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Author: Siamak Molazadeh

Title: Phylogenetic analysis of Ehrlichia species isolated from horses and dogs

Summary:

The present study was conducted to determine the genomic detection of the genus Ehrlichia and its species in blood samples collected from horses and dogs in the provinces of West Azerbaijan, Gulistan, Kerman, Khuzestan and Mazandaran. 400 blood samples were randomly collected from horses (200 samples) and dogs (200 samples) in these provinces. The blood samples were collected in 2019, as were the age and sex of the animals. Bacterial genomic DNA was extracted from all blood samples using a commercial kit. PCR was then used to determine the genus based on the 16S rRNA gene. The semi-nested PCR method was also used to amplify the dsb, TRP36 and gltA genes. Positive samples were identified based on the 16S rRNA gene for species detection with specific primers of the mentioned genes. The results showed that 18 samples (5/4) percent, (95 percent Cl: 7/2 - 2/7 percent) of the blood samples examined were 6 samples (3 percent) of horse samples and 12 samples (6 percent) of dog samples contained the genome of the positive Ehrlichia, a significant difference was observed based on the species (p < 0.05). A significant difference between the age groups 4 to 8 years and 8 years and older was observed in the horse, a significant correlation was also observed between the dog and the presence of *Ehrlichia* in the blood (p < 0.05). The highest prevalence of bacteria in blood was observed in the regions of Kerman and Khuzestan, and the lowest prevalence in the regions of West Azerbaijan and the northern provinces of Gulistan and Mazandaran (p < 0.05). All samples positive for the 16S rRNA gene were also positive for the gene determinants (gltA, dsb and TRP36). The phylogenetic tree constructed on the basis of the sequence of the 16S rRNA and dsb genes among the Ehrlichia samples diagnosed in the present study showed very little difference from the reports in the gene bank. Based on the results, it can be concluded that the blood samples from dogs and horses were contaminated with the Ehrlichia genome and should be considered as the source of *Ehrlichia*. It was also found that the population of horses and dogs in the country should be considered as an important factor in the epidemiology of Ehrlichiosis and therefore in public health. The study finally found that the species E. canis and E. ruminatum were dominant study aera based on the dsb gene, and according to BLAST at the gene Bank, the gene has a lot in common with recorded sequences from other parts of the world.

Key words: Ehrlichia, Molecular detection, horse, dog