SHORT COMMUNICATION

Mycobacterium bovis detection from milk of skin test negative cows

C. F. O. Zarden, C. D. Marassi, E. E. E. S. Figueiredo, W. Lilenbaum

Context
The purpose of this study was to investigate the presence of Mycobacterium bovis in milk of single intradermal test (SIT)-negative dairy cows during an outbreak of bovine tuberculosis (TB).

Main conclusion
During an outbreak of bovine TB, intradermal tuberculin tests were not sufficient for the identification of all infected cows. Additional tests, such as PCR of milk collected from SIT-negative cows, could reduce spread of M bovis to human beings ingesting raw contaminated milk, or to other cattle.

Approach
After a 90 day interval period and the slaughtering of SIT-positive cows, all of the herd was tested by SIT. Eight SIT-negative cows were lactating. Milk samples from these cows were tested by a multiplex-PCR (m-PCR) that used two sets of primers simultaneously: the RvD1Rv2031c (500 bp) specific for M bovis and IS6110 (245 bp) sequence present in all members of Mycobacterium tuberculosis complex (MTC). Primers used were: INS1 (5’-GTGAGGGCATCGAGGTGGC-3’) and INS2 (5’-GCCGTAGGGCTCCTGAGACAAA-3’) for MTC and JB21 (5’-TCGTCGCTGATGCAAGTGC-3’) and JB22 (5’-CGTCGGTCGCTCAAGAAAG-3’) for M bovis.

Results
Using SIT, 17 (22.1 per cent) cows were positive, whereas seven had inconclusive results (9.1 per cent). Five (62.5 per cent) of eight milk samples collected from negative-SIT cows were positive to M bovis; three by PCR, one by culture and one by both tests. Positive-SIT cows were immediately slaughtered and milk samples were not collected.

Significance of findings
The results suggest the need for a more comprehensive diagnostic approach in an outbreak situation, where SIT-negative cows could be infected and remaining in the herd, contributing to the maintenance of the disease.
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Veterinary Record 2013 172: 130 originally published online January 3, 2013
doi: 10.1136/vr.101054

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