Knowledge, Attitudes and Practices of Licensed Dog Breeders in Kansas Regarding Canine Brucellosis

by

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Table of Contents

Abstract

Background: Canine brucellosis is an infectious reproductive disease of dogs and is zoonotic. Cases of disease in dogs and humans are reportable to the Kansas Department of Agriculture – Division of Animal Health (KDA-DAH) and the Kansas Department of Health and Environment (KDHE), respectively. People with occupational exposures, such as dog breeders, are more likely to be exposed to this bacterium. It can be difficult to diagnose in humans; the true incidence is not well understood. The objectives of this study were to understand the knowledge, attitudes and practices (KAP) of breeders regarding canine brucellosis and to determine the human incidence of infection in Kansas.

Methods: A KAP survey was mailed to all 294 licensed breeders in Kansas. We evaluated all case reports of human brucellosis reported to KDHE from 1997-2012.

Results: The response rate was 25.5% (75/294). Eighty-eight percent of breeders (66/75) had knowledge of canine brucellosis. Only 66.7% (44/66) of those knew it was zoonotic; they were not more likely to wear gloves during whelping ($p = 0.4156$). Seven breeders (9.33%) reported cases of brucellosis in dogs; no respondents reported symptoms of canine brucellosis. Eleven confirmed, sixteen probable and twenty-eight suspect human cases of brucellosis have been reported in Kansas; 81% (22/27) did not report a specific species of *Brucella*.

Conclusions: Breeders are knowledgeable about canine brucellosis; however, they were not more likely to take appropriate personal protective measures. Public and animal health officials should be aware that dogs can serve as a source of brucellosis infection to people.

Background

Brucella canis

Brucellosis was first identified in the year 450 by Hippocrates, who described cases of recurring fever and death in his book Epidemics.[1] In 1887, David Bruce isolated an organism from the spleen of a human patient and named the bacteria *Micrococcus melitensis.*[2] A decade later, a Danish physician and veterinarian named Bernhard Bang isolated the organism causing contagious abortion in cattle and called it *Bacterium abortus*. It was not until 1917 that an American bacteriologist named Alice Evans realized that the two species were related and the bacteria were renamed *Brucella melitensis* and *Brucella abortus*, respectively.[1] Other *Brucella* species were discovered later, including *B. suis* in 1914, *B. ovis* in 1953, and finally *B. canis* in 1966.

Canine brucellosis is an infectious reproductive disease of dogs that can lead to prostatitis and epididymitis in males, late-term abortions and stillbirths in females, and infertility in both sexes. Also a zoonotic disease, *Brucella canis* can cause flu-like illness in humans and in rare cases may cause serious sequelae including meningitis and endocarditis. Canine brucellosis is thought to be present worldwide except in Australia and New Zealand.[3] Prevalence estimates in infected dogs in the United States are 1-8% of the dog population.[4] Canine brucellosis in humans is considered rare and estimated to represent only 1% of human brucellosis cases reported annually.[5] The Centers for Disease Control and Prevention (CDC) receives reports of just over 100 human cases of brucellosis each year.[4] Since data collection began in 1973, the CDC has recorded approximately 50 confirmed cases of canine brucellosis in humans.[3] The apparent rarity of the disease in humans likely contributes to the anonymity of *B. canis* relative to the other *Brucella* species. However, these estimates are thought to be falsely low due to the nonspecific clinical presentation and the lack of a reliable serologic test to detect human infection.[6]

The Bacterium

All species in the genus *Brucella* are Gram-negative, aerobic, intracellular coccobacilli.[7] *Brucella* species can affect multiple host species, but each known bacterial species has a preferred animal host. The genus *Brucella* has five main species: *Brucella abortus* (cattle), *B. suis* (swine), *B. melitensis* (sheep and goats), *B. ovis* (sheep), and *B. canis* (dogs).[7] *Brucella canis* is not considered as virulent in humans as are *B. melitensis* and *B. suis.*[8] Colony morphology in culture varies: some species form smooth colonies (B. *abortus, B. suis,* and *B. melitensis*), while others grow rough phase, mucoid colonies (*B. canis* and *B. ovis)*.[4] The implications of this difference with regard to testing for canine brucellosis will be explored in the diagnosis section below.

Infection begins when the bacteria enter the body through mucous membranes of the respiratory, oral or reproductive tract.[7] Bacteria initially localize in lymph nodes, and then disseminate in the blood to other organs such as the uterus, prostate and testicles. Bacteremia in dogs is first detectable two to three weeks post-infection. The bacteria can enter host phagocytic cells and prevent cellular degranulation, contributing to the persistence of the disease.[7]

Transmission

B. canis can enter the body through inhalation, ingestion and direct contact with mucous membranes or broken skin.[4] The organism is present in most bodily fluids and reproductive tissues. The highest risk of transmission involves direct contact with contaminated placenta, aborted fetus, and vaginal discharge from an infected bitch during estrus, whelping or abortion. Semen and urine from male dogs can be a source of infection, even years after initial diagnosis and attempted treatment. Live puppies born to an infected mother can be infected *in utero* across the placenta, during birth from ingestion of birthing fluids, or through milk.[4] The organism may survive for months on fomites or in water and soil in an environment of high humidity, low temperature and no sunlight.[3] Veterinarians have the additional occupational risk of direct contact with blood from a bacteremic patient. There is no documentation of person-to-person transmission of canine brucellosis.

Testing and Diagnosis

Serology can be used as a screening tool in dogs because the veterinary tests have a high sensitivity and low specificity. A negative screening test can rule out disease but a positive result should be followed up with a more specific confirmatory test. Failure to use a confirmatory test may result in unnecessary euthanasia of breeding dogs with an initial false positive screening test result. Screening tests include the rapid slide agglutination test (RSAT), modified RSAT with 2-mercaptoethanol test (ME-RSAT), and tube agglutination test (TAT).[9] The RSAT test uses a *B. ovis* antigen, which cross reacts with antibodies to *B. canis,* while the modified RSAT uses *B. canis* antigen.

Confirmatory tests include an agar-gel immunodiffusion (AGID) test using cytoplasmic protein antigens (AGIDcpa), polymerase chain reaction (PCR), and serial blood cultures.[9] A positive blood culture is considered definitive for a suspect case, but a single negative culture cannot rule out disease. The *Brucella canis* organism is very fastidious and may take weeks to grow in culture. This can produce a false negative result if the growth medium is insufficient or if the plate is read and discarded prematurely.[4] Bacteremia in dogs begins two to three weeks after exposure and can last for several years, but may occur in low levels. To increase the likelihood of collecting a positive sample, it is advisable to collect and submit three blood samples taken a minimum of 24 hours apart.[10] It is also important to take baseline blood samples before beginning any empiric treatment because antibiotics will decrease bacteremia.[9] Specimens from vaginal discharge, aborted fetus, milk, semen and urine can also be used for culture.[7] *Brucella* species are highly infectious in a laboratory environment, so it is critically important to clearly label the specimen as a brucellosis suspect before submission.[11]

The Kansas State Veterinary Diagnostic Laboratory (KSVDL) offers both blood culture and PCR testing services. A sample of whole blood in a citrate tube can be used for culture ($2.50 per sample) or for PCR ($28.50 per sample). Vaginal swabs are also an acceptable sample for PCR testing.[12]

In humans, routine screening tests for *Brucella* species are ineffective at detecting canine brucellosis. Human serology tests use the smooth-coated *B. abortus* antigen to detect *B. abortus, B.*

melitensis and *B. suis.* This test does not react with antibodies formed against the rough-coated *B. canis* organism, and thus produces a false negative result.[4] The definitive diagnostic test for canine brucellosis is identification of the colonies in culture, but this test has a significant false negative rate for the same reasons mentioned for culturing canine samples. Empiric antibiotic treatment will lower bacteremia and increase the likelihood of a false negative culture as well. On a more encouraging note, there are recent reports in the literature of an indirect ELISA with promising initial results for the diagnosis of canine brucellosis in both dogs and humans.[6,13]

Treatment

Treatment is not recommended for dogs in breeding facilities because antibiotics have limited effect and therapy is often unrewarding. Extensive use of antibiotics will reduce bacteremia but cannot reliably eliminate the intracellular organism from the body. Relapses are common. Spaying or neutering the animal can reduce but not eliminate the transmission risk.[7] In males, the bacteria remain sequestered in the prostate and can be shed intermittently in urine for years. The only reliable method of complete elimination of canine brucellosis in a breeding facility involves isolation, testing, and euthanasia of positive animals. There is no vaccine or prophylactic treatment for dogs.

