**Abstract**

Several growth curves for selected pathogens and hygiene indicators alone and as selected dairy starter cultures (LAB) and commercial probiotics have been performed. All strains for LAB and commercial probiotics were inoculated as pure cultures into skim milk to get an initial coccibacilli:enterococci ratio of 2:1:1 and a concentration of approx. 10^7 cfu mL^{-1} until challenge tests were carried out. Selected cultures were grown aerobically in Nutrient Broth (NB, Oxoid CM0001, incubated at 37°C for 24 h) at 24 h was approximately 1x10^9 cfu mL^{-1}.

**Materials and methods**

**Starter cultures and probiotics**

Bacterial strains used as starter cultures were taken from the culture collection of the Laboratorio di Ispezione degli Alimenti di Origine Animale: Lactococcus lactis ssp. lactis, strain 340; L. lactis ssp. lactis, strain 16; Lactobacillus casei ssp. casei, strain 208 and Enterococcus faecium UBEF-41. Lactococci and lactobacilli were isolated from traditional cheeses manufactured in small-scale dairy plants in Umbria, Italy, while Enterococcus faecium UBEF-41 was provided by Bromatech Srl (Milano, Italy). The morphological, biochemical and physiological characterization, together with the acidifying activity of the three lactic acid bacteria has been reported by the authors in previous papers (Cenci Goga et al., 1995; Clementi et al., 1998), while characterization of Enterococcus faecium UBEF-41 is available at Unique Biotech Ltd. (http://uniquebiotech.com/products/animal_healthcare/enterococcus_faecium). Growth curves for all starter cultures alone and as selected pathogens have been recorded throughout the whole experiment. Before challenge-test, freeze-dried strains of the starter cultures were grown aerobically in Nutrient Broth (NB, Oxoid CM0001, Basingstoke, UK) at 37°C for 24 h. Each strain was then sub-cultured in Skim Milk (BD Difco, Franklin Lakes, NJ, USA, 232100) at 37°C for 24 h. Total viable cells (TVC) count (on Nutrient Agar, NA, Oxoid CM0003, incubated at 37°C on air for 24 h) at 24 h was approximately 1x10^9 cfu mL^{-1}.

**Selected microorganisms**

Selected pathogens and hygiene indicators came from the culture collection of the Laboratorio di Ispezione degli Alimenti di Origine Animale or were reference strains: Escherichia coli, CSH26 K12, Staphylococcus aureus, 27R, Salmonella Derby 27, Pseudomonas fluorescens ATCC 13525, Listeria innocua ATCC 33090.

Before challenge-test, freeze-dried strains of the starter cultures were grown aerobically in Nutrient Broth (NB, Oxoid CM0001, Basingstoke, UK) at 37°C for 24 hrs. Each strain was then sub-cultured in Skim Milk (BD Difco, Franklin Lakes, NJ, USA, 232100) at 37°C for 24 h. Total viable cells (TVC) count (on Nutrient Agar, NA, Oxoid CM0003, incubated at 37°C on air for 24 h) at 24 h was approximately 1x10^9 cfu mL^{-1}.

**Characterization of starter cultures with acidification and challenge growth curves**

**Acidification**

All strains were inoculated as pure cultures into skim milk (BD Difco) to get an initial concentration of about 10^7 cfu mL^{-1}. All strains were incubated at 37°C. The association was...
inoculated into skim milk (BD Difco) to get an initial concentration of approximately 10^6 cfu mL^-1. The association was incubated at 37°C and pH was measured with a Foodtrade electrode (Hamilton Company, Reno, NV, USA) hooked to an Eutech pH 2700 (Eutech Instrument Europe B.V. Nijkerk, Netherlands) which recorded pH values continuously with CyberComm 6000 (Eutech Instrument) every minute. The following fourth degree polynomial was used as an empirical model to fit the experimental data:

\[ y = a + bx + cx^2 + dx^3 + ex^4 \]  
(eq. 1)

where \( y \) is pH; \( x \) is time; \( a, c, d, e \) are the generic regression coefficients of the polynomial under study, as determined by the statistical package Stat Graphics, ver. 6.1 (Manugistics Inc. Rockville, MD, USA).

