Targeted Elk Brucellosis Surveillance Project
2017 Annual Report

Executive Summary

Montana Fish, Wildlife & Parks (MFWP) is conducting a multi-year targeted elk brucellosis surveillance project to evaluate 1) prevalence and spatial extent of brucellosis exposure in elk populations, 2) elk spatial overlap with livestock and interchange between elk populations, 3) risk of seropositive elk shedding and potentially transmitting *Brucella abortus*, and 4) effects of brucellosis management hazing and lethal removal on elk distributions and spatial overlap with livestock. This report is an annual summary of the 2017 targeted elk brucellosis surveillance project. In January and February 2017, we sampled a total of 63 elk from populations in the Red Lodge and Paradise Valley areas and screened blood serum for exposure to *B. abortus*. We found elk exposure to *B. abortus* in the Sixmile Creek area of Paradise Valley (29%, n = 42), but did not detect elk exposure to *B. abortus* in the central or northern Red Lodge area (n = 21). Brucellosis surveillance efforts in the Red Lodge area during the past 2 years confirmed the presence of *B. abortus* in the Clarks Fork elk herd that winters along the border of Montana and Wyoming, but not in adjacent Red Lodge area elk herds. We collared a sample of elk in each study area and are currently collecting elk movement information. To evaluate the risk of seropositive elk shedding *B. abortus* during abortion or birth events, we recaptured and assessed the pregnancy status of 14 seropositive elk originally captured and collared in southwest Montana elk populations during 2014 and 2015. We found that 10 of the 14 seropositive elk were pregnant. We outfitted these pregnant elk with vaginal implant transmitters (VITs) to monitor birth events and sampled birth sites for *B. abortus*. We did not detect any abortion events. Two elk lost their VITs due to mechanical failure (i.e., VIT fell out), 1 elk expelled her VIT but no birth site was located, and 2 elk retained their VITs and no birth event was documented. We identified and sampled 5 live birth events and *B. abortus* was detected in a calf carcass at 1 birth site. Following 5 years of monitoring, we euthanized, necropsied, and sampled 1 seropositive elk to estimate the prevalence of active *B. abortus* infections in seropositive elk. In addition, we sampled 1 seropositive elk that was euthanized during capture due to a broken leg. We submitted a comprehensive assortment of tissue samples from these 2 elk for culture testing and *B. abortus* was not detected in either seropositive elk.

Introduction

Montana Fish, Wildlife & Parks (MFWP) has conducted surveillance for brucellosis in elk populations since the early 1980s. Surveillance consists of screening blood serum for antibodies signifying exposure to *Brucella abortus*, the bacteria that causes the disease brucellosis. Brucellosis typically causes abortion in pregnant elk from February through May (Cross et al. 2015) and is primarily transmitted through contact with infected fetuses, birthing fluids and material. Elk that test positive for exposure to *B. abortus* (seropositive) may or may not be actively infected with the bacteria. Although not a true indicator of infection or the ability of an animal to shed *B. abortus* on the landscape, detection of seropositive elk indicates brucellosis is present in the area and indicates the potential for elk to transmit the disease to livestock or other elk.

In efforts to increase understanding of brucellosis in elk populations, MFWP initiated a targeted elk brucellosis surveillance project in 2011. The goals of the project are to 1) evaluate the prevalence and spatial extent of brucellosis exposure in elk populations, 2) document elk movements to evaluate the extent of spatial overlap with livestock and interchange between elk herds, 3) evaluate the risk of seropositive elk shedding and potentially transmitting *B. abortus*, and 4) evaluate the effects of brucellosis management actions, such as hazing and lethal removal, on elk distributions and spatial overlap with livestock. In order to achieve these goals, MFWP has conducted intensive sampling efforts focused on 1 – 2 elk populations per year each year since 2011. Study areas are selected based on their proximity to the known distribution of brucellosis and/or significant livestock concerns. Surveillance areas are identified through collaborative discussions between MFWP, the Montana Department of Livestock (DOL), and landowners. Surveillance areas are both inside and outside of the State of Montana brucellosis designated surveillance area (DSA).

Study areas

Since 2011, we have sampled elk populations from 11 study areas (Figure 1). In February 2017, we sampled elk from 4 herds in the Red Lodge Area (Crow Line, Dry Creek, Silver Run, Grove Creek) and the northern portion of the North Yellowstone population in the Sixmile Creek area of the Paradise Valley. We sampled Red Lodge area elk in 2017 to supplement our 2016 data from this area and increase sample size.

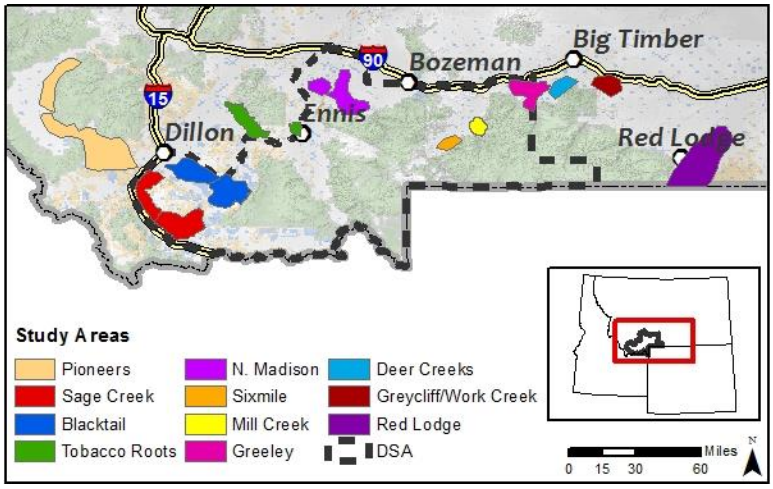

Figure 1. Study areas sampled during the 2011 – 2017 targeted elk brucellosis surveillance project.

Methods

To evaluate *B. abortus* presence and prevalence in the Sixmile Creek and Red Lodge study areas, we captured elk using helicopter netgunning and collected a blood sample to screen animals for exposure. We also opportunistically collected blood samples from hunter harvested animals. Exposure was determined by the presence of antibodies to *B. abortus* in an animal's blood serum. Blood serum samples were tested at the Montana Department of Livestock Diagnostic Lab (Diagnostic Lab). Samples were screened utilizing the Rapid Automated Presumptive (RAP) and Flourescence Polarization Assay (FPA) plate tests. Suspect or reactors to these screening tests were further tested with the FPA tube test. Final classification of serostatus (i.e., seropositive or seronegative) was based on test results received from the Diagnostic Lab.

