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Guidelines for Improving the Performance of PCR Assays for Routine Diagnosis of Bovine Mastitis

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The application of DNA-based molecular diagnostic such as PCR assay are increasingly implemented as a routine method for mastitis control programs in the last few years. Unlike traditional methods, they provides quick, quantitative and offer large-scale routine mastitis diagnosis. In addition, they have important role in identification of udder pathogens at the subspecies level for epidemiological studies on animal and herd levels [1,2]. Most of research studies confirmed the higher performance, sensitivity and specificity of PCR assays over the traditional methods for mastitis pathogens detection. Therefore, increasing the application of PCR assays in udder health management is a crucial necessity for improving the efficiency of mastitis prevention and control programs. On the other hand, bacterial culture is the most common diagnostic method for isolation and identification of mastitis pathogens. Bacterial culture has been regarded as the gold standard for mastitis diagnosis by most conservative laboratories worldwide. The bacterial culture is widely used for many purposes such as (1) specific control programs or for surveillance to detect the presence of new or emerging udder pathogens, (2) evaluate treatment of mastitis efficacy and (3) establish susceptibility patterns to aid in the development of rational treatment strategies [3,4].

Bacterial culture and PCR assays are commonly used for routine diagnosis of mastitis in dairy herds however; there are some neglected or vague key difference points between the precautions of application of these methods and their results interpretation. In routine sampling procedures for bacterial culture, the milk samples must be collected aseptically following the National Mastitis Council guidelines [5]. Such procedures are necessary to ensure that the isolated and identified pathogen on the culture plate is definitely from inside the mammary glands and therefore, it accurately reflect the intramammary microbiological condition. On the other hand, there is no specific guidelines or recommendations for sample collection for PCR assays especially at routine milk recordings (DHI).

Therefore, the results of the PCR assay could be affected resulting in false positive results from bacterial pathogens colonizing and/or contaminate the teat apex [6] and carryover of bacterial DNA [7,8] and subsequently misdiagnosis.

For traditional microbiological assays such as bacterial culture, the National Mastitis Council has set up a recommendations and guidelines for accurate identification of mastitis causing pathogens including criteria for definition of contamination [5]. Such criteria defined as growth of three or more different colony species on the culture plate regarded as contamination and must be excluded from the analysis and resampling is required. As for PCR assay, there are no available guidelines or recommendations for the accurate definition of the contamination based on results of PCR assay used for identification the udder pathogens at routine milk recording. Consequently, the clinical relevance of some PCR results can be questionable. Hence, many recent studies concluded that results from any microbiological analysis in general and PCR assay on particular should always be interpreted with all available information, such as clinical signs and history of the cows, SCC information, California mastitis test and milking order of the cows [8-10]. Using such information will definitely, improve the decision-making for antimicrobial treatment and culling purposes.

In conclusion, DNA-based molecular diagnostic are a promising tools for mastitis diagnosis and control in dairy herds. PCR assay provide quick mastitis diagnosis however, it is necessary to combine PCR results with other available information such as clinical history of the cow, milking order and SCC to improve the overall confidence about the presence or absence of particular organisms and their impact on mammary health. Therefore, accurate interpretation of PCR results requires establishing (1) specific guidelines/precautions for collection of milk samples, and (2) specific guidelines for defining the contamination of milk samples and, their type and sources at routine milk recordings to avoid the misidentification of intramammary infections and subsequently, misinterpretation of the results.

Bibliography

1. Mweu MM., *et al*. "Annual incidence, prevalence and transmission characteristics of Streptococcus agalactiae in Danish dairy herds". *Preventive Veterinary Medicine* 106.3-4 (2012): 244-250.

2. Mahmmod YS., *et al*. "Molecular epidemiology and strain-specific characteristics of Streptococcus agalactiae at the herd and cow level". *Journal of Dairy Science* 98.10 (2015): 6913-6924.

3. Ruegg PL. "Milk Quality and Mastitis Tests" (2002): 1-33.

4. Persson Waller K. "Microbiological diagnostics of udder infections" The 29th NKVet Symposium "New knowledge on diagnostics and control on modern dairy farms" (2013).

5. NMC. "Laboratory Handbook on Bovine Mastitis". National Mastitis Council, Madison, WI (1999).

6. Mahmmod YS., *et al*. "Effect of presampling procedures on real-time PCR used for diagnosis of intramammary infections with Staphylococcus aureus in dairy cows at routine milk recordings". *Journal of Dairy Science* 96.4 (2013): 2226-2233.

7. Mahmmod YS., *et al*. "Effect of carryover and presampling procedures on the results of real-time PCR used for diagnosis of bovine intramammary infections with Streptococcus agalactiae at routine milk recordings". *Preventive Veterinary Medicine* 113.4 (2014): 512-521.

8. Mahmmod YS., *et al*. "DNA carryover in milk samples from routine milk recording used for PCR-based diagnosis of bovine Staphylococcus aureus mastitis". *Journal of Dairy Science* 100.7 (2017): 5709-5716.

9. Keane OM., *et al*. "Increased detection of mastitis pathogens by real-time PCR compared to bacterial culture". *Veterinary Record* 173.11 (2013): 268.

10. Steele NM., *et al*. "Using polymerase chain reaction to identify Streptococcus uberis in bovine milk: How does it compare with bacterial culture?". *Proceedings of the New Zealand Society of Animal Production* 75 (2015): 20-23.

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