically higher NaCl content in UFA-cheese
- a more pungent aroma

Tendency (*P<0.15)* was recorded for the creamy texture, bitterness and milky aroma.

Statistical **effects of salt content** can be seen on:
-the mushroom aroma, which is reduced with reduced salt content
-the flexibility (higher with higher NaCl content)
-the salty taste
-the aromatic richness (higher with standard salt content)

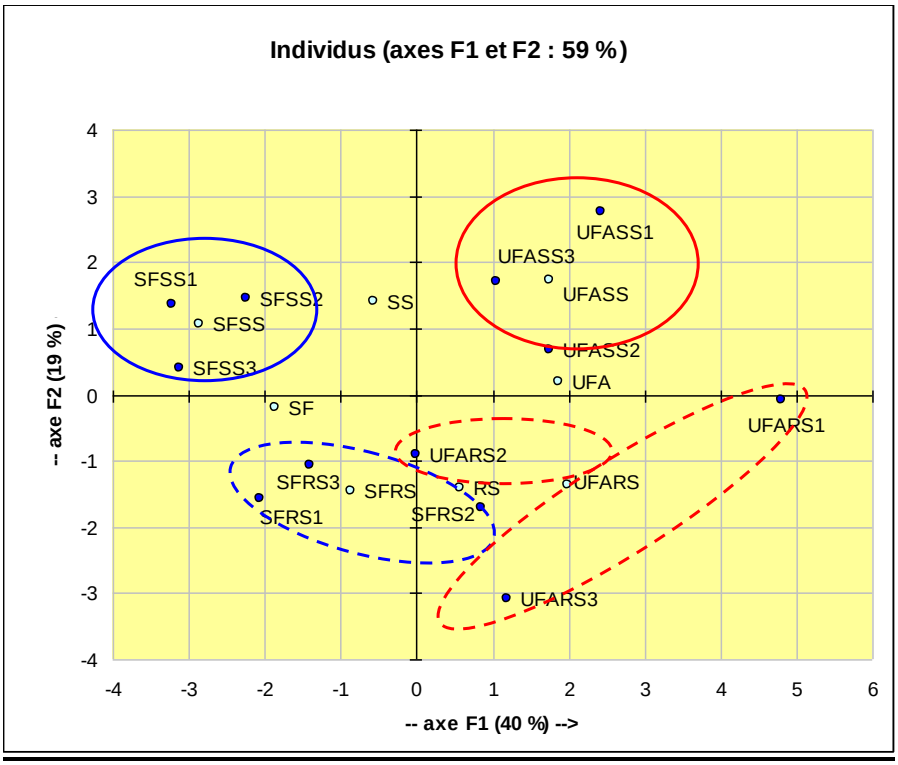
Tendency (*P<0.15)* was recorded for the global taste intensity and the mushroom aroma (low values for reduced salt cheeses). Despite a strong increase of lipolysis in reduced-salt cheeses, there was no significant effect of salt on metallic, pungent or rancid items.

An **interaction between salt and fat** composition can be seen for bitterness. UFA-fat with standard salt content and Standard-fat, reduced salt cheese were the more bitter cheese. Standard-fat with standard salt content was the less bitter cheese.

The **Principal Component Analysis** clearly discriminates the experimental treatments on the basis of sensory attributes showing significant differences or tendency in ANOVA (Fig 2). The first factorial plan explains 59% of the variability: 30% for the first axis and 19% for the second principal component. The first principal component correlated positively with pungent aroma (r=0.88), creamy texture (0.74), the colour of the paste (0.73) and the runny aspect (0.70). It was negatively related to the mushroom odour (r=-0.85) and firmness in mouth (- 0.80). The second axis correlated positively with aromatic richness (r=0.79), flexible texture (0.78), salty taste (0.72) and global taste intensity (0.61).

The first axis tends to discriminate the cheese according to their fat composition and the second axis according their salt content. Standard Salted (identified SS on the figure) cheeses were well discriminated, fat standard cheeses (FS) being mapped in the high-left quarter of the first principal plan and the UFA-enriched cheeses (UFA) in the high-right quarter. These Brie-cheese were well separated from Reduced Salt (RS) located in the low moiety of the map. Again, the FS cheeses were positioned on the left and the UFA-cheese on the right but the discrimination was not perfect due to both cheeses from Day 2 which are very similar.

Fig 2: Mapping of the Brie-cheeses on the first plan of the PCA of sensory analysis made with descriptors showing p value< 0.20 in ANOVA. SFSS=standard fat, standard salt; SFRS=standard fat, reduced salt; UFASS= UFA-enriched fat, standard salt; UFARS = UFA-enriched fat, reduced salt. b) Mapping of the descriptors.


