Chapter

MONITORING OF THE REGULATORY CRITERIA FOR RAW MILK COLLECTED FROM DAIRY FARMS IN CENTRAL ITALY

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ABSTRACT

The milk from dairy animals has a good nutritional value because it represents an indispensable source of high quality proteins. Moreover, it is the raw material for many dairy products consumed worldwide such as fresh and ripened cheeses, butter, cream and fermented derivatives like yoghurt, kefir, koumiss, etc. According to the European legislation (Regulation EC No 853/2004), the food business operators collecting raw milk intended to the production of milk and dairy products must ensure compliance with some health requirements of the animals. More in detail, the animals must not show any symptoms of infectious diseases for humans, or signs of diseases of the udder or the genital tract that could

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contaminate milk. Furthermore, they must belong to a holding free or officially free of tuberculosis and brucellosis and no unauthorized substances or authorized drugs without respect of the withdrawal period must have been administered.

In addition to these requirements, raw milk coming from cows must meet some criteria with regards to the plate count at 30°C (ufc/ml) and somatic cell count (cells/ml). The bacterial contamination of milk can adversely affect its quality and safety and high values of somatic cell count can be considered an important indicator of mastitis. The results for these two regulatory parameters, i.e., the plate count at 30°C and somatic cell count, must be expressed as a rolling geometric average over a two- or three-month period, with at least two samples or one per month, respectively.

The aim of this study was the evaluation of compliance with the mentioned criteria in milk samples collected from 100 different dairy farms located in Central Italy. The plate count at 30°C was exceeded in 12 dairy farms while the somatic cell count just only in 2. These results demonstrated a good management of hygiene practices during milking and collection of raw milk. This approach could include some important elements such as the definition of primary udder health parameters, the detection of cows causing the problem and the implementation of a good herd management plan.

**INTRODUCTION**

Milk is the product of the mammary gland of many different animals such as cows, buffaloes, sheep, goats and donkeys. It is virtually sterile when secreted into the alveoli of the udder, but it can be contaminated during milking and/or handling with personnel, equipment and environmental sources (Sarkar, 2015). Due to its high nutrient content and low acidity, it represents an excellent growth medium for different bacteria, which can multiply based on temperature, other competing microorganisms and their metabolic products. Therefore, it is a very perishable commodity (Claeys et al., 2013; Ritota et al., 2017).

The microbiological quality of milk can be affected by the health status of dairy animals. In fact, they can show an inflammation of the mammary gland, as clinical or subclinical mastitis, determining the alteration of milk composition (Yu et al., 2017). Besides an animal welfare problem, mastitis
can be considered also a food safety concern as well as one of the biggest economic damage sources for the herd management. It can be caused by a wide spectrum of pathogens (i.e., *Staphylococcus aureus*, *Streptococcus agalactiae*, *Corynebacterium bovis*, *Mycoplasma* spp.) spreading from animal to animal, primarily during milking. Their presence tend to result in chronic infections with flare-ups of clinical events (Abebe et al., 2016). This disease can be classified as clinical or subclinical. The usual signs of inflammation are present in the first form, characterized by red, hot and swollen mammary glands, while the subclinical mastitis is rarely diagnosed because no visible signs are seen, but generally milk production decreases and the somatic cell count (SCC) increases (Khan and Khan, 2006). The milk somatic cells include 75% leukocytes (i.e., neutrophils, macrophages, lymphocytes and erythrocytes), produced by the animals’ immune system to fight the inflammation of the mammary gland, and 25% epithelial cells. The white blood cells can increase up to 99% in animals with mastitis (EFSA, 2015). The main factors affecting the SCC other than mastitis can be: i) stage of lactation; ii) age and breed of dairy animals; iii) season, stress and diurnal variation (Sharma et al., 2011). The increase of the SCC can cause the reduction in casein, fat and lactose of milk, affecting consequently the quality and yield of the derived dairy products (Malek dos Reis et al., 2013).

Another important indicator of milk quality is the total aerobic mesophilic bacteria count. It can be increased by several conditions, such as unhygienic practices during milking, dirty cow udders, teat injuries due to inadequate stall or platform design and late or insufficient milk cooling (Ürkek et al., 2017).

The European legislation (Regulation EC No 853/2004) set the criteria for raw cows’ milk as follows:

- plate count at 30°C (ufc/ml) ≤ 100 000
- somatic cell count (cells/ml) ≤ 400 000

The value of the plate count at 30°C must be ≤ 1 500 000 ufc/ml for raw milk collected from other species, but if it is intended for the manufacture of
products made by a process that does not involve any heat treatment, this value must be \( \leq 500,000 \) ufc/ml.

In addition, the plate count at 30°C must be calculated as rolling geometric average over a two-month period, with at least two samples per month, whereas for the SCC the rolling geometric average over a three-month period, with at least one sample per month, must be considered. The geometric mean calculation generally produces a value somewhat less than the arithmetic mean for the same data set. This method has been chosen because a single high count in a data set has a greater impact on the arithmetic mean than the geometric mean. Moreover, only one very high value will not trigger regulatory action when using the geometric mean procedure (Sharma et al., 2011).

The aim of this paper was the evaluation of compliance with the regulatory criteria in cows’ raw milk samples collected from 100 different dairy farms, located in 5 regions of Central Italy during the year 2017.