The process of eliminating canine brucellosis from a breeding facility can be daunting and expensive. An information sheet published by the State of Georgia in 2008 provided the following recommendations.[11] All breeding dogs in the facility should be housed individually to halt transmission. All dogs are tested with a screening test and a confirmatory test. Those dogs testing positive on both tests should be euthanized. Puppies under 6 weeks of age should be tested using three blood cultures taken 24 hours apart and any puppy with a positive culture should be euthanized. All dogs that initially tested negative on both tests should be retested 4 weeks later to identify any acutely infected dogs that were not identified during the first round of testing. Continue to test all the remaining dogs every 4

weeks until all dogs are negative on two consecutive serologic tests.[11] Clearly, fulfilling the above recommendations can be financially draining and may be too much for a breeder to withstand.

It is common for humans, in contrast to dogs, to completely clear the bacteria following antibiotic therapy. Some patients may recover spontaneously, while others can develop a chronic disease with fluctuating severity of symptoms if not treated with a course of antibiotics. The CDC recommendation for post-exposure prophylaxis in cases of *Brucella* exposure is a combination of rifampin with either doxycycline or trimethoprim-sulfamethoxazole.[11] Case mortality rate is very low (2-5%) and most deaths are associated with rare complications such as endocarditis and meningitis.

Brucellosis Reporting in Humans

Brucellosis, regardless of bacterial species, is also considered a reportable disease when it occurs in humans. Human cases are reported through a chain of agencies that eventually reaches a national level. In Kansas, the diagnosing healthcare provider or laboratory reports the case to the Kansas Department of Health and Environment (KDHE) Bureau of Epidemiology and Public Health Informatics (BEPHI). The case is then entered into the Kansas Electronic Disease Reporting System (EpiTrax) and assigned to the local public health jurisdiction in which the case-patient resides. All information requested on the disease reporting forms must be obtained and entered into EpiTrax within seven days of receiving notification of the case. The process of disease investigation is outlined in the "Brucellosis Investigation Guideline" document maintained by KDHE.[16] The local health department initiates an investigation consisting of the following components: use current case definition to confirm diagnosis, conduct a case investigation to identify possible sources of infection, conduct a contact investigation to identify additional cases, and initiate control and prevention measures to prevent spread of disease. The investigation guidelines detail how to conduct a standard case investigation, as well as how to manage special situations such as acts of bioterrorism, exposure to *Brucella*-containing vaccine and laboratory exposure to *Brucella* isolates.[16] Data from standard case investigations is

gathered by KDHE and sent to the Centers for Disease Control and Prevention (CDC) in Atlanta, GA on an annual basis. However, multiple brucellosis cases that are temporally/spatially clustered or are suspect acts of terrorism must be reported to CDC by telephone within 4 hours.

Brucella canis infections in humans are thought to be rare because approximately fifty cases have been reported to CDC since the discovery of the bacteria in 1966.[4] Licensed dog breeders likely represent a group of individuals with a higher risk of acquiring canine brucellosis than the general public because they have the potential occupational risk of contact with infectious materials such as aborted fetuses or vaginal discharge from an infected bitch.

Brucellosis Reporting in Dogs

The Kansas Department of Agriculture's Animal Health Division (KDA-AHD) inspects and regulates the licensing of all canine and feline breeding premises in Kansas. There are four types of breeding licenses: Hobby (sells 3-5 litters a year), Animal Breeder (sells 6+ litters a year, primarily for wholesale), Retail Breeder (sells 6+ litters a year, primarily at retail), and Animal Breeder and Distributor.[14]

The Animal Health Division is also tasked with tracking and maintaining records of reportable or contagious disease cases that occur in the state. Kansas Administrative Regulations (KAR 9-27-1) includes "all species of brucellosis" in the list of diseases that "shall be designated as reportable infectious or contagious animal diseases and shall be reported in accordance with KSA 47-622."[15] According to KSA 47-622, it is the duty of "any person who discovers the existence of any such contagious or infectious disease among the domestic animals of any person to immediately report this information to the animal health commissioner."[15] The Animal Health Division has recorded between 75 and 100 culture confirmed cases of canine brucellosis in the past ten years in Kansas, with an average of 6 to 12 reported cases annually.

Study Objectives

- To determine Kansas licensed dog breeders' knowledge, attitudes and practices regarding *Brucella canis* infection in both dogs and humans,
- To screen dog breeders for a history of symptoms that could suggest human cases of canine brucellosis
- To assess the human incidence of *Brucella* infection in Kansas.
- To identify current case investigation guidelines in place for investigating canine brucellosis cases in dog breeding facilities.

Methods

Knowledge, Attitudes and Practices Survey

A Knowledge, Attitudes and Practices (KAP) survey was developed and administered to licensed dog breeders in Kansas. The survey consisted of 44 multiple-choice and free-text questions covering topics including knowledge of the disease, testing schedules, personal protective behaviors, history of symptoms in dogs and humans, and finally demographics of the responder and space for additional comments. The survey was completely anonymous and no questions were asked that could identify any individual breeder or facility. The survey was granted an exemption by the Institutional Review Board at the Kansas Department of Health and Environment

Contact information was obtained from records of licensed dog breeders maintained by the Kansas Department of Agriculture's Animal Health Division (KDA-AHD). There were 294 licensed breeding facilities in Kansas during 2012. This population was considered small enough to facilitate a complete population survey, so the survey was sent to all 294 licensed breeders. Using an online calculator (Custom Insight), it was determined that 167 respondents (56.8% response rate) were needed at a 95% confidence interval for a 5% error level in interpreting the hypothesis tests.[17]

The decision to send a paper survey was multifactorial. The KDA-AHD had physical addresses for all 294 licensed facilities but had e-mail addresses for less than 50% of the breeding facilities, therefore using physical addresses allowed the inclusion of the entire desired population, as compared with sending an e-mail copy of the survey to only 150 breeders. The increased pool of potential responders was desired because canine brucellosis is a sensitive topic among breeders and a low response rate was anticipated. Additionally, based on past experiences with surveys conducted by KDHE, a paper survey has been shown to increase the survey response rate over distribution via e-mail.[18]

Surveys were mailed on Friday July 27, 2012 and respondents were asked to return the surveys via the enclosed pre-paid envelope by Monday August 6, 2012. Surveys were accepted through Wednesday September 12, 2012. No follow-up actions were taken to improve survey response due to budget restrictions.

Data from the surveys were entered into Microsoft® Excel 2010 for analysis. Surveys were securely stored until all information had been tabulated in the Excel file, after which the paper surveys were destroyed. Results were analyzed in Excel® using both basic and descriptive statistics. Answers for each question were tabulated so that counts and frequencies of responses could be calculated. Calculation of frequencies took into account skip patterns, and surveys that did not answer that specific question were not included in the denominator. Answers for demographic factors such as age, level of education, and household income were arranged in numerical order and a cut-off point was chosen where a natural break seemed to occur to divide the factor into two roughly equal categories. For example, age was divided into above and below 60 years because 42 respondents were below that age while 33 respondents were above 60 years of age. These categories were used when grouping data into 2x2 tables for hypothesis testing to determine if perceived associations between select responder characteristics were statistically significant. The null hypothesis is that there is no association. If the p-

value for the two-tailed Fisher's exact test was less than the significance level of 0.05, the null hypothesis was rejected and it was determined that there was an association between the two factors.

Brucellosis Reporting in Humans

A search for human brucellosis cases in Kansas was performed using the Kansas Electronic Disease Surveillance System (KS-EDSS) and EpiTrax electronic surveillance system maintained by the Kansas Department of Health and Environment. A new reporting system, EpiTrax, was implemented in 2012; cases reported prior to 2012 are maintained in KS-EDSS. The search included all species of *Brucella* and was not specific to *Brucella canis*. Table 1 defines each case classification used by KDHE, adopted directly from the National Notifiable Disease Surveillance System (NNDSS).[19] This case definition was last updated January 2010. Cases reported in KS-EDSS and EpiTrax were classified according to these case definitions.

Table 1 – Case Classification for Human *Brucella* Infection

Confirmed	• Clinically compatible illness* • Definitive laboratory evidence† of *Brucella* infection

Probable	• Clinically compatible illness with at least one of the following:
	  ○ Epidemiologically linked to a confirmed human or animal brucellosis case
	  ○ Presumptive laboratory evidence††

Suspect	• Laboratory results only • No clinical information

Clinically compatible illness is defined as a fever with one or more of the following: night sweats, arthralgia, headache, fatigue, anorexia, myalgia, weight loss, arthritis/spondylitis, meningitis, or focal organ involvement (endocarditis, orchitis/epididymitis, hepatomegaly, splenomegaly).