Individus (axes F1 et F2 : 59 %)
-- axe F2 (19 %)
-- axe F1 (40 %) -->
UFASS1
UFASS3
UFASS
UFASS2
UFA
SFSS1
SFSS2
SS
SFSS
SFSS3
SF
UFARS1
UFARS2
SFRS3
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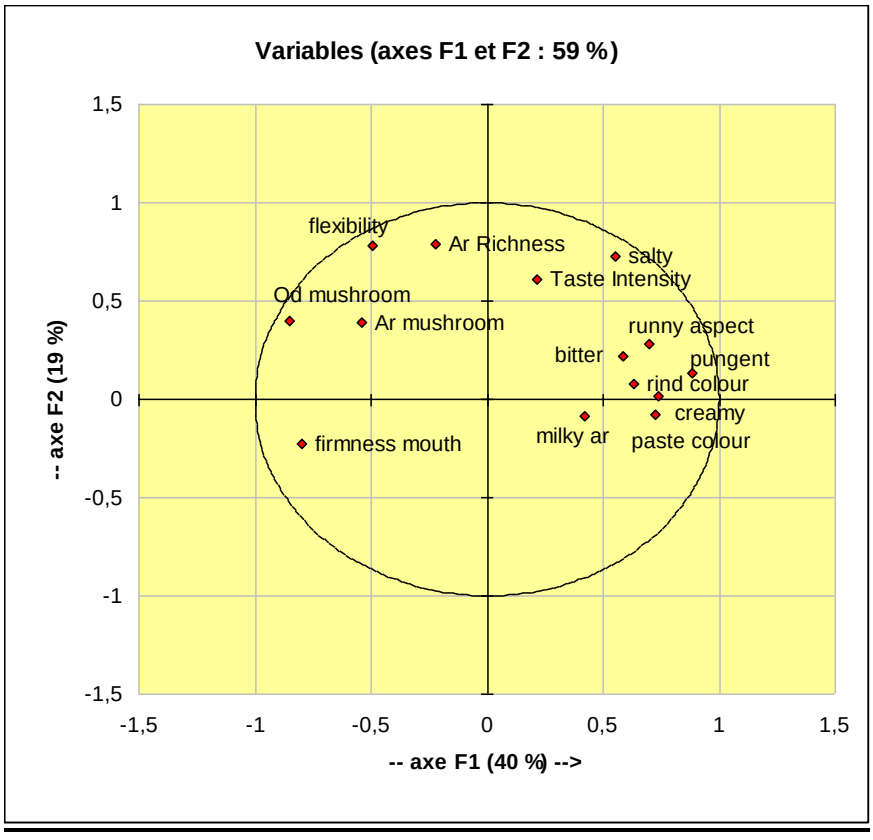
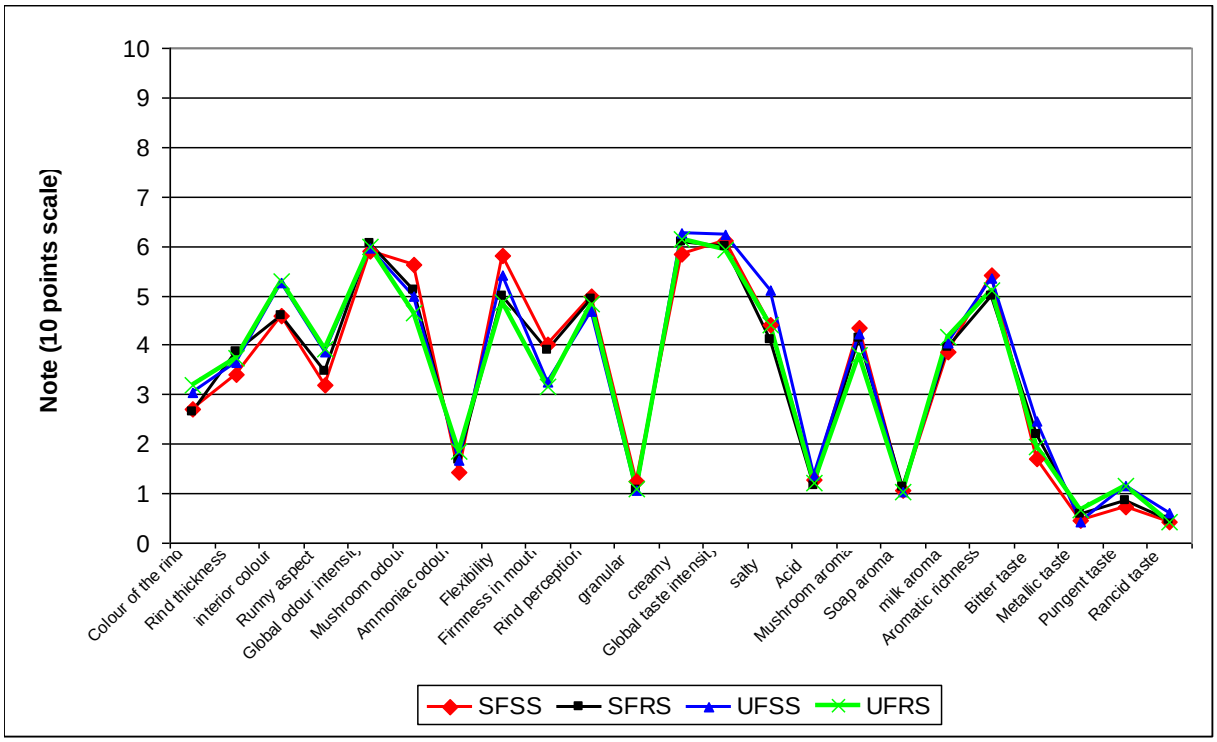

Variables (axes F1 et F2 : 59 %)
-- axe F2 (19 %)
-- axe F1 (40 %) -->
flexibility
Ar Richness
salty
Taste Intensity
Od mushroom
Ar mushroom
runny aspect
bitter
pungent
rind colour
creamy
milky ar
paste colour
firmness mouth

Fig. 3: Comparison of the sensory profile of four Brie type cheeses.



# 4. Conclusion

Stabilized Brie cheeses were made with reduced saturated fat milk and reduced salt content. The differences in saturated acids content were realistic and correspond to unsaturated fatty acids enriched commercial milks.

These reductions had a little influence on the sensory profile. Similar conclusions can be drawn from the technological results.

The conclusion of this trials it that it is possible to make correct Brie-type cheese with low-salt and improved fat composition (UFA enrichment = SFA lowering).

