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MORPHOLOGICAL CHANGES IN *BABESIA GIBSONI* IN *IN VITRO* CULTURE
AND ITS RELATION TO HEMOLYSIS OF INFECTED ERYTHROCYTES

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For clarifying the pathogenesis of anemia induced by *Babesia gibsoni* infection in dogs, the morphological changes in the parasite and its relationship to the destruction of infected erythrocytes were investigated *in vitro*. Erythrocytes infected with *B. gibsoni* were incubated in α -medium supplemented with L-glutamine (0.3 mg / ml), sodium pyruvate (0.11 mg / ml), sodium bicarbonate (2 mg / ml), penicillin G (100 units / ml), streptomycin (0.1 mg / ml) and 40 % normal canine serum at 37C under a humidified atmosphere with 5 % CO₂ for 4–15 days.

The parasites were morphologically classified into 7 forms on the basis of microscopical observations: the ring, dot, willow leaf, paired pears, amoeboid, multiple, and petaroid forms. When the parasite began to increase in number in the culture, almost all of the parasites in erythrocytes were the ring form. In contrast, as parasite number began to decrease, the percentages of dot, amoeboid, and willow leaf forms, especially of the dot form, increased. At the end of the incubation period, almost all the parasites were of the dot forms. These morphological changes in *B. gibsoni* during *in vitro* culture corresponded well with those seen in the peripheral blood of *B. gibsoni*-infected dogs.

These results suggest that the ring form of the parasite is a multiplication form, and that both the dot and amoeboid forms may be the degenerated or inactivated forms. In addition, the number of erythrocytes in the culture gradually decreased with hemolysis during multiplication of the parasites, indicating a destruction of parasitized erythrocytes. This destruction of erythrocytes progressed even after the parasite number in the culture had decreased. These results suggest that the multiplication of the parasite within erythrocytes may be not only the direct cause of the destruction of host cells, but also the cause of hemolysis of non-parasitized cells.