The first derivative of the equation gives the instantaneous acidification rate and its maximum value \((V_m)\) which corresponds to the inflection point of the acidification curve, whereas the second derivative gives the acceleration and its roots give the \( x \) values \((t_m)\) at the inflection points.

**Challenge test**

Several growth curves for selected pathogens and hygiene indicators alone and as the selected dairy starter cultures and commercial probiotics have been performed. All strains for the selected dairy starter cultures and commercial probiotics were inoculated as pure cultures into skim milk (BD Difco) and incubated at 37°C to get an initial concentration of about 10^9 cfu mL^-1 after 24 h of incubation. The association was then inoculated into skim milk (BD Difco) to get an initial cocci:bacilli:enterococi ratio of 2:1:1 and a concentration of about 10^7 cfu mL^-1. The inoculated strains were tested aseptically to 225 mL of sterile, buffered, peptone water (Oxoid, CM1049), and homogenised in a stomacher (PBI International) for 1 min at low speed and 1 min at high speed at room temperature. Serial decimal dilutions in buffered peptone water were prepared and triplicate 1 mL or 0.1 mL samples of appropriate dilutions were poured or spread on total count and selective agar plates. Total lactic microbiota was determined on de Man, Rogosa, Sharpe (MRS) Agar (Oxoid) acidified to pH 5.5, incubated at 30°C for 72 h under anaerobic conditions (Gas generating kit, Oxoid, BR0038); Enterococcus faecium on Slanetz and Bartley (SB) Agar (Oxoid, CM0377), incubated at 37°C for 48 h; Pseudomonas fluorescens on Pseudomonas agar base (Oxoid, CM0559) added with Pseudomonas CFC Supplement (Oxoid, SR0103), incubated at 25°C for 48 h. S. aureus were determined on Rabbit Plasma Fibrinogen (RFP) Agar (Oxoid, CM0961, with RFP Supplement (Oxoid, SR0122), incubated at 37°C for 48 h. Escherichia coli CSH26 K12 on Sorbitol MacConkey agar (McS) Agar (Oxoid, CM0813), overlaid with 5 mL of the same medium and incubated at 30°C for 24 h. The ISO 6579 method was used (ISO, 2007) for Salmonella Derby 27. Briefly, after a pre-enrichment step (buffered peptone water, Oxoid), 18 h at 37°C, and the inoculum on two selective enrichments, Rappaport Vassiliadis Soya peptone broth (RVS, Oxoid CM0866), 24 h at 42°C, and Muller-Kaufmann Tetraionate-Novobiocin Broth (MKTN, Oxoid, CM1048), 24 h at 37°C, a loopful of broth was spread onto Xylose-Lysine-Desoxycholate Agar (XLD, Oxoid, CM0469) and Salmonella Chromogenic Agar (Oxoid, CM1007) with Salmonella selective supplement, Oxoid, SR0194). The ISO 11290 method (ISO, 1996) was used for Listeria innocua ATCC 35909. Briefly, after a primary (Fraser, Oxoid, CM 0895 with half Fraser selective supplement, Oxoid, SR0166), 24 h at 30°C, and a secondary enrichment medium (Fraser with Fraser supplement, Oxoid, SR0156), 48 h at 35°C, a loopful of broth was spread onto Oxford agar (Listeria selective agar, Oxoid, CM0856 with Listeria selective supplement, SR0140) and PALCAM agar (PALCAM agar base, Oxoid, CM0877 with PALCAM selective supplement, Oxoid, SR0150).

