Shelf life evaluation of *ricotta fresca* sheep cheese in modified atmosphere packaging

Carlo Pala,[1] Christian Scarano,[1] Massimiliano Venusti,[2] Daniela Sardo,[2] Daniele Casti,[1] Francesca Cossu,[1] Sonia Lamon,[1] Vincenzo Spanu,[1] Michela Ibba,[1] Michela Marras,[1] Antonio Paba,[3] Carlo Spanu,[1] Enrico Pietro Luigi De Santis[1]

[1]Department of Veterinary Medicine, University of Sassari, Sassari; [2]LAORE Sardinia Agency, Zootechnical Production Department, Sassari; [3]AGRIS Sardinia Agency, Department of Research on Animal Production, Sassari, Italy

Abstract

Ricotta fresca cheese is the product of Sardinian dairy industry most exposed to microbial post-process contamination. Due to its technological characteristics, intrinsic parameters, pH (6.10-6.80) and water activity (0.974-0.991), it represents an excellent substrate for the growth of spoilage and pathogenic microorganisms, which are usually resident in cheese-making plants environments. Generally, *ricotta fresca* has a shelf life of 5-7 days. For this reason, at industrial level, modified atmosphere packaging (MAP) is used to extend the durability of the product. However, few investigations have been conducted to validate the use of MAP in *ricotta fresca*. The aim of this work is to evaluate the shelf life of *ricotta fresca* under MAP. A total of 108 samples were collected from three Sardinian industrial cheese-making plants and analysed within 24 h after packaging and after 7, 14 and 21 days of refrigerated storage. Aerobic mesophilic bacteria, mesophilic and thermophilic cocci and lactobacilli, *Enterobacteriaceae* and *E. coli*, *L. monocytogenes*, *Pseudomonas* spp., *Bacillus cereus*, yeasts and moulds, and the chemical-physical parameters and composition of the product were determined. At the end of the shelf life, *Pseudomonas* spp. and *Enterobacteriaceae* reached high concentrations, 5 to 7 and 3 to 6 $\log_{10}$ colony forming unit g^{-1}, respectively. The presence of environmental contaminants indicates that the use of MAP without the appropriate implementation of prerequisite programmes is not sufficient to extend the durability of *ricotta fresca*. Gas mixture and packaging material should be selected only on the basis of scientific evidence of their effectiveness.