**MATERIALS AND METHODS**

Raw milk samples were collected from 100 different dairy farms, located in the following regions of Central Italy: Abruzzo, Lazio, Marche, Molise, Umbria (Figure 1). The sampling was carried out during the first two quarters (from January to March and from April to June) of the year 2017. More in detail, the samples were collected twice a month and calculated as rolling geometric average over a two-month period for the plate count at 30°C, for a total of 1,200 samples in the first two quarters of the year, whereas they were collected one sample per month and calculated as rolling geometric average over a three-month period for the SCC, for a total of 600 samples.

All farms used automatic milking. The samples were put into 10 ml sterile tubes and immersed in icebox until arrival at the laboratory.

The analyses were performed according to Regulation EC No 2074/2005 following the reference methods: EN/ISO 4833 for the plate count at 30°C and ISO 13366-1 for the SCC.
The statistical analysis of the results was carried out using statistical software program (GraphPad Software Version 3.0 (Inc, San Diego, CA, USA). The ANOVA test was used to evaluate the effect of the collecting period.

Figure 1. Map of the sampling sites located in 5 regions of Central Italy.
RESULTS AND DISCUSSION

The data reported in this study resumed the rolling geometric average of the plate count at 30°C and the SCC in cows’ raw milk samples collected from a total of 100 dairy farms, located in 5 regions of Central Italy. The results showed a general good hygienic condition of all dairy farms, indicating a suitable management system. The regulatory limits were exceeded in 12% of farms, independently from the investigated quarter of the year. The maximum value reached about 300 000 ufc/ml (Figure 2). Regarding to the SCC, only 2% of dairy farms exceeded the limit of 400 000 cells/ml up to about 700 000 cells/ml (Figure 3). The statistical analysis showed a significant difference ($p < 0.01$) between the two examined quarters of 2017 from all regions for the plate count at 30°C, and only between Abruzzo and Marche regions for the SCC.

![Figure 2. The plate count at 30°C related to the investigated dairy farms in the first two quarters of the year 2017.](image-url)
This variability could be due to the different management of the investigated dairy farms, with regards above all to milking hygiene, such as wearing of gloves during milking and adopting a post-milking teat disinfection, but also to other measures referred to the treatment of mastitis during non-lactating period and the implementation of regular mastitis prevention (Abebe et al., 2016). The increase of the plate count at 30°C could be related to mastitis, environmental microorganisms, dirty milking equipment or failure of refrigeration (Kelly et al., 2009). The spread of microorganisms among cows can occur through vectors, such as milking machines, human hands or flies (Vanderhaeghen et al., 2015). Moreover, an indirect contamination may arise from the faecal matter of the individual itself, or other cows contaminating the udder and teats (EFSA, 2015). Also milk storage temperature and length, season of production, herd sizes, farm geographical location and hygiene practices can affect the microbial milk quality (Ramsahoi et al., 2011).
The microbial contamination of cows’ udder and teats can be associated with the skin of animals, as well as the environment in which cows are housed and milked. So, pre-milking udder hygiene techniques could affect the plate count at 30°C of milk and a good cleaning of the teat with the sanitizing solution followed by drying with a clean towel is effective in reducing this value. In addition, the stress on the cow’s teat produced by the action of milking machine can open the teat canal allowing the entry of bacteria capable of infecting the mammary glands (Ledenbach and Marshall, 2009). Also, the cleaning of milking apparatus can influence the total bacteria count. For instance, milk residue left on the contact surfaces of used equipment can favor the growth of microorganisms.

Other authors (Bolzoni et al., 2015) reported that 29% out of 5 200 dairy farms exceeded the SCC limit one or more times a year, but they returned under this limit within 3 months; another 27% among the positive dairy farms remained non-compliant after this period. Bogdanovičová et al. (2016) evaluated the microbiological quality of cow’s, goat’s and sheep’s milk in the Czech Republic collected from 41 dairy farms. The plate count at 30°C exceeded the regulatory limits in 13% of the samples, whereas the highest SCC value in cow’s milk was 990 000 cells/ml.

According to Regulation EC No 853/2004, when raw milk fails to meet the legislative criteria, the food business operator must inform the competent authority and take measures to correct this situation. If these parameters do not return to be regular within the following three months, the delivery of raw milk from the production dairy farm will be suspended or subjected to requirements concerning its treatment and use necessary to protect public health (Regulation EC No 854/2004). Therefore, milk collection from the positive dairy farms of this study was excluded until these criteria returned to be regular. In that occasion, the farmers improved milking hygiene and in the meantime, they resorted to veterinary care for all the suspected ill cows.

**CONCLUSION**
The microbiological load of raw milk produced at dairy farm level can affect quality, shelf life and safety not only of this product itself, but also of the derived dairy products. The exceeding of the regulatory criteria for raw milk generally depends on animal’s health status with regards to the SCC, even if the farmers are not able to recognize animals with mastitis. Instead, the high values of the plate count at 30°C can reflect poor hygienic conditions as a consequence of an improper management system. The application of sanitary practices on dairy farm, such as teat disinfection pre- and post-milking, correct handling, collection and storage of milk and staff hygiene represent important strategies to reduce the contamination of this commodity. In addition, the implementation of standard mastitis prevention and/or control programs can result in the reduction of prevalence and incidence of this disease in cows’ herds.

REFERENCES


P. K.