**Microbiological analysis**

For the analysis about 25 mL of sample were transferred aseptically to 225 mL of sterile, buffered, peptone water (Oxoid, CM1049), and homogenised in a stomacher (PBI International) for 1 min at low speed and 1 min at high speed at room temperature. Serial decimal dilutions in buffered peptone water were prepared and triplicate 1 mL or 0.1 mL samples of appropriate dilutions were poured or spread on total count and selective agar plates. Total lactic microbiota was determined on de Man, Rogosa, Sharpe (MRS) Agar (Oxoid) acidified to pH 5.5, incubated at 30°C for 72 h under anaerobic conditions (Gas generating kit, Oxoid, BR0038); Enterococcus faecium on Slanetz and Bartley (SB) Agar (Oxoid, CM0377), incubated at 37°C for 48 h; Pseudomonas fluorescens on Pseudomonas agar base (Oxoid, CM0559) added with Pseudomonas CFC Supplement (Oxoid, SR0103), incubated at 25°C for 48 h. S. aureus were determined on Rabbit Plasma Fibrinogen (RFP) Agar (Oxoid, CM0961, with RFP Supplement (Oxoid, SR0122), incubated at 37°C for 48 h. Escherichia coli CSH26 K12 on Sorbitol MacConkey agar (McS) Agar (Oxoid, CM0813), overlaid with 5 mL of the same medium and incubated at 30°C for 24 h. The ISO 6579 method was used (ISO, 2007) for Salmonella Derby 27. Briefly, after a pre-enrichment step (buffered peptone water, Oxoid), 18 h at 37°C, and the inoculum on two selective enrichments, Rappaport Vassiliadis Soya peptone broth (RVS, Oxoid CM0866), 24 h at 42°C, and Muller-Kaufmann Tetraionate-Novobiocin Broth (MKTN, Oxoid, CM1048), 24 h at 37°C, a loopful of broth was spread onto Xylose-Lysine-Desoxycholate Agar (XLD, Oxoid, CM0469) and Salmonella Chromogenic Agar (Oxoid, CM1007) with Salmonella selective supplement, Oxoid, SR0194). The ISO 11290 method (ISO, 1996) was used for Listeria innocua ATCC 35909. Briefly, after a primary (Fraser, Oxoid, CM 0895 with half Fraser selective supplement, Oxoid, SR0166), 24 h at 30°C, and a secondary enrichment medium (Fraser with Fraser supplement, Oxoid, SR0156), 48 h at 35°C, a loopful of broth was spread onto Oxford agar (Listeria selective agar, Oxoid, CM0856 with Listeria selective supplement, SR0140) and PALCAM agar (PALCAM agar base, Oxoid, CM0877 with PALCAM selective supplement, Oxoid, SR0150).

Sensitivity for spread plate was 10^2 cfu g^-1 and for pour plate was 10 cfu g^-1 and the 95% confidence limit, as given by the classic formula 2s=2\(\sqrt{s}\) (Adams and Moss, 2000), ranged between ±37% and ±12% (i.e. plates with a number of cfu ranging from 30 to 300). Therefore, no plates with less than 30 cfu were used for data analysis, and when this applied to the lowest dilution, the results were recorded as <300 for pour plate and <3000 for spread plate. Samples without typical colonies were recorded as negative and samples with at least one typical colony in the lowest dilution were recorded as positive, regardless the number of colonies (Cenci Goga et al., 2005).

**Analysis of the results**

For each sampling the arithmetic means of three subsamples of each of the three sausages was calculated and then all the values (converted to log for microbiological analyses) were analysed using GraphPad InStat, version 3.0b for Mac OS X and graphs were obtained with GraphPad Prism version 6.0d for Mac OS X.

**Results and discussion**

**Characterization of starter cultures with acidification and challenge growth curves**

**Addifying activity of selected starter cultures**

Three species of lactic acid bacteria previously isolated from among the most representative ones from sheep milk, curd and cheese in cheese making trials carried out previously (Cenci Goga et al., 1995) along with a commercial probiotic were chosen to be used as a starter. These strains had been previously identified by API 50 CHL and some of them had already been used as a starter in the manufacturing of salami (Cenci Goga et al., 2008; Clementi et al., 2012). The acidifying activity of the different species had been preliminarily tested (Cenci Goga et al., 1995; Clementi et al., 1998) as pure cultures and as different associations in sterilized cow milk to assess their suitability to be used as a starter. The parameters describing the acidification kinetics are reported in Table 1 together with the regression coefficients of