We collared a sample of elk in the Red Lodge study area to track movements and evaluate risk of brucellosis transmission to livestock and other elk populations. We deployed collars that have a timed-release mechanism that releases the collar after 52 – 72 weeks so that collars may be retrieved and location data downloaded. We deployed 40 collars in the Sixmile study area to evaluate the effects of brucellosis management actions on elk to livestock brucellosis transmission risk. These collars collect a GPS location every 30 minutes during December through April when management actions are most likely to occur, and every 11 hours during May through November in order to conserve battery power, and transmit location data through a satellite service. Collars are expected to function for 3 years. All collars have a mortality sensor that detects if the collar is stationary for > 6 hours.

We recaptured seropositive elk initially detected and collared during the 2011 – 2015 portion of this project. The purpose of maintaining a collared sample of seropositive animals is to monitor serostatus and birth events for 5 years to understand the epidemiology of the disease post-infection, and determine the level of risk associated with exposed elk through time. We retest seropositive elk annually for exposure to determine if elk experience antibody titer loss following exposure. While testing blood serum annually determines if an elk has antibodies for *B. abortus*, lethal removal is the most reliable way to determine if an elk is infected (i.e., capable of transmitting the disease brucellosis) because reproductive organs and lymph nodes need to be collected to culture *B. abortus*. We euthanize seropositive elk following 5 years of monitoring and sample to detect *B. abortus* bacteria using culture testing of tissues. In March 2017, we recaptured and euthanized 1 seropositive elk from the Sage Creek herd that had been monitored for 5 years during 2012 – 2016. The Diagnostic Laboratory performed a necropsy and collected extensive tissue samples (e.g., lymph nodes, organs). Samples were submitted to the National Veterinary Services Lab (NVSL) for culture testing to detect *B. abortus*

bacteria. In addition, 1 seropositive elk from N. Madison broke a leg and was euthanized during capture, and we conducted a full necropsy and tissue sampling for culture testing.

At each of the seropositive elk recapture events, we assessed pregnancy status and outfitted pregnant elk with a VIT to track seropositive elk birth events. VITs are programmed to emit a slow pulse when the temperature is 32^0 C or higher (i.e., inside the body), and emit a fast pulse once the temperature cools below 28^0 C (i.e., expelled outside the body during an abortion or live birth). VITs have a precise event transmitter (PET) code which indicates the time since the VIT was expelled and cooled to a temperature below 28^0 C. We monitored the pulse rate and PET code to determine if an implant had been expelled and the timing of expulsion. To identify birth events, we tracked elk outfitted with VITs every 1 – 2 days from time of capture until the VITs were expelled.

We investigated each birth site to determine if an abortion or live birth occurred and sampled the birth site to determine if *B. abortus* bacteria were shed. We collected birth site samples from the VIT, soil, vegetation, and any available tissue or fluid. We also collected swabs of the VIT and any moist surface or material. All samples were submitted to the Diagnostic Lab to culture (i.e., grow) and identify any bacteria present in the sample. If bacteria cultured from the samples are suspected to be *B. abortus* they are forwarded to the National Veterinary Services Laboratory (NVSL) for final identification. In addition, we submitted a swab of the VIT to the Wyoming State Veterinary Lab for a polymerase chain reaction (PCR) test that detects *B. abortus* DNA and can detect bacteria that is no longer viable (i.e., died from exposure before sampling). The PCR method allows for detection of dead bacteria that would not be detected in culture testing of tissues. The PCR test is a new method of detecting *B. abortus* that was unavailable before

2015. Detection of *B. abortus* from any sample, via culture or PCR, led to the classification of detected for that event. We categorized each birth site as *B. abortus* "detected" or "not detected" based on culture and PCR results. We considered elk that gave birth on or after May 15 to have carried their calf to full term, unless evidence of an abortion event was detected at the birth site (Barbknecht et al. 2009, Cross et al. 2015). We monitored the adult elk post-calving to confirm the presence of a live calf whenever possible. We categorized birth events as a confirmed abortion, suspected abortion, confirmed live birth, suspected live birth, or unknown. We defined a confirmed abortion as a birth event when the fetus was located and a suspected abortion as a birth event occurring outside of the normal calving period (May 15 – June 30) when no fetus was located at the birth site. We defined a confirmed live birth as a birth event where a live calf was located at the birth site or observed with the adult female, and a suspected live birth as a birth event occurring during the normal calving period (May 15 – June 30) where no fetal material or live calf was observed. Unknown events were restricted to cases where the VIT was lost due to a malfunction (i.e., stopped transmitting), the VIT was expelled but not at a birth site (i.e., mechanical failure of the VIT), or when no birth event was detected and the elk retained the VIT.

To evaluate the effects of brucellosis management hazing and lethal removal on elk distributions and spatial overlap with livestock, we monitored both elk movements and brucellosis management actions in the Sixmile Creek area. During 2017, brucellosis management included hazing elk from high-risk areas. Hazers conducting brucellosis management carried GPS units and recorded track logs during each elk hazing event. We will evaluate the effects of brucellosis management actions on elk movements to determine the distance and amount of time elk stayed away from high-risk areas.

Results

Brucellosis surveillance

In February 2017, we sampled 20 elk from the Red Lodge area. This area includes 5 elk herds, with semi-distinct core winter ranges (Figure 2). Core winter ranges were defined based on a combination of elk movement data, annual winter surveys, and knowledge of the regional biologist. In the Crow Line herd, 0 of 8 elk tested positive for exposure to *B. abortus* and we deployed collars on 4 elk (Table 1). In the Dry Creek herd, 0 of 5 elk tested positive for exposure to *B. abortus* and we deployed collars on 3 elk. In the Silver Run herd, 0 of 4 elk tested positive for exposure to *B. abortus* and we deployed collars on 0 elk. In the Grove Creek herd, 0 of 3 elk tested positive for exposure to *B. abortus* (Table 1) and we deployed collars on 3 elk. Together with surveillance data collected in 2016, estimated seroprevalence and 95% confidence intervals ranged from 0% in the Grove Creek, Silver Run, Dry Creek, and Crow Line herds to 43% (95% CI = 21-67%) in the Clarks Fork herd (Figure 2, Table 1). Location data for these Red Lodge area elk is limited to flights every 1 – 2 months until the collars drop off in Spring 2018. To date, collared elk from the Silver Run and Dry Creek herds have remained in the same general area of their capture sites, and elk captured near Crow Line have moved northwest. We sampled 42 elk from the Sixmile Creek area in the Paradise Valley