Introduction

Ricotta fresca is a traditional cheese produced by heat coagulation of ovine whey. In order to enhance product texture and increase yield, *ricotta fresca* can be obtained from a blend of whey with milk or cream milk. In Sardinia it is typically produced from the whey remaining after the production of sheep's milk cheeses and it is also named *ricotta fresca ovina* or *ricotta gentile* (Ministerial Decree 8 September 1999, n.350; Italian Republic, 1999). In industrial cheese-making plants, the traditional batch process is applied. The whey is filtered, preheated to about 65°C, sent to storage tanks and then to large open kettles, where, by steam injection or by indirect heating, the temperature rises to 80-82°C. As the flocculated proteins start floating on the whey surface the heating is interrupted and the mixture is held for 15-20 min. The clotted proteins are collected using perforated ladle and transferred into conic plastic baskets to drain. *Ricotta* baskets are then placed in cold rooms (3±1°C) where they are allowed to drip and to rapidly cool down within 12 h. The average weight of the final product is approximately 1.7 kg, with pH ranging from 6.10 to 6.80 and water activity (a_w) from 0.974 to 0.991 (De Santis *et al.*, 1999). The mean composition is 70-80% moisture, 10-25% fat and 8-10% protein. Depending on the target market, *ricotta* can be wrapped in food paper or packaged in modified atmosphere packaging (MAP). *Ricotta* cheese wrapped in food paper is usually sold in proximity market, gourmet and specialty food stores, while the product in MAP in large-scale retailers. *Ricotta fresca* can be consumed as it is or as ingredient in sweet and savoury dishes. Durability defined by food business operators (FBO) of *ricotta fresca* differs depending on the packaging methods, being for the wrapped one 5-7 days while for *ricotta* in MAP up to 21 days. As a consequence of high temperatures applied during production, *ricotta fresca* cheese has naturally a poor competitive microflora (Pintado *et al.,* 2002). At clotting it is mainly represented by bacterial spores survived at high temperatures, like *Bacillus cereus* spores (De Santis *et al.*, 1999). The open nature of batch production, which includes handling of the curd after floating, exposes *ricotta* to secondary contamination originating from the equipment and the processing environment (Greenwood *et al.,* 1991). An elevated risk of contamination exists in environments downstream curd flocculation such as moulding, chilling and packaging that are held in an environment not always equipped with adequate hygienic protection systems, like air filtration (Kousta *et al.,* 2010). Sources of contamination are represented by food contact (*i.e.* drainage tables and utensils) and non-food contact surfaces (*i.e.* floors, drains, outer parts of processing equipment). The contaminants can persist within niches in the processing environment protected by a structured biofilm ecosystem (Simões *et al.,* 2009). From niches microorganism find their way into exposed food through direct contact, aerosol, dripping or water splashes and by means of operators. Even with the application of good hygiene practice (GHP) and good manufacturing practice (GMP), environmental contamination of *ricotta* cannot be avoided but only limited. *Ricotta* composition and its intrinsic properties, and the absence of preservatives in the formulation make this product an excellent substrate for the growth of spoilage and pathogen microorganisms (De Santis and Mazzette, 2002). The mean aerobic mesophilic bacteria counts 24 h after the production usually ranges between <2.0 and 3.0 $\log_{10}$ CFU g^{-1}. During cold storage different groups of microorganisms such as coliforms, coagulase-negative staphylococci and psychotrophic *B. cereus* strains are able to grow even 1-2 $\log_{10}$ in 24 h (Pintado and Malcata, 2000). However, at refrigeration temperature psychotropic microorganisms belonging to the *Pseudomonas* genera can overgrow the other microflora and cause food spoilage (Champagne *et al.*, 1994; Carrascosa *et al.*, 2015). MAP is a packaging technique frequently used in the food industry in order to control the microbial growth (Peelman *et al.,* 2014). After extraction of the air present in the headspace of the package, a mixture of CO_2, O_2 and

Correspondence: Carlo Spanu, Department of Veterinary Medicine, University of Sassari, via Vienna 2, 07100, Sassari, Italy.
Tel: +39.079.229454 - Fax: +39.079.229458.
E-mail: cspanu@uniss.it

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N_2 in different proportion is usually introduced before the packaging sealing. In order to have a good increase of the shelf life and maintain the desired CO_2 concentration it is important to use a high-barrier packaging film and apply the vacuum as well as possible. MAP has been successfully used to extend the shelf life of a wide variety of ready-to-eat foods (Gonzalez-Fandos *et al.,* 2000). Several studies demonstrated the effectiveness of MAP in shelf life extension of hard and fresh cheeses (Eliot *et al*, 1998). Moir *et al.* (1993) demonstrated that MAP packaging can inhibit *Pseudomonas* spp. growth in soft cheese stored at 5 and 15°C.

Due to the always-increasing demand by the large distribution of *ricotta fresca* with extended durability, in the last years Sardinian industrial cheese-making plants adopted MAP. The shelf life of MAP *ricotta fresca* is defined by the FBO and generally varies from 15 to 21 days. The use of MAP for the shelf life extension was previously evaluated in different *ricotta* cheeses from sheep (Del Nobile *et al*., 2009) or cow whey milk and in similar products such as *Myzithra Kalathaki* made in Greece (Dermiki *et al.,* 2008). The aim of this work is to conduct a durability study to evaluate the evolution of the microbiological profile, physicochemical characteristics and composition of industrial sheep *ricotta fresca* cheese in MAP during refrigerated storage up to 21 days.

Materials and Methods

Ricotta fresca cheese in modified atmosphere packaging samples

The study was conducted in Sardinia (Italy) during the year 2014 enrolling three sheep milk industrial cheese-making plants (respectively A, B and C). The selected plants were representative of Sardinian sheep milk cheese-making production sector, characterised by similar *ricotta* manufacturing process and by the use of MAP. In each cheese-making plant data about process, packaging technologies, and GMPs and GHPs enforcement level were collected. From each plant *ricotta fresca* cheese samples randomly selected from 3 different production batches, which were representative of the seasonal sheep's milk production (from November/December to July), were obtained. Therefore, samples were collected in February/March, April/May and June/July and identified as batch 1, 2 and 3, respectively. A total of 108 *ricotta fresca* samples were collected, 36 from each plant and 12 from each batch. All *ricotta fresca* samples were packaged the day after the production. The chosen gas mixtures were composed of 30% CO_2 and 70% N_2 for two cheese-making plants (A, B) and 100% N_2 for the third one (C) (Figure 1).

