Analysis of host defense immunity and development of recombinant vaccines against Babesia microti infection

その他（別言語等）のタイトル	バベシアマイクロティ感染に対する宿主免疫の解析と組換えワクチンの開発
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学位授与機関	帯広畜産大学
学位授与年度	2017
学位授与番号	10105甲第80号
URL	http://id.nii.ac.jp/1588/00001396/

Form No.5

Abstract of Thesis/Dissertation

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Master's/Doctoral Program in Animal and Food Hygiene

Graduate School of Animal Husbandry

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(バベシアマイクロティ感染に対する宿主免疫の解析と組換えワクチンの開発)

Abstract

Babesia organisms are tick-transmitted hemoprotozoan parasites belonging to the phylum Apicomplexa, class Piroplasmea and order Piroplasmida. Pathogenesis of *Babesia* species results from the asexual erythrocytic stage, where the parasite invades, replicates and consequently causes destruction of host cells, haemoglobinuria and anemia. The increasingly emergence of human babesiosis has resulted in demands for urgent preventive strategies to control the *Babesia* infection.

In chapter 1, I investigated the host immunity involved in the cross-protection between *B. rodhaini* and *B. microti*. I found that mice which had recovered from *B. rodhaini* infection by drug treatment were completely protected against *B. rodhaini* reinfection and *B. microti* challenge infection, which characterized by extremely reduced parasitemia, higher hematocrit value and no mortality compared to the control mice. In contrast, mice immunized with dead *B. rodhaini* did not show any protection against *B.*

rodhaini and *B. microti* challenge. High level of antibody response and low levels of cytokines (INF-γ, IL-4, IL-12, IL-10) were detected in the protected mice. The resulting protection should not be attributed to antibodies and cytokines induced by the live parasites, because protected mice produced low level of cytokines and there are no cross-reacting antibodies between *B. rodhaini* and *B. microti*. Therefore, these findings indicate a possible role of innate immune cells in the case of this cross-protection.

In chapter 2, I evaluated the protective effect of two *B. microti* antigens, AMA1 and RON2, as subunit vaccines. The genes encoding for predicted BmAMA1 domain I and domain II (DIDII) and the gene encoding for predicted BmRON2 transmembrane region 2 to 3 (TM2-TM3) were expressed and purified. I found that immunization with rBmAMA1+rBmRON2 conferred partial protection against *B. microti* challenge infection in hamsters, which characterized by significantly reduced parasitemia and higher hematocrit value after challenge infection. However, immunization with rBmAMA1 and rBmRON2 alone did not show any significant protection compared to the control group. In addition, there is no significant difference in the total amount of antibodies against rBmAMA1 and rBmRON2 between the groups immunized with single and combined proteins. These results suggest that antibodies targeting on the key epitopes of both antigens are required for the protective immunity.

In chapter 3, a heterologous prime-boost strategy using plasmid pBmAMA1 and recombinant adenovirus Ad5BmAMA1 for immunizing hamsters against *B. microti* infection was valuated. The result showed that the heterologous prime-boost strategy stimulates a strong Th1-bias immune response. Hamsters immunized with pAMA1/Ad5AMA1 exhibited a degree of protection against *B. microti* infection,

characterized by lower parasitemia and higher hematocrit values at the acute stage of infection compared to the control group. These results indicate that the heterologous DNA priming and recombinant adenovirus boost strategy could improve the protective efficacy of vaccination against *B. microti*.

Overall, the present study analyzed the host immune response against *Babesia* parasites and evaluated the protective effect of several types of vaccines against *B. microti* infection. These results may provide useful information for developing effective preventive strategies against babesiosis.