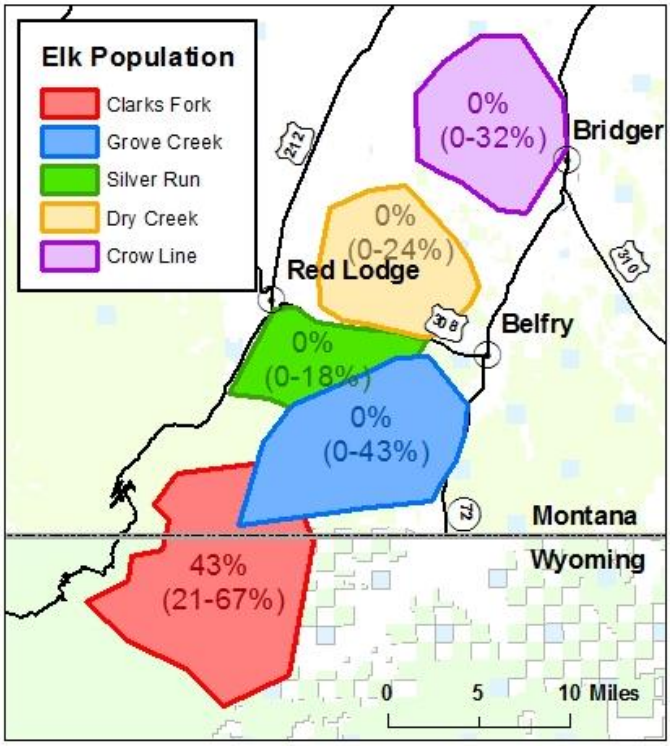

Figure 2. Estimated brucellosis seroprevalence and 95% confidence interval in the five Red Lodge area elk herds sampled during 2016 – 2017. Herd polygons reflect approximate core winter range based on collar data as well as different ecologies of these elk herds.

(Table 1). Twelve of 42 elk tested positive for exposure to *B. abortus* and we deployed collars on 40 elk.

Table 1. The study areas where elk were screened for exposure to *B. abortus* during February 2016 – 2017, sample size, number of elk testing seropositive for exposure, and the estimated seroprevalence with 95% confidence intervals.

Study Area	Herd	Hunting Districts	Sample Size	Number Seropositive	Estimated Seroprevalence
Red Lodge	Crow Line	502	8	0	0 (0, 0.32)
Red Lodge	Dry Creek	502	12	0	0 (0, 0.24)
Red Lodge	Silver Run	520	18*	0	0 (0, 0.18)
Red Lodge	Grove Creek	520	5	0	0 (0, 0.43)
Red Lodge	Clarks Fork	520	14	6	0.43 (0.21, 0.67)
Sixmile	N. Yellowstone	313, 317	42	12	0.29 (0.17, 0.44)

*Includes 4 hunter-harvest samples

Combining data from the last 10 years of hunter harvest samples and sampling from brucellosis surveillance captures of elk, we estimate brucellosis seroprevalence in elk varies spatially across southwest Montana and ranges from 0 – 38% (Figure 3).

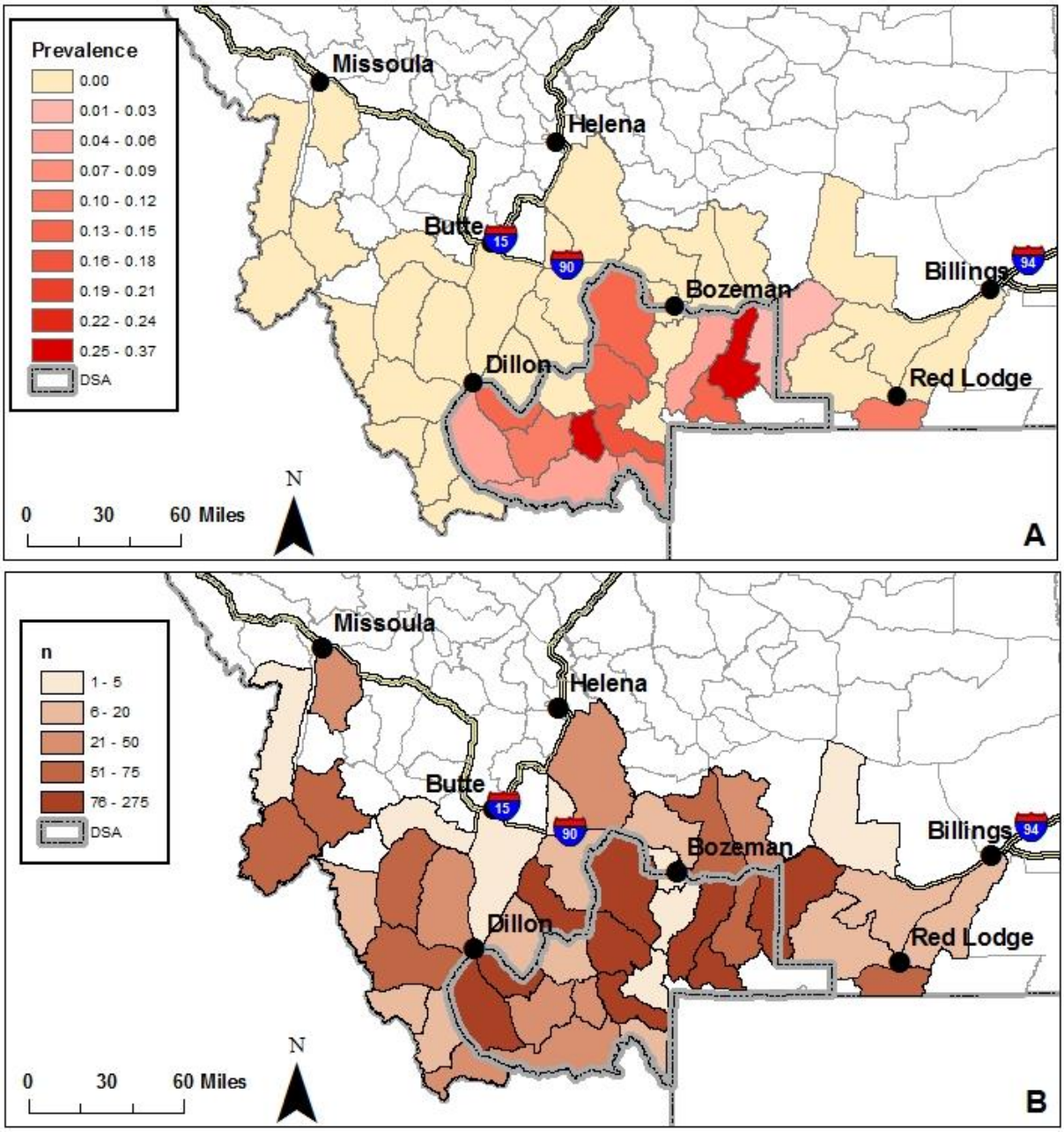

Figure 3. The estimated brucellosis seroprevalence (Panel A) and number of samples screened (n, Panel B) for adult female elk by hunting district* during 2007 – 2016. Samples include those collected during winter research captures and fall hunter harvest. Note some seroprevalence estimates are derived from a low number of samples. The gray line denotes the boundary of the Montana designated brucellosis surveillance area (DSA). *Hunt district 520, west of Red Lodge, is divided in two along a legally defined sub-district boundary to reflect the limited sampling in the northwestern portion of the district.

