INVENTORY AND MONITORING OF AMPHIBIANS AND Reptiles in the Powder River Basin Area of Wyoming

FINAL REPORT 2010

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EXECUTIVE SUMMARY

Coal Bed Natural Gas (CBNG) development in the Powder River Basin (PRB), Wyoming, continues to expand. The effect of CBNG development on local amphibians and reptiles is largely unknown, but possible impacts could result from changes in water quality, changes in water flow regimes, and increases in road densities and traffic volume. The Aquatic Task Group (ATG) is an inter-agency working group focused on studying and mitigating impacts of CBNG development in northeastern Wyoming and southeastern Montana. The Wyoming Natural Diversity Database (WYNDD) has been working with the Buffalo Field Office of the BLM, a member of the ATG, since 2008 to conduct an inventory and develop a monitoring plan for amphibians and reptiles in the PRB. The goal of this project is to determine the current status of amphibians and reptiles in the PRB so that future monitoring can detect changes, if any, in species composition, reproductive success, and occupancy rates due to CBNG development. We conducted several types of surveys to inventory amphibians and reptiles from 2008-2010 and to began monitoring efforts in 2010. Survey methods included visual encounter surveys of water bodies, roadkill/basking surveys, nocturnal call surveys, and visual encounter surveys of rock outcrops. We also measured water quality and/or habitat characteristics at all sites.

Over the course of the project, we have documented 19 (6 amphibian, 13 reptile) of the 21 species thought to possibly occur in the PRB, with species detection varying by survey method. The only species not confirmed that might occur in the PRB are the Bullfrog, an invasive species in Wyoming, and the Northern Prairie Lizard. In 2010, we detected 2 Pale Milksnakes, a species for which confirmed observations previously were lacking in the PRB. Tiger Salamanders and Boreal Chorus Frogs were the most common amphibian species detected, but Northern Leopard Frogs were also estimated to occupy 42% of water bodies. Bullsnake and Prairie Rattlesnakes were the most common snakes detected on roads and at rock outcroppings. Gartersnakes were typically found during VES surveys along water bodies. Northern Sagebrush Lizards were detected throughout the PRB at most rock outcrops surveyed.

We recorded road mortality along set routes at previously identified "roadkill hotspots" as well as incidental findings of amphibian and reptiles killed by vehicles. Bullsnakes and Prairie rattlesnakes were most frequently found dead on roads and average roadkill rate was 3.2 mortalities/mile at the 5 "hotspots". However, 88% of mortalities were documented on 2 stretches of SR338 next to the Welch Management Area.

We continued to collect data on known amphibian pathogens in the PRB. Large numbers of dead Tiger Salamanders have been observed in the PRB since surveys began in 2008. In 2009, diagnostic tests confirmed that salamanders sent for testing had died of a ranavirus infection. Tiger Salamander mortality was documented at 50% of lentic (standing water) sites in 2010, including the Fortification Creek area and near Ice Cave west of Kaycee. Chytrid fungus was also detected in amphibians in 2010, however, the infection rate in 2010 (7.7%) was much less than in 2009 (23%). The decreased infection rate could indicate a decrease in the prevalence of the fungus in the PRB or could result from annual variation in site selection or local weather patterns. Continued monitoring of chytrid infection rates is necessary to assess the nature of the observed trend.

In addition to monitoring efforts, we also conducted inventories for amphibians and reptiles at 10 areas of special interest to the BLM. Of these, the Fortification Creek area and the Welch

Management area contained the most species detected. We identified 8 species at Fortification Creek in 2010 and 11 species in the Welch Management Area across 2009 and 2010 field seasons.

At the conclusion of the report, we provide locations of potential long-term monitoring sites. We recommend that the BLM and ATG to review sites to make sure they meet the needs of the group. We also recommend survey protocols for the different types of surveys proposed for long-term monitoring, including recommended modifications to improve the ability to assess trends in populations and reproductive success using occupancy modeling.

