Safety status and physical-chemical composition of Plaisentif Cheese.

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Abstract. This is the first attempt to describe Plaisentif cheese. Plaisentif is a traditional cheese produced in the Western Alps and obtained from the raw milk of cows fed on pastures during the violet flowering period. This cheese has never been studied. The aim of this work was evaluate the safety status of milk and cheese and some basic physical-chemical parameters . Samples of milk, and of cheese at the end of ripening (60 days), were collected from nine Alpine dairy farms. Content of dry matter, protein, fat and ash, as well as pH and water activity (Aw) were evaluated. The microbiological parameters considered were: coagulase-positive Staphylococci, *Enterobacteriaceae*, *Salmonella* spp and *Listeria monocytogenes*. The final mean values of pH and Aw for cheeses were 5.39 and 0.963, respectively. *Salmonella* spp was found neither in milk nor in cheese. *Listeria monocytogenes* was found in one sample of milk. *Enterobacteriaceae* varied between <10 and $3.26x10^4$ CFU/ml in milk and between <10 and 8.1×10^4 CFU/gr in cheese. Coagulase-positive Staphylococci varied from <100 to 3.1×10^4 CFU/ml for milk. Three ripened cheeses from different producers showed counts higher than those permitted by the European Regulations. The rise in *Enterobacteriaceae* could be due to environmental contamination, because some cheese factories, located above 1400 m a.s.l., are used only during the summer period, when sufficient hygiene standards could be difficult to obtain.

Keywords. Plaisentif – cheese – microbiology – chemical composition.

Qualité sanitaire et compostion physico-chimique du fromage Plaisentif.

Résumé. Ce travail représente la première tentative de caractérisation du fromage Plaisentif. Plaisentif est un fromage traditionnel produit dans les Alpes occidentales et obtenu à partir de lait cru de vaches nourries sur les pâturages pendant la période de floraison des violettes. Il n'as jamais été caractérisé. L'objectif de ce travail était de évaluer la qualité sanitaire microbiologique des laits et des fromages et de déterminer leur composition physico- chimique. Des échantillons de laits et de fromages à la fin de l'affinage (60 jours) ont été collectés dans neuf fermes laitières alpines. Les teneurs en matière sèche, protéines, graisses et cendres, ainsi que le pH et l'activité de l'eau (Aw) ont été mesurées. Les staphylocoques à coagulase positive, entérobactéries, *Salmonella* spp et *Listeria monocytogenes* ont été dénombrés. Les valeurs moyennes finales de pH et Aw pour les fromages ont été de 5.39 et 0.963, respectivement. *Salmonella* spp n'était détectée ni dans les laits, ni dans les fromages. *Listeria monocytogenes* n'était trouvée que dans un échantillon de lait. La charge en Entérobactéries était comprise entre <10 et $3.26x10^4$ UFC / ml dans les laits et entre < 10 et 8.1×10^4 UFC/g dans les fromages. Dans les laits, la charge en Staphylocoques à coagulase positive a varié de <100 à 3.1×10^4 UFC/ml. Trois fromages affinés provenant de différents producteurs ont montrés des valeurs supérieures que celles autorisées par la Réglementation Européenne. Les valeurs élevées en entérobactéries pourraient être dues à la contamination de l'environnement, puisque certaines fromageries sont situées au-dessus de 1400 m d'altitude et ne sont utilisées que pendant l'été. Cela peut rendre difficile le maintient de forte pratiques d'hygiène.

Mots-clés. *Plaisentif – fromage – microbiologie – composition chimique*

I – Introduction

Plaisentif cheese is a bovine cheese produced with raw milk and generally without using starters from cows fed with pasture grass. Pastures and cheese factories are located respectively at a minimum height of 1400 and 1800 m a.s.l. The peculiarity of this cheese is the fact that it is produced from pastures during the violet flowering period (from June to July). Briefly, the cheese is produced by adding the milk obtained from the morning milking to that from the previous evening's milking and warmed to 33-36 °C. Rennet is added and clotting time is established visually by the cheese-maker (usually takes around one hour). The curd is cut into blocks of varying sizes, depending on the producer's preferences. After 5-10 minutes, the curd is mixed, collected, pressed and drained for 12 hours. The cheese can be dry-salted or in brine, and then ripened for 60 days. The disciplinary contain no specifications regarding temperature and humidity for this phase. At the end of the ripening phase, the cheese is marked with the typical logo, the producer name and the day of marking. The aim of this work was to evaluate the microbial hygiene and safety status and bromatological composition of milk and Plaisentif cheese.