Elk movements

In January 2016, we deployed 11 collars in the southern portion and 13 collars in the central portion of the Red Lodge study area (Figure 4). We assigned animals to herd based on a combination of the capture location (i.e., which core winter range the animal was captured within) and movement patterns (i.e., location data collected from collars). We recovered data from 19 of 24 collars from Red Lodge. Two collared elk died in the Red Lodge area, one from Clarks Fork in April 2016 and 1 from Silver Run in early June 2016. These mortality events resulted in limited movement data, and these individuals are not included in summaries of movement data. Five collars are still deployed on Red Lodge elk due to failure of the timed-release drop-off mechanisms.

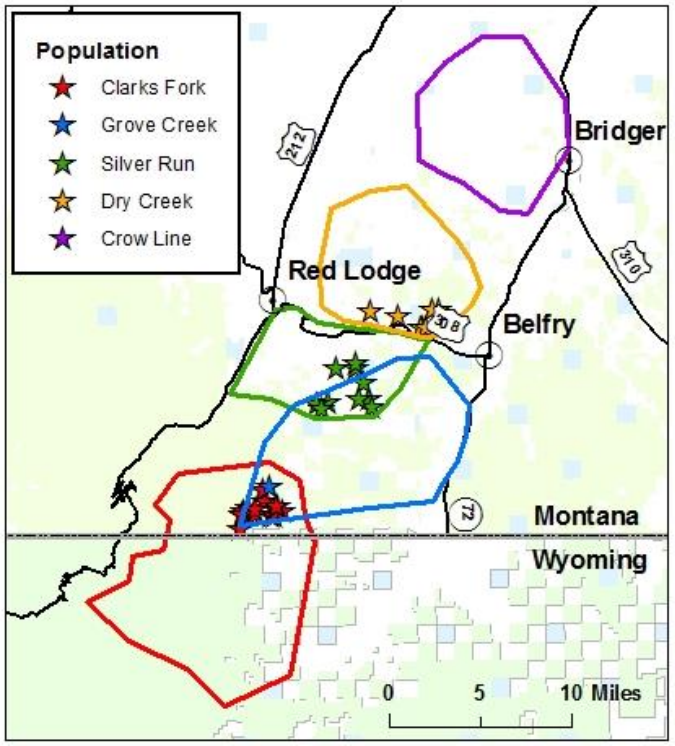

Figure 4. Red Lodge area collar deployment locations in January 2016 (stars) and approximate boundaries of elk population winter ranges (polygons).

In the Red Lodge area, we recovered collar location data from 8 Clarks Fork, 2 Grove Creek, 5 Silver Run, and 2 Dry Creek elk. In general, Clarks Fork elk winter in the foothills of the Beartooth Mountains on both sides of the MT-WY border near Line Creek (Figure 5A). All 8 collared Clarks Fork elk migrated south into Wyoming for the summer. Five of those elk spent portions of the summer in Montana within 3 miles of the MT-WY border near Line Creek. Movement data from 2 collared elk captured within the Clarks Fork herd winter range indicate that they are from the Grove Creek herd. These 2 Grove Creek elk moved northeast after capture and summered along Grove Creek in Montana (Figure 5B). Grove Creek elk are nonmigratory and associated with agricultural lands throughout the year, whereas Clarks Fork elk are migratory. One Grove Creek elk moved farther north into the Dry

Creek and then Crow Line areas in fall 2016, where she remained until her collar dropped off in March 2017.

Silver Run elk typically wintered east of Red Lodge and south of MT-308. In the spring, all 5 collared Silver Run elk migrated southwest into the Beartooth Mountains between West Fork Rock Creek and Lake Fork Creek (Figure 5C). Most remained west of highway US-212 into the fall. Both collared elk from Dry Creek are non-migratory and remained east of US-212 year-round (Figure 5D). These elk stayed primarily north of MT-308 near the headwaters of both the North and South Fork of Dry Creek, venturing north to Sand Creek in early Spring and late Fall. Both spent portions of February and March 2016 south of MT-308. Dry Creek elk tend to remain east of Red Lodge and north of MT-308 year-round.

