عنوان مقاله:

Molecular typing of Iranian strains of Brucella abortus and Brucella melitensis using RAPD-PCR

محل انتشار:

بیستمین کنگره بین المللی میکروب شناسی ایران (سال: 1398)

تعداد صفحات اصل مقاله: 1

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خلاصه مقاله:

Introduction and Objectives: Molecular Typing of pathogenic bacteria are introduced to identify local and regional strains, tracing the source of infection, probable changes in native strains, and even an evolutionary study of phylogenic trees. The objective of this study was to identify DNA polymorphisms and genomic fingerprinting of Iranian Brucella strains using RAPD-PCR technique. Materials and Methods: 101 strains of Brucella, including seven strains of reference (S.99, 16M., RB51, S.19 544, Rev.1, H.38) and 94 Iranian field strains from different geographical regions during 1340 to 1382 were investigated. Brucella strains were cultured on Brucella broth, Brucella agar according to recommended methods. And the purity of strains cultivated with standard methods was assured. Then, all species were determined by phage-typing. Total DNA was extracted from the strains using three methods, including a phenol, Chlorophorm and Iso amilalchol. Optimization of RAPD-PCR conditions with nine primers of 13 single individual primers and 13 pair individuals primers with two different strains of abortus and melitenesis were performed separately. The best result was obtained with the AP4 peptide primer with a sequence of 3 \\ 5 \\ -TCA CGC TGC A-A and the primer pair REP- AP4 with 15-mer sequence, 5-CGC TTA TCG GCC TAC-3 and AP4 for Brucella species for the first time. Results: 101 strains of Brucella strain with AP4 primer were evaluated with optimum RAPD-PCR conditions. The final PCR product in 5.1% electrophoresis gel with ethidium bromide staining revealed 72 different patterns of DNA from 101 strains of Brucella. This method, even among strains within a biotype of the phenomenon of polymorphism, was shown to be repeatable. Within the five genotypes, two to four different biotypes were also included. Conclusion: RAPD can be useful method to distinguish related bacterial species, Despite the presence of high polymorphism in DNA. in this study, there was a presence of infection among animals of different Brucella biotypes, Brucella abortus, Biotype III, and Brucella melitensis Biotype 1, which are still dominant and indigenous Iranian strains in the past four decades. This technique appears to be a simple, quick and sensitive for the epidemiological investigation of brucellosis.

کلمات کلیدی:

RAPD-PCR, Brucella, Molecular typing, Brucella strains.