# 5. Annexes : Tables and graphs

| Table I: Solubilised soft cheese flow chart | |
|---|---|
| Milk ripening | |
| Temperature | 39°C |
| Duration | to reach pH 6.30 (XXXXXXXXXXmin) |
| $CaCl_2$ | 0,15 g/kg |
| GDL | 0,9 g/kg |
| Starter culture (Danisco, Dangé St Romain, France) | |
| *Streptococcus salivarius subsp. thermophilus* - TA 054 | 30 DCU/100kg |
| Ripening cultures (Danisco, Dangé St Romain, France) | |
| *Geotrichum candidium* - Geo17 | 2 D/kL |
| *Penicillium candidium* - PC Neige | 8 D/kL |
| *Staphylococcus xylosus* – MVA | 2 D/kL |
| Coagulation | |
| Room temperature | 33°C |
| Milk Temperature | 38°C |
| pH | 6,30 |
| Amount of Chymax Plus (Chr Hansen, Arpajon) | 22 mL/100 kg |
| Setting time | 6.5 min |
| Set to cut time | 10 min |
| **Coagulation** | |
| Mean curd size | 1.5 x 1.5 x 1.7 cm |
| **Stirring** | 6 intermittent stirrings: 5-10-15-20-25 and 30 min after cutting |
| **Whey drawn off** | 28% |
| **Moulding** | |
| Temperature | 36°C |
| Mould | ▫ 11 cm |
| **Draining** | |
| Temperatures | 3h at 33°C and overnight at 18°C |
| Curd turning over | 3 times (30, 90, 180 min after moulding) |
| **Brining** | |
| Temperature | 12°C |
| Brine density | Saturated |
| pH | 5 |
| Duration | 20 min (Control) or 10 min ( 30% reduced) |
| **Ripening** | |
| *Penicillium candidium* - PC VS | Sprayed onto the curd surface |
| Temperature | 12°C |
| Humidity | 96% |
| Duration | 14 days |
| **Cold storage** | |
| Temperature | 4°C |
| Duration | 6 weeks |
| Packing paper | Paraffin wax coating paper + wood b |

**Table III: analyses**

| Analysis | Method |
|---|---|
| **Milk** | |
| Total Solids | Oven drying (NFV 04-367) |
| Fat | Butyrometric method (NFV 04-210) |
| Total Nitrogen | Kjeldahl (FIL 20B) |
| Non Casein Nitrogen | Kjeldahl on pH 4.6 filtrate (FIL 29) |
| Non Protein Nitrogen | Kjeldahl on TCA12% filtrate (FIL 20B) |
| Total Calcium | Complexometry |
| Soluble Calcium | Complexometry |
| Lactose | Enzymatic |
| Fatty Acid Composition of fat | |
| **Curd and cheese** | |
| Total solids | Oven drying (NFV 04-282) |
| Fat | Butyrometric acid (NF V 04-287#) |
| Total Nitrogen | Kjeldahl (FIL 20B) |
| pH 4.6 Soluble Nitrogen | Kjeldahl (adapted from FIL 20B) |
| 12%TCA Soluble Nitrogen | Kjeldahl (adapted from FIL 20B) |
| Casein hydrolysis | Urea PAGE of pH 4.6 insoluble N |
| Galactose | Enzymatic |
| L(+) and D(-) Lactate | Enzymatic |
| NaCl | Chloride meter |
| Total Calcium | Complexometry |
| Fatty Acid Composition of fat | Gaz Chromatogrpahy (ITERG) |
| Free Fatty acids | |

Table IV: Composition of the cheesemilk (n=12)

| | | Standard fat | | UFA enriched fat | |
|---|---|---|---|---|---|
| | unit | M | sd | M | sd |
| Crude protiens | g/kg | 42,8 | 0,2 | 39,7 | 0,3 |
| True proteins | g/kg | 41,0 | 0,1 | 38,1 | 0,2 |
| (TN – NCN) x 6,38 | g/kg | 37,3 | 0,1 | 34,7 | 0,2 |
| denaturated whey proteins | g/kg | 1,9 | 0,1 | 2,0 | 0,1 |
| casein | g/kg | 35,4 | 0,0 | 32,7 | 0,2 |
| Fat | g/kg | 69,0 | 0,9 | 67,3 | 0,6 |
| Fat/(caseins+ denatured whey proteins) | (-) | 1,85 | 0,0 | 1,94 | 0,0 |
| Saturated fatty acids | % of fatty acids | 68,28 | 0,0 | 61,32 | 0,1 |
| Mono-unsaturated fatty acids | % of fatty acids | 27,00 | 0,0 | 32,14 | 0,2 |
| poly-unsaturated fatty acids | % of fatty acids | 3,93 | 0,0 | 5,36 | 0,0 |
| omega 3 fatty acids | % of fatty acids | 0,66 | 0,0 | 1,32 | 0,0 |
| omega 6 fatty acids | % of fatty acids | 2,19 | 0,0 | 1,90 | 0,0 |
| trans fatty acids | % of fatty acids | 3,51 | 0,0 | 6,10 | 0,0 |