INTRODUCTION

The Powder River Basin (PRB) in northeastern Wyoming is a surprisingly complex landscape marked by plains and large forested escarpments in the north and highly eroded ridges and breaks along the river to the south. The PRB encompasses sagebrush and grassland dominated vegetation types and is part of the Great Plains. The complex topography and sedimentary soils support a diverse array of plant and animal species. The PRB also is an important area for the extraction of coal bed natural gas (CBNG). Currently, over 18,000 CBNG wells exist in the PRB and numerous additional leases are pending. CBNG development entails establishment of new roads, construction of wells and other infrastructure related to the extraction of CBNG, and the release of groundwater extracted from coal seams into surface streams, rivers, or holding ponds. In 2003, the Buffalo Field Office (BFO) of the Bureau of Land Management (BLM) prepared an Environmental Impact Statement (EIS) for CBNG development in Wyoming and Montana which outlines potential impacts to hydrology, wildlife, and other natural resources in the Powder River Basin (BFO BLM 2003). The EIS identified 2 primary areas of concern with respect to amphibians and reptiles; (1) changes in water quality and timing due to release of CBNG product waters into surface waters, and (2) road mortalities from increased vehicular traffic.

The Aquatic Task Group (ATG) was formed in response to the lack of knowledge regarding how CBNG could impact local natural resources. The ATG is an inter-agency and inter-state working group focused on studying and mitigating impacts of energy development on aquatic ecosystems in northeastern Wyoming and southeastern Montana. In 2008, the Wyoming Natural Diversity Database (WYNDD) began a 3-year study of amphibians and reptiles in the PRB under the guidance of the ATG. The purpose of the study was to conduct an inventory of reptiles and amphibians in the PRB and design a monitoring program to track population trends as CBNG development continues to increase in the region. The majority of funding for this work was provided by the Buffalo Field Office of the BLM with some matching funds from WYNDD.

In 2008 and 2009, WYNDD conducted inventories for reptiles and amphibians across the PRB to determine the presence and relative abundance of species in the region. In 2010, our objectives focused on establishing a feasible monitoring plan with standardized survey protocols and collecting the first year of data under this plan. Because new long-term riparian monitoring reaches along the Powder River were in the process of being established, WYNDD was instructed to exclude riparian monitoring until sites were agreed upon. Thus, monitoring in 2010 focused on upland and lentic (pond) sites, with additional inventories in areas of particular interest to the Buffalo Field Office of the BLM.

Objectives

This project had 4 overall objectives:

- 1. Complete a literature review of impacts to amphibians and reptiles from vehicular traffic and changes in water quality.
- 2. Determine the general distribution and characterize the status and habitat of all amphibians and reptiles occurring in the PRB below 1,370 m.

To do this, locations of all herptofauna observations will be combined with data from the WGFD (Turner, 2007) and WYNDD's database to produce a distribution map for each species. Overall site occupancy measures and current literature will be used to categorize species into general groups such as 'common in PRB', 'moderately abundant in PRB', and 'rare in PRB'. Qualitative descriptions of habitat by species will be produced by combining standardized variables collected at survey sites (including water quality) and those presented in the literature.

3. Identify potential current and long-term impacts of CBNG activities on amphibians and reptiles and suggest possible mitigation measures. This will be achieved by measuring road mortality rates and water quality tolerance levels.

Note: The impact of CBNG activities on amphibians and reptiles can only be *roughly estimated* within the framework of this project.

4. Establish a long-term, repeatable monitoring protocol that can be used to track population trends and impacts from CBNG activities in the future.

The literature review (Objective 1) was completed in 2008, and surveys to determine the distribution and habitat use of amphibians and reptiles in the PRB were conducted in 2008 and 2009 (Objective 2 & 3; Estes-Zumpf et al. 2010). Draft distribution maps for each species were provided in the Year 2 progress report (Estes-Zumpf et al. 2010) and final distribution maps are available free of charge upon request from WYNDD

(<u>https://survey.uwyo.edu/TakeSurvey.aspx?SurveyID=120Klp4K</u>). This report details the first year of formal monitoring efforts (site choice, protocols, etc.) as well as additional inventories at key sites identified by the BLM.