Sampling plan

Ricotta fresca cheese samples under refrigeration conditions (3±1°C) were transported to the Department of Veterinary Medicine, University of Sassari, Sassari, and stored for laboratory analysis. Samples were analysed the day of the delivery (T_0), at 7 (T_7), 14 (T_{14}) and 21 (T_{21}) days after the production for the determination of the headspaces gas concentration, composition, intrinsic properties, and microbiological analysis.

Headspace gas composition

Headspaces gas mixture composition was determined using the Dansensor gas analyser (PBI- Dansensor, Ringsted, Denmark). Determinations were conducted before all other analysis by piercing the surface of sealed *ricotta fresca* MAP samples with a sterile needle connected to the Dansensor. Measures of combined residual O_2% and CO_2% were directly read on the instrument, while N_2 was obtained by difference.

Microbiological analysis

All microbiological analyses were conducted according to international standard methods and included the following parameters: total aerobic mesophilic counts (ISO 4833; ISO, 2003), *Enterobacteriaceae* (ISO 21528-2; ISO, 2004a), *Listeria monocytogenes* (ISO 11290-1/2; ISO, 1996, 1998), *Pseudomonas* spp. (ISO/TS 11059:2009; ISO, 2009), *B. cereus* (ISO 7932; ISO, 2004b), yeast and moulds (ISO 6611/IDF 94; ISO, 2004c). Presumptive mesophilic and thermophilic cocci were enumerated on M17 agar (Microbiol, Cagliari, Italy) incubated anaerobically for 72 h at 30°C and at 45°C, respectively. Presumptive mesophilic lactobacilli were enumerated on FH agar (Isolini *et al*. 1990) and anaerobically incubated at 37°C for 72 h. Presumptive thermophilic lactobacilli were enumerated on MRS agar (Microbiol) adjusted to pH 5.4, anaerobically incubated at 45°C for 72 h. The KAA agar medium (Lab M Limited, Heywood, UK), aerobically incubated at 42°C for 18 h, was used for presumptive enterococci enumeration. From each samples two 25 g aliquots were aseptically collected and homogenised, one with 225 mL of fraser broth base (Biolife, Milan, Italy) for the detection of *L. monocytogenes* and one with 225 mL of buffered pepton water (BPW) (Biolife) for all other parameters. After homogenisation, serial decimal dilutions were prepared in BPW solution and inoculated onto agar plates containing the culture media appropriate for each of the parameter to be investigated.

Physico-chemical analysis and composition

From each *ricotta fresca* cheese sample 40 g, representative of the whole products, were sampled. Intrinsic properties (pH and a_w) were measured using pH meter GLP22 (Crison Instruments, Barcelona, Spain) and a_w-meter Aqualab 4TE (Decagon, Pullman, WA, USA), respectively. Fat, moisture, protein were analysed using a near infrared transmittance compositional analyser (FOSS, Eden Prairie, MN, USA).

Statistical analysis

Differences among average microbiological counts [colony forming unit (CFU) g^{-1}], headspace gas concentration (%), intrinsic proper-