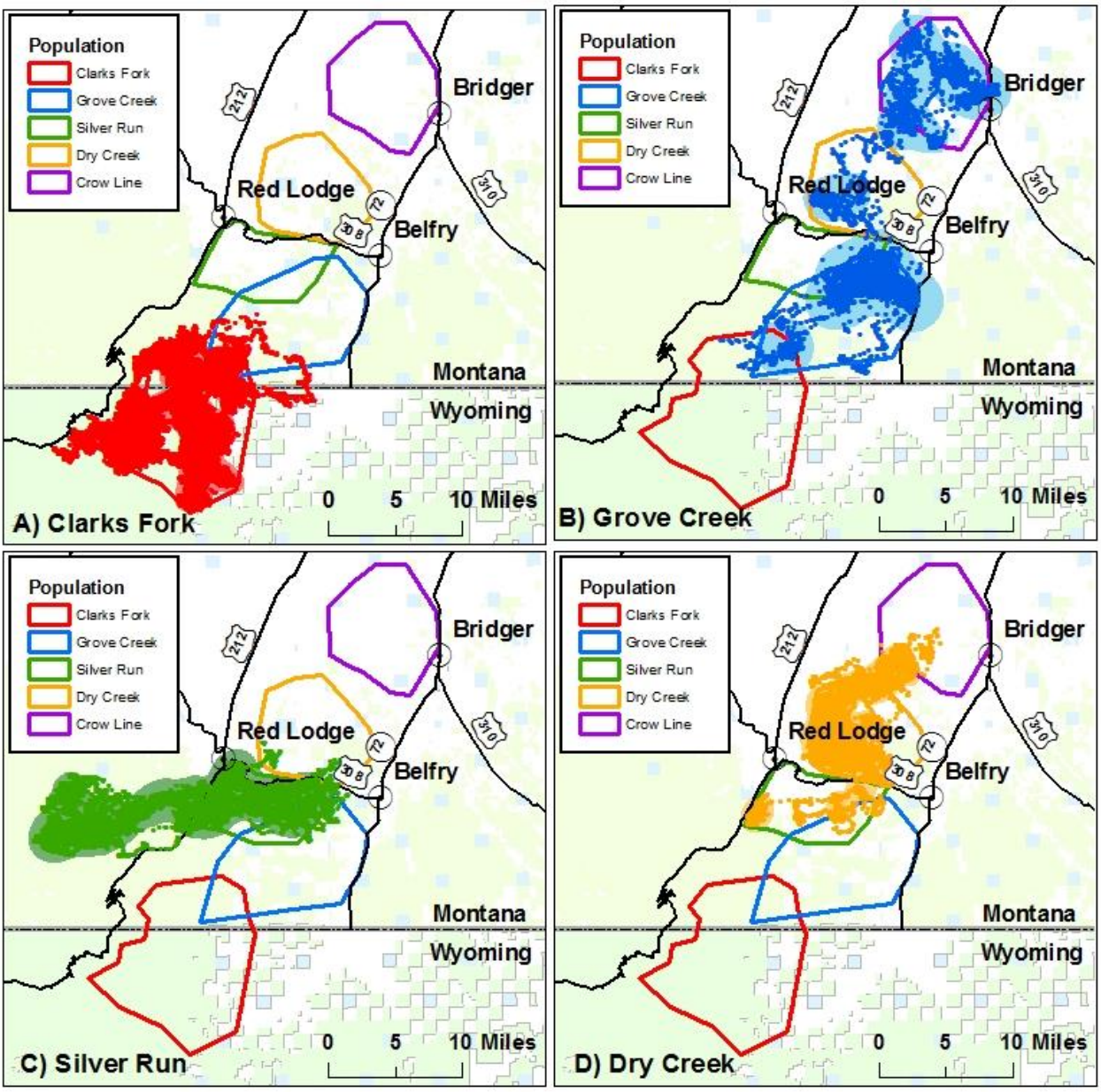

Figure 5. Annual locations (circles) and a 95% kernel utilization distribution (shaded area) of elk from the Clarks Fork (Panel A), Grove Creek (Panel B), Silver Run (Panel C), and Dry Creek (Panel D) herds in the Red Lodge study area. Elk in the Crow Line herd are currently collared and movement data will be available in 2018.

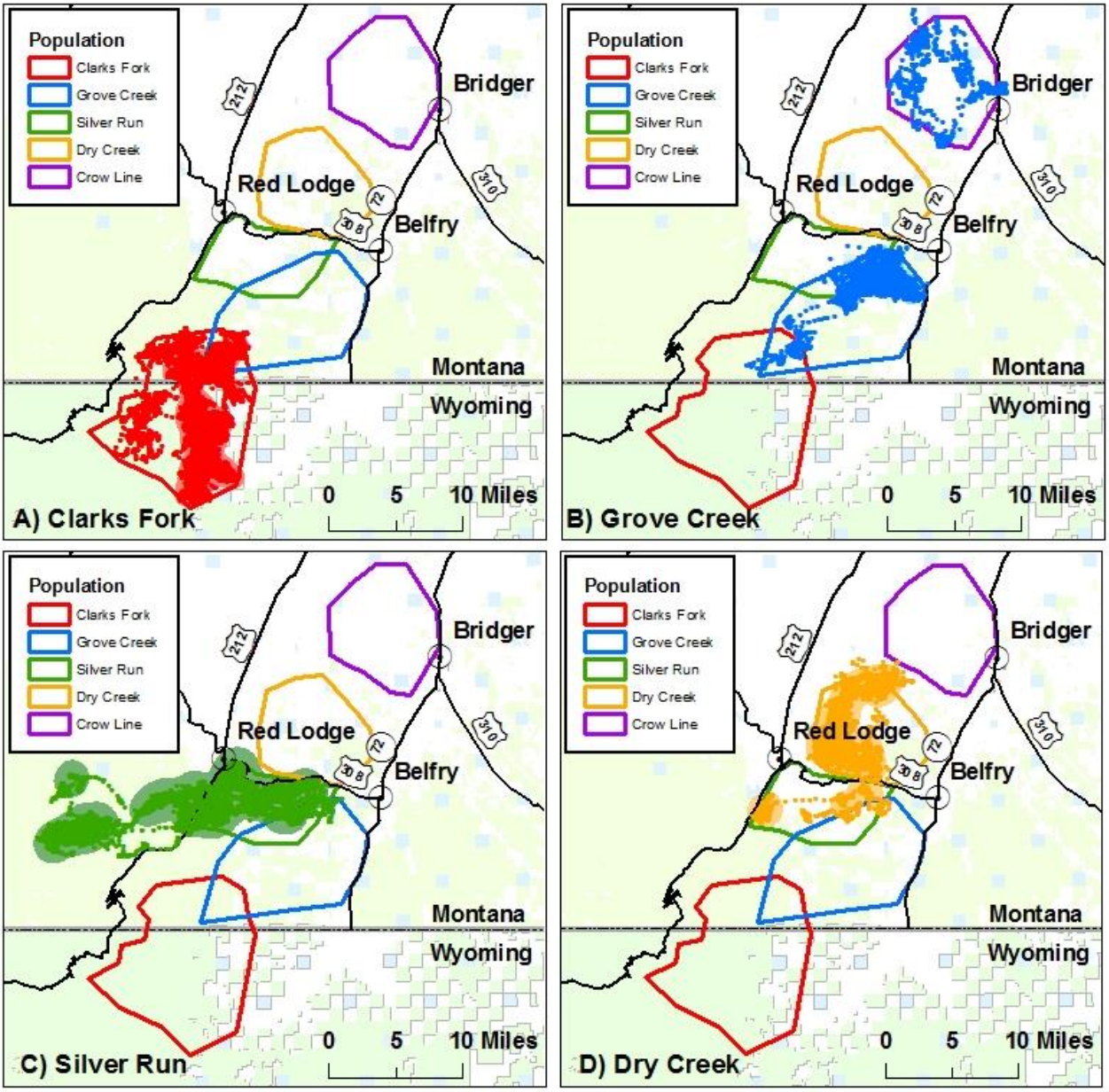

Figure 6. Risk period (Feb-June) locations (circles) and a 95% kernel utilization distribution (shaded area) of elk from the Clarks Fork (Panel A), Grove Creek (Panel B), Silver Run (Panel C), and Dry Creek (Panel D) herds in the Red Lodge study area.

During the February through June risk period, Clarks Fork elk were located in Wyoming and up to 4 miles north into Montana (Figure 6A). Grove Creek elk spent most of the risk period along Grove Creek and Wolf Creek near Belfry, MT (Figure 6B). One of the 2 collared elk moved southwest towards Line Creek, but returned to the Grove Creek area. The other collared elk from the Grove Creek herd moved northwest of Belfry, MT in the fall of 2016 and remained there into the 2017 risk season. Silver Run elk resided on their winter and early summer range during the risk season, spreading from Wolf Creek to the West Fork of Rock Creek (Figure 6C). Dry Creek elk spent most of the risk season north of MT-308, but did spend part of February and March south of Red Lodge and MT-308 (Figure 6D).