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the fourth degree polynomial (equation 1) which was used as an empirical model. This adequately fitted the experimental data, since the $r^2$ values varied from 0.997 to 0.9998 and the actual values were almost exactly superimposed on the empirical model curves (Figure 1). The values of the maximum instantaneous acidification rate ($V_m$) were of the same order posed on the empirical model curves (Figure 1). The actual values were almost exactly superimposed on those (-5.5 $\times 10^{-3}$). These values were not very different from those of strain 16 which had the highest $V_m$ (-3.14 $\times 10^{-3}$). Table 1 shows the kinetic parameters of the acidifying activity of several lactic acid bacteria with a similar procedure (Pique et al., 1992). The final pH values of milk inoculated with the different strains as pure cultures (Figure 1) were lower than those previously obtained in cow milk (Cenci Goga et al., 1995; Clementi et al., 1998) except for Lb. casei spp casei which reached the same value in both types of milk. After 900 min, the two strains of Lc. lactis spp lactis reached pH values equal to or lower than those obtained in cow and sheep milk inoculated with Lc. lactis spp lactis strains isolated from cheeses (Ledda et al., 1994). Enterococcus faecium gave a one fold pH decrease within about 500 min, like some commercial thermophilic lactobacilli (Chamba and Prost, 1989). The association of the four strains performed well with a final pH of 3.74 and showed the fastest pH drop, within the first 24 h along with a steady descent subsequently. The association mimicked the behaviour of the two Lc. lactis spp lactis strains in the first 16 h and then, when the lactococci acidification activity decreased (steady pH values after 24 h), mirrored the Lb. casei spp casei performance for a continuous pH drop up to 70 hrs. This synergism is of the utmost importance in the production of fermented food such as cheeses and dry salami because the LAB activity is desirable throughout the whole fermentation process (Cenci Goga et al., 2012, Cizekieni et al., 2013).