Table V: Cheesemaking parameters and composition of D1 cheeses (=12 vats)

| | Mean | SD | Standard | UFA + | *P* |
|---|---|---|---|---|---|
| Milk in ripening vat | 52 | 0 | 52 | 52 | NS |
| pH | 6,68 | 0,02 | 6,68 | 6,69 | 0,222 |
| Ripening time (min) | 42,33 | 1,86 | 42 | 42,7 | 0,355 |
| Température (°C) | 39,53 | 0,59 | 39,3 | 39,7 | 0,234 |
| pH renneting | 6,32 | 0,01 | 6,32 | 6,31 | 0,100 |
| T°renneting (°C) | 37,78 | 0,37 | 37,83 | 37,73 | 0,332 |
| Setting time (min) | 6,91 | 0,34 | 7,05 | 6,78 | 0,083 |
| Set to cut (min) | 9,98 | 0,07 | 10,00 | 9,96 | 0,170 |
| pH at moulding | 6,17 | 0,02 | 6,16 | 6,18 | 0,083 |
| T° at moulding (°C) | 35,47 | 0,48 | 35,35 | 35,58 | 0,214 |
| pH 30min after moulding | 6,13 | 0,02 | 6,14 | 6,13 | 0,366 |
| T°C at moulding +30 min | 34,90 | 0,77 | 34,62 | 35,18 | 0,108 |
| pH at M+90 min | 5,90 | 0,07 | 5,92 | 5,89 | 0,261 |
| T°C at M+90 min | 33,50 | 0,88 | 33,02 | 33,98 | 0,026 |
| Whey (Kg) at M+90 min | 15,98 | 0,23 | 15,98 | 15,97 | 0,454 |
| pH at M+3h | 5,60 | 0,07 | 5,62 | 5,58 | 0,171 |
| T°C at M+3h | 32,32 | 0,43 | 32,22 | 32,42 | 0,222 |
| Whey (Kg) at M+3h | 17,30 | 0,18 | 17,30 | 17,31 | 0,469 |
| Moulding to cooling time (h) | 4,05 | 0,55 | 4,17 | 3,93 | 0,212 |
| pH at cooling | 5,44 | 0,02 | 5,44 | 5,44 | 0,314 |
| Cheese weight (kg) at D1 | 5,70 | 0,16 | 5,84 | 5,56 | 0,000 |
| pH at D1 | 5,16 | 0,03 | 5,17 | 5,14 | 0,027 |
| whey (Kg) at day 1 | 18,44 | 0,21 | 18,34 | 18,53 | 0,052 |
| Fat (g/100g) at D1 | 29,23 | 0,31 | 29,08 | 29,38 | 0,320 |
| Total Solids (g/100g) at D1 | 48,38 | 0,50 | 48,45 | 48,31 | 0,005 |
| FDM (%) at D1 | 60,42 | 0,58 | 60,02 | 60,81 | 0,220 |
| MNFS (%) at D1 | 72,94 | 0,58 | 72,69 | 73,19 | 0,069 |

NS : not significant at p<00.5

Table VI: Composition of the cheese at packaging (14days)

| | | Fat | | Salt | | Stand fat,stand salt | Stand fat, red salt | UFA +, stand salt | UFA +, red salt |
|---|---|---|---|---|---|---|---|---|---|
| | unit | Stand | UFA + | Stand | Red | | | | |
| TS | g/100g | 51,64 | 51,76 | 51,25 | 52,04 | 50,75 | 52,30 | 51,74 | 51,77 |
| Fat | g/100g | 31,31 | 31,67 | 31,13 | 31,50 | 30,42 | 31,50 | 31,83 | 31,50 |
| FDM | % | 60,63 | 61,18 | 60,73 | 60,53 | 59,93 | 60,23 | 61,52 | 60,83 |
| MNFS | % | 70,40 | 70,60 | 70,79 | 70,02 | 70,77 | 69,63 | 70,80 | 70,41 |
| Protein | g/100g | 16,75 | 16,63 | 16,61 | 16,90 | 16,65 | 17,10 | 16,57 | 16,69 |
| **SN/TN** | **%** | **43,80** | **43,16** | **42,35b** | **45,25a** | **43,13** | **45,76** | **41,58** | **44,74** |
| NPN/TN | % | 22,06 | 21,69 | 21,66 | 22,47 | 22,44 | 22,44 | 20,88 | 22,50 |
| NPN/SN | % | 47,38 | 47,05 | 48,05 | 46,71 | 48,97 | 46,45 | 47,13 | 46,96 |
| Ca | mg/100g | 352 | 348 | 350 | 353 | 340 | 370 | 360 | 336 |
| Ca/SNF | % | 1,73 | 1,73 | 1,74 | 1,72 | 1,68 | 1,78 | 1,81 | 1,66 |
| **Salt** | **g/100g** | **1,37** | **1,38** | **1,53a** | **1,22b** | **1,55** | **1,18** | **1,50** | **1,27** |
| **S/M** | **%** | **2,84** | **2,87** | **3,13a** | **2,54b** | **3,15** | **2,46** | **3,11** | **2,63** |

The letters a and b mean that the results are significantly different by the Newman-Keuls test ($p<0.05$)