†***Definitive laboratory evidence*** requires the culture and identification of *Brucella* from a clinical specimen, or evidence of a fourfold or greater rise in *Brucella* antibody titer between acute and convalescent serum samples taken at least two weeks apart.

††***Presumptive laboratory evidence*** can include a *Brucella* total antibody titer of greater than or equal to 160 by standard tube agglutination test (SAT) or *Brucella* microagglutination test (BMAT) in one or more serum specimens obtained after onset of symptoms, or the detection of *Brucella* DNA in a clinical specimen by PCR assay.

Brucellosis Reporting in Dogs

A search for case investigation guidelines for canine brucellosis in dogs was performed by searching current Kansas Statutes, the KDA-AHD website and personal correspondence with Dr. Paul Grosdidier from KDA-AHD.

Results

Descriptive Analysis of Knowledge, Attitudes and Practices Survey

Of the 294 surveys sent, 78 surveys were returned. Three of those surveys were unsuitable for analysis: two were from cat breeders, and one was returned blank. Thus, 75 surveys were analyzed for a response rate of 25.5%.

Respondent Demographics

Respondents were asked a series of demographic questions to characterize the survey group. The only available demographic data for the entire population of licensed breeders in Kansas was the county where the kennel was registered. A chi-square test was performed to compare the distribution of survey respondents to the distribution of all 294 licensed breeders by region of Kansas where the breeding facility is located. The state was divided into nine regions; the individual counties included in each region are listed in Table 2. The null hypothesis was not rejected and it was concluded that licensed breeders that responded to the survey were not statistically different from non-responding licensed breeders in Kansas by geographical region (chi-square = 4.19; df = 8; p-value = 0.8396).

Table 2 – Geographic regions of Kansas and associated counties

Northwest	Barton, Cheyenne, Decatur, Ellis, Gove, Graham, Logan, Ness, Norton, Osborne, Phillips, Rawlins, Rooks, Rush, Russell, Sheridan, Sherman, Smith, Thomas, Trego, Wallace
Southwest	Barber, Clark, Comanche, Edwards, Finney, Ford, Grant, Gray, Greeley, Hamilton, Haskell, Hodgeman, Kearny, Kiowa, Lane, Meade, Morton, Pawnee, Pratt, Scott, Seward, Stafford, Stanton, Stevens, Wichita

North Central	Clay, Cloud, Dickinson, Ellsworth, Geary, Jewell, Lincoln, Marion, McPherson, Mitchell, Morris, Ottawa, Republic, Rice, Riley, Saline, Washington
South Central	Chase, Cowley, Harper, Kingman, Reno
Northeast	Atchison, Brown, Doniphan, Douglas, Marshall, Nemaha, Pottawatomie
Southeast	Allen, Anderson, Bourbon, Chautauqua, Cherokee, Coffey, Crawford, Elk, Greenwood, Labette, Lyon, Montgomery, Neosho, Wilson, Woodson
Kansas City Metropolitan Area	Franklin, Johnson, Leavenworth, Linn, Miami, Wyandotte
Topeka Metropolitan Area	Jackson, Jefferson, Osage, Shawnee, Wabaunsee
Wichita Metropolitan Area	Butler, Harvey, Sedgwick, Sumner

The majority of respondents were women (59 breeders ; 78.7%) compared to men (14 breeders; 18.7%). The median age of respondents was 56 years old, with a range of 26 to 80 years. No respondents reported being of Hispanic or Latino ethnicity. Seventy (89.7%) respondents listed their race as "White," 4 (5.1%) listed their race as "American Indian or Alaska Native" and 1 (1.3%) respondent listed "Black or African American." Tables 3 and 4 show the distribution of respondents by annual household income and level of education, respectively. The majority of respondents earn an annual household income under $75,000 (n = 42; 56.0%), but many respondents preferred not to answer the question (n = 31; 28.0%). The level of education was roughly split among respondents between a high school diploma or less (n = 35; 46.7%), and higher education (n = 37; 49.4%). Table 5 shows an analysis of the sizes of breeding facilities owned by the survey respondents. There was a wide range between respondents in the number of breeding females, breeding males, and number of litters produced per year. While some breeders reported numbers in the hundreds for each of the parameters, the median among responding breeding facilities was 15 breeding females, 6 breeding males and 10 litters produced per year.

Table 3 – Annual Household Income

Annual Household Income	N (%)
Under $25,000	16 (21.3%)
$26,000 - $50,000	14 (18.7%)
$51,000 - $75,000	12 (16.0%)
$76,000 - $99,000	6 (8.0%)
$100,000 or above	5 (6.7%)
Prefer not to answer	21 (28.0%)
No answer	1 (1.3%)
Total	75 (100%)

Table 4 – Highest level of education completed

Highest level of education	N (%)
Elementary school	2 (2.7%)
High school diploma	33 (44.0%)
Undergraduate college degree	17 (22.7%)
Higher education	12 (16.0%)
Apprenticeship or trade school	8 (10.7%)
Prefer not to answer	3 (4.0%)
No answer	0 (0.0%)
Total	75 (100%)

Table 5 – Kennel size by number of breeding females, breeding males, and litters

	Median	Minimum	Maximum
Breeding females	15	2	624
Breeding males	6	0	175
Litters produced per year	10	2	250

Knowledge of Disease

According to the survey results, 66 (88.0%) breeders had heard of canine brucellosis and 9 (12.0%) breeders had not heard of the disease. Those who had heard of canine brucellosis then answered questions #2 – 9 regarding their knowledge of disease transmission and treatment in dogs and humans. Many respondents correctly identified the three most common modes of disease transmission between dogs: sexual contact during breeding (89.4%), contact with infected vaginal discharge (78.8%) and infection passed from bitch to pups in utero or during whelping (59.1%). Forty-four (66.7%) breeders said that canine brucellosis in dogs could not be cured, while 10 (15.2%) believed it could be cured and 11 (16.7%) were unsure. Three of those who responded that canine brucellosis could not be cured also left comments that there is speculation and that the true answer to the question has not been reliably demonstrated scientifically.

Table 6 - Responses of breeders to survey questions about *Brucella canis* transmission and treatment

Question	Response
Have you heard of canine brucellosis?	Yes: **66 (88.0%)** No: **9 (12.0%)**
In what ways can canine brucellosis be spread from dog to dog? *(Please choose all that apply)*	Contact with vaginal discharge or product of abortion from an infected bitch: **52 (78.8%)** Contact with urine from an infected male: **37 (56.1%)** Shared food and water bowls: **15 (22.7%)** Infection passed from infected mother to puppies during pregnancy or whelping: **39 (59.1%)** Tick bite: **4 (6.1%)** Sexual contact during natural breeding: **59 (89.4%)** Use of artificial insemination: **26 (39.4%)** Not sure: **5 (7.6%)**
Can canine brucellosis in dogs be cured?	Yes: **44 (66.7%)** No: **10 (15.2%)** Not sure: **11 (16.7%)**

Breeders were also asked questions regarding their knowledge of canine brucellosis in humans. Of the 66 respondents, 44 (66.7%) said that the disease could spread to humans, 12 (18.2%) said the disease could not spread to humans, 8 (12.1%) were unsure and 2 (3.0%) did not respond. Among those who knew the disease was zoonotic, there was more uncertainty regarding knowledge of disease transmission and treatment in humans compared to dogs. Only eleven (23.9%) breeders correctly answered that the disease could not be spread from person to person while 10 (21.7%) thought that it could and 24 (52.2%) were unsure. The percentage distribution of answers was very similar for the question asking if the disease in humans could be cured.

Table 7 - Responses of kennel owners to survey questions about *Brucella canis* in humans

Question	Response
Can a dog infected with canine brucellosis spread the disease to humans?	Yes: **44 (66.7%)** No: **12 (18.2%)** Not sure: **8 (12.1%)**
Can canine brucellosis be spread from person to person?	Yes: **10 (21.7%)** No: **11 (23.9%)** Not sure: **24 (52.2%)**
Can canine brucellosis in humans be cured?	Yes: **15 (32.6%)** No: **8 (17.4%)** Not sure: **22 (47.8%)**

The relationship between knowledge of disease and various demographic factors were analyzed using Fisher exact tests. Surveys were counted and organized into two-by-two tables according to answers provided. Surveys that did not include answers to one or both of the questions of interest for each table were discarded. Because of this, the total number (N) in each two-by-two table varies. There was not a statistically significant relationship ($p < 0.05$) between knowledge of the disease in dogs and any of the demographic factors analyzed, including annual household income, level of education, age of respondent or a history of abortion within the respondent's facility. Results are shown in Table 8.