METHODS

Study Area

The Powder River watershed is located in northeastern Wyoming (Figure 1) and has an area of approximately 25,000 km². The main stem of the river flows north into Montana where it joins the Yellowstone River. Within Wyoming, tributary streams that originate in the Bighorn Mountains to the west generally have perennial flow fed by snowmelt, whereas ephemeral tributaries originating in the plains to the south and east are characterized by short duration flows from rainstorms (Davis et al., 2006). The majority of CBNG development in the PRB is below 1,370 m elevation and, thus, defines the study area for this project. Topography is complex and includes floodplains, escarpments, upland plains, and highly eroded breaks. The climate is semi-arid and land cover is dominated by two ecological systems; Inter-mountain Basins Big Sagebrush Steppe and Northwestern Great Plains Mixed-grass Prairie. Riparian vegetation along rivers and streams is usually composed of willows and tall grasses with cottonwood forests restricted to the larger rivers, but there are very few natural lentic (standing water) wetlands in the Basin. Twenty-one species of reptiles and amphibians potentially occur in the Powder River Basin of Wyoming (Table 1). Most of these species belong to the suite of Great Plains species

whose far western range occurs in northeastern Wyoming. See Table 2 for a list of acronyms for species names used in this document.

Over 50% of land ownership in the PRB is private, limiting efforts to establish long-term monitoring sites on accessible public lands. Livestock grazing dominates land use with irrigated agriculture restricted to areas immediately adjacent to perennial rivers (Davis et al. 2006). CBNG production has increased greatly on both public and private lands in the last 10 years, especially in the eastern half of the PRB where natural gas is most easily recoverable. Gas field development often entails building and maintaining extensive road networks to serve well pads, pipelines, and compression stations. Deep aquifer water is pumped to the surface and discharged into ephemeral drainages or reservoirs in order to release natural gas trapped in coal seams. The aquifer water is usually colder than surface water and often has higher concentrations of sodium bicarbonate and other salts (Davis et al. 2006). For more background information about the potential impacts of water quality and road network changes on amphibians and reptiles, see Griscom et al. (2009; Appendix A).

Selection of Monitoring Sites

Selection of monitoring sites was guided by the following criteria.

- 1. Sites should be on accessible public lands to ensure access for future surveys.
- 2. Sites should be located in or near habitat for sensitive reptile and amphibian species (water bodies and/or rock outcrops).
- 3. Sites should be selected in areas of high and low potential CBNG impact.
- 4. Sites are known to contain one or more sensitive amphibian and/or reptile species.

It is important to note that much of the PRB is private land and was excluded in most cases from our monitoring plan. Limitations due to access issues and presence of habitat (especially water) further restricted site selection. As a result, sites selected for long-term monitoring were strategically chosen rather than randomly selected and much of the PRB was not sampled. Therefore, inferences about changes in amphibian and reptile populations from this monitoring plan are limited to changes observed at monitoring sites and should not be extrapolated to the entire PRB.

Surveys Protocols

Nocturnal Call Surveys

Nocturnal call surveys can be an efficient and effective way to survey for anurans (frogs and toads), especially in areas where to habitat is restricted. Males of all anuran species in the PRB vocalize under suitable weather conditions to attract females to breeding sites. Calls can be used to identify individuals to species and, depending on environmental conditions, can be heard up to an estimated 2 km away. Road-based nocturnal call surveys are an effective way of detecting species presence across relatively large distances without requiring physical access to adjacent land.

