Study of *Mycobacterium bovis* genotypes in human and bovine isolates using spoligotyping, MIRUVNTR and RFLP-PCR

Pooyan Khaleghian a,b,*, Parisa Farnia a, Keivan Tadayon b, Nader Mosavari b, Mohadeseh Mozafari a, Zahra Derakhshani Nejad a, Rouollah Keshaavaz b, Shojaat Dashtipour b, Mohamad Reza Masjedi a, Ali Akbar Vellayati a, Rainak Ghaderi b, Samed Boroumandfar c

a Mycobacteriology Research Center, NRITLD, Masih Daneshvari Hospital, Shahid Beheshti University of Medical Science, Tehran, Iran
b TB Department, Razi Vaccine & Serum Research Institute, Karaj, Iran
c Office of Health and Animal Diseases Management, Iranian Veterinary Organization, Tehran, Iran

**Abstract**

**Aims and objectives:** The aim of this study is to investigate and detect the prevalence of *Mycobacterium bovis* subtypes (*Mycobacterium bovis* subtype *bovis* and *Mycobacterium bovis* subtype *caprae*) in humans and compare the genetic diversity of *Mycobacterium bovis* in humans and cattle with spoligotyping methods, as well as pyrazinamide susceptibility study of subtypes.

**Methods:** Examining these subtypes with molecular epidemiology techniques is particularly important due to different treatment of *M. bovis* diverse subtypes in humans. Culture tests were performed on clinical samples that were isolated from Lowenstein-Jensen culture medium. Identification tests were performed to differentiate *Mycobacterium bovis* from *Mycobacterium tuberculosis*. DNA was extracted and spoligotyping (spacer oligonucleotide typing) was performed using the DRb and DRA primers.

**Results:** The results were analyzed with the SPOLDB4 site. PCR-RFLP method of pncA gene was used to evaluate the resistance to pyrazinamide and pncA gene polymorphism.

**Conclusions:** *Mycobacterium bovis* subtype *bovis* in the Iranian population was reported with a frequency of 0.6% which was below the average of the previous reviews. In this study, all the strains were *M. bovis* subtype *bovis* resistant to pyrazinamide. No *Mycobacterium bovis* subtype *caprae* was detected. The only shared ST between humans and cattle was ST694. *Mycobacterium bovis* subtype *bovis* with ST 595 was reported as human bovis index.

© 2014 Asian-African Society for Mycobacteriology. Published by Elsevier Ltd. All rights reserved.