Table 8 – Breeders who have heard of canine brucellosis, stratified by select demographics

		Heard of Disease		
		Yes	*No*	Total
Household Income	*> $50K*	20	3	23
	≤ $50K	27	3	30
	Total	47	6	53

Fisher's exact test (two tailed): 1

		Heard of Disease		
		Yes	*No*	Total
Level of Education	*College*	33	4	37
	H. S.	30	5	35
	Total	63	9	72

Fisher's exact test (two tailed): 0.7318

		Heard of Disease		
		Yes	*No*	Total
Age	*≥ 60 yrs*	28	3	31
	< 60 yrs	36	6	42
	Total	64	9	73

Fisher's exact test (two tailed): 0.7243

		Heard of Disease		
		Yes	*No*	Total
History of Abortion	*Yes*	32	2	34
	No	31	7	38
	Total	63	9	72

Fisher's exact test (two tailed): 0.1578

A second set of two-by-two tables were created to evaluate potential relationships between knowledge of brucellosis as a zoonotic disease and the same four demographic factors used to analyze knowledge of the disease in dogs. Only one relationship was shown to be statistically significant; that of an association between the level of education of the breeder and knowledge of canine brucellosis as a zoonotic disease (Fisher value = 0.0134). Breeders with a level of education exceeding completion of high school are 1.57 times more likely to know that canine brucellosis is a zoonotic disease than breeders who did not receive formal education past the completion of high school.

Table 9 – Breeders who know that canine brucellosis is zoonotic, stratified by select demographics

		Knew of Zoonosis		
		Yes	*No*	Total
Household Income	*> $50K*	15	4	19
	≤ $50K	14	12	26
	Total	29	16	45

Fisher's exact test (two tailed): 0.1181

		Knew of Zoonosis		
		Yes	*No*	Total
Level of Education	*College*	26	5	31
	H. S.	16	14	30
	Total	42	19	61

Fisher's exact test (two tailed): 0.0134
Prevalence Ratio: 1.57

		Knew of Zoonosis		
		Yes	*No*	Total
Age	*≥ 60 yrs*	19	8	27
	< 60 yrs	23	12	35
	Total	42	20	62

Fisher's exact test (two tailed): 0.7876

		Knew of Zoonosis		
		Yes	*No*	Total
History of Abortion	*Yes*	22	9	31
	No	20	10	30
	Total	42	19	61

Fisher's exact test (two tailed): 0.7863

Brucellosis Testing – Attitudes and Practices

Of the 75 survey respondents, 43 (57.3%) indicated that they tested dogs for brucellosis, while 31 breeders (41.3%) did not perform testing and 1 (1.3%) did not respond. When asked why they decided not to test, the most common reasons were "Do not consider my dogs at risk of getting the disease (15 breeders; 45.5%), "No fertility problems in my kennel" (15 breeders; 45.5%) and "Did not know about the disease" (7 breeders; 21.2%). Nineteen of the thirty-one breeders who do not test report using only their own puppies for breeding. The other twelve breeders purchase dogs or semen from sources outside of their facility. The majority of breeders that test their dogs report testing new animals before co-mingling them with current dogs (36 breeders; 78.3%). Other common reasons were testing dogs with fertility problems (17 breeders, 37.0%) and testing dogs based on a veterinarian's recommendation (12 breeders, 26.1%). Responses are shown in Table 10.

Table 10 – Responses to questions regarding brucellosis testing

Question	Response
Do you test dogs in your kennel for canine brucellosis?	Yes: **43 (57.3%)** No: **31 (41.3%)**
What reasons made you decide to not test your dogs for canine brucellosis? *(Choose all that apply)*	Did not know about the disease: **7 (21.2%)** Do not consider my dogs at risk of getting the disease: **15 (45.5%)** No fertility problems in my kennel: **15 (45.5%)** Do not want to know if a dog is positive for the disease: **0 (0.0%)** Testing takes too much time and effort: **0 (0.0%)** Testing is too expensive: **5 (15.2%)**
When do you test your dogs for brucellosis?	Test dogs that have fertility problems: **17 (37.0%)** Test dogs on a veterinarian's recommendation: **12 (26.1%)** Test all breeding stock once a year: **0 (0.0%)** Test dogs prior to each breeding event: **2 (4.3%)** Test new animals before co-mingling with other dogs: **36 (78.3%)**

Fisher's exact tests were again used to assess the relationships between breeders that performed testing and those that knew canine brucellosis was zoonotic or those that had a history of dogs with reproductive problems. There was a statistically significant relationship between breeders that tested and those that had a history of infertility ($p = 0.0478$). Respondents who knew that brucellosis is zoonotic, and those with a history of abortion in the facility, tended to test more frequently ($p = 0.0988$ and $p = 0.0577$, respectively).

Table 11 – Comparison of breeders who test dogs, stratified by select characteristics

		Perform testing: *Yes*	Perform testing: *No*	Total	
Knew of zoonosis	*Yes*	31	12	43	Fisher's exact test (two tailed): 0.0988
	No/Unsure	10	10	20	
	Total	41	22	63	

		Perform testing: *Yes*	Perform testing: *No*	Total	
History of Abortion	*Yes*	24	10	34	Fisher's exact test (two tailed): 0.0577
	No	18	20	38	
	Total	42	30	72	

		Perform testing: *Yes*	Perform testing: *No*	Total	
History of Stillbirth	*Yes*	30	17	47	Fisher's exact test (two tailed): 0.2183
	No	12	13	25	
	Total	42	30	72	

		Perform testing: *Yes*	Perform testing: *No*	Total	
History of Infertility	*Yes*	18	6	24	Fisher's exact test (two tailed): 0.0478 Prevalence Ratio: 1.50
	No	24	24	48	
	Total	42	30	72	

Attitudes toward an Aborting Dam in the Facility

Licensed breeders were asked what they would do if a pregnant dam aborted in their facility. The question did not imply a suspicion of brucellosis as the cause of the abortion. The majority of respondents (49; 65.3%) would take the dog to a veterinarian. Nine respondents (12.0%) would spay the dog and sell her as a pet, while seven respondents (9.3%) would keep her and try to rebreed her later. Two breeders (2.7%) responded that they would choose to euthanize the dog.

Estimates of Canine Disease Burden

Respondents were asked to recall the number of canine brucellosis tests they had performed, the number of dogs that tested positive and the number of dogs that had been diagnosed with brucellosis by a veterinarian over the past 5 years, and then just in the past 12 months. These questions

were intended to collect information that could be used to estimate testing frequency and disease burden in dogs in Kansas. The raw numbers collected are shown in Table 12.

Table 12 – Number of tests performed and number of dogs diagnosed with brucellosis

	Past 5 years	Past 12 months
Total tests performed	2086	697
Total test-positive dogs	63	18
Total diagnosed dogs	63	18

The results indicate that there were 18 dogs diagnosed with canine brucellosis within the past 12 months and 63 dogs within the last five years among the population of dogs owned by those breeders who responded to the survey. For comparison, KDA-AHD receives an average of 6-12 cases annually reported in Kansas. The majority of breeders (90.6%) did not respond or reported zero dogs tested positive or were diagnosed in the past 5 years or 12 months. Only seven breeders (9.3%) reported dogs that tested positive or were diagnosed in the past 5 years or 12 months. In each of those seven surveys, the reported number of dogs that tested positive was exactly the same as the number of dogs that were diagnosed. At least two of those respondents wrote on their surveys that they were confused about the distinction between dogs that "tested positive" and dogs that were "diagnosed" with canine brucellosis. The impact of this confusion on interpretation of the data will be assessed in the Discussion section.

Sources of Breeding Dogs

One of the principal ways that canine brucellosis can be introduced into a breeding facility is through purchasing replacement dogs that are infected and then introduced into the facility. When asked about sources of replacement animals, almost all breeders responded that they keep some of their own puppies (72 breeders; 96.0%), but 52 (69.3%) breeders also purchase dogs from other breeders and 16 (21.3%) breeders purchase dogs from auction events. Of the seven breeders with positive dogs, five of those purchase dogs from auction events. A Fisher's exact test was used to compare breeders that purchase dogs from auction events with those who reported having at least one

positive dog in the facility in the past five years. Breeders who buy dogs from auctions are 9.2 times as likely to have a positive dog as breeders who do not purchase dogs from auction events.

Table 13 – Comparison of breeders who buy dogs from auction, stratified by those with positive dogs

		Positive Dog(s)		
		Yes	*No*	Total
Buy from auction	*Yes*	5	11	16
	No	2	57	59
	Total	7	68	75

Fisher's exact test (two tailed): 0.0040
Prevalence ratio: 9.2

Performance of Risk or Protective Behaviors

An important means of reducing risk of transmission from dogs to humans is the use of appropriate personal protective equipment while in direct contact with infected dogs or infectious material such as an aborted fetus or vaginal discharge. While gloves were worn about 50% of the time during various cleaning activities, there was almost no report of anyone wearing a mask, goggles or any other protective equipment while engaging in behaviors that could lead to disease transmission, such as assisting with whelping or cleaning whelping areas. Responses are included in Table 14.