Nocturnal call surveys involved a two person crew starting at a fixed location on a public road and driving a predefined distance to each subsequent listening point. Results from nocturnal call surveys conducted in the PRB in 2006 by the Wyoming Game and Fish Department (WGFD; Turner 2007) and by WYNDD in 2008 and 2009 reveal that shorter distance between listening points tends to increase detection of anurans with softer calls that don't carry well over larger distances (e.g., Northern Leopard Frogs (*Lithobates pipiens*)). The number of species detected along routes with listening points at every 0.1 miles was greater than the number of species detected when listening points were separated by 0.5 miles. Based on these findings, in 2010 we chose to maximize both distance covered and species detections by conducting nocturnal call surveys for anurans every 0.2 miles along chosen 2-mile stretches of roads.

All surveys were conducted after dark and detailed weather information was collected at the beginning and end of the survey (wind speed, barometric pressure, relative humidity, cloud cover, and air temperature). At each listening point, surveyors got out of the vehicle and listened for 3 minutes, recording the coordinates, species, calling intensity, direction and distance to caller(s), and ambient noise. Most calling routes were surveyed twice between late May and early July to increase the likelihood of detecting pulse breeders such as Plains Spadefoot Toads (*Spea bombifrons*). Because weather patterns influence the annual onset and conclusion of amphibian breeding seasons, we excluded from analyses all surveys conducted after the last survey during which amphibians were heard calling. This is based on the assumption that breeding season concluded shortly after that date and all surveys after the conclusion of breeding had a zero probability of detecting calling anurans.

Results from nocturnal call surveys were used to estimate occupancy rates for each species based on this survey method. Occupancy rates can be used to monitor changes across time. Naïve occupancy rates are simply the number of sites occupied by a species divided by the number of sites surveyed. Because our ability to detect an amphibian species is rarely perfect, non-detection could mean that the species was not present, or the species was present but we failed to detect it. Occupancy modeling helps to correct for imperfect detection by using repeat surveys during a season to estimate the probability of detecting a species (MacKenzie et al. 2006). Occupancy rates for each species are then corrected for this bias in detection. We used program PRESENCE (Hines 2006) to estimate occupancy rates at nocturnal call survey sites. We tested if day of year, moon phase, time since last rain, barometric pressure, or air temperature influenced calling behavior and, thus, our ability to detect species. We determined the best model(s) given our set of candidate models using Akaike's Information Criteria (AIC; Burnham and Anderson 2002). In the event that a species was not detected enough times to allow for estimation of detection probabilities, we present naïve (uncorrected) occupancy rates.

Water Body and Riparian Surveys

Visual encounter surveys (VES) allow surveyors to visually detect species and their life stages present at a site. We conducted VES surveys along the banks of standing (lentic) and some flowing (lotic) water in the PRB. This survey method primarily targets amphibians, though reptiles are often found. Surveys followed protocols described in the ATG Monitoring Plan for Amphibians and Reptiles. In previous years, two observers worked together to conduct surveys at each site. In 2010, we modified protocols to facilitate estimating detection probabilities using the dual observer method. Under this method each observer surveys the site independently and does not discuss results with the other observer. This method provides 2 independent repeat surveys for each visit to the site. At lentic sites, observers waited 15 minutes between surveys to allow animals time to settle between surveys. At lotic sites (typically narrow stream channels), observers surveyed different banks. Although this method is not as ideal as multiple return visits to a site, it is an accepted technique to obtain multiple independent surveys at sites across large landscapes or with limited access.

Surveyors searched at a consistent rate around the edge of lentic water bodies or along riparian reaches on either side of stream channels. At each site, surveyors collected extensive data on habitat, water chemistry (pH, temperature, conductivity, TDS), species observed, and life stage. Surveys were timed to record search effort, and timers were stopped while animals were being processed. Dead amphibians were collected, labeled, and preserved in 95% ethanol for later analysis. Frogs and toads were caught by hand or dip net, when possible, and swabbed for chytrid fungus (*Batrachochytrium dendrobatidis*; see below).

