

Real-Time PCR in Mastitis Diagnostics

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Introduction Mastitis remains the most frequent and costly disease of dairy cattle. Accurate, rapid etiological diagnosis is essential for targeted therapy, antimicrobial stewardship, and herd level control. Since June 2010, Finland has implemented routine real-time multiplex PCR (qPCR) for mastitis diagnostics at national scale, replacing most bacteriological culture (BC) and generating a unique, longitudinal dataset to inform udder-health management. This large-scale adoption provides a unique basis for combining diagnostic, milk quality, and antimicrobial use data to evaluate udder health and management outcomes at population level after the radical change.

Material & methods We summarize operational experience from Finland's centralized diagnostic workflow involving Valio Oy's laboratory network, Movet IDEXX, and the University of Helsinki, which collectively process over 130,000 mastitis PCR samples annually, compared with approximately 2,000 bacterial culture (BC) samples analyzed mainly at the University of Helsinki and Movet. Samples include aseptically collected quarter milk samples from clinical and subclinical mastitis cases, taken predominantly by farmers under veterinary guidance. In addition, national Dairy Herd Improvement (DHI) data from over 180,000 cows are analyzed regularly for somatic cell count (SCC), allowing integration of herd-level inflammation and infection trends with PCR pathogen profiles. PCR analyses employ commercial qPCR panels (15 targets including β -lactamase resistance gene). All laboratories follow harmonized procedures for aseptic sampling, DNA extraction, amplification, and automated Ct-value based result interpretation. Antimicrobial use data originate from FINRES-Vet national surveillance, enabling linkage of diagnostic adoption with trends in veterinary antimicrobial consumption.

Results & Discussion Annual qPCR throughput exceeds 130,000 samples, demonstrating the national scale and robustness of Finland's udder-health surveillance system. The median turnaround time decreased from ≥ 48 h for BC (and 6–10 days for *Mycoplasma bovis*) to same-day reporting (~ 4 h analytical time). Across multiple Finnish and international studies, qPCR consistently detected mastitis pathogens more frequently than BC and achieved pathogen detection rates of 83–92% in routine diagnostics. Sample quality was acceptable at large scale: $\sim 13\%$ classified as contaminated (> 2 targets), and 12–18% PCR-negative, indicating reliable farmer sampling. National DHI SCC data show a gradual decline in herd-average SCC over the past decade, corresponding with enhanced diagnostic coverage and targeted herd management. Importantly, antimicrobial usage in Finnish dairy herds has decreased steadily since 2010, contrary to initial concerns that PCR adoption would increase prescriptions. According to FINRES-Vet 2022, both overall antimicrobial consumption and resistance indicators declined, supporting improved stewardship outcomes. **Conclusion** Finland's nationwide qPCR implementation demonstrates that molecular diagnostics can be successfully integrated into large-scale udder-health management without increasing antimicrobial use. The key lessons include:

1. Embedding diagnostics within herd-health strategy - clear sampling indications, guidelines and aseptic quarter-based sampling technique.
2. Combining molecular and production data - integration of PCR results with DHI SCC data enables herd-specific pathogen profiling and longitudinal follow up of the cows and benchmarking.
3. Interpreting Ct-values contextually - Ct data should be routinely reported and evaluated alongside the pathogen information, SCC and clinical findings.
4. Monitoring stewardship metrics - routine PCR use facilitates evidence-based antimicrobial decision-making, reducing unnecessary treatments. Finland's experience shows that qPCR, when combined with systematic SCC monitoring and national antimicrobial surveillance, forms a powerful, objective, and data-driven foundation for precision mastitis management. The Finnish model provides an international benchmark for linking diagnostics, prevention, and responsible medicine use in sustainable dairy production.