**Challenge growth curve**

The results are shown in Figure 2, which displays the different growth curves of pathogens or hygiene indicators vs the association of selected dairy starter cultures and commercial probiotics. Figure 2a shows the evolution of E. coli with an even faster drop (<10^5 cfu mL^-1) after 30 h. S. aureus proved to be more resilient and a substantial reduction in total cell numbers was observed. Panel 2c shows Staphylococcus aureus, strain 27R; panel 2d shows Listeria innocua ATCC 33090; panel 2e shows Pseudomonas fluorescens ATCC 12983 and panel 2f shows the evolution of LAB and E. faecium. For the challenge E. coli vs LAB, level of E. coli in milk dropped, after 48 h, below 10^5 cfu mL^-1, and was no longer detectable at 72 h, while in BHI the growth curve of E. coli was parallel to control, indicating an effect of pH rather than a direct activity of LAB and/or bacteriocins. A similar behaviour was shown by Salmonella Derby with an even faster drop (<10^5 cfu mL^-1) after 30 h. S. aureus proved to be more resilient and a substantial reduction in total cell numbers was observed. Panel 2c shows Staphylococcus aureus, strain 27R; panel 2d shows Listeria innocua ATCC 33090; panel 2e shows Pseudomonas fluorescens ATCC 12983 and panel 2f shows the evolution of LAB and E. faecium. For the challenge E. coli vs LAB, level of E. coli in milk dropped, after 48 h, below 10^5 cfu mL^-1, and was no longer detectable at 72 h, while in BHI the growth curve of E. coli was parallel to control, indicating an effect of pH rather than a direct activity of LAB and/or bacteriocins. A similar behaviour was shown by Salmonella Derby with an even faster drop (<10^5 cfu mL^-1) after 30 h. S. aureus proved to be more resilient and a substantial reduction in total cell numbers was observed. Panel 2c shows Staphylococcus aureus, strain 27R; panel 2d shows Listeria innocua ATCC 33090; panel 2e shows Pseudomonas fluorescens ATCC 12983 and panel 2f shows the evolution of LAB and E. faecium. For the challenge E. coli vs LAB, level of E. coli in milk dropped, after 48 h, below 10^5 cfu mL^-1, and was no longer detectable at 72 h, while in BHI the growth curve of E. coli was parallel to control, indicating an effect of pH rather than a direct activity of LAB and/or bacteriocins. A similar behaviour was shown by Salmonella Derby with an even faster drop (<10^5 cfu mL^-1) after 30 h. S. aureus proved to be more resilient and a substantial reduction in total cell numbers was observed. Panel 2c shows Staphylococcus aureus, strain 27R; panel 2d shows Listeria innocua ATCC 33090; panel 2e shows Pseudomonas fluorescens ATCC 12983 and panel 2f shows the evolution of LAB and E. faecium. For the challenge E. coli vs LAB, level of E. coli in milk dropped, after 48 h, below 10^5 cfu mL^-1, and was no longer detectable at 72 h, while in BHI the growth curve of E. coli was parallel to control, indicating an effect of pH rather than a direct activity of LAB and/or bacteriocins. A similar behaviour was shown by Salmonella Derby with an even faster drop (<10^5 cfu mL^-1) after 30 h. S. aureus proved to be more resilient and a substantial reduction in total cell numbers was observed. Panel 2c shows Staphylococcus aureus, strain 27R; panel 2d shows Listeria innocua ATCC 33090; panel 2e shows Pseudomonas fluorescens ATCC 12983 and panel 2f shows the evolution of LAB and E. faecium. For the challenge E. coli vs LAB, level of E. coli in milk dropped, after 48 h, below 10^5 cfu mL^-1, and was no longer detectable at 72 h, while in BHI the growth curve of E. coli was parallel to control, indicating an effect of pH rather than a direct activity of LAB and/or bacteriocins. A similar behaviour was shown by Salmonella Derby with an even faster drop (<10^5 cfu mL^-1) after 30 h. S. aureus proved to be more resilient and a substantial reduction in total cell numbers was observed. Panel 2c shows Staphylococcus aureus, strain 27R; panel 2d shows Listeria innocua ATCC 33090; panel 2e shows Pseudomonas fluorescens ATCC 12983 and panel 2f shows the evolution of LAB and E. faecium. For the challenge E. coli vs LAB, level of E. coli in milk dropped, after 48 h, below 10^5 cfu mL^-1, and was no longer detectable at 72 h, while in BHI the growth curve of E. coli was parallel to control, indicating an effect of pH rather than a direct activity of LAB and/or bacteriocins. A similar behaviour was shown by Salmonella Derby with an even faster drop (<10^5 cfu mL^-1) after 30 h. S. aureus proved to be more resilient and a substantial reduction in total cell numbers was observed.
viable cells was observed between 72 and 120 h (<10^5 cfu mL^-1 after 120 h) with a complete elimination after 168 hrs. *L. innocua* was no longer detectable in milk after 120 h, with a steady drop after 24 hrs (<10^4 cfu mL^-1 after 72 h). LAB had an effect on *L. innocua*, also in BHI, with levels below 10^6 cfu mL^-1 just after 30 h. The direct effect of certain LAB strains on *Listeria* spp. rather than the indirect effect of pH drop has been described (Winkelströter and De Martinis, 2015). *P. fluorescens* had a similar drop both in milk and in BHI and was no longer detectable after only 24 h, proving that the LAB formulation tested in this study is able to limit the growth of this typical spoilage microorganism even in absence of pH drop (Folawe Okorhi, 2014). Figure 2f shows the correct evolution of the LAB formulation and of the probiotic strain both in milk and in BHI. The probiotic strain of *E. faecium* showed a certain susceptibility to pH drop, in fact, after 48 h of incubation in milk as pure culture, a rapid descent was recorded and counts reached values below 10^6 cfu mL^-1 after 72 h and below 10^4 cfu mL^-1 after 120 h, when pH had reached levels below 5.5. A similar behaviour has been described in cheese (Reale et al., 2010).