Results from lotic and lentic water body surveys were used to estimate occupancy rates for each species. We used program PRESENCE (Hines 2006) to estimate occupancy rates after first correcting for bias due to imperfect detection rates. We tested if day of year and amount of cloud cover significantly influenced species detectability and if the type of site (lotic or lentic) influenced occupancy by a species. We determined the best model(s) given our set of candidate models using AIC. As with nocturnal call surveys, in the event that a species was not detected enough times to allow for estimation of detection probabilities, we present naïve (uncorrected) occupancy rates.

Rock Outcrop Surveys

Rocky outcrops are important landscape features for many reptiles because they provide protection from predators, basking surfaces, and shade from the midday sun, and are often used as hibernacula by multiple species of snakes. Therefore, we conducted visual encounter surveys for reptiles along south-facing rock outcrops. Rock outcrops typically occur in more upland areas and often are not associated with aquatic habitat. Thus, these surveys were used to inventory reptiles and search for snake hibernacula in the PRB but will likely not be part of a long-term monitoring program unless specifically requested by the ATG or the BLM.

Rock outcrops were indentified with aerial photographs and field reconnaissance. Surveys were conducted primarily during morning hours or evening hours, when reptiles are most likely to be active. Surveys involved searching under and around rocks and organic debris. We photographed species encountered and described the general habitat in which the species was found. We recorded search effort by timing all surveys, stopping the timer during data recording and identification of species. We also recorded all non-reptile species observed during rock-outcrop surveys.

Roadkill and Basking Surveys

Roads serve as basking surfaces for reptiles and amphibians. Because animals basking on or crossing roads are often killed by vehicles, surveys along roads are commonly used to inventory local reptiles and amphibians (Heyer et al. 1994). Roadkill 'hotspots', where multiple

individuals are found in a short distance, can signify proximity to hibernacula or migration corridors and the location of hotspots can be used to guide management plans. Encounter rates or mortality rates along fixed stretches of road can also be calculated and compared across seasons or years. In 2008 and 2009 we used roadkill/basking surveys primarily to inventory species present in the PRB; thus we targeted different types of roads (paved, gravel, dirt, etc.) near or adjacent to rock outcrops or water bodies. We incorporated roadkill hotspots identified in 2008 and 2009, along with stretches of the Powder River Road that occur near reptile and amphibian habitat into our monitoring plan in 2010. The Powder River Road is the major north-south access road in the region and is heavily used by residents and CBNG industry workers.

We conducted roadkill/basking surveys along 1-mile fixed stretches of roads. Technicians walked on opposite sides of the road searching for dead or basking reptiles and amphibians. Animals found were photographed and their location was recorded. In addition to fixed roadkill surveys, technicians also recorded all incidental sightings of dead or basking reptiles and amphibians found while driving between survey sites.

Monitoring Pathogens

Chytrid Analyses

Chytrid fungus (*Batrachochytrium dendrobatidis*) has been implicated in amphibian declines around the world, especially in concert with other environmental stressors, and infected animals have been documented in several amphibian species in the PRB (Turner 2007, Griscom et al. 2009, Estes-Zumpf et al. 2010). Because chyrid fungus occurs in the PRB and the ATG is concerned about potential environmental stressor associated with CBNG (changes in water temperature, increased levels of sodium bicarbonate, etc.), any amphibian monitoring program in the region should include monitoring of chytrid prevalence in local amphibian populations.

To identify whether amphibians were infected with chytrid fungus, we collected epithelial tissue samples from a subset of all amphibians found at each site during surveys. Sample collection followed established procedures (Livo 2003). Amphibians were systematically swabbed with sterile cotton swabs to collect epidermal DNA. Swabs were immediately stored in sterile microcentrifuge tubes containing 95% ethanol and labeled with unique specimen numbers. We stored samples in a -20°F freezer until shipping. Samples were sent to Dr. John Wood at Pisces Molecular LLC in Boulder, Colorado, for analysis via PCR test to determine if the fungus was present.