Table VII: Composition of the day-35 cheeses

| | | Fat | | Salt | | Stand fat | Stand fat | UFA + | UFA + |
|---|---|---|---|---|---|---|---|---|---|
| | Unit | Standard | UFA + | Standard | Reduced | Stand salt | Red salt | stand salt | red salt |
| TS | g/100g | 55,01 | 54,55 | 54,70 | 54,69 | 55,34 | 54,68 | 54,39 | 54,59 |
| Fat | g/100g | 33,33b | 33,71a | 33,45 | 33,63 | 33,33 | 33,33 | 33,50 | 33,75 |
| FDM | % | 60,60b | 61,80a | 61,16 | 61,48 | 60,24 | 60,96 | 61,60 | 61,83 |
| MNFS | % | 67,49b | 68,57a | 68,06 | 68,27 | 67,00 | 67,98 | 68,58 | 68,55 |
| pH | (-) | 6,24b | 6,50a | 6,40 | 6,36 | 6,23 | 6,25 | 6,53 | 6,37 |
| TN | | 2,94 | 2,82 | 2,86 | 2,89 | 2,94 | 2,94 | 2,81 | 2,85 |
| NS | | 1,84 | 1,74 | 1,71 | 1,87 | 1,78 | 1,90 | 1,64 | 1,89 |
| NPN | | 1,09 | 1,00 | 1,00 | 1,10 | 1,05 | 1,13 | 0,92 | 1,12 |
| Protein | g/100g | 18,34 | 18,01 | 18,27 | 17,99 | 18,77 | 17,90 | 17,96 | 18,21 |
| SN/TN | % | 62,6 | 61,8 | 59,8 | 64,9 | 60,6 | 64,6 | 58,3 | 66,3 |
| NPN/TN | % | 37,0 | 35,4 | 34,7 | 38,1 | 35,7 | 38,3 | 32,8 | 39,3 |
| NPN/SN | % | 59,1 | 57,0 | 57,3 | 58,6 | 58,9 | 59,3 | 56,3 | 59,2 |
| NH3 | mg/100g | 163 | 158 | 155 | 164 | 159 | 165 | 153 | 162 |
| Ca | mg/100g | 376 | 355 | 369 | 360 | 378 | 374 | 363 | 352 |
| Ca/SNF | % | 1,74 | 1,70 | 1,74 | 1,71 | 1,72 | 1,75 | 1,74 | 1,69 |
| Salt | g/100g | 1,45 | 1,54 | **1,60a** | **1,38b** | 1,57 | 1,33 | 1,64 | 1,39 |
| S/M | % | 3,23 | 3,38 | **3,54a** | **3,05b** | 3,52 | 2,94 | 3,60 | 3,06 |
| FFA | mg/100g | 1495 | 1449 | **1106b** | **1786a** | 1170 | 1820 | 1146 | 1718 |
| SFA | % of total fatty acids | 73,4a | 66,3b | | | | | | |
| MUFA | | 22,6b | 27,8a | | | | | | |
| PUFA | | 3,2b | 4,6a | | | | | | |
| oméga 3 | | 0,6b | 1,2a | | | | | | |
| oméga 6 | | 1,9 | 1,8 | | | | | | |
| trans FA | | 4,2b | 7,1a | | | | | | |

The letters a and b mean that the results are significantly different by the Newman-Keuls test ($p<0.05$)

Table VIII : Sensory evaluation

| | Fat | | Salt | | Standard fat | | UFA + fat | |
|---|---|---|---|---|---|---|---|---|
| Descriptor | Standard | UFA + | Standard | Reduced | Standard salt | Reduced salt | standard salt | reduced salt |
| Colour of the rind | **2,67b** | **3,11a** | 2,87 | 2,92 | 2,7 | 2,64 | 3,04 | 3,2 |
| Rind thickness | 3,63 | 3,73 | 3,54 | 3,82 | 3,41 | 3,85 | 3,66 | 3,75 |
| interior colour | 4,59b | 5,28a | 0,92 | 4,95 | 4,58 | 4,6 | 5,26 | 5,3 |
| Runny aspect | **3,31b** | **3,89a** | 3,51 | 3,69 | 3,18 | 3,45 | 3,85 | 3,93 |
| Global odour intensity | 5,97 | 5,98 | 5,93 | 6,02 | 5,91 | 6,04 | 5,95 | 6 |
| Mushroom odour | **5,37a** | **4,82b** | **5,3a** | **4,88b** | 5,61 | 5,1 | 4,99 | 4,64 |
| Ammoniac odour | 1,55 | 1,77 | 1,55 | 1,77 | 1,42 | 1,68 | 1,68 | 1,85 |
| Flexibility | 5,4 | 5,13 | **5,61a** | **4,91b** | 5,81 | 4,98 | 5,42 | 4,85 |
| Firmness in mouth | **3,93a** | **3,21b** | 3,64 | 3,49 | 4,01 | 3,884 | 3,26 | 3,15 |
| Rind perception | 4,97 | 4,75 | 4,84 | 4,88 | 5 | 4,93 | 4,68 | 4,83 |
| granular | 1,16 | 1,08 | 1,16 | 1,08 | 1,26 | 1,06 | 1,06 | 1,1 |
| creamy | 5,97 | 6,2 | 6,05 | 6,12 | 5,84 | 6,09 | 6,26 | 6,14 |
| Global taste intensity | 6,05 | 6,18 | 6,18 | 5,97 | 6,11 | 5,99 | 6,22 | 5,94 |
| salty | **4,27b** | **4,76a** | **4,7a** | **4,27b** | 4,42 | 4,11 | 5,1 | 4,42 |
| Acid | 1,21 | 1,3 | 1,33 | 1,17 | 1,28 | 1,14 | 1,38 | 1,21 |
| Mushroom aroma | 4,24 | 4,01 | 4,29 | 3,96 | 4,36 | 4,12 | 4,22 | 3,81 |
| Soap aroma | 1,09 | 1,03 | 1,04 | 1,08 | 1,05 | 1,13 | 1,03 | 1,04 |
| milk aroma | 3,89 | 4,09 | 3,95 | 4,04 | 3,86 | 3,94 | 4,04 | 4,15 |
| Aromatic richness | 5,21 | 5,23 | **5,38a** | **5,06b** | 5,42 | 5 | 5,34 | 5,12 |
| Bitter taste | 1,94 | 2,2 | 2,07 | 2,07 | **1,7c** | **2,18ab** | **2,45a** | **1,95bc** |
| Metallic taste | 0,52 | 0,54 | 0,43 | 0,63 | 0,45 | 0,59 | 0,42 | 0,66 |
| Pungent taste | **0,79b** | **1,16a** | 0,94 | 1,01 | 0,72 | 0,86 | 1,15 | 1,16 |
| Rancid taste | 0,45 | 0,52 | 0,53 | 0,44 | 0,43 | 0,47 | 0,62 | 0,42 |