Table 14 – Use of Personal Protective Equipment

	Rubber or disposable gloves	Face mask that covers nose and mouth	Goggles that cover eyes
Routine cleaning of cages/runs	35 (46.7%)	2 (2.7%)	0
Helping whelp puppies	37 (49.3%)	0	0
Cleaning the area after whelping	35 (46.7%)	0	0

The relationships between knowledge of the zoonotic potential of canine brucellosis and use of gloves while in contact with potentially infective materials were analyzed using Fisher exact tests to determine if knowledge of zoonotic potential was associated with an increased use of gloves to reduce possible disease transmission. Breeders who knew that brucellosis is zoonotic are not more likely to wear gloves while performing routine cleaning ($p = 0.0971$), helping to whelp puppies ($p = 0.4156$), or when cleaning after whelping ($p = 0.0536$).

Table 15 – Number of respondents who wear gloves, stratified by knowledge of zoonosis

Use gloves – routine cleaning

		Yes	*No*	Total
Knowledge of Zoonosis	*Yes*	25	18	43
	No/Unsure	6	13	19
	Total	31	31	62

Fisher's exact test (two tailed): 0.0971

Use gloves – assist with whelping

		Yes	*No*	Total
Knowledge of Zoonosis	*Yes*	23	19	42
	No/Unsure	8	11	19
	Total	31	30	61

Fisher's exact test (two tailed): 0.4156

Use gloves – clean after whelping

		Yes	*No*	Total
Knowledge of Zoonosis	*Yes*	26	17	43
	No/Unsure	6	13	19
	Total	32	30	62

Fisher's exact test (two tailed): 0.0536

Estimates of Human Disease

As previously discussed, the symptoms of canine brucellosis in humans are vague and may be easily overlooked or misdiagnosed. The survey asked respondents to report any history of symptoms of canine brucellosis in themselves that occurred within two months of any stillbirth or abortion at the breeding facility. The question specifically included the following symptoms: fever, fatigue, night sweats, headache, painful joints, unexplained weight loss, weakness, arthritis or back pain. The question was not applicable if the respondent chose "We have never had a stillbirth or abortion in the kennel" (n = 22) or "I had none of these symptoms" (n = 46). Only one respondent noted having fatigue, but later in the survey revealed a diagnosis of sleep apnea. The survey revealed no evidence of any symptoms that could suggest human illness in those breeders that responded to the survey.

Table 16 – Report of clinical symptoms of canine brucellosis among survey respondents

Question	Response
Within two months of any stillbirth or abortion in your kennel, did you notice any of the following symptoms in yourself? (*Please choose all that apply)*	We have never had a stillbirth or abortion in the kennel: **22 (29.7%)** I had none of these symptoms: **46 (62.2%)** No response: **5 (6.8%)** Fatigue: **1 (1.4%)** Other symptoms*: **0 (0.0%)**

* Zero respondents indicated having fever, night sweats, painful joints, weakness, headache, unexplained weight loss, arthritis or back pain

Response to Symptoms of Human Disease

Respondents were provided with a list of common clinical signs of canine brucellosis infection in humans, and asked what they would do if they noticed any of those symptoms in themselves. Fifty-four respondents (72.0%) would visit a doctor or medical professional. Less commonly, 8 respondents (10.7%) would contact both their doctor and their veterinarian and four people (5.3%) would wait for symptoms to resolve over time (Table 17). One respondent reported that he/she would seek medical care, but preferred holistic remedies to traditional medicine.

Table 17 – Forms of Healthcare Sought for Symptoms of Canine Brucellosis

Question	Response
What would you do if you thought you had symptoms of brucellosis?	Wait for symptoms to resolve over time: **4 (5.3%)** Visit a doctor: **54 (72.0%)** Contact your veterinarian: **1 (1.3%)** Contact both doctor and veterinarian: **8 (10.7%)** Other: **1 (1.3%)**

Brucellosis Reporting in Humans

A total of eleven confirmed, sixteen probable and twenty-eight suspect cases of brucellosis have been reported in Kansas from 1997 to 2012. Only five confirmed cases listed a specific *Brucella* species; four reported *B. melitensis* and one reported *B. abortus* as the specific causative agent.

Analysis of Current Case Investigation Guidelines for *Brucella canis* infection in dogs

KDA-AHD does not currently have a written protocol guiding case investigation upon report of a brucellosis-positive dog in a breeding facility. It is the responsibility of the attending veterinarian and the owner of the breeding facility to develop a plan to eliminate brucellosis from the facility. This plan must be reviewed and approved by KDA-AHD. Additionally, the Animal Health Division does have legal authority to mandate action in response to a case of canine brucellosis under Kansas Statutes (KSA) 47-610 through 47-635.[23] According to Statue 47-610, the "state animal health commissioner is directed to protect the health of domestic animals of the state from all contagious and infectious diseases and for this purpose is hereby authorized and empowered to establish, maintain and enforce such quarantine, sanitary and other regulations as necessary." The commissioner may impose mandatory "disinfection of the premises where a diseased animal or animals" have been housed under Statute 47-634.

Discussion

Knowledge, Attitudes and Practices Survey

Knowledge of canine brucellosis in humans is not nearly as robust as knowledge of the disease in dogs. Nearly all of the respondents (66, 88.0%) had heard of canine brucellosis but only 44 (66.7%) of those who had heard of the disease knew that it was zoonotic. Knowledge of disease transmission and prognosis in dogs was considerable with over 50% of respondents correctly answering each question. However, knowledge of the disease in humans was not nearly as striking with nearly half of breeders responding "Not sure" when asked if canine brucellosis could spread from person to person or if it could be cured in humans. This indicates that breeders are knowledgeable about the disease in dogs but are unaware of the potential health consequences in humans.

Relationships between knowledge of the disease and various demographic factors (household income, education, age, history of abortion) showed no statistically significant relationships. Comparisons of knowledge of the disease in humans and those same four demographic factors showed

one significant association. Respondents with formal education beyond high school were 1.57 times more likely to know that canine brucellosis can be spread to humans than respondents whose formal education ceased upon graduation of high school or earlier.

A Fisher exact test was performed to determine if there was any relationship between knowledge of the zoonotic potential of canine brucellosis and the decision to wear gloves while performing activities that could result in disease transmission, such as assisting in whelping or cleaning after an abortion or stillbirth. Breeders who know the disease is zoonotic are more likely to wear gloves while doing routine cleaning or cleaning after whelping, but this was not statistically significant. It was interesting to note that there was not a difference in proportions ($p = 0.4156$) between breeders who knew that canine brucellosis is zoonotic and those that wore gloves while helping whelp puppies. This could suggest that while breeders are aware of the zoonotic potential, they do not feel that they are at a high enough risk to merit extra measures to protect themselves from infection.

Eighty-one percent of breeders test new dogs before introducing them into the facility, showing that many breeders view brucellosis testing as a valuable preventive measure to avoid introduction of brucellosis into their dog population. There was not a statistically significant relationship between breeders with a history of abortion or stillbirth in the kennel and those who test dogs for brucellosis. Additionally, only 37% of breeders will test dogs that develop fertility problems. This suggests that breeders are more likely to test new dogs than they are to test currently owned dogs. Testing new dogs is beneficial to the breeder because it is a relatively low cost method of preventing the introduction of brucellosis into the kennel. However, testing dogs that are currently in the facility carries a higher risk to the breeder because of the regulatory implications of a positive dog, including mandatory quarantine and testing. These repercussions likely play a role in reluctance to test current breeding animals.

The results indicated that none of the respondents had experienced any of the symptoms of canine brucellosis in themselves within two months of any abortion or stillbirth at their breeding facility.

The flu-like symptoms of canine brucellosis are vague and may be mild and self-limiting in some people. The non-specific clinical presentation could result in underreporting if the symptoms did not merit a high level of concern and thus were not remembered. The incubation period is variable, ranging from 5 to 60 days, with most illnesses occurring within one month of exposure.[17] It may have been several years since the last abortion or stillbirth occurred in a respondent's breeding facility and they may not remember having symptoms or associated them with being around dogs with infertility, stillbirth or abortions.