Additionally, we deployed 1 collar in Work Creek, 2 collars in Greycliff, Creek and 4 collars in Deer Creeks in 2016 and recovered data from these collars in 2017. We recovered data from 1 collar from Greycliff Creek and 2 collars from Deer Creeks. One Deer Creeks collar failed shortly after deployment resulting in no movement data, and another failed to drop off. One collar failed to drop off in both Greycliff Creek and Work Creek. Deer Creeks elk spend most of the year between the Boulder River and the West Fork Upper Deer Creek, from Happy Jack Gulch south to Enos Creek, generally moving farther south in summer (Figure 7A). In October, both collared elk spent portions of October west of the Boulder River and 1 elk moved to Coal Mine Rim west of the Boulder River in December and stayed in that area until her collar fell off in March 2017. During the risk period (Figure 7B), both Deer Creeks elk stayed east of the Boulder River. Movement data from 1 collar in Greycliff Creek indicate a non-migratory population that resides year-round south of I-90 between Lower Deer Creek and Bridger Creek.

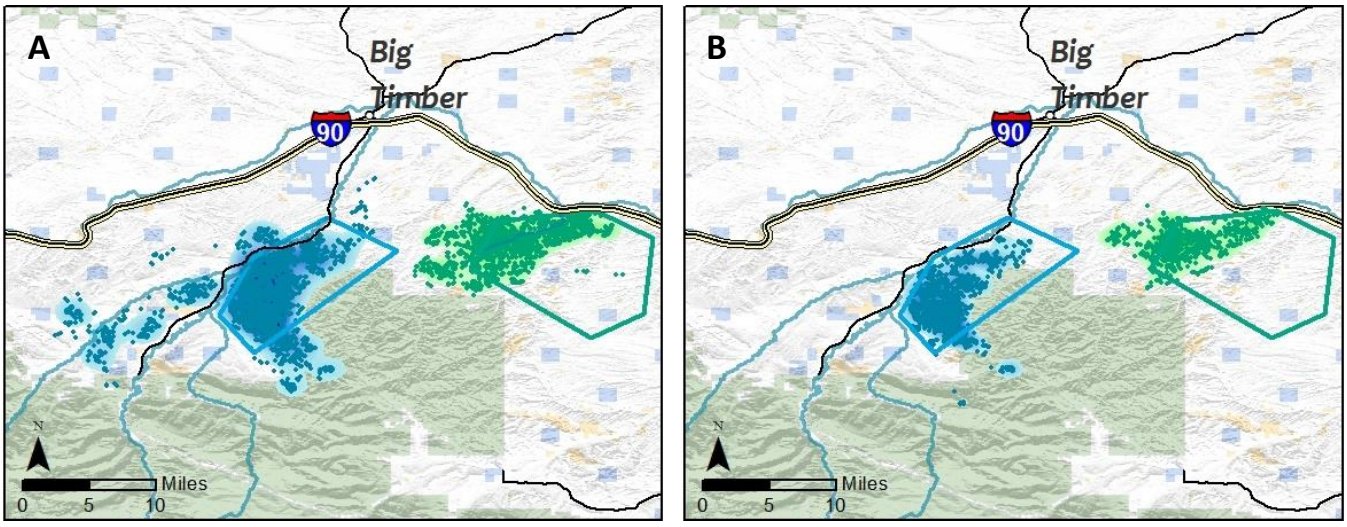

Figure 7. Annual (Panel A, Feb 2016 – Mar 2017) and risk period (Panel B, 15 Feb – 30 Jun) locations and a 95% kernel utilization distribution of 2 Deer Creeks (blue) and 1 Greycliff Creek (green) elk. Polygons represent core population level winter use areas.

We outfitted 40 Sixmile Creek elk with satellite uplink GPS collars that provide real time location data. One elk died of winter kill on 3/29/17 and the GPS function on 4 collars failed in May and June. We are currently collecting location data from 35 elk (Figure 8). Of these 35 elk, 4 elk moved west of highway US-89 in March, while the remaining 31 wintered around Dailey Lake through April. Migration began in May and continued through June. Three elk remained west of Highway US-89 near Rock Creek and the Yellowstone River in June, and another 3 elk remained near Sixmile Creek in the foothills. Four elk migrated to Gardiner, and 1 elk migrated southwest to Hebgen Lake. The remaining 24 elk migrated farther south, primarily deeper into

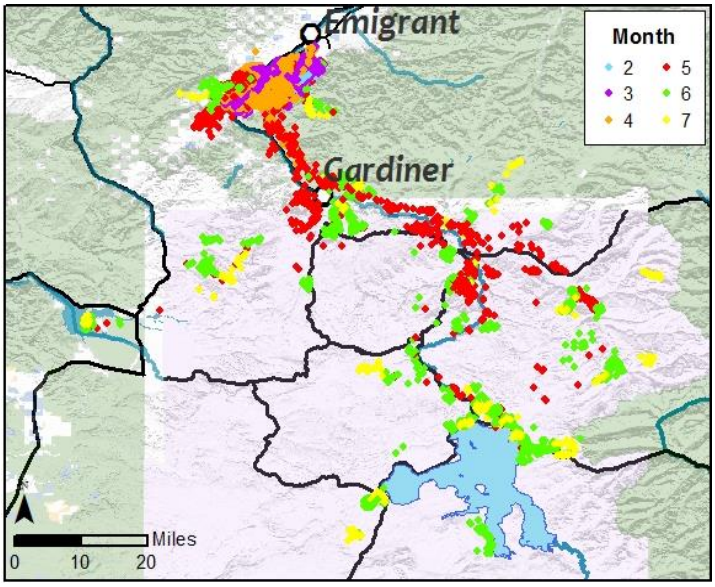

Figure 8. Sixmile elk collar locations by month through early July 2017.

Yellowstone National Park, spreading out from the Washburn Range near Tower Junction to the Mirror Plateau and Yellowstone Lake.

Management hazing in the Sixmile Creek area occurred throughout the winter to move elk off private property with cattle (Table 2). No brucellosis hunts were implemented during winter-spring 2016 – 2017. We will continue to monitor brucellosis management actions and elk responses to management actions throughout the next two years.

Table 2. Number of brucellosis management hazing events in the Sixmile Creek area by month for winter 2016 – 2017.