Ranavirus

WYNDD technicians documented over 700 dead or dying Tiger Salamanders (*Ambystoma mavortium*) at lentic sites in the PRB during surveys in 2008 and 2009. Diagnostic tests of samples sent to the National Wildlife Health Center in Madison, Wisconsin in 2009 revealed that the salamanders tested had died of a ranavirus infection. Because we have found no evidence of a ranavirus infection in frogs and toads in the PRB, it is possible that the ranavirus is *Ambystoma tigrinum* virus (or ATV), which only affects Tiger Salamanders. Tiger Salamanders are currently common throughout Wyoming, however, the current spread of ranavirus through populations in eastern Wyoming (and Montana) warrants monitoring Tiger Salamander populations to determine their ability to rebound from this lethal and highly contagious virus. Thus we continued to document Tiger Salamander mortalities at lentic sites during VES surveys.

RESULTS & DISCUSSION

Monitoring Sites

We identified monitoring sites largely on accessible areas of public land and several sections of state land, thereby ensuring continued access to most sites in the future. Several sites surveyed in 2010 as special interest areas to the BLM were incorporated into the monitoring plan. We identified 21 VES survey routes at water bodies, 5 roadkill/basking routes, and 12 nocturnal call survey routes (Figure 2, Table 3). The ATG is also working with the USGS to identify a number of long-term monitoring reaches above and below major tributaries and CBNG discharge points along the Powder and Tongue Rivers. These sites will also be incorporated into the long-term monitoring plan.

2010 Survey Results

Nocturnal Call Surveys:

We conducted nocturnal call surveys at 15 sites in the PRB (Figure 3), twelve of which are part of the proposed monitoring plan. Three sites were surveyed to inventory species at areas of interest to the BLM, but likely will not be included in a long-term monitoring plan. Surveys were conducted from 19 May to 15 July, however, calling dropped off in early July and the last survey during which anurans were heard calling was on 12 July. Thus, we excluded 1 survey that occurred after 12 July.

We detected at least 3 species at most sites and detected all 5 anuran species at one site along Echeta Road (site NOC011). Boreal Chorus Frogs (*Pseudacris maculata*) were the most common species recorded and were detected at 87% (\pm 8.8) of sites (Table 4). Detectability of Boreal Chorus Frogs varied between repeat surveys, however, likely resulting from repeat surveys being conducted later in the season and closer to the end of the breeding season. Plains Spadefoot Toads and Woodhouse's Toads (*Anaxyrus woodhousii*) also were common and estimated to occur at about 60% of sites. Detectability of Woodhouse's toads decreased with day of year, again indicating that detectability was lower later in the season. Great Plains Toads (*Anaxyrus cognatus*) and Northern Leopard Frogs (*Lithobates pipiens*) were the least common and were only documented at 20% of sites (Table 4).

We were unable to correct nocturnal call survey occupancy rates for detection bias due to low power resulting from having only 2 surveys at all sites. To compensate for this, future implementation of this monitoring plan should visit several sites ≥ 3 times in order to improve the ability to estimate detection rates (see Recommendations section for further discussion).

Water Body and Riparian Surveys:

We conducted VES surveys at 29 sites in 2010. Surveys were conducted in 11 riparian areas along streams and rivers (lotic systems), and at 18 stockponds and CBNG ponds (lentic systems) throughout the PRB (Figure 3). We documented 5 species of amphibians (Table 5) and 9 species of reptiles (Table 6) during riparian surveys. Tiger Salamanders and Boreal Chorus Frogs were

the most common species detected, and Boreal Chorus Frogs were often detected audibly rather than by sight. Tiger Salamanders were more common at lentic sites than lotic sites, and the top occupancy model for this species had separate occupancy rates for lentic (0.788 ± 0.117) and lotic (0.307 ± 0.152) sites (Table 5). For all other amphibians the simple model (1 group, constant occupancy, constant detectability) performed better than more complex alternative models. Thus, occupancy and detectability estimates from the simple model are provided (Table 5). Northern Leopard Frogs, a species currently being petitioned for listing as threatened under the Endangered Species Act, were estimated to occupy 42% of sites surveyed in 2010. Plains Spadefoot Toads and Great Plains Toads were the least common amphibian species detected during VES surveys, likely due in part to their habitat preferences and breeding behavior (pulse breeders).

