

## Analysis of microbiological variation in PDO Vastedda della valle del Belice cheese during the storage period

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### Abstract

The PDO “Vastedda della valle del Belice” is a Sicilian *pasta filata* sheep cheese, made from raw milk without starter addition. It is a small round cheese without rind, weighing about 500-700 g. It is cheese is marketed also out of Sicily to allow its marketing and to prolong its shelf-life.

The aim of this work was to evaluate the variation of microbiological parameters during the shelf-life period. Then 162 Vastedda cheeses from 18 cheese-making processes in 7 farms have been analysed at different times of storage at 4°C (0, 15, 30, 45, 60, 75, 90, 105, 120 days).

Coliforms and *E. coli* were detected into 7/18 cheese-making processes and their count decreased during the storage period. Enterococci were more resistant to high temperature achieved during the stretching and their were rather stable during storage period ( $10^5$  cfu/g).

The concentration of total Bacterial Count and Mesophilic Lactococci were around  $10^7$  cfu/g, while the concentration of Thermophilic Lactococci was higher ( $10^8$  cfu/g) probably due to the stretching at 90°C.

In conclusion, this study shows the good sanitary conditions of cheeses. Overall, the lactic flora was kept alive and vital at high concentration ( $> 10^7$  cfu/g) until 120 days of storage.

Keywords: PDO Vastedda della valle del Belice cheese, microbiological variation, shelf-life.

### Introduction

The PDO “Vastedda della valle del Belice” cheese is an unripened cheese obtained from raw ovine milk of the Valle del Belice breed ewes reared in the Belice area. This historical cheese is the only *pasta filata* cheese produced in Italy with raw sheep milk. The metabolic activity of the lactic microflora in the raw milk determines the dropping of pH in the curd, that happens after 6-48 h of curd maturation according to different environmental temperatures. Near to pH 5.5-5.2, the curd is stretched using water or whey at 90-95°C and the shape is due to ceramics plates. It is a small round cheese without rind, weighing about 500-700 g. The PDO “Vastedda della valle del Belice” is marketed also out of Sicily, so to allow its marketing and to prolong its shelf-life, the cheese after 24-36 h of brine salting is sealed under vacuum with water vapour impermeable plastic film and stored at 4°C up to 90 days. The microbiological content of the PDO “Vastedda della valle del Belice” was investigated by Reale et al. (2007) and Scatassa et al. (2007). Authors reported the dominant populations of microflora present in the Vastedda cheese and their variations during the cheese-making process. No data are available on microbiological variations during the storage until the sale; the aim of this work was to evaluate this variations during the shelf-life period.

### Material and methods

A total of 162 Vastedda cheeses from 18 cheese-making batches in 7 farms were analysed at different times of storage at 4°C (0, 15, 30, 45, 60, 75, 90, 105, 120 days).

The samples were subjected to microbiological analyses using the following procedures: Coagulase-positive staphylococci (ISO 6888-1:1999/A1:2003 and ISO 6888-2:1999/A1:2003 and confirmation with API Staph); detection of Staphylococcus enterotoxin according to ELFA: VIDAS (Staph enterotoxin II e SET-RPLA (Oxoid) / ELISA test - Diffchamb); Enumeration of microorganisms at 30°C (ISO 4833:2003); Coliforms (ISO 4832:1991); *Escherichia coli* (ISO 16649-2:2001); Enterococci on Rapid *Enterococcus* Agar (REA) incubated at 44°C for 48 h, whose suspect colonies were tested for confirmation and biochemical identification (API 20Strep); Sulphite reducing bacteria growing under anaerobic conditions (ISO 15213:2003); *Salmonella spp.* (ISO 6579:2002); *Listeria monocytogenes* (ISO 11290-1:1996); mesophilic and thermophilic Lactococci on M17 medium, incubated at 22°C for 48 h and at 44°C for 72 h respectively; *Lactobacilli* on acidified MRS medium and incubated under micro-aerobic conditions (5% CO<sub>2</sub>) at 37°C for 72 h. For statistical analysis, a two factors ANOVA model was utilised to estimate the means relative to each class of storage period, moreover least square means were plotted to observe the variations of microflora. Data were statistically analysed by GLM procedure of the SAS 9.1.2 software (2004).

