Short time variation in daily shedding of contagious mastitis pathogens

Line Svennesen¹, Torben W. Bennedsgaard², Karl Pedersen³ and Ilka C. Klaas⁴

¹Department of Large Animal Sciences, University of Copenhagen, Frederiksberg, Denmark
²Dyr lægerne Himmerland Kvæg, Aars, Denmark
³National Veterinary Institute, Technical University of Denmark, Copenhagen, Denmark

With rapidly increasing herd sizes, focus on effective control of contagious mastitis pathogens as Staphylococcus aureus and Streptococcus agalactiae becomes important. Real-time PCR tests may be efficient diagnostic tools for early and accurate diagnosis of contagious mastitis. Inconsistent shedding based on bacteriological culture (BC) is reported for Staph. aureus and Strep. agalactiae, but knowledge about shedding patterns based on PCR is sparse. Our objective was to evaluate the daily shedding pattern of Staph. aureus and Strep. agalactiae determined with PCR test (Mastit4, DNA diagnostic), bacteriological culture (BC) and somatic cell count (SCC).

In two dairy herds with a history of Staph. aureus and Strep. agalactiae (PCR Ct-value in bulk tank milk ≤ 32), 589 cows were screened for Staph. aureus and Strep. agalactiae. Aseptic composite foremilk samples were collected in bronopol preserved vials for PCR test. Aseptic quarter foremilk samples were then collected daily from cows positive for Staph. aureus (Ct ≤ 37) or Strep. agalactiae (Ct < 40) at screening (n = 43 quarters). All samples were taken after routine preparation of the udder by the milking staff, following NMC recommendations. Quarters that were positive at screening but negative the following 8 days were omitted (n = 8). Each sample was split into; a bronopol preserved sample for PCR test, a bronopol preserved sample for SCC, and a non-preserved sample for BC. After BC was carried out, the non-preserved sample was frozen and later on used for a second PCR test round. PCR test and SCC were carried out by a commercial lab (Eurofins, Vejen) and BC was carried out by the first author following NMC recommendations. The approximate CFU count (up to 300 CFU/10 µL) was determined.

Preliminary results including BC, SCC and PCR results from the second test round show different types of shedding patterns with large variation. Twenty-one Staph. aureus infected quarters and 14 Strep. agalactiae infected quarters were followed, with a median Ct-value within quarter of 27 (range 20-40) and 22 (range 14-40), respectively. The mean number of PCR positive days (Ct < 40) was 18 for Staph. aureus and 15 for Strep. agalactiae. The mean number of BC positive days (CFU ≥ 1 per 10 µL) was 19 for Staph. aureus and 10 for Strep. agalactiae. There was no visual correlation between SCC and Ct-values.

In conclusion, we see inconsistent shedding patterns in both Strep. agalactiae and Staph. aureus infected quarters, determined with either BC or PCR test. For inconsistently shedding quarters, repeated sampling may be recommended. There appears to be no difference between BC and PCR test for Staph. aureus infected quarters (mainly consistent shedding). However, in Strep. agalactiae infected quarters there appears to be a larger change of detection with the PCR test compared to BC, based on the higher number of test positive days. Further analysis will be carried out in 2017.

The project is funded by the Danish Milk Levy Fund.