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Enumeration of clostridia in goat milk using an optimized membrane filtration technique

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Abstract

A membrane filtration technique developed for counting butyric acid bacteria in cow milk was further developed for analysis of goat milk. Reduction of the sample volume, prolongation of incubation time after addition of proteolytic enzyme and detergent, and a novel step of ultrasonic treatment during incubation allowed filtration of goat milk even in the case of somatic cell counts (SCC) exceeding 10^6 /mL. However, filterability was impaired in milk from goats in late lactation. In total, spore counts were assessed in 329 farm bulk goat milk samples. Membrane filtration technique counts were lower than numbers revealed by the classic most probable number technique. Thus, method-specific thresholds for milk to evaluate the risk of late blowing have to be set. As expected, the spore counts of milk samples from suppliers not feeding silage were significantly lower than the spore counts of milk samples from suppliers using silage feeds. Not only were counts different, the clostridial spore population also varied significantly. By using 16S

rRNA gene PCR and gene sequencing, 342 strains from 15 clostridial species were identified. The most common *Clostridium* species were *Clostridium tyrobutyricum* (40.4%), *Clostridium sporogenes* (38.3%), *Clostridium bifermentans* (7.6%), and *Clostridium perfringens* (5.3%). The 2 most frequently occurring species *C. tyrobutyricum* and *C. sporogenes* accounted for 84.7% of the isolates derived from samples of suppliers feeding silage (n = 288). In contrast, in samples from suppliers without silage feeding (n = 55), these species were detected in only 45.5% of the isolates.



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Key words

clostridia; butyric acid bacteria; goat milk; membrane filtration; PCR

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