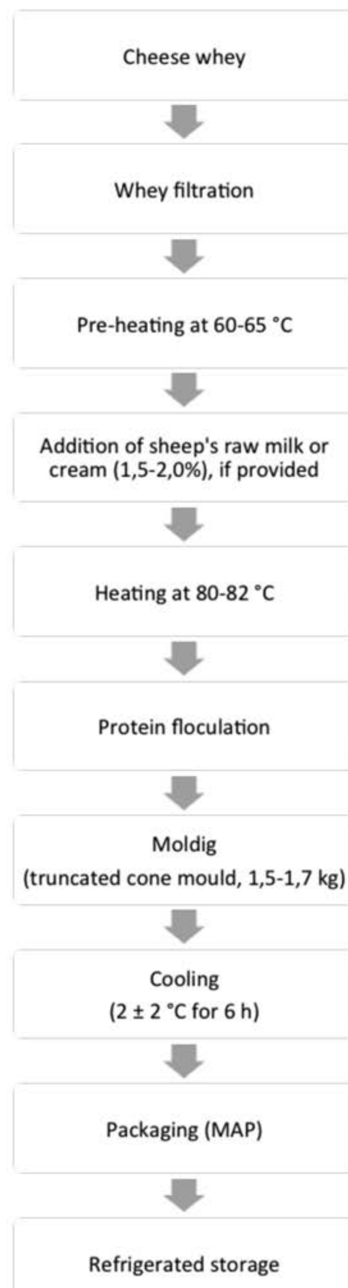

Figure 1. Process flow diagram of modified atmosphere packaged *ricotta fresca.*

ties (x) and centesimal composition (%) over time (T_0, T_7, T_{14} and T_{21}) were compared using Fisher's least significant difference test. Statistical analyses were performed with Statgraphics Centurion XVI software (Stat Point Technologies, Warrenton, VA, USA).

Results

Headspace gas composition

The headspace gas composition values for each cheese-making plant at each sampling time are reported in Table 1. The gas mixtures composition used in *ricotta fresca* cheese packaging lines was 30% CO_2 and 70% N_2 for two cheese-making plants (A and B) and 100% N_2 for the third one (C). In samples from A and B plants the CO_2% at T_0 was 10.96±0.93 and 8.50±2.92 respectively, *i.e.* values three times lower than in the gas mixtures. A further considerable CO_2% decrease was detected at T_7, when it reached values as low as 3.38±1.85 (A) and 4.11±1.85 (B). More differences were detected in CO_2% between products from A and B cheese-making plants at T_{14} and T_{21} during refrigerated storage. The O_2% ranged between 1.38±2.52% (C) and 4.48±3.0 (B) at T_0. The O_2% shows a progressive reduction until T_{21}. In samples from cheese-making plant C, where only N_2 was used as gas for MAP, slight decrease was observed in N_2%, the O_2 residual % detected at T_0 showed a decrease during storage, while CO_2 that was not detected at T_0 increased until 1.87±1.43 % at T_{21}.

Physico-chemical parameters and composition

Slight pH differences at T_0 were observed in samples from cheese-making plants (6.47-6.65) with very low variations after storage at T_{21} (6.50-6.62). A_w levels were usually >0.990 with very low variations among samples and storage times. Composition values (mean±standard deviation) in all samples were: protein 8.48±0.66%, fat 11.94%±2.37, and moisture 76.36±2.44. Higher fat content in samples from cheese-making plant B (P<0.001) was detected in other samples due to cream addition. Table 2 reports the evolution of pH and a_w, while Table 3 the evolution of chemical parameters during shelf life.

Microbiological profile

The aerobic mesophilic bacteria count at T_0 in the samples where it was detectable (positive sample) was between 2.4-2.7 $\log_{10}$ CFU g^{-1}, while higher variability was shown by the positive rate ranging from 44.4 to 100% depending on cheese-making plants (Table 4). During storage, counts progressively increased (P<0.001) reaching levels >7 $\log_{10}$ CFU g^{-1} at T_{14} and T_{21} in samples from B and C dairies, while in samples from A plant they were about 6 $\log_{10}$ CFU g^{-1} (P<0.001) and were detected only at T_{21}. *Enterobacteriaceae* were detectable only in a sample in dairy B at T_0 and during the whole shelf life period (from T_7 to T_{21}) in sam-

Table 1. Evolution of O_2, CO_2 and N_2 percentage (mean±standard deviation) in the headspace of *ricotta fresca* cheese manufactured in three cheese-making plants (three different batches).