**Conclusions**

There are certain criteria that lactic acid bacteria starter cultures should meet when being included in food products, such as to compete effectively with indigenous lactic acid bacteria, to produce adequate quantities of lactic acid, to grow in the presence of up to 6% NaCl, to tolerate NaN_2O_2 to grow between 10°C and 40°C, with an optimum between 30°C and 37°C, to be homo-fermentative and catalase-positive, to be non-proteolytic and to produce only very small quantities of H_2O_2 to reduce nitrate, to enhance flavour of the finished sausage, not to produce biogenic amines, not to produce slime, to be antagonistic to pathogenic and other undesirable micro-organisms and tolerant of, or synergistic with, other starter components. Various species have been, and are, used as inoculants in sausage fermentation (Campbell-Platt, 1987). The fermentation of carbohydrates leads to several desirable results: production of organic acids, which lead to the reduced pH value; production of favourable organoleptic compounds; coagulation of meat proteins, thereby decreasing the water holding capacity and thus, facilitating the drying process, which affects the texture and firmness of the end product, and red colour formation due to the reaction of nitrogen monoxide with the heme group in myoglobin at pH 5.4 to 5.5. The association of selected starter cultures and probiotics tested in this research was able to limit the growth of pathogens and hygiene indicators in an in-vitro setting. According to the strong inhibitory activity, our results show that the proposed formulation of LAB and probiotics, could be applied as food preservative and as starter cultures. The LAB application is wide-ranging and most promising if one considers that the indicator microorganisms used in this experiment were suppressed. These paper along with another study on the use of this formulation in salami production (Cenci-Goga et al., 2015) highlights the possibility that food safety and food quality can dramatically be improved by using novel formulations of LAB with antimicrobial activities as starter for food fermentation. It must be considered that in the production of traditional southern European style sausages, the fermentation profile must phase in order to ensure the growth of the added starter culture at the expense of the background flora. Additionally, the acidification profile must not reach values below pH 4.8-5.0 at any time. This will ensure that autochthonous microbiota maintain its activity over a longer period of time; foremost reductase and
flavour-forming activities (Lebert et al., 2007; Marco et al., 2008; Barbut, 2010; Reale et al., 2010). The starter cultures should be specifically selected for traditional fermentation profiles applying temperatures not higher than 24°C (Holley et al., 1988; Scannell et al., 2001; Cenci Goga et al., 2012). On the other hand, the production of north European and US style sausages the fermentation profile have a very short lag phase in order to rapidly show a fast drop in pH to below 5.3 within 30 hours as a minimum. This ensures an efficient inhibition of background flora and the production time is typically less than 2 weeks at the cost of a general lower consumer acceptability because the faster the acidification the less enzymes enhancing colour and aroma formation are produced (Marco et al., 2008; Barbut, 2010).

The proposed formulation is an additional tool in the production of low-acid fermented dry salami that are nowadays the preferred consumer’s choice given that the low speed of fermentation is a further asset because the acidity acidification of the less enzymes enhancing colour and aroma formation are produced (Marco et al., 2008; Barbut, 2010). Enterococci are, in fact, capable of modulating the aroma by means of the conversion of amino acids and free fatty acids (González-Fernández et al., 2006; Leroy et al., 2006; Corbiere Morot-Bizot et al., 2007; García Fontán et al., 2007).

References

Adams, M.R., Moss, M.O., 2000. Food microbiology. Royal Society of Chemistry Publ., Cambridge, UK.

Barbut S. 2010. Color development during natural fermentation and chemical acidification of salami-type products. J. Muscle Food. 21:499-508.

Campbell-Platt, G., 1987. Fermented foods of the world. Butterworth, London, UK.

Cenci Goga, B.T., Clementi, E., Di Antonio, E., 1995. Behaviour of lactic acid non lactic microflora during production and ripening of farm manufactured Pecorino cheese. Ann. Microbiol. 45:219-236.

Cenci Goga, B.T., Ortenzi, R., Bartocci, E., Codega de Oliveira, A., Clementi, E., Vizzani, A., 2005. Effect of the implementation of HACCP on the microbiological quality of meals at a university restaurant. Foodborne Pathog. Dis. 2:138-145.

Cenci Goga, B.T., Ranucci, D., Miraglia, D., Cioffi, A., 2008. Use of starter cultures of dairy origin in the production of Salame nostrano, an Italian dry-cured sausage. Meat Sci. 78:381-390.

Cenci Goga, B.T., Rossitto, P.V., Sechi, P., Parmegiani, S., Cambiotti, V., Cullor, J.S., 2012. Effect of selected dairy starter cultures on microbiological, chemical and sensory characteristics of swine and venison (Dama dama) nitrite-free dry-cured sausages. Meat Sci. 90:599-606.

Cenci-Goga, B.T., Karama, M., Sechi, P., Iulietto, M.F., Mattei, S., Novelli, S., 2015. Effect of a novel starter culture and specific ripening conditions on the characteristics of Italian dry salami. Meat Sci. (In press).