The letters a and b mean that the results are significantly different by the Newman-Keuls test ($p<0.05$)

Annexe A: Detailed composition of milk fat

(NS: not significant for p<0.05)

| | Standard | | UFA + | | *P* |
|---|---|---|---|---|---|
| % of fatty acids | M | sd | M | sd | |
| c4 | 3,7 | 0,0 | 3,7 | 0,1 | 0,41406578 |
| c5 | 0,03 | 0,00 | 0,03 | 0,00 | NS |
| c6 | 2,25 | 0,01 | 2,11 | 0,04 | 0,002 |
| c7 | 0,03 | 0,00 | 0,02 | 0,00 | NS |
| C8 | 1,34 | 0,00 | 1,19 | 0,04 | 0,001 |
| C9 | 0,03 | 0,00 | 0,03 | 0,00 | NS |
| C10:0 | 3,03 | 0,01 | 2,55 | 0,07 | 0,000 |
| C10:1 | 0,36 | 0,00 | 0,33 | 0,01 | 0,003 |
| C12:0 | 3,48 | 0,01 | 2,89 | 0,04 | 0,000 |
| C12:1c | 0,10 | 0,00 | 0,08 | 0,01 | 0,004 |
| C13:0 | 0,09 | 0,00 | 0,08 | 0,00 | 0,000 |
| Ic13:0 | 0,03 | 0,00 | 0,04 | 0,00 | NS |
| aC13:0 | 0,09 | 0,00 | 0,08 | 0,00 | 0,000 |
| C14:0 | 11,31 | 0,04 | 9,91 | 0,04 | 0,000 |
| iC14:0 | 0,10 | 0,01 | 0,09 | 0,00 | 0,058 |
| C14:1t | 0,01 | 0,00 | 0,01 | 0,00 | NS |
| C14:1 9c | 0,94 | 0,01 | 0,92 | 0,01 | 0,007 |
| C15:0 | 1,04 | 0,01 | 1,01 | 0,01 | 0,002 |
| iC15:0 | 0,23 | 0,01 | 0,26 | 0,01 | 0,004 |
| aC15:0 | 0,45 | 0,04 | 0,52 | 0,01 | 0,016 |
| C15:1 10c | 0,01 | 0,00 | 0,01 | 0,00 | NS |
| C16:0 | 28,94 | 0,02 | 22,92 | 0,05 | 0,000 |
| iC16:0 | 0,27 | 0,01 | 0,30 | 0,00 | 0,008 |
| C16:1 9c | 1,49 | 0,00 | 1,37 | 0,00 | 0,000 |
| C16:7t | 0,06 | 0,00 | 0,11 | 0,00 | 0,000 |
| C17:0 | 0,54 | 0,04 | 0,53 | 0,02 | 0,259 |
| iC17:0 | 0,38 | 0,01 | 0,43 | 0,00 | 0,000 |
| aC17:0 | 0,45 | 0,01 | 0,47 | 0,00 | 0,004 |
| C17:1 10c | 0,23 | 0,00 | 0,21 | 0,01 | 0,001 |
| C18:0 | 9,97 | 0,01 | 11,65 | 0,14 | 0,000 |
| C18:0 iso | 0,06 | 0,00 | 0,06 | 0,01 | 0,187 |
| C18:1 4t | 0,02 | 0,00 | 0,02 | 0,00 | NS |
| C18:1 5t | 0,01 | 0,00 | 0,02 | 0,00 | NS |
| C18:1 6t+8t | 0,25 | 0,02 | 0,36 | 0,02 | 0,001 |
| C18:1 9t | 0,17 | 0,03 | 0,30 | 0,03 | 0,004 |
| C18:1 10t | 0,52 | 0,16 | 0,91 | 0,16 | 0,020 |
| C18:1 11t | 1,05 | 0,13 | 1,83 | 0,15 | 0,001 |
| C18:1 12t | 0,30 | 0,01 | 16,67 | 28,00 | 0,184 |
| C18:1 13t | 0,00 | 0,00 | 0,00 | 0,00 | NS |
| C18:1 16t | 0,33 | 0,01 | 0,61 | 0,01 | 0,000 |
| C18:1 9c | 20,17 | 0,04 | 23,27 | 0,07 | 0,000 |
| C18:1 11c | 0,43 | 0,01 | 0,41 | 0,00 | 0,001 |