Brucellosis Reporting in Humans

Brucella canis infections in humans are thought to be rare because approximately fifty cases have been reported in the United States since the discovery of the bacteria in 1966.[4] However, this number is thought to be falsely low for a number of reasons. First, the flu-like, non-specific nature of the clinical disease may not be severe enough to cause patients to visit a healthcare provider. Additionally, patients will always test negative on a standard brucellosis screening test and the healthcare provider may rule out brucellosis as a differential without realizing the need to request a culture to specifically test for *B. canis* infection. Healthcare providers may not realize that dogs can transmit brucellosis. Finally, reports made by state health departments to the CDC are not required to include the etiologic species of *Brucella,* so it is impossible to estimate the true prevalence of *B. canis* specifically from national brucellosis data.[4] The Council of State and Territorial Epidemiologists (CSTE) released a memorandum in 2012 addressing these issues, advocating the development of a serologic assay that can reliably identify antibodies that are specific to *B. canis* in human serum, and recommending that all reports to CDC be required to include the species of *Brucella*.[4] These steps would help to reveal the true burden of disease, as well as guide protocols for disease investigation and response.

Brucellosis Reporting in Dogs

The Kansas Statutes Annotated KSA 47-610 to KSA 47-635 grant the state animal health commissioner authority over regulation of premises housing dogs infected with *Brucella canis* because canine brucellosis is considered a reportable infectious disease. This includes the authority to mandate quarantine and disinfection of the affected premises. Currently there is no case investigation protocol in place to guide management of a case of canine brucellosis in a dog or breeding facility. Creation of such a protocol would help standardize the investigation process and could lead to more complete information collection during the investigation.

Limitations

The survey response rate was 25.5%; this was lower than the desired response rate of 56.8%. The main concern with a low response rate is the introduction of error due to nonresponse, which is the likelihood that there are significant differences between the responders and the non-responders that would bias the survey results. There was very little demographic data on licensed breeders available from the KDA-AHD. Respondent counties were grouped into regions of Kansas and compared to the regions of all 294 licensed breeders. There was not a statistically significant difference, supporting the assertion that the responding population was representative of the entire population of Kansas breeders. The strength of this assertion could have been improved by having more demographic data such as age or sex of the licensed breeders for use in comparing responders and non-responders.

The small number of survey respondents may have also limited the power of the study to determine if there were significant differences between the selected populations compared in the 2x2 tables. A number of the associations had p-values between 0.05 and 0.10 and were deemed not significant. Increasing the number of participants by increased follow-up measures to improve response or involving a larger population of breeders across multiple states could improve the power of the study to determine significant associations in the data. The small size of the study may have been insufficient to find a case of human infection with *Brucella canis*, since the disease is considered rare in humans.

The survey included many retrospective questions, asking respondents to recall past events that may have occurred months or years ago such as abortions in dogs, brucellosis tests performed and symptoms of illness in themselves. Under-reporting could occur if respondents did not perfectly remember a past event or symptoms because they did not seem very significant at the time. Because symptoms of canine brucellosis in humans can be so vague, under-reporting may have played a significant role in the fact that there were no reported symptoms related to canine brucellosis among the respondents. Imperfect recall could also influence the accuracy of the number of tests performed and number of brucellosis positive dogs reported by the respondents.

As with any survey, there is the potential for questions to be misinterpreted by the respondents. There was note of a problem among a few respondents regarding questions 13 through 18 about history of testing dogs for canine brucellosis. Two respondents wrote that they were unsure of the difference between the phrases “tested positive” and “diagnosed” and wrote the same number for each question (questions 14 and 15, and then 17 and 18). The purpose of the distinction in the survey was the fact that screening tests can have a significant false positive rate, so not all dogs that initially test positive on a screening test will ultimately be diagnosed with canine brucellosis. As a result of this confusion, the data is likely incorrect. First, there may be more dogs that tested positive on a screening test than were reported because respondents only reported the number of dogs that were ultimately diagnosed. Second, there may be fewer dogs that are diagnosed as a confirmed case of canine brucellosis because respondents reported all of the dogs that had a positive test result as “diagnosed.” All seven kennel owners that responded wrote the same number for “tested positive” and “diagnosed,” leading to the conclusion that the questions were likely confusing to all the respondents. This unfortunately makes the data from these questions unreliable and the data should not be used alone to determine disease burden in this population.

Respondents reported a total of 18 positive dogs in the past year, and 63 positive dogs over the past 5 years. These numbers are in excess of the estimated 6-12 cases of canine brucellosis that are reported to the KDA-AHD annually. However, KDA-AHD does not have an exact case count of how many suspect or confirmed cases have been reported, so there is no way to truly compare the number of cases reported by the respondents to the number of cases reported to KDA-AHD.

Conclusions

Licensed dog breeders in Kansas are very knowledgeable about canine brucellosis in dogs, but less knowledgeable about the disease in humans. While many breeders know that canine brucellosis is zoonotic, this knowledge does not translate into the use of appropriate personal protective actions such as wearing gloves when in contact with potentially infected dogs. Attitudes toward testing are favorable regarding testing of newly purchased dogs, but testing is much less commonly performed on dogs that are already a part of that breeding facility, even if the dog develops problems with fertility.

Recommendations

We recommend that KDHE and KDA-AHD develop a joint protocol to follow-up on reported cases of canine brucellosis in dogs that may have resulted in human exposure. The protocol should include information about managing infected dogs, but should also make provisions for addressing possible human exposures. A fact sheet should be created that could be provided to breeders by KDA-AHD staff during an investigation that includes symptoms of canine brucellosis in humans, the time period from exposure in which symptoms may develop, what to do if you have any of these symptoms and what to tell your doctor. An example of a possible information sheet is included in this report (Appendix B). An additional fact sheet providing a concise summary about canine brucellosis in both dogs and humans is also included (Appendix C).

Although canine cases are reportable to the KDA-AHD and human cases are reportable to KDHE, there is often an information gap between these two entities, as each agency is responsible for its own investigation. Improved communication between the two departments could help to improve reporting and management of the disease in Kansas. Specifically, KDA-AHD should contact KDHE upon receiving a report of a confirmed or suspect canine brucellosis case in a dog. KDHE would then collaborate with the Animal Health Department on the investigation to interview potential human exposures and follow up with those contacts to determine if they developed symptoms of canine brucellosis in the subsequent weeks to months following exposure.

We recommend that further studies be done to more accurately characterize the true incidence of human infection with canine brucellosis. Performance of serologic testing would be the most accurate way to assess prior exposure to canine brucellosis. Licensed dog breeders are the best population for this study due to their relatively high risk of direct contact with infected dogs shedding high levels of the bacteria, such as occurs during estrus, whelping or abortion. A cohort study could be designed that used positive cases of canine brucellosis in dogs as diagnosed at KSVDL or other laboratories to then trace the human contacts involved in those cases and follow them for disease development.

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Appendix A – Knowledge, Attitudes and Practices Survey

Place an "X" in the ☐ next to your answer choices. The survey should take less than 10 minutes of your time.

Please fill out the survey completely and return it in the enclosed pre-paid envelope by **Monday, August 6th, 2012.** For questions or for more information, please contact Allison Crow at acrow@kdheks.gov or at (785) 296-1059.

1. Have you heard of canine brucellosis?

- ☐ No - - > *skip to question # 10*
- ☐ Yes - - > *continue to question #2*

2. In what ways can canine brucellosis be spread from dog to dog? *(Please choose all that apply)*

- ☐ Contact with vaginal discharge or products of abortion from an infected bitch
- ☐ Contact with urine from an infected male
- ☐ Shared food and water bowls
- ☐ Infection passed from infected mother to puppies during pregnancy or whelping
- ☐ Tick bite
- ☐ Sexual contact during natural breeding
- ☐ Use of artificial insemination
- ☐ Not sure

3. Can dogs that are spayed or neutered get infected with canine brucellosis?

- ☐ No
- ☐ Yes
- ☐ Not sure

4. Is it possible for dogs that have never been bred to have canine brucellosis?

- ☐ No
- ☐ Yes
- ☐ Not sure

5. Can canine brucellosis in dogs be cured?

- ☐ No
- ☐ Yes
- ☐ Not sure

6. Can a dog infected with canine brucellosis spread the disease to humans?

- ☐ No - - > *skip to question #10*
- ☐ Yes - - > *continue to question #7*
- ☐ Not sure - - > *skip to question #10*

7. **How can humans become infected by canine brucellosis?** *(Please choose all that apply)*
   - ☐ Petting or touching a dog's fur
   - ☐ Breathing in the bacteria by nose or mouth
   - ☐ Getting licked on the face or mouth by an infected dog
   - ☐ Direct contact with abortion material or vaginal discharge from an infected bitch
   - ☐ Not sure

8. **Can canine brucellosis spread from person to person?**
   - ☐ No
   - ☐ Yes
   - ☐ Not sure