Chamba, J.F., Prost, F., 1989. Mesure de l’activité acidifiant de bactéries lactiques thermophiles utilisées pour la fabrication des fromages à pâte cuite. Lait 69:417-431.

Cizeikiene, D., Juodeikiene, G., Paskevicius, A., Bartkien, E., 2013. Antimicrobial activity of lactic acid bacteria against pathogenic and spoilage microorganism isolated from food and their control in wheat bread. Food Control 31:539-545.

Clementi, E., Cenci Goga, B.T., Trabalza-Marinucci, M., Di Antonio, E., 1998. Use of selected starter cultures in the production of farm manufactured goat cheese from thermized milk. Ital. J. Food Sci. 10:41-56.

Corbiere Morot-Bizot, S., Leroy, S., Talon, R., 2007. Monitoring of staphylococcal starters in two French processing plants manufacturing dry fermented sausages. J. Appl. Microbiol. 102:238-244.

Folawe Okorhi, B., 2014. Anti-pseudomonas activity of organic acids produced by lactic acid bacteria. Issues Biol. Sci. Pharm. Res. 2:106-114.

Garcia Fontán, M.C., Lorenzo, J.M., Parada, A., Franco, I., Carballo, J., 2007. Microbiological characteristics of “androlla”, a Spanish traditional pork sausage. Food Microbiol. 24:52-58.

Gonzalez-Fernandez, C., Santos, E.M., Rovira, J., Jaime, I., 2006. The effect of sugar concentration and starter culture on instrumental and sensory textural properties of chorizo-Spanish dry-cured sausage. Meat Sci. 74:467-473.

Holley, R.A., Lammerding, A.M., Tittiger, F., 1988. Occurrence and significance of streptococci in fermented Italian type dry sausage. Int. J. Food Microbiol. 7:63-72.

ISO, 1996. Microbiology of food and animal feeding stuffs. Horizontal method for detection and enumeration of Listeria monocytogenes. Norm ISO 11290-1:1996. International Organization for Standardization Publ., Geneva, Switzerland.

ISO, 2007. Microbiology of food and animal feeding stuffs. Horizontal method for the detection of Salmonella spp. Norm ISO 6579:2002/Amd 1:2007. International Organization for Standardization Publ., Geneva, Switzerland.

Lebert, I., Leroy, S., Giammarinaro, P., Lebert, A., Chacornac, J.P., Bover-Cid, S., Vidal-Barou, M.C., Talon, R., 2007. Diversity of microorganisms in the environment and dry fermented sausages of small traditional French processing units. Meat Sci. 76:112-122.

Ledda, A., Sciunto, M.F., Pirisi, A., Mannu, L., 1994. Caratterizzazione tecnologica di ceppi di lattococchi e di enterococchi per la produzione di formaggio pecorino fiore-sardo. Sci. Tecn. Latt. Cas. 45:443-456.

Leroy, F., Verluyten, J., De Vuyst, L., 2006. Functional meat starter cultures for improved sausage fermentation. Int. J. Food Microbiol. 106:270-285.

Marco, A., Navarro, J.L., Flores, M., 2008. The sensory quality of dry fermented sausages as affected by fermentation stage and curing agents. Eur. Food Res. Technol. 226:449-458.

Pique, D., Perret, B., Latrille, W., Corrigeu, G., 1992. Characterisation et classification de bactéries lactiques à partir de la mesure de leur cinétique d’acidification. Lebensm.-Wiss. Technol. 25:181-186.

Reale, S., Vitale, F., Scatassa, M.L., Caracappa, S., Currò, V., Todaro, M., 2007. Molecular characterization of dominant bacterial population in “Vastedda della Valle del Belice” cheese: preliminary investigation. Ital. J. Anim. Sci. 6(Suppl.1):595-597.

Scannell, A.G.M., Schwarz, G., Hill, C., Ross, R.P., Arendt, E.K., 2001. Pre-inoculation enrichment procedure enhances the performance of bacteriocinogenic Lactococcus lactis meat starter culture. Int. J. Food Microbiol. 64:151-159.

Winkelströter, L.K., De Martinis, E.C., 2015. In vitro protective effect of lactic acid bacteria on Listeria monocytogenes adhesion and invasion of Caco-2 cells. Beneficial Microbes 11:1-8.