Plant	O_2				CO_2				N_2			
	T_0	T_7	T_{14}	T_{21}	T_0	T_7	T_{14}	T_{21}	T_0	T_7	T_{14}	T_{21}
A	1.69±0.94[a]	3.08±1.41[ab]	3.82±2.54[b]	2.10±0.39[a]	10.96±0.93[a]	3.38±1.85[b]	3.00±1.18[b]	3.18±1.11[b]	87.35±1.02[a]	93.55±1.29[bc]	93.18±2.12[b]	94.72±0.96[c]
B	4.48±3.00[a]	5.08±2.97[a]	1.25±1.46[b]	0.0±0.0[b]	8.50±2.92[a]	4.11±1.85[b]	5.99±2.47[ab]	8.12±3.49[a]	87.02±2.58[a]	90.81±2.70[b]	92.76±2.10[b]	91.88±3.59[b]
C	1.38±05[a]	1.57±0.24[a]	0.02±0.03[b]	0.04±0.12[b]	0.00±0.00[a]	0.18±0.27[a]	0.59±0.55[a]	1.87±1.45[b]	98.62±0.57[ab]	98.25±0.22[a]	99.39±0.55[b]	98.09±1.40[a]

The sampling times T_0, T_7, T_{14} and T_{21} refer to the days (0, 7, 14, and 21, respectively) elapsed during the shelf life. [a-c]Means in the same row with different superscript letters are significantly different (P<0.05).

Table 2. Evolution of pH and water activity (mean of three batches±standard deviation) of *ricotta fresca* manufactured in three cheese-making plants during shelf life.

Plant	pH				a_w			
	T_0	T_7	T_{14}	T_{21}	T_0	T_7	T_{14}	T_{21}
A	6.47±0.08[a]	6.46±0.15[a]	6.57±0.11[a]	6.50±0.07[a]	0.994±0.002[a]	0.993±0.003[ab]	0.990±0.005[bc]	0.989±0.005[c]
B	6.74±0.31[a]	6.73±0.09[a]	6.62±0.32[bc]	6.53±0.26[b]	0.989±0.003[a]	0.991±0.005[a]	0.989±0.011[a]	0.989±0.011[a]
C	6.65±0.14[a]	6.71±0.06[ab]	6.82±0.09[b]	6.62±0.25[a]	0.992±0.003[a]	0.993±0.002[a]	0.990±0.006[a]	0.991±0.006[a]

a_w, water activity. The sampling times T_0, T_7, T_{14} and T_{21} refer to the days (0, 7, 14, and 21, respectively) elapsed during shelf life. [a-c]Means in the same row with different superscript letters are significantly different (P<0.05).

Table 3. Evolution of *ricotta fresca* cheese composition (mean±standard deviation) from three batches produced in three cheese-making plants during shelf life.

Composition	Plant	T_0	T_7	T_{14}	T_{21}
Moisture	A	78.49±1.76	76.81±1.96	76.68±1.71	76.69±2.63
	B	72.88±3.17	73.26±3.47	72.15±4.33	73.00±1.86
	C	79.22±2.43	78.14±1.54	78.99±1.51	80.03±2.92
Fat	A	9.82±1.59	11.13±1.69	10.50±2.25	11.44±2.82
	B	15.29±3.63	15.22±1.69	16.11±4.83	15.82±2.15
	C	8.96±1.31	10.11±1.90	9.89±1.92	9.01±2.60
Protein	A	8.13±0.50	8.70±0.38	8.91±0.78	8.67±0.88
	B	8.70±0.88	8.50±0.85	8.74±0.93	8.76±0.87
	C	8.08±0.50	8.48±0.47	7.88±0.23	8.23±0.59

The sampling times T_0, T_7, T_{14} and T_{21} refer to the days (0, 7, 14, and 21, respectively) elapsed during shelf life.

ples from cheese-making plants B and C, reaching 6.10±0.81 (B) and 5.83±1.55 (C) log_{10} CFU g^{-1} (T_{21}). *Enterobacteriaceae* contamination levels in samples from A plant were lower than in B and C and those microorganisms were detected only at T_{21} (2 out 9 samples) when they reached 3.46±1.32 log_{10} CFU g^{-1} (Table 4).