9. **Can canine brucellosis in humans be cured?**
   - ☐ No
   - ☐ Yes
   - ☐ Not sure

10. **Do you test dogs in your kennel for canine brucellosis?**
    - ☐ No - - > *continue to question #11*
    - ☐ Yes - - > *skip to question #12*

11. **What reasons made you decide to not test your dogs for canine brucellosis?** *(Choose all that apply)*
    - ☐ Did not know about the disease
    - ☐ Do not consider my dogs at risk of getting the disease
    - ☐ No fertility problems in my kennel
    - ☐ Do not want to know if a dog is positive for the disease
    - ☐ Testing takes too much time and effort
    - ☐ Testing is too expensive
    - ☐ Other (please specify) _____

Any answer - - > skip to question 19

12. **When do you test your dogs for brucellosis?** *(Please choose all that apply)*
    - ☐ Test dogs that have fertility problems (such as abortion, stillbirth, failure to conceive)
    - ☐ Test dogs on a veterinarian's recommendation
    - ☐ Test all breeding stock once a year
    - ☐ Test dogs prior to each breeding event
    - ☐ Test new animals before co-mingling them with the other dogs
    - ☐ Other (please specify) _____

13. **In the past 5 years, <u>how many times</u> have you tested your dogs for brucellosis?**
(Please count total number of tests performed, even if multiple tests were performed on the same dog. For example, testing 10 dogs, twice a year, for 5 years would be 10 x 2 x 5 = 100 tests)
_____ tests

14. **In the past 5 years, how many dogs <u>tested positive</u> for canine brucellosis?**
(If no dogs have had a positive test, please answer with the number "0")
_____ dogs

15. **In the past 5 years, how many dogs have been <u>diagnosed</u> with canine brucellosis by a veterinarian?**
(If no dogs have been diagnosed with brucellosis, please answer with the number "0")
_____ dogs

<u>Questions #16-18 are the same as questions #13-15, but apply to only the last 12 months.</u>

16. **In the past 12 months, <u>how many times</u> have you tested your dogs for brucellosis?**
(Please count total number of tests performed, even if multiple tests were performed on the same dog. For example, testing 10 dogs, twice a year, for 5 years would be 10 x 2 x 5 = 100 tests)
_____ tests

17. **In the past 12 months, how many dogs <u>tested positive</u> for canine brucellosis?**
(If no dogs have had a positive test, please answer with the number "0")
_____ dogs

18. **In the past 12 months, how many dogs have been <u>diagnosed</u> with canine brucellosis by a veterinarian?**
(If no dogs have been diagnosed with brucellosis, please answer with the number "0")
_____ dogs

19. **What are the sources of your adult breeding animals?** *(Please choose all that apply)*
- ☐ Buy dogs from other breeders
- ☐ Buy dogs from auction events
- ☐ Keep some of own puppies that are raised for breeding
- ☐ Buy semen and use artificial insemination
- ☐ Other (please specify) _____

20. **Do you regularly quarantine any new dogs before mixing them with current dogs?**
- ☐ No, new dogs are not quarantined - - > *skip to question #22*
- ☐ Some incoming dogs are quarantined - - > *continue to question #21*
- ☐ Yes, all incoming dogs are quarantined - -> *continue to question #21*

21. How long is the quarantine period?

_____ days

22. Are your adult breeding animals (over 6 months) up to date on rabies vaccination?

- ☐ Yes, all dogs are current on rabies vaccination
- ☐ Some dogs are current; others are not current
- ☐ None of the dogs are current on rabies vaccination

23. How many <u>breeding females</u> currently live at your facility? _____ females

"Breeding female" is any female dog over 6 months of age that is currently or is intended to be used for breeding.

24. How many <u>breeding males</u> currently live at your facility? _____ males

"Breeding male" is any intact male dog over 6 months of age that is currently or is intended to be used for breeding.

25. How many litters of puppies are born in one calendar year at your facility? _____ litters

"Litter" can include normal births, stillbirths and abortions

26. Do you use any of the following personal protective equipment while performing the following activities?

(Please place an "X" in the corresponding boxes where applicable)

	Rubber or disposable gloves	Face mask that covers nose and mouth	Goggles that cover eyes	I do not wear any protective equipment
Routine cleaning of cages/runs				
Helping whelp puppies				
Cleaning the area after whelping				

27. Have any of the pregnant females at your kennel ever had an abortion or stillbirth?

"Abortion" is defined as premature delivery of dead puppies.

"Stillbirth" is defined as the delivery of dead puppies after a full-term pregnancy.

- ☐ No
- ☐ Yes
- ☐ I prefer not to answer

28. **Have any of the pregnant females at your facility ever delivered a litter with a mix of dead and live puppies, or delivered live puppies that died within days after birth?**
    - ☐ No
    - ☐ Yes
    - ☐ I prefer not to answer

29. **Have any of the dogs at your facility (males or females) ever developed problems with fertility?**
    - ☐ No
    - ☐ Yes
    - ☐ I prefer not to answer

30. **If a pregnancy were to result in an abortion or stillbirth, what would you do with the bitch after the abortion?**
    - ☐ Have a veterinarian examine the bitch to determine the cause of the abortion
    - ☐ Keep the bitch and try to rebreed her later
    - ☐ Keep the bitch, but no longer use her for breeding
    - ☐ Spay the bitch and sell her or give her away as a pet
    - ☐ Euthanize the bitch
    - ☐ Other (please specify) _____

31. **Within two months of any stillbirth or abortion in your kennel, did you notice any of the following symptoms in <u>yourself</u>?** *(Please choose all that apply)*

    ☐ Fever ☐ Night sweats ☐ Painful joints ☐ Weakness
    ☐ Fatigue ☐ Headache ☐ Unexplained weight loss ☐ Arthritis/ back pain
    ☐ We have never had a stillbirth or abortion in the kennel --> skip to question #36
    ☐ I had none of these symptoms --> skip to question #36

32. **If you developed any of the above symptoms, did you visit a healthcare provider about your symptoms?**
    - ☐ No - - > *skip to question #36*
    - ☐ Yes - - > *continue to question #33*

33. **Did your healthcare provider give you a diagnosis?**
    - ☐ No
    - ☐ Yes, I was diagnosed with _____ *(please fill in diagnosis)*
    - ☐ I don't remember

34. **Were you given or prescribed antibiotics?**
    - ☐ No
    - ☐ Yes
    - ☐ I do not remember

35. Did you completely recover from your illness?

- ☐ No
- ☐ Yes

36. The following are possible symptoms of brucellosis in humans. What would you do if you thought you had symptoms of brucellosis?

Fever	*Night sweats*	*Painful joints*	*Weakness*
Fatigue	*Headache*	*Unexplained weight loss*	*Arthritis or back pain*

- ☐ Wait for symptoms to resolve over time
- ☐ Visit a doctor or other healthcare provider
- ☐ Contact your veterinarian
- ☐ Other (please specify) _____

The remaining questions are about you, the responder. We will pool the responses so that no one individual, or kennel, will be identified.

37. What is your current age? _____ years

38. What is your gender?

- ☐ Male
- ☐ Female
- ☐ Prefer not to answer

39. Are you Hispanic or Latino?

- ☐ Yes
- ☐ No
- ☐ Don't know/ Not sure
- ☐ Prefer not to answer

40. Which one or more of the following would you say is your race? *(Please choose all that apply)*

- ☐ White
- ☐ Black or African American
- ☐ Asian
- ☐ Native Hawaiian or Other Pacific Islander
- ☐ American Indian or Alaska Native
- ☐ Other (please specify) _____
- ☐ Prefer not to answer

41. What is your annual household income?

- ☐ Under $25,000
- ☐ $26,000 - $50,000
- ☐ $51,000 - $75,000
- ☐ $76,000 - $99,000
- ☐ $100,000 or above
- ☐ Prefer not to answer

42. What is the highest level of education you have completed?

- ☐ Elementary school
- ☐ High school diploma
- ☐ Undergraduate college degree
- ☐ Higher education (professional or post-graduate)
- ☐ Apprenticeship or trade school
- ☐ Prefer not to answer