We detected 5 snake, 3 turtle, and 1 lizard species during formal VES surveys (Table 6), however detections were too few for most species to allow probability of detection to be estimated. Thus, occupancy rates for most species are naïve (not corrected for detection bias). Gartersnakes were the most common type of snakes detected during VES surveys, likely due to their close association with water. We estimate that Wandering Gartersnakes (*Thamnophis elegans vagrans*) occupied 18% of sites, and Plains Gartersnakes (*Thamnophis radix*) occupied 11% of sites (Table 6). All other snake species detected using this method occurred in very low numbers, likely because they prefer rocky habitat typically found in more upland areas. We documented Western Painted Turtles (*Chrysemys picta*) at 10% and only documented Snapping Turtles (*Chelydra serpentina*) and Spiny Softshell Turtles (*Apalone spinifera*) at one site each. Although turtles likely occur in more sites, the probability of detecting turtles using VES surveys is relatively low. Use of turtle traps is a more effective means of surveying for turtles (Turner 2007), but this technique is time consuming and should only be employed if management goals require monitoring trends in turtle populations at specific sites.

Two lentic sites on state land in Campbell County had the highest species diversity (i.e., Spotted Horse Reservoir and LS Bar Pond). We detected 4 amphibian species at both sites. No amphibians sampled tested positive for chytrid fungus, but dead salamanders were found at Spotted Horse. We also documented 6 reptile species, including Snapping Turtles and numerous Western Painted Turtles, at Spotted Horse. Most evidence of turtles, however, were from shells of dead turtles. We detected 5 reptile species as LS Bar Pond. CBNG development occurs in the area of both sites, however, we were unable to assess influence of any upstream CBNG development, if any, on these ponds.

Two sites with direct input of CBNG product water also had high species diversity. We documented 4 species of amphibians, including Plains Spadefoot Toad, at Fallen Eagle Reservoir. Burger Draw also had 3 species of amphibians and 1 reptile (Northern Sagebrush Lizard (*Sceloporus graciosus*)). Chytrid fungus was not documented at either site, though dead salamanders were reported at Fallen Eagle.

Rock Outcrop Surveys

We conducted rock outcrop surveys at 19 sites across the PRB (Figure 3). We detected 86 individual herps from 7 species. Five species of reptiles (4 snakes, 1 lizard) were documented during surveys. The most notable species detected was a Pale Milksnake (*Lampropeltis*)

triangulum), as species suspected to occur in the PRB but for which observations are lacking. One Pale Milksnake was found under a thin slab of layer rock at survey site ROC01001 just south of Schoonover Rd and about 2km east of the Powder River. Northern Sagebrush Lizards were the most common reptile found, accounting for 83% of observations, and were observed at most rock outcrops (84%). Prairie Rattlesnakes (Crotalus viridis) were the second most common reptile found on rock outcrops and were documented at 26% of sites. Bullsnakes (Pituophis catenifer sayi) and Eastern Yellowbelly Racers (Coluber constrictor flaviventris) were detected at 11% of rock outcrops. We also documented 2 amphibian species during rock outcrop surveys. Woodhouse's Toads were found at 2 sites and a Boreal Chorus Frog was found at 1 site. Other species found at rock outcropping included bushy-tailed woodrats (Neotoma cinerea), cottontail rabbits (Sylvilagus spp.), mule deer (Odocoileus hemionus), pronghorn (Antilocapra americana), coyotes (Canis latrans), swallows, magpies