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First isolation of Anaplasma phagocytophilum AAIK from Apodemus agrarius in Korea

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uman granulocytic anaplasmosis, an emerging infectious disease in the Republic of Korea (ROK), is caused by an obli-H gate intracellular tick-borne bacterium of the family Anaplasmataceae. Anaplasma phagocytophilum has been found in a variety of animal species including wild deer, cats, dogs, gray squirrels, horses, and mice. The objective of the present study was to isolate and characterize the strains of A. phagocytophilum in black-striped field mice (Apodemus agrarius) and Korean water deer (Hydropotes inermis argyropus) in the ROK. Anaplasma infection based on the 16S rRNA genes were detected by conventional polymerase chain reaction (PCR) and species-specific nested PCR assays with DNA from Korean water deer (KWD) blood and black-striped field mice (BSFM) spleen. For isolation of A. phagocytophilum, BALB/c mice were used for propagation. White blood cells from 4-5 infected BALB/c mice were inoculated into 3×10⁴ HL60 and THP-1 cell line, respectively. Eight KWD and 27 BSFM were captured. Seven (87.5%) blood samples of KWD and 12 (44.4%) spleens of BSFM were positive for A. phagocytophilum according to PCR. However, 7 white blood cell samples of KWD were not propagated in HL60 and THP-1 cell line. Twelve spleen suspensions of BSFM were pooled for inoculum, and 0.3 ml of spleen suspension was intraperitoneally inoculated into 20 BALB/c mice (5 in each group; total 4 groups). When tested for 10 days post-inoculation, inoculated mice were positive for A. phagocytophilum by PCR. Three of 4 groups were not propagated and 1 of 4 groups was propagated in THP-1 cell line. A. phagocytophilum was observed in Wright-Giemsa stain preparations at 9 days after inoculation of cell cultures. Morulae were found on 70% of THP-1 cells at 12 days post-inoculation. Cultured A. phagocytophilum were confirmed by indirect immunofluorescence assay. PCR was performed using 16S rRNA, ankA, groEL, and msp2 gene primers were used to amplify the genes of A. phagocytophilum. All of the nucleotide sequences from cultured isolate were identical with A. phagocytophilum. We isolated the strain of A. phagocytophilum (AAIK isolate) in black-striped field mice (Apodemus agrarius) in the ROK. This strain may be expected to contribute to public health and veterinary medicine.

Biography

Joon-seek Chae has completed his DVM, MS, PhD from Chonbuk National University and postdoctoral studies from Texas A&M University and University of California-Davis. He is the Professor of College of Veterinary Medicine, Seoul National University. He has published more than 160 papers in reputed journals and serving as an editorial board member of repute. His recent interesting research areas are tick-borne zoonotic pathogens (*Anaplasma, Ehrlichia, Rickettsia, Bartonella*, TBE and SFTS virus etc.) and equine stem cell therapy.

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