	Dec	Jan	Feb	Mar	Apr	May	Jun
Hazing	9	20	15	6	3	7	0

Seropositive elk recapture and sampling

During February and March 2017, we recaptured 14 seropositive elk from Northern Madison (n = 4), Mill Creek (n = 8), and Greeley (n = 2). We were unable to recapture 1 seropositive elk from Mill Creek. Ten of these recaptured, seropositive elk were pregnant and were outfitted with VITs, while the remaining 4 recaptured, seropositive elk were open and not outfitted with VITs. Two VITs fell out shortly after capture due to structural failure of the VIT. We monitored 8 seropositive elk pregnancies through the entire parturition season and documented 4 confirmed live births, 1 suspected live birth, and 3 unknowns (1 VIT expelled but no birth site detected or calf subsequently seen, 2 elk retained the VIT and no birth event detected; Table 3). We investigated 5 birth sites within 3 days of the birth event, with 3 of 5 events investigated within 24 hours. PCR testing of VITs at the 5 birth sites did not detect *B. abortus*. Culture testing of grass, soil and fluids at the 5 birth sites did not detect *B. abortus*. One calf was born alive, but died within 24 hours. The carcass was collected and submitted for necropsy and tissue sampling. We detected *B. abortus* in the lung tissue of this newborn calf,

although *B. abortus* was not detected in the environmental samples from the birth site submitted for culture or the PCR test of the VIT.

Table 3. The total number of 2017 seropositive elk pregnancies monitored by study area, and the number and type of birth events documented. The 2 VITs that fell out due to structural failure of the VIT are not included.

Study Area	Total Monitored	Abortion		Live Birth		Unknown
		Confirmed	Suspected	Confirmed	Suspected	
N. Madison	1	0	0	0	0	1*
Mill Creek	5	0	0	3	0	2^
Greeley	2	0	0	1	1	0
TOTAL	**8**	**0**	**0**	**4**	**1**	**3**

* The VIT antenna broke and the signal does not transmit.
^ One elk retained her VIT throughout the calving period. The other VIT was found on a game trail with no sign of a birth event.

B. abortus was not detected in the tissue sampling of the 1 Sage Creek elk or the 1 North Madison elk that were euthanized and collected for *B. abortus* culture testing. Culture testing was conducted on 28 samples from the Sage Creek elk and 26 samples from the North Madison elk. Tissue samples from both elk submitted for culture testing included: lymph nodes (supramammary, popliteal, prefemoral, prescapular, iliac, hepatic, mesenteric, bronchial, parotid, mandibular, retropharyngeal), organs (kidney, liver, spleen, ileum), reproductive tract (ovary, mammary gland, placental cotyledon, fetus, placenta, amniotic fluid), swabs (vaginal, rectal, uterine) and feces. Additional samples submitted from the Sage Creek elk included abomasal fluid, uterine wall and cervix. Two distinct samples of amniotic fluid with different coloration were submitted from the North Madison elk. The annual serology results for these elk show that both remained seropositive throughout their monitoring period (Table 4). From 2012 – 2016, we documented a combination of live births and years of being not pregnant for these elk (Table 5). No abortions were documented and *B. abortus* was not detected at birth sites associated with either of these elk.

Table 4. Annual serology results for two euthanized seropositive elk by individual and year, 2012 – 2017.

ElkID	Study Area	2012	2013	2014	2015	2016	2017
SC50	Sage Creek	Pos	Pos	Pos	Pos	Pos	Pos
BF04	N. Madison	---	---	Pos	Pos	Pos	Pos

Table 5. Annual pregnancy and/or birth event results for two euthanized seropositive elk by individual and year, 2012 – 2017.

ElkID	Study Area	2012	2013	2014	2015	2016	2017
SC50	Sage Creek	Live Birth	Open	Live Birth	Live Birth	Open	Preg
BF04	N. Madison	---	---	Live Birth	Open	Live Birth	Preg

Discussion

Our targeted brucellosis surveillance efforts in the Red Lodge area during 2016 and 2017 confirmed the presence of brucellosis in the portion of the Clarks Fork elk herd that winters along the border of Montana and Wyoming (Scurlock and Edwards 2010). However, to date, brucellosis has not been detected in the adjacent elk herd units in the Red Lodge area of Montana. Elk movement data shows that there is interchange among elk herds in the Red Lodge area, and some spatial overlap between the Clarks Fork and Grove Creek elk herds during the risk period. These movements identify the potential for northward brucellosis expansion in Red Lodge area elk herd units, and continued surveillance efforts in this area are warranted. MFWP will continue to collect blood samples and will encourage hunters to participate in collection of samples from harvested antlerless elk in this area.

Brucellosis surveillance efforts confirmed the presence of *B. abortus* in elk from the Sixmile Creek area in Paradise Valley. *B. abortus* was first documented in elk in this area in 1991 and subsequent testing has documented an apparent increase in seroprevalence. During the 1990's brucellosis seroprevalence in the northern Yellowstone elk population within HD313 was estimated at 1-5% annually (n = 176 to 527). In the early 2000's, testing estimated prevalence at 5-14% annually, (n = 70 to 287). In 2016, this area had an estimated seroprevalence of 9% (n = 32, 95% CI: 3 – 24%).

Estimates from specific years vary and different segments of the northern Yellowstone elk population may have different levels of seroprevalence. Our prevalence estimate of 29% (n = 42, 95% CI = 17 – 44%) from elk in the northern portion of this population's winter range in 2017 is higher than the 2016 estimate. The difference in seroprevalence estimates from the northern portion of the population sampled in 2017 and the overall population sample in 2016 suggests that seroprevalence is not uniform across the entire population. In 2016, samples were distributed across the entire range of the population, including areas adjacent to Yellowstone National Park that were not sampled in 2017. Seroprevalence estimates generated from samples collected from different segments of the population may not be directly comparable.

Our epidemiological results from 2017 are similar to results from 2011 – 2016 and suggest that only a small proportion of seropositive elk are shedding *B. abortus* bacteria and pose a risk for transmitting the disease to livestock or other elk. We have observed 3 abortion events out of a total of 61 (4.9%) known-fate birth events from 30 individual elk, and *B. abortus* was present at each of these 3 abortion sites. The abortion events occurred on 30 March 2014, 20 April 2012 and 14 May 2012. These dates fall within the riskiest time of year, March through mid-May (Cross et al. 2015). Additionally, since 2011, *B. abortus* was detected at 2 of 58 live birth events (3.4%) from 29 individual elk, suggesting that live births pose some limited risk for transmission, although these cases are rare. The Wyoming Game and Fish Department (WGFD) have similarly detected *B. abortus* at 5 out of 118 (4%) live birth events (B. Scurlock, personal communication, August 2016). Although time to detection and sampling efforts did not differ between abortions and live birth events, typical female elk behavior during live birth events (i.e., consumption of birth material and vegetation) may remove some of the *B. abortus* shed at a live birth event.