II – Materials and methods

Samples of pooled milk (sampled in the whole herd when it was in the vat) and 60-day ripened cheese were taken from nine producers. pH and water activity (Aw) were measured using an HI 99163 pH meter (Hanna instruments, Italy) and an Acqualab 3TE water activity meter (Decagon Devices, Inc., Pullman, WA), respectively. Milk and cheese samples were analyzed to determine dry matter, ash by ignition to 550°C and crude protein content according to the AOAC method (AOAC, 2000) while the crude fat content was determined according to the method described by Hara and Radin (1978).

Regarding microbiology, *Enterobacteriaceae* were analysed using the AFNOR V08-054 protocol, Salmonella with the ISO 6579-1993 protocol, *Listeria monocytogenes* with ISO11290-1/Amd 1:2004E and Coagulase-positive *Staphylococci* with the AFNOR V08-14 protocol modified using Baird-Parker Agar with RPF in order to detect coagulase activity (Liophilchem, Italy). The other mediums were purchased from Oxoid (Oxoid, UK).

III – Results and discussion

A mean pH value of 6.77±0.06 and 5.39±0.19 was found for milk and cheese, respectively; cheese showed a mean Aw of 0.963±0.009. The results of the bromatological composition are reported in Table 1. The analytical results showed that, even in the presence of a production regulation, there is a great difference among producers, as far as shape, cheese-making process, and ripening conditions are concerned. This may explain the high degree of variability observed among milk and cheese composition. For example, the use of partially-skimmed milk from the evening milking, mixed with milk from the morning milking, could affect the fat content of the ripened cheese. The same conditions have been found in other typical dairy products, such as Toma Piemontese cheese, which is produced in three different types according to fat content that can vary between 17 % and 28 % (Manzi *et al.*, 2007; Zeppa, 2004).

Regarding microbiological analysis, *Salmonella* was not found in the milk, while *Listeria monocytogenes* was only found once. Neither *Salmonella* nor *Listeria monocytogenes* were found in the cheese. *Enterobacteriaceae* varied from <10 to 3.26×10^4 CFU/ml (mean: 7.39×10^3) and from <10 to 8.1×10^4 CFU/g (mean: 1.65×10^4) in milk and in cheese, respectively. The *Enterobacteriaceae* count distributions in milk and cheese among producers are shown in Figures 1 and 3, respectively. Coagulase-positive *Staphylococci* varied from <100 to 3.1×10^4 CFU/ml (mean 5.26×10^3) for milk (Figure 2). Only three ripened

samples out of nine (one = 500 and two = 100 CFU/g) from different producers showed counts higher than those permitted by European Regulations (Figure 4). Some producers had low EC counts in milk and high EC counts in cheese. This may be ascribed to the poor hygienic conditions of cheese factories located at high altitude, which determine the *Enterobacteriaceae* contamination of environment, water and milk (Bintis *et al.*, 2008; Bhatt *et al*., 2012; Coton *et al.*, 2012).

Table 1. Bromatological composition (% of fresh matter) of milk (n=18) and Plaisentif cheese (n=18) from nine Piedmont farms (mean ± SD†)

	Dry matter	Protein	Fat	Ash
Milk	12.74±0.75	3.09±0.31	2.62±0.54	0.69±0.03
Cheese	58.97±2.87	22.55±2.14	21.94±3.46	2.47±0.15

†SD: standard deviation

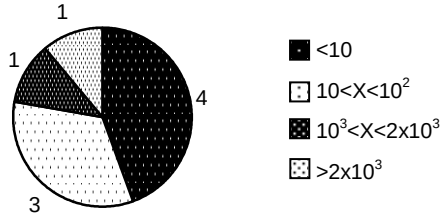

Fig. 1. *Enterobacteriacee* count in milk samples (CFU/ml).