| | | | | | |
|---|---|---|---|---|---|
| C18:1 12c | 0,21 | 0,00 | 0,28 | 0,00 | 0,000 |
| C18:1 13c | 0,07 | 0,00 | 0,09 | 0,00 | 0,000 |
| C18:1: 15c | 0,07 | 0,01 | 0,18 | 0,01 | 0,000 |
| C18:2 9t12c | 0,01 | 0,00 | 0,01 | 0,00 | NS |
| C18:2 t | 0,73 | 0,00 | 1,34 | 0,01 | 0,000 |
| C18:2 9c12c | 1,70 | 0,00 | 47,27 | 79,44 | 0,188 |
| C18:2 9c11t | 0,38 | 0,31 | 1,14 | 0,01 | 0,007 |
| C18:2 10t12c | 0,02 | 0,01 | 0,02 | 0,00 | 0,187 |
| C18:3 n-6 | 0,18 | 0,25 | 0,02 | 0,01 | 0,177 |
| C18:3 n-3 | 0,33 | 0,25 | 0,96 | 0,01 | 0,006 |
| C18:3 t | 0,19 | 0,30 | 0,09 | 0,01 | 0,286 |
| C19:0 | 0,09 | 0,01 | 0,09 | 0,02 | 0,398 |
| C20:0 | 0,12 | 0,01 | 0,12 | 0,01 | 0,443 |
| C20:1 9c | 0,12 | 0,00 | 0,14 | 0,00 | 0,000 |
| C20:1 11c | 0,04 | 0,01 | 0,05 | 0,00 | 0,058 |
| C20:2 n-6 | 0,02 | 0,00 | 0,02 | 0,00 | NS |
| C20:3 n-6 | 0,08 | 0,00 | 0,06 | 0,00 | 0,000 |
| C20:3 n-3 | 0,01 | 0,00 | 0,02 | 0,01 | 0,058 |
| C20:4 n-6 | 0,13 | 0,01 | 0,08 | 0,01 | 0,000 |
| C20:5 n-3 | 0,04 | 0,00 | 0,08 | 0,00 | 0,000 |
| C21:0 | 0,01 | 0,00 | 0,01 | 0,00 | NS |
| C22:0 | 0,05 | 0,01 | 0,07 | 0,01 | 0,024 |
| C22:1 n-9 | 0,01 | 0,01 | 0,01 | 0,00 | 0,187 |
| C22:2 n-6 | 0,02 | 0,00 | 0,04 | 0,01 | 0,001 |
| C22:5 n-3 | 0,06 | 0,00 | 0,09 | 0,00 | 0,000 |
| C22:6 n-3 | 0,00 | 0,00 | 0,00 | 0,00 | NS |
| C23:0 | 0,02 | 0,00 | 0,02 | 0,00 | NS |
| C24:0 | 0,03 | 0,00 | 0,03 | 0,00 | NS |
| C24:1 n-9 | 0,00 | 0,01 | 0,00 | 0,00 | 0,187 |

Annexe B: detailed composition of cheese fat (D+35)

NS : not significant at p<00.5

| % of total fatty acids | Standard fat | | UFA + fat | | |
|---|---|---|---|---|---|
| | Mean | SD | Mean | SD | *P* |
| c4 | 2,78 | 0,19 | 2,73 | 0,22 | 0,40 |
| c5 | 0,03 | 0,01 | 0,02 | 0,01 | 0,36 |
| c6 | 1,82 | 0,07 | 1,70 | 0,17 | 0,17 |
| c7 | 0,02 | 0,00 | 0,02 | 0,00 | NS |
| C8 | 1,19 | 0,03 | 1,05 | 0,02 | 0,00 |
| C9 | 0,03 | 0,00 | 0,02 | 0,00 | NS |
| C10:0 | 2,86 | 0,04 | 2,41 | 0,01 | 0,00 |
| C10:1 | 0,32 | 0,01 | 0,29 | 0,01 | 0,00 |
| C12:0 | 3,58 | 0,01 | 2,99 | 0,05 | 0,00 |
| C12:1c | 0,11 | 0,01 | 0,09 | 0,00 | 0,00 |
| C13:0 | 0,08 | 0,00 | 0,05 | 0,04 | 0,14 |
| iC13:0 | 0,03 | 0,00 | 0,31 | 0,46 | 0,18 |
| aC13:0 | 0,09 | 0,00 | 0,42 | 0,59 | 0,19 |
| C14:0 | 12,38 | 0,18 | 10,91 | 0,20 | 0,00 |
| iC14:0 | 0,10 | 0,00 | 0,09 | 0,01 | 0,06 |
| C14:1t | 0,04 | 0,03 | 0,01 | 0,00 | 0,08 |
| C14:1 9c | 0,83 | 0,02 | 0,84 | 0,02 | 0,34 |
| C15:0 | 1,12 | 0,01 | 1,11 | 0,02 | 0,14 |
| iC15:0 | 0,25 | 0,00 | 0,31 | 0,06 | 0,08 |
| aC15:0 | 0,54 | 0,06 | 0,56 | 0,02 | 0,28 |
| C15:1 10c | 0,01 | 0,00 | 0,01 | 0,00 | NS |
| C16:0 | 32,94 | 0,73 | 26,02 | 0,52 | 0,00 |
| iC16:0 | 0,31 | 0,01 | 0,34 | 0,01 | 0,00 |
| C16:1 9c | 1,27 | 0,04 | 1,25 | 0,03 | 0,26 |
| C16:7t | 0,08 | 0,02 | 0,13 | 0,02 | 0,01 |
| C17:0 | 0,38 | 0,22 | 0,54 | 0,02 | 0,13 |
| iC17:0 | 0,42 | 0,01 | 0,50 | 0,04 | 0,02 |
| aC17:0 | 0,50 | 0,02 | 0,51 | 0,01 | 0,14 |
| C17:1 10c | 0,18 | 0,01 | 0,16 | 0,01 | 0,14 |
| C18:0 | 11,57 | 0,31 | 13,82 | 0,32 | 0,00 |
| C18:0 iso | 0,06 | 0,00 | 0,07 | 0,01 | 0,06 |
| C18:1 4t | 0,02 | 0,00 | 0,03 | 0,01 | 0,06 |
| C18:1 5t | 0,02 | 0,01 | 0,02 | 0,00 | 0,08 |
| C18:1 6t+8t | 0,21 | 0,00 | 0,35 | 0,02 | 0,00 |
| C18:1 9t | 0,22 | 0,01 | 0,29 | 0,04 | 0,03 |
| C18:1 10t | 0,50 | 0,06 | 0,95 | 0,15 | 0,08 |
| C18:1 11t | 1,08 | 0,07 | 1,92 | 0,20 | 0,00 |
| C18:1 12t | 0,32 | 0,00 | 0,58 | 0,06 | 0,00 |
| C18:1 13t | 0,64 | 0,06 | 0,77 | 0,68 | 0,38 |
| C18:1 16t | 0,42 | 0,03 | 0,72 | 0,02 | 0,00 |
| C18:1 9c | 15,50 | 0,68 | 18,26 | 1,67 | 0,03 |
| C18:1 11c | 0,38 | 0,00 | 0,38 | 0,01 | 0,19 |
| C18:1 12c | 1,10 | 0,90 | 0,28 | 0,00 | 0,09 |