### Results and discussion

Coliforms and *E. coli* were observed in 7/18 cheese-making batches and they decreased during the storage period (Fig. 1). Only in two cheese-making batches, *E. coli* presented concentrations between  $10^5$  and  $10^6$  cfu/g. *Enterococci* were more resistant to the high temperature achieved during the stretching and their counts were

rather stable during the storage period ( $10^5$  cfu/g). Coagulase-positive staphylococci showed a decrease until 60-75 days to lightly go up later (Fig. 1).

The concentrations of Total Bacterial Count and Mesophilic *Lactococci* were around  $10^7$  cfu/g, while the concentration of Thermophilic Lactococci was higher ( $10^8$  cfu/g). This is probably due to bacterial selection during the cheese-making process of Vastedda and in particular during the stretching that uses hot water at 90°C. Counts of CBT remained constant such as mesophilic *Lactobacilli*, while Lactococci (LATTOCM and LATTOCT) showed a rapid decline of their concentration during storage (Fig. 2).

Otherwise, no pathogenic organisms (including sulfite reducing Clostridia, *Listeria monocytogenes* and *Salmonella*) were observed in the samples analysed. *S. aureus* was found in 5/18 cheese-making batches with maximum concentration around to  $10^5$  cfu/g but no enterotoxaemia activity was found.

Figure 1: Variations of Coliforms (COLIFOR), *E. coli* (ECOLI), Coagulase-positive staphylococci (STAFF) and Enterococci bacterial (ENTERO) during the shelf life period

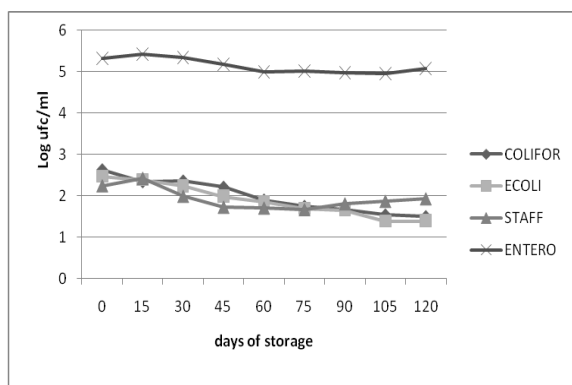
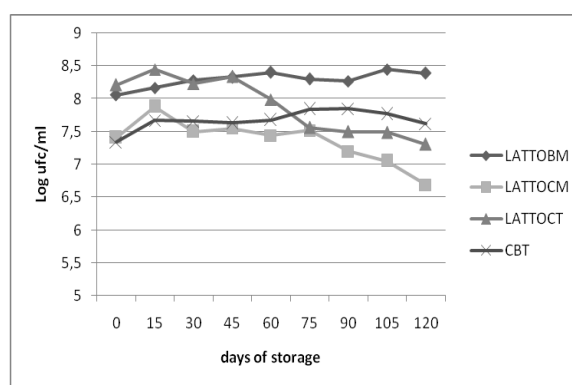


Figure 2: Variations of Mesophilic Lactobacilli (LATTOBM), Mesophilic Lactococci (LATTOCM), Thermophilic Lactococci (LATTOCT) and Total Bacterial Count (CBT) during the shelf life period



## Conclusions

In conclusion this study has evidenced the good sanitary conditions of the PDO “Vastedda della Valle del Belice” cheese, without the presence of pathogenic bacteria. Overall, the non lactic microflora decreased during the storage ( $< 10^2$  cfu/g) with the exception of *Enterococci*, that remain around  $10^5$  cfu/g; the lactic flora was kept alive and vital at high concentration ( $> 10^7$  cfu/g) until 120 days of storage.

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