The sampling and culture testing of the 2 euthanized, seropositive elk in 2017 did not detect *B. abortus* in any tissues. Full necropsy and testing has been performed on a total of 9 seropositive elk since 2016 and we have examined a total of 214 tissue samples. In 2016, we detected *B. abortus* in 1 of 22 tissue samples from 1 seropositive elk from the N. Madison study area, and we have been unable to culture *B. abortus* from all other tissue samples. Our limited detection from tissues of seropositive elk suggests that (1) *B. abortus* is difficult to detect using culture, and (2) seropositive individuals do not harbor widespread infections of *B. abortus*. Even if *B. abortus* is difficult to culture, given the considerable number of samples collected and tested from these seropositive elk, it is likely that at least some of them were not actively infected at the time of their death. It should be noted that this does not mean these elk posed no transmission risk over the previous 5 years, or prior to inclusion in this study. They could have been actively infected in previous years.

Data from GPS collars has improved our understanding of elk movement and potential routes for the spatial spread of brucellosis or other diseases among elk populations (Figure 9). Elk movements will be used to determine the timing and degree of spatial overlap between elk and livestock in future focused analyses.

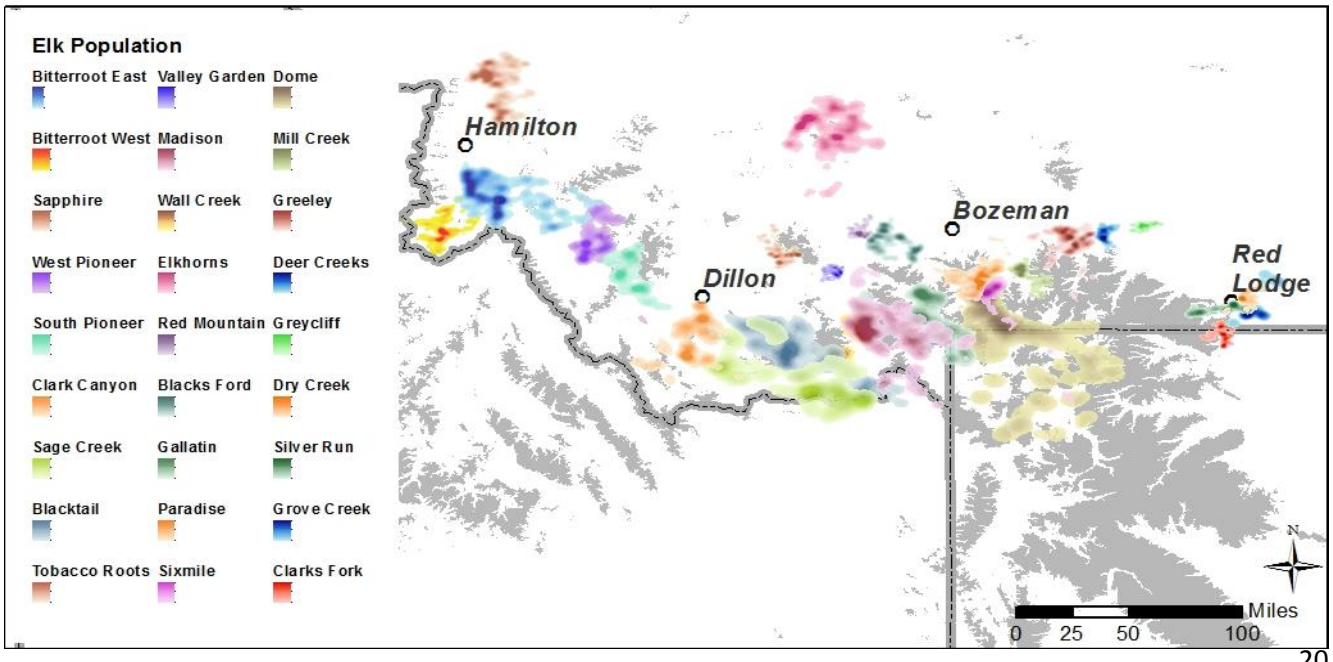

Figure 9. Annual kernel density distributions of elk herds in SW Montana with GPS collar data showing the potential overlap and interchange between herds. Gray polygons represent

Next Steps

Over the next two years, we plan to continue brucellosis surveillance efforts in the Tendoy Mountains area south of Dillon, Montana in Hunt Districts (HD) 300, 302 and 328 just west of the DSA boundary (Figure 10). The focus of the next 3 years of effort will be to 1) continue to document the spatial extent of the disease, 2) to integrate the exposure, movement and epidemiology data to predict the risk of transmission from elk to livestock, and 3) to evaluate the effectiveness of elk management actions designed to affect elk distribution and elk-cattle spatial overlap at reducing transmission risk within the DSA. For seropositive elk captured prior to 2016, we will continue to monitor their serology, movement and birth events. After five years, seropositive elk will be euthanized and tissues cultured to determine if they are actively infected with brucellosis. Seropositive elk in the remaining areas will be euthanized in 2019 (N. Madison) and 2020 (Mill Creek, Greeley).

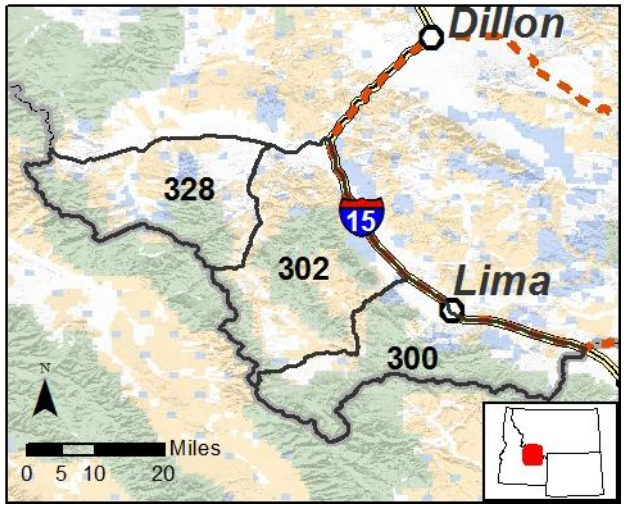

Figure 10. Planned sampling areas for 2018 and 2019 in the Tendoy Mountains west of Lima, MT near the Montana – Idaho border.

The primary goal of this project is to provide wildlife and livestock managers with information useful for designing strategies to reduce the risk of brucellosis transmission from elk to livestock. Transmission risk is a complex combination of elk seroprevalence, infection, population size, pregnancy rates, associated risk of shedding from abortions and live births, and the spatial overlap of elk and livestock during the risk period. Seroprevalence, epidemiology and elk movement data collected during the first five years of this project will be integrated with livestock distribution maps to develop a risk model that will quantify the actual risk of transmission across space and time within the

DSA. With this model, the riskiest areas based on spatial and temporal overlap between elk and livestock can be identified and prioritized for management actions designed to reduce transmission risk. Management actions can then target these risky areas for more effective resource allocation.

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