| | | | | | |
|---|---|---|---|---|---|
| C18:1 13c | 0,07 | 0,00 | 0,09 | 0,01 | 0,00 |
| C18:1: 15c | 0,09 | 0,00 | 0,21 | 0,02 | 0,00 |
| C18:2 9t12c | 0,01 | 0,00 | 0,01 | 0,00 | NS |
| C18:2 t | 0,65 | 0,02 | 1,23 | 0,01 | 0,00 |
| C18:2 9c12c | 1,38 | 0,08 | 1,18 | 0,09 | 0,02 |
| C18:2 9c11t | 0,38 | 0,08 | 0,92 | 0,15 | 0,00 |
| C18:2 10t12c | 0,02 | 0,02 | 0,03 | 0,01 | 0,14 |
| C18:3 n-6 | 0,03 | 0,00 | 0,02 | 0,00 | NS |
| C18:3 n-3 | 0,34 | 0,04 | 0,73 | 0,05 | 0,00 |
| C18:3 t | 0,04 | 0,01 | 0,06 | 0,02 | 0,10 |
| C19:0 | 0,55 | 0,45 | 0,10 | 0,02 | 0,08 |
| C20:0 | 0,19 | 0,04 | 0,18 | 0,02 | 0,47 |
| C20:1 9c | 0,11 | 0,01 | 0,13 | 0,01 | 0,01 |
| C20:1 11c | 0,04 | 0,01 | 0,05 | 0,01 | 0,04 |
| C20:2 n-6 | 0,02 | 0,00 | 0,02 | 0,00 | NS |
| C20:3 n-6 | 0,08 | 0,00 | 0,07 | 0,01 | 0,08 |
| C20:3 n-3 | 0,01 | 0,00 | 0,01 | 0,00 | NS |
| C20:4 n-6 | 0,15 | 0,00 | 0,09 | 0,00 | 0,00 |
| C20:5 n-3 | 0,07 | 0,03 | 0,10 | 0,01 | 0,05 |
| C21:0 | 0,01 | 0,01 | 0,01 | 0,01 | 0,36 |
| C22:0 | 0,06 | 0,01 | 0,07 | 0,01 | 0,01 |
| C22:1 n-9 | 0,00 | 0,00 | 0,01 | 0,00 | NS |
| C22:2 n-6 | 0,02 | 0,00 | 0,05 | 0,00 | 0,00 |
| C22:5 n-3 | 0,07 | 0,01 | 0,11 | 0,01 | 0,00 |
| C22:6 n-3 | 0,00 | 0,00 | 0,01 | 0,01 | 0,06 |
| C23:0 | 0,02 | 0,01 | 0,01 | 0,00 | 0,08 |
| C24:0 | 0,05 | 0,01 | 0,05 | 0,03 | 0,38 |
| C24:1 n-9 | 0,00 | 0,00 | 0,00 | 0,01 | 0,19 |

Annexe C: detailed composition of free fatty acids in cheese (D35)

| FFA (mg/100g) | Fat | | Salt | | Standard fat | | UFA + fat | |
|---|---|---|---|---|---|---|---|---|
| | Standard | UFA + | Standard | Reduced | Stand salt | Red salt | stand salt | red salt |
| C4:0 | 14 | 13 | 11 | 16 | 11 | 16 | 11 | 13 |
| C6:0 | 12 | 12 | 10 | 13 | 10 | 14 | 10 | 12 |
| C8:0 | 11 | 12 | 9b | 14a | 9 | 13 | 9 | 13 |
| C10:0 | 41 | 37 | 27b | 48a | 32 | 49 | 27 | 46 |
| C12:0 | 55 | 43 | 34b | 61a | 42 | 68 | 33 | 58 |
| C14:0 | 146 | 129 | 101b | 168a | 111 | 181 | 103 | 154 |
| C14:1 | 34 | 36 | 25a | 43a | 24 | 43 | 28 | 42 |
| C15:0 | 15 | 14 | 11b | 17a | 11 | 18 | 11 | 16 |
| C16:0 | 344 | 259 | 224b | 361a | 271 | 418 | 215 | 311 |
| C16:1 | 61 | 50 | 42 | 67 | 48 | 73 | 40 | 60 |
| C17:1 | 7 | 7 | 5 | 9 | 6 | 9 | 5 | 10 |
| C18:0 | 109 | 123 | 84b | 142a | 79 | 138 | 101 | 141 |
| C18:1 | 540 | 577 | 432 | 673 | 435 | 644 | 453 | 680 |
| C18:2 | 79 | 88 | 62 | 106 | 57 | 101 | 65 | 106 |
| C18:3 | 30 | 51 | 30 | 50 | 24 | 35 | 38 | 57 |
| FFA | 1495 | 1449 | 1106b | 1786a | 1170 | 1820 | 1147 | 1718 |

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