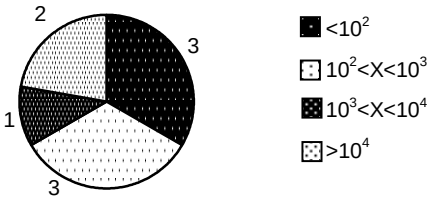

Fig. 2. Coagulase-positive *Staphylococci* count in milk samples (CFU/ml).

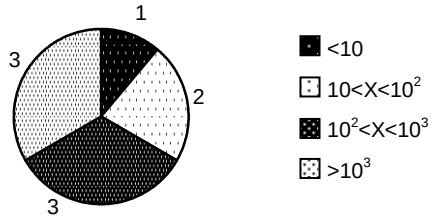

Fig. 3. *Enterobacteriacee* count in cheese samples (CFU/g).

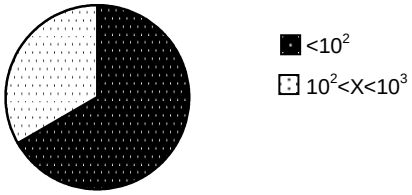

Fig. 4. Coagulase-positive *Staphylococci* count in cheese samples (CFU/g).

IV – Conclusions

Preserving typical food products provides valuable support for the natural environment and economic revitalization of local communities. In this sense, Plaisentif turns out to be a very interesting model. This work is thus the first attempt to evaluate the hygienic status of Plaisentif cheese and its chemical composition.

From these data, the cheese appears to be considered as safe. There is clearly, however, considerable variability in the data due to the craft-based nature and to the difficult conditions of the cheese-making process. This could be considered positive giving a proof in traditional and artisanal products.

From microbiological point of view further investigations are necessary in order to clarify the source of *Enterobacteriaceae*.

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References

AOAC, **2000.** Official Method of Analysis, 17th Edn. Association of Official Analytical Chemists, AOAC, Arlington, VA.

Bhatt, V. D., Ahir, V. B., Koringa, P. G., Jakhesara, D. N., Rank, D. N., Naurial, D. S., Kunjadia, A. P. and Joshi, C. G., 2012. Milk microbiome signatures of subclinical mastitis affected cattle analysed by shotgun sequencing. In: *Journal of Applied Microbiology* 112, p. 639-650.

Bintsis, T., Angelidis, A. S. and Psoni, L. 2008. Modern Laboratory Practices – Analysis of Dairy Products In: *Advanced Diary Science and Technology* (eds T.J. Britz and R.K. Robinson) Oxford (UK): Blackwell Publishing Ldt, pp 312.

COMMISSION REGULATION (EC), 2007. No 1441/2007 of 5 December 2007 amending Regulation (EC) No 2073/2005 on microbiological criteria for foodstuffs. GUCE L322/12, 23.

Coton, M, Delbés-Paus, C., Irlinger, F., Desmasures, N., Le Fleche, A., Stahl, V., Montel, M. C. and Coton, E., 2012. Diversity and assessment of potential risk factors of gram-negative isolates associated with French cheeses. In: *Food Microbiology,* 29, p. 88-89.

Hara, A. and Radin, N. S., 1978. Lipid extraction of tissues with a low-toxicity solvent. In: *Analytical Biochemistry*, 90, p. 420-426.

Manzi, P., Marconi, S., Di Costanzo, M. G. and Pizzoferrato, L. 2007. Composizione di formaggi DOP italiani. In: *La Rivista di Scienza dell’Alimentazione*, 36, p. 9-22.

Zeppa, G., 2004. Toma Piemontese. [on line]. Available from: http://www./diaryscience.info/cheese-of-piedmont-region-of-italy/72-toma-piemontese.htlm (accessed: 27 January, 2014).