43. Using the county chart below, indicate the region of Kansas where your kennel is located.

Northwest	Barton, Cheyenne, Decatur, Ellis, Gove, Graham, Logan, Ness, Norton, Osborne, Phillips, Rawlins, Rooks, Rush, Russell, Sheridan, Sherman, Smith, Thomas, Trego, Wallace
Southwest	Barber, Clark, Comanche, Edwards, Finney, Ford, Grant, Gray, Greeley, Hamilton, Haskell, Hodgeman, Kearny, Kiowa, Lane, Meade, Morton, Pawnee, Pratt, Scott, Seward, Stafford, Stanton, Stevens, Wichita
North Central	Clay, Cloud, Dickinson, Ellsworth, Geary, Jewell, Lincoln, Marion, McPherson, Mitchell, Morris, Ottawa, Republic, Rice, Riley, Saline, Washington
South Central	Chase, Cowley, Harper, Kingman, Reno
Northeast	Atchison, Brown, Doniphan, Douglas, Marshall, Nemaha, Pottawatomie
Southeast	Allen, Anderson, Bourbon, Chautauqua, Cherokee, Coffey, Crawford, Elk, Greenwood, Labette, Lyon, Montgomery, Neosho, Wilson, Woodson
Kansas City Metropolitan Area	Franklin, Johnson, Leavenworth, Linn, Miami, Wyandotte
Topeka Metropolitan Area	Jackson, Jefferson, Osage, Shawnee, Wabaunsee
Wichita Metropolitan Area	Butler, Harvey, Sedgwick, Sumner

- ☐ Northeast
- ☐ Southeast
- ☐ North Central
- ☐ South Central
- ☐ Northwest
- ☐ Southwest
- ☐ Kansas City Metropolitan Area
- ☐ Topeka Metropolitan Area
- ☐ Wichita Metropolitan Area
- ☐ Prefer not to answer

44. Please share any additional comments here.

_____
_____
_____
_____
_____

Thank you for completing this survey! Your responses are very valuable to our study.

A summary of the results of our survey will be publically available on the KDHE website later this year. Go to http://www.kdheks.gov/epi/index.html and click on the "Investigative Reports" section if you are interested.

Appendix B – Risk Assessment Tool for Canine Brucellosis

Human cases of canine brucellosis are rare, but there have been cases of dogs infected with *Brucella canis* spreading the disease to humans. Please use the following handout to lower your risk of being infected with canine brucellosis, and to watch for any symptoms of the disease that you may develop.

Transmission

The highest risk of transmission is direct contact with an infected dog that has had an abortion or stillbirth, or has a green, mucoid vaginal discharge. These fluids contain very high levels of bacteria. Always wear gloves and protective clothing when helping to whelp puppies or when cleaning, especially if there were complications with the birth such as abortion or delivery of dead puppies that looked swollen, partially decomposed or otherwise abnormal. If you use a high powered water sprayer when cleaning runs, it is advised to wear goggles and a mask, as bacteria can become aerosolized in the water particles and be inhaled. Basic hygiene can go a long way in preventing the spread of this disease from dogs to humans.

Cleaning

The bacteria can be killed by many disinfectants. Cleaning hard surfaces with quaternary ammonium compounds such as Roccal® or iodide solutions can kill the bacteria. Wear gloves whenever you clean.

Symptoms of Disease

Symptoms of disease in humans usually begin 2-4 weeks after exposure to the bacteria and may include fever (above 100^0F), fatigue, night sweats, headache, painful joints, unexplained weight loss, weakness, arthritis or back pain. Use this sheet to note any symptoms you may develop and the dates when they occurred. Continue to monitor for symptoms for at least six months after you were exposed.

Date	Description of Symptoms

If You Develop Symptoms

If you develop any of these symptoms within the next two months, contact your healthcare provider. Take this sheet with you, and let your health care provider know that you have been exposed to dogs that are infected with canine brucellosis.

Appendix C – Canine Brucellosis (*Brucella canis*) for Dog Owners

Importance: Canine brucellosis is caused by the bacterium *Brucella canis*. It can result in significant reproductive and financial losses if introduced into a breeding facility. While rarely reported, canine brucellosis can also cause disease in humans. In Kansas, canine brucellosis in dogs is reportable to the Kansas Department of Agriculture's Animal Health Division (KDA-AHD), and disease in humans is reportable to the Kansas Department of Health and Environment (KDHE).

Signs and Symptoms: Clinical signs can vary widely, contributing to the difficulty in diagnosing this disease. Dogs will test positive within 2 weeks of being infected, but may or may not show clinical signs.

Female dogs: The most commonly recognized symptom is abortion in an otherwise healthy bitch that occurs between 7 and 9 weeks of gestation. Abortion is often followed by a mucoid vaginal discharge lasting 4-6 weeks. Brucellosis can also cause very early embryonic death that can be mistakenly interpreted as a failure to conceive.

Male dogs: The *Brucella* organism is found in the epididymis, testicles and prostate gland and can remain hidden in these tissues for months to years. Sperm abnormalities cause decreased fertility. There may be inflammation of the testicles, or one or both testicles may shrink. This can cause significant pain and discomfort, prompting the dog to frequently lick and irritate the skin of the scrotum.

Non-specific signs in dogs: In addition to reproductive problems, both sexes can exhibit swelling of lymph nodes, loss of libido, and lethargy. Infected dogs may show no clinical signs at all. Rarely, serious complications can occur including inflammation of the eyes, heart disease, or meningitis that could progress to serious injury or death.

Humans: *B. canis* infection in humans causes a flu-like illness with symptoms that include fever, night sweats, weakness, fatigue, headache and painful joints. This disease is rare in humans, but some cases may go undiagnosed because the clinical signs are nonspecific. Symptoms typically begin within 3-4 weeks of exposure, but some infected individuals may show no signs at all.

Transmission: The bacteria can enter the body through inhalation, ingestion and direct contact with mucous membranes or broken skin. The highest risk of transmission involves direct contact with contaminated placenta, aborted fetus, and vaginal discharge from an infected bitch during estrus, whelping or abortion. Semen and urine from male dogs can be a source of infection, even years after initial diagnosis and attempted treatment. Live puppies born to an infected bitch can be infected across the placenta, during birth from ingestion of birthing fluids, or possibly through milk. The bacteria can survive for months on surfaces or in dust and soil in an environment of high humidity, low temperature and no sunlight. There is no evidence that canine brucellosis in humans can be spread from person-to-person.

Other Diseases: There are many causes of abortion in dogs, including bacteria, parasites and viruses. Canine brucellosis should always be a consideration when female dogs have an abortion or stillbirth late in gestation, or male dogs develop inflammation or swelling of the scrotum. Dogs exhibiting those symptoms should always be seen by a veterinarian for proper diagnosis and management.

Diagnosis: Diagnosing canine brucellosis in a dog can be a complex process. No test is perfect, so the results of two (or more) different tests must be used. The first test is an agglutination test that checks for specific antibodies in the dog's blood. Dogs that test negative are considered truly negative at that time. Dogs that test positive may be infected, or may have a false positive result. These dogs will need additional testing to confirm if the dog is infected.

Humans: A special test that is specific for *Brucella canis* must be used. If you develop symptoms of brucellosis or have a history of brucellosis in your kennel, be sure to tell your healthcare provider so that they order the most effective test.

Treatment: Treatment is generally not recommended for dogs in breeding kennels due to poor success rate and the potential for continued spread of the bacteria to other dogs or humans. Extensive antibiotic therapy will reduce the level of bacteria in the blood, but cannot completely eliminate the organism from the body. Many treated animals will appear to recover and have a negative blood test initially, but then relapse and test positive again less than a year later because the bacteria has remained in the body the whole time. Spaying or neutering the animal can reduce but not eliminate the transmission risk. In males, the bacteria remain in the prostate and can be shed intermittently in urine for years.

Current recommendations for elimination of canine brucellosis in a breeding kennel include isolation, testing, and removal of positive animals. Breeders should consult a veterinarian to create a plan to eliminate the disease while minimizing losses.

Humans: Unlike in dogs, it is common for humans to completely eliminate the bacteria after taking antibiotics. Some people may recover without treatment, while others can develop chronic disease, such as heart valve problems, if not properly treated. Fatalities are rare and most deaths are associated with complications such as heart disease and meningitis.

Prevention: Only purchase new dogs from a Brucellosis-free kennel. Isolate and test new dogs twice at least 4 weeks apart before allowing them to join the other dogs. Test all dogs yearly, as well as before any breeding event that occurs outside the facility. Practice good sanitation to prevent disease transmission. The brucellosis bacteria can be killed with disinfectants such as quaternary ammonium compounds and iodides.[7] Always wear personal protective equipment such as gloves and a mask when cleaning after an abortion event or working with infected dogs.

For More Information about Brucellosis in Dogs
Kansas Department of Agriculture
Animal Health Division
(785) 296-2326

For More Information about Brucellosis in Humans
Kansas Department of Health and Environment
Bureau of Epidemiology and Public Health Informatics
(785) 296-1059

Centers for Disease Control and Prevention website: http://www.cdc.gov/brucellosis/
Iowa State PDF: http://www.cfsph.iastate.edu/Factsheets/pdfs/brucellosis_canis.pdf