

## Preventing Pathogenic Bacteria in Milk and in Dairy Farms: The Usefulness of Molecular Biology Tools

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### Abstract

Milk and dairy products have an important role in human nutrition, especially in the first years of life, once these products are sources of high quality protein, carbohydrates, fat and mineral salts necessary for growth. Therefore, it is essential to ensure the integrity and intrinsic quality of milk and dairy products for human consumption. Raw milk represents an ideal growth medium for several microorganisms that have been identified as food borne pathogens or as causative agents of bovine mastitis, such as *Staphylococcus aureus*, *Escherichia coli* and *Listeria monocytogenes*. In dairy cattle, the inflammation of the mammary gland in response to bacterial attachment and growth is an important cause of milk contamination. Contaminated milking equipment and the hands of the milkers are also common sources of transmission of pathogenic bacteria in the dairy farm environment, leading to potential health risks. Thus, under an epidemiological point of view, it is essential to use analytical methodologies to identify not only the pathogenic microorganisms in dairy farms, but also the respective sources of contamination in the milking environment. In dairy and other food industries, the determination of pathogenic micro-organisms has been performed routinely by molecular techniques, mainly the polymerase chain reaction (PCR), in addition to automated PCR systems such as BAXTM [1]. These methods are very useful for the identification of bacterial species, but they do not differentiate the lineages of bacteria from the same species, which is required in the case of epidemiological investigations. DNA-based methods are generally recommended for typing of pathogens such as *L. monocytogenes* and *S. aureus*, since they make possible to evaluate genetic relationships of strains from different sources, thus serving as guidance for the bacterial control in the environment. Although DNA sequencing is the most accurate method to evaluate genetic relationships between microorganisms, this technique is limited to specialized laboratories; therefore its application in the routine analysis in the food industry is still expensive and not practical. As an alternative to sequencing methods, several molecular techniques have been used to differentiate lineages of pathogens, especially the rDNA typing and the Pulsed-Field Gel Electrophoresis (PFGE). PFGE is considered the most effective technique for typing because of its simplicity, relative

low cost, time efficiency and high discriminatory power, depending on the restriction enzymes employed. In recent years, the application of molecular techniques in dairy farms has markedly increased, thus providing several data on the incidence of pathogen isolates found in milkers, equipment, individual cow's milk and bulk tank milk that were epidemiologically related. Those reports indicated that *S. aureus* and other pathogenic bacteria may not only be transmitted between cows in the same farm, but also disseminated among farms and different, unrelated regions. In a study conducted in Brazil, PFGE was used as a tool for identifying the diversity of *S. aureus* strains among different sites inside the dairy farms, and how they contribute for contamination of the bulk milk [2]. PFGE identification of clusters and pulsotypes at different periods of sampling shows the persistence of strains in the environment. Therefore, the use of molecular identification of pathogens in the milking environment could be useful for addressing specific control strategies, including regular monitoring of the sanitizing procedures in the milking environment of dairy farms. In conclusion, although PFGE would be useful to screen for pathogenic bacteria in milk and in the milking environment, further studies are needed to determine if the cost-benefit relationship of such technique would be appropriate for dairy operations worldwide. In addition, future research should focus on the development of other molecular techniques that could be applicable for rapid, robust analysis of microorganisms in the environment of dairy farms, an area that currently seems to be a priority in the broad domain of the dairy science research..

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