Pseudomonas spp. was detectable at T_0 only in samples from cheese-making plant B, while it was always recovered in all samples from T_7 onwards. In samples collected from B and C cheese-making plants similar levels of *Pseudomonas* spp. were found with a significant increase from T_7 to T_{14}, where maximum values of 7.43±0.67 and 7.26±0.69 log_{10} CFU g^{-1} were reached. The concentration of *Pseudomonas* spp. in dairy A samples was lower ($P<0.001$) than B and C, and it reached values of 5.99±1.24 log_{10} CFU g^{-1} at T_{21} (Table 4). Mesophilic and thermophilic lactobacilli were never detected. Mesophilic cocci were found in 67 out 108 (62.0%) samples with a prevalence of 54.0% (A), 87.5% (B) and 41.6% (C). The mesophilic cocci counts increased during the shelf life in all samples from 0.99±1.31 log_{10} CFU g^{-1} at T_0 to 3.33±2.74 log_{10} CFU g^{-1} at T_{21}. Thermophilic cocci were detected in 27 samples (25%) with levels of 2.79±0.90. Enterococci were detected in 5 samples (4.63%) collected from B and C plants with concentration of 3.32±1.04 log_{10} CFU g^{-1}. In all samples *L. monocytogenes*, *E. coli* and *B. cereus* were always below the detection limit.

Discussion

MAP has been widely used to extend the shelf life of several ready-to-eat foods including dairy products. Literature reports several studies validating the use of MAP packaging in *ricotta* cheese, hard and soft cheeses such as mozzarella, *stracciatella*, *fior di latte*, cottage and *myzithra kalathaki* with various gases mixture (Moir *et al.,* 1993; Eliot *et al.,* 1998; Dermiki *et al.*, 2008; Conte *et al.*, 2011; Del Nobile *et al.*, 2009; Gammariello *et al.*, 2009; Mastromatteo *et al.*, 2014). The increasing demand of *ricotta fresca* cheese by large-scale retail with extended durability drives the use of MAP or of alternative production technologies, like the packaging of hot homogenized products (De Santis *et al*., 1999). MAP industrial *ricotta fresca* produced in Sardinia has already been on the market for several years, with a shelf life extended by FBO until 21 days. However, no scientific evidence supports the definition of this durability. The first evidence obtained with the survey is that differences exist in the initial gas composition used by different operators. Two cheese-making plants used a mixture of 30% CO_2 and 70% N_2 (A, B), while the third cheese-making plant used only N_2 (C). No justification on the use of a mixture is given by FBO. Another observation is that even with similar gas injection the actual initial composition in the headspace is different and changes over time. Samples from cheese-making plants A and B at T_0 showed a different CO_2 levels variability (B>A) as a result of different packaging procedures. In plant B insufficient headspace was left and inaccuracy of gas injection occurred. At T_0 CO_2 concentration in headspace differs from level provided in the gas mixture as a result of prompt gas solving on the product. The further CO_2 reductions after the first storage week could be related to packaging permeability, as confirmed by evidence collected during visits at cheese-making plants showing non-compliance of packaging materials with the requirement of high density barrier (the O_2 transfer rate at 20°C and 65% RH was about 30 cc/m^2). In samples where poor microbiological characteristics were observed at T_{14} and T_{21} (dairy B) a further increase in CO_2 is associated with high aerobic mesophilic bacteria level (>log 7 CFU g^{-1}) resulting in CO_2 release in the headspace, as a microbial fermentation product. The same evolution could be observed in samples from dairy C with a slight CO_2 increase in the last part of storage. The O_2% at T_0 was higher than 1%, and in dairy B lacking in control of residual level of this gas as showed by concentration >4%. The O_2 reduction during storage is associated with the increase of aerobic mesophilic microbial count. The changes in the relative concentrations of CO_2 and O_2 during storage resulted in higher variability of N_2 levels, obviously lower in product from dairy C, where it was the only component of the initial gas mixture. The residual O_2 combined with intrinsic properties and absence of competitive microflora makes *ricotta fresca* an excellent substrate for growth of aerobic psychrotrophic spoilage and pathogen microorganism. In analysed samples pathogens like *L. monocytogenes* and *B. cereus* were never detected, while spoilage psychrotrophic microorganism such as *Pseudomonas* spp. can overgrow other microflora reducing the risk of pathogens growth (Buchanan and Bagi, 1999). Furthermore, the combination of low temperatures and long storage time represents a selective advantage for *Pseudomonas* spp. (Carrascosa *et al*., 2015). *Pseudomonas* spp. as high as 7 log CFU g^{-1} in *ricotta* cheese should be regarded as values reached in spoiled products. High concentration of *Pseudomonas* spp. negatively affects the flavour and the texture of the product (Leriche *et al.,* 2004). An important feature of some *Pseudomonas* spp. is the secretion of the yellow-green pyoverdin (Meyer *et al.,* 2002) or blue pyocyanin (Cantoni *et al.*, 2003) that induce important discoloration of the product. Microbial levels in *ricotta fresca* cheese at production occurred mostly as a post-process contamination, depending from GMP and GHP enforcement at production and packaging. During the preliminary inspection in cheese-making plants B and C, evidence of poor implementation of hygiene management (*e.g.*, sanification, personnel hygiene and training, *etc*.), process control and packaging procedures were observed. Failure in hygienic management resulted in higher

