Screening canine sera for smooth brucella strain antibodies via *Brucella abortus* fluorescent polarization assay

Alyssa Helms,[a] Orsolya Balogh,[a] Rebecca Franklin-Guild,[b] Kevin Lahmers,[c] Clayton Caswell,[c] Julie Cecere[a]

[a]Department of Small Animal Clinical Sciences, Virginia-Maryland College of Veterinary Medicine, Blacksburg, VA; [b]Department of Population Medicine and Diagnostic Sciences, College of Veterinary Medicine Cornell University, Ithaca, NY; [c]Department of Biomedical Sciences and Pathobiology, Virginia-Maryland College of Veterinary Medicine, Blacksburg, VA

Dogs can serve as hosts for all 4 of the most common zoonotic strains of *brucella*: the smooth strains (i.e. *Brucella abortus* [*B. abortus*], *B. melitensis*, *B. suis*, and the rough strains [i.e. *B. canis*]). However, due to differences in cell wall morphologies between the smooth and rough strains, the only validated serologic test currently available for brucellosis screening in dogs is limited to the identification of the rough strains (i.e. *B. canis*) alone. Recently, the USDA added the fluorescent polarization assay (FPA) as an approved test for the confirmation of brucellosis infection in cattle, bison, and pigs, due to the demonstration of sufficient cross reactivity between *B. abortus*, *B. melitensis*, and *B. suis*. Thus, the goal of our study was to utilize the FPA test to identify antibodies to smooth brucella strains in canine sera, and to compare the results of the FPA test to the commonly utilized *B. abortus* card agglutination (BCA) test. Sera from 95 clinically healthy dogs, including 45 hog hunting dogs, were screened for circulating antibodies utilizing a combination of canine *brucella* slide agglutination test (CBSA), *Brucella canis* agar gel immunodiffusion II test (AGID), and BCA and FPA tests. Suggested test interpretation results yielded a 0% (0/95) smooth *brucella* strain seropositivity rate, with 38/95 (40%) BCA positive results, and 0/95 (0%) FPA positive results. Rough brucella strain serology yielded an inconclusive result (0 - 2% rough strain seropositivity rate) in 2% (2/95) of dogs. Additionally, a retrospective portion of the study was performed to identify sera containing circulating antibodies to any of the smooth strains of *brucella* via *Brucella abortus* FPA by testing banked canine serum samples that had been submitted to Cornell's veterinary diagnostic laboratory between 2018 - 2019 and previously screened by CBSA and AGID for *B. canis* with a positive or inconclusive test interpretation result. Of the 769 serum samples tested, 30/769 (4%) yielded a positive FPA test result, 13/769 (1.7%) yielded an inconclusive result, 725/769 (94.2%) were negative, and 1/769 (0.1%) sample excluded due to insufficient sample remaining to perform the diagnostic test. Of the 30 FPA suspect positive canine serum samples, 97% (29/30) also tested positive on CBSA. Additionally, the signalment of FPA suspect positive dogs was much more likely ($p < 0.0001$) to be spayed or neutered compared to intact, and mixed breed compared to purebred.

Keywords: Brucellosis, canine, abortion, smooth strain, fluorescence polarization test

Fetal loss at time of elective cesarean section in a dog

Brittany Shumack

Village Center Veterinary Care, Colorado Springs, CO

A 3-year-old Greater Swiss Mountain dog was presented exhibiting prolonged signs of stage 1 labor. Abdominal radiographs obtained 6 days prior revealed 8 fetuses appearing viable with heartbeats >180 bpm on ultrasonographic examination. Owner elected for cesarean section. Surgical site was prepared while the patient was given supplemental oxygen. An anesthetic protocol of alfaxalone for induction and isoflurane for maintenance were utilized. Time from anesthetic induction to removal of the first puppy from the uterus was ~ 8 minutes. The first, fourth, and seventh puppy delivered resuscitated in < 2 minutes. Five remaining puppies did not respond to any resuscitation efforts including accordion technique, GV26 acupuncture point, and epinephrine. When GV26 acupuncture points were performed substantial bleeding was noted from the site. On physical examination the 5 puppies were also judged to have distended abdomens. Paracentesis was performed and abdomen of each puppy was full of what was suspected to be frank blood. Blood was sent for CBC/coagulations panels. Radiographic images of the neonates were obtained and 4 of 5 puppies had fluid in their lungs. Three of the 5 struggling puppies had faint heartbeats. After 40 minutes of resuscitation efforts with no improvement the owner elected to discontinue the efforts. Blood had markedly decreased platelets with very minor platelet clumping observed. Many of the platelets were enlarged, suggesting accelerated platelet production of the bone marrow. A neutropenia was present. Prothrombin time and APTT were markedly increased, fibrinogen was markedly decreased, and D-Dimer was severely elevated. Three fetuses that resuscitated quickly had no abnormalities on physical examination and there were no concerns going forward. Multiple consults and discussions about this case did not provide answers as to why the fetal loss occurred. P2Y12 is a platelet disorder present in greater Swiss Mountain dogs. The sire was negative, and the dam was negative or a carrier. It is unclear if a natural whelping would have yielded better results. The purpose of this case is to present an occurrence of multiple unexplained fetal losses and bring awareness to a possible phenomenon. Despite much criticism from the greater Swiss Mountain dog breed club for isoflurane use during cesarean sections due to a supposed breed sensitivity, we have not changed our anesthetic protocol. Multiple cesareans have been performed on greater Swiss Mountain dogs since this case with a 100% fetal resuscitation rate.

Keywords: Fetal loss, cesarean section, clotting disorder