ABSTRACT

Borrelia burgdorferi (BB) vaccines are commonly administered to prevent BB infection and clinical Lyme disease in endemic areas. Numerous published studies have documented that assays based on BB VlsE derived C6 peptide do not react with sera from vaccinated dogs. However, because of variability in vaccine formulations, studies to provide variability in interpretation of confirmatory assays, evaluation of samples derived from a controlled experimental vaccination study was warranted.

Twelve purebred specific-pathogen-free beagles were utilized. Groups of three dogs were assigned to a vaccine group and administered one of four vaccines [Recombikin™ (Merial), Merck6, Fort Dodge Laboratories], and the Lyme vaccine (Schering-Plough and Novibac Lyme (Merck)). The first three vaccines were administered on weeks 0, 2, 33, and 39 to generate high titer serum. The Novibac Lyme Vaccine was administered on weeks 0 and 3. Blood samples collected before and after vaccinations at all time points were processed and serum was tested utilizing 80IFA (IDEXX Reference Laboratories), Quant C6 ELISA (IDEXX Reference Laboratories) and SNAP®AX Plus.

Sera from all dogs had positive IFA titers, ranging from 1:800 five weeks post-vaccination to 1:6400 following additional vaccinations. The Lyme Quant C6 Test and the SNAP 4dx Plus test were negative for all samples from all vaccinated dogs at all time points indicating a lack of IFA titers. The results of this study document that the BB vaccines studied did not induce antibodies detectable in C6 based immunoassays, even when dogs are hyper-vaccinated.

RESULTS

SNAP 4dx Plus and Quant C6 Assays: In the Recombikin™, Lymavelx™, and Galvax™ and Novibac Lyme vaccine studies, sera from vaccinated dogs were tested by the SNAP 4dx Plus and Quant C6 Assays. All dogs were negative for antibody to B. burgdorferi in both assays throughout the study (Tables 3-4).

Immunofluorescence Assay: In the Recombikin™, Lymavelx™, and Galvax™ Lyme vaccination study, sera from all of the vaccinated dogs had significant IFA titers (individual titers ranged from 1:800 to 1:3600) when tested 5 weeks following the initial vaccination (Tables 1-3). Serum antibody titers decreased to 16 fold (individual titer range: 1:1500 to 1:4000) by week 33. Antibody titers increased substantially following administration of the additional dose of vaccine on weeks 36 and 37 with the highest levels (individual titer range 1:1600 to 1:5400) one week following the fifth vaccination (week 40). In the Novibac Lyme vaccination study, sera from all of the vaccinated dogs had significant IFA titers (individual titers ranged from 1:1600 to 1:6400) when tested 4 weeks following the fifth vaccination (Table 4).

Western Blot assay: The Western Blot results at week 5 for sera from dogs immunized with the whole bacterial vaccines (Lymavelx™, Galvax™) had prominent bands at positions corresponding to B. burgdorferi proteins with molecular weights of 31,000 (OspA) and 34,000 (OspB). Sera from dogs immunized with the recombinant OspA vaccine (Recombikin™) had a single prominent band corresponding to the B. burgdorferi-specific OspA protein (31 kDa). In the Novibac Lyme vaccination study, sera from all of the vaccinated dogs induced antibody responses to both OspA and OspC. B. burgdorferi proteins, the latter not commonly associated with other commercial Lyme vaccines.

INTRODUCTION

Introduction of Lyme disease vaccines for use in canines has led to the widespread use of vaccine in Lyme disease-endemic areas of the United States. The SNAP 4dx Plus Test Kit uses Borrelia-specific peptide (C6 peptide) for detection of antibody to Borrelia burgdorferi. Numerous published scientific articles have documented that these immunoassays incorporating the C6 peptide as the target diagnostic antigen failed to react with sera from vaccinated dogs. The purpose of the study was to test sera from vaccinated dogs using the C6-peptide-based microliter format ELISA and SNAP 4dx Plus test to demonstrate that these tests are non-reactive with samples from dogs receiving repeated vaccinations.

METHODS

Vaccine Study: Experimental vaccinations were performed in collaboration with Covance Research Products (Denver, PA) and Colorado State University using standard vaccination protocols. Groups of 3 dogs each received following vaccines: Recombikin™ (Merial), Lymavelx™ (Fort Dodge Laboratories), Galvax™ (Schering-Plough) and Novibac Lyme Vaccine (Merck dog health). Vaccine was administered on weeks 0, 2, 33, and 39. The Novibac Lyme Vaccine (Merck dog health) was administered on weeks 0 and 3. Blood samples were drawn at weekly intervals, from weeks 0 through week 11.

B. burgdorferi IFA and Quantitative C6 ELISA: An indirect IFA and the Lyme Quant C6 Test were performed on each sample at IDEXX Reference Laboratories.

B. burgdorferi Western Blot (WB) Assay: Samples were sent to University of Connecticut Veterinary Medicine Diagnostic Testing Lab for Lyme Western Blot testing.

SNAP®AX Plus Test: All samples were blinded and run on the SNAP 4x Plus Test Kit and read by one operator visually, per the kit insert protocol.

SUMMARY AND CONCLUSIONS

The results of the experimental vaccination study demonstrate the absence of reactivity of C6 based immunoassays with serum antibodies from Lyme-vaccinated dogs known to be free of B. burgdorferi infection. The SNAP 4dx Plus Test Kit B. burgdorferi assay detects antibody induced as a result of natural infection with the organism and not following REPEATED immunization with the following commercial vaccines: Recombikin™, Lymavelx™, Galvax™; and Novibac Lyme.