Table 4. Microbiological profile (log_{10} colony forming unit g^{-1}; mean of three batches±standard deviation (*ricotta fresca*) of I manufactured in three different cheese-making plants during shelf life.

Parameters	Plants	T_0	T_7	T_{14}	T_{21}
Aerobic mesophilic bacteria	A	2.43±0.21[a] (4/9)	2.41±0.48[a] (6/9)	4.70±1.18[b] (8/9)	5.91±1.14[c] (9/9)
	B	3.56±0.69[a] (8/9)	5.51±1.38[b] (9/9)	7.22±0.45[c] (9/9)	7.09±0.75[c] (9/9)
	C	2.70±0.38[a] (9/9)	4.16±1.81[b] (9/9)	6.91±0.89[c](9/9)	7.43±0.50[c] (9/9)
Enterobacteriaceae	A	nda (0/9)	nda (0/9)	nd[a] (0/9)	3.46±1.32[b] (2/9)
	B	3.09±0.00[a](1/9)	3.53±1.48[a] (7/9)	5.07±1.11[b] (9/9)	6.10±0.81[c] (9/9)
	C	nd[a] (0/9)	2.66±0.94[b] (6/9)	5.06±1.03[c] (9/9)	5.83±1.55[c](9/9)
Pseudomonas spp.	A	nd[a] (0/9)	3.08±0.18[b] (2/9)	5.06±1.15[c] (6/9)	5.99±1.24[c] (6/9)
	B	3.41±0.71[a] (3/9)	5.47±1.43[b] (9/9)	7.26±0.69[c] (9/9)	7.21±0.43[c] (9/9)
	C	nd[a] (0/9)	5.92±0.80[b] (9/9)	7.43±0.67[c](9/9)	7.28±1.05[c] (9/9)

nd, not detectable (below the detection limit). The sampling times T_0, T_7, T_{14} and T_{21} refer to the days (0, 7, 14, and 21, respectively) elapsed during shelf life. The number of positive samples is within brackets.
[a-c]Means in the same row with different superscript letters are significantly different ($P<0.05$).

contamination levels of *ricotta* cheese at T_0 in a faster increase of microorganisms since 4-5 log at T_7, and high level of aerobic mesophilic counts, *Pseudomonas* spp. and *Enterobacteriaceae* at T_{14}. Efficacy of MAP is conditioned by several factors, like the relationship between the product volume and the headspace, the residual O_2 rate contained in the product, the level of vacuum and the correct selection of packaging materials (Khoshgozaran *et al.,* 2012). MAP needs specific shelf life studies with controlled experimental parameters in order to assess the efficacy in controlling spoilage and pathogen microorganisms and to define the durability of *ricotta fresca* based on scientific evidences.

Conclusions

Considering the microbial evolution and high prevalence of contaminants, particularly in *Pseudomonas* spp., in two of three dairies, it is reasonable to assume that results confirmed the different good hygiene and manufacturing practices levels evaluated during the preliminary visits in each dairy plant. Despite MAP packaging, the *ricotta fresca* shelf life is significantly influenced by GMP and GHP applied during production, packaging and storage. Furthermore, the extended durability of chilled products, especially with little or no presence of competitive microflora, exposes the product to the growth of spoilage and pathogen microorganisms. There is no evidence that the actual MAP conditions used in Sardinian cheese-making plants allow to extend the *ricotta fresca* shelf life to 21 days.

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