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Molecular Identification and Genotyping of Human Brucella Strains Isolated in Kuwait

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Abstract : Brucellosis is a zoonotic disease endemic in Kuwait. Human brucellosis can be caused by several Brucella species with Brucella melitensis causing the most severe and Brucella abortus the least severe disease. Furthermore, relapses are common after successful chemotherapy of patients. The classical biochemical methods of culture and serology for identification of Brucellae provide information about the species and serotypes only. However, to differentiate between relapse and reinfection/epidemiological investigations, the identification of genotypes using molecular methods is essential. In this study, four molecular methods [16S rRNA gene sequencing, real-time PCR, enterobacterial repetitive intergenic consensus (ERIC)-PCR and multilocus variable-number tandem-repeat analysis (MLVA)-16] were evaluated for the identification and typing of 75 strains of Brucella isolated in Kuwait. The 16S rRNA gene sequencing suggested that all the strains were B. melitensis and real-time PCR confirmed their species identity as B. melitensis. The ERIC-PCR band profiles produced a dendrogram of 75 branches suggesting each strain to be of a unique type. The cluster classification, based on ~ 80% similarity, divided all the ERIC genotypes into two clusters, A and B. Cluster A consisted of 9 ERIC genotypes (A1-A9) corresponding to 9 individual strains. Cluster B comprised of 13 ERIC genotypes (B1-B13) with B5 forming the largest cluster of 51 strains. MLVA-16 identified all isolates as B. melitensis and divided them into 71 MLVA-types. The cluster analysis of MLVA-16-types suggested that most of the strains in Kuwait originated from the East Mediterranean Region, a few from the African group and one new genotype closely matched with the West Mediterranean region. In conclusion, this work demonstrates that B. melitensis, the most pathogenic species of Brucella, is prevalent in Kuwait. Furthermore, MLVA-16 is the best molecular method, which can identify the Brucella species and genotypes as well as determine their origin in the global context. Supported by Kuwait University Research Sector grants MI04/15 and SRUL02/13.

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