学位論文内容の要旨
Abstract of the dissertation

博士の専攻分野の名称：博士（感染症学）　　氏名：小方　昌平

学位論文題名
The title of the doctoral dissertation

Studies on genetic diversity and transmission dynamics of *Spiroplasma* in ixodid ticks
（マダニにおける *Spiroplasma* の遺伝的多様性と伝播動態に関する研究）

Ixodid ticks are blood sucking ectoparasites of vertebrates with about 700 species distributed in the world. They serve as vectors of many pathogens and cause significant public health and veterinary health problems globally. Symbiosis is a general term used to describe two or more different species living in close association with each other. In nature, symbiotic relationships between bacteria and arthropods are well known and have been studied extensively. Understanding the molecular and biochemical mechanisms that underpin these relationships is a notable focus of symbiont research. Members of the genus *Spiroplasma* are gram-positive bacteria without cell walls. They are known as symbionts of arthropods and plants. *Spiroplasma* is one of the most common endosymbionts with a wide range of hosts, including insects, arachnids, crustaceans, and plants. It is estimated that 5–10% of insect species harbour this symbiont group. *Spiroplasma* has a wide range of fitness effects and transmission strategies. Some *Spiroplasma* species affect the sex ratio by inducing male killing in hosts such as flies, butterflies, and ladybird beetles. Several *Spiroplasma* species are known to cause disease in arthropods such as bees and plants. On the other hand, some flies infected with *Spiroplasma* can develop resistance to other pathogens. This characteristic of *Spiroplasma* is not only biologically interesting but also useful for symbiotic control applications among host individuals.

Chapter I focused on the infection status and genetic diversity of *Spiroplasma* in ticks. A total of 712 ticks were tested for *Spiroplasma* infection by PCR targeting 16S rDNA, and *Spiroplasma* species were genetically characterized based on 16S rDNA, ITS, *dnaA*, and *rpoB* gene sequences. A total of 109 samples originating from eight tick species were positive for *Spiroplasma* infection, with infection rates ranging from 0% to 84% depending on the species. A linear mixed model indicated that tick species was the primary factor associated with *Spiroplasma* infection. Moreover, certain *Spiroplasma* alleles that are highly adapted to specific tick species may explain the high infection rates in *Ixodes ovatus* and *Haemaphysalis kitaokai*. A comparison of the alleles obtained suggests that horizontal

transmission between tick species may not be a frequent event. These findings provide clues to understand the transmission cycle of *Spiroplasma* species in wild tick populations and their roles in host ticks.

Chapter II focused on vertical transmission potential of *Spiroplasma* in ticks. Two species of *Siproplasma*, *S. ixodestis* and *S. mirum*, were experimentally inoculated into laboratory colonies of *Haemaphysalis longicornis*. The presence of *Spiroplasma* was examined in the eggs and larvae originating from *Spiropalsma*-inoculated ticks by PCR. The results indicated that only *S. ixodetis* was transmitted into the eggs and larvae when ticks were inoculated with the concentration of 5×10^{11} bacteria per individual. There was no significant difference in engorged weight, egg weight and hatching rate between *Spiroplasma*-inoculated and control groups, indicating that *Spiroplasma* infection does not affect the reproduction of ticks. The data obtained are the first experimental evidence to demonstrate the vertical transmission potential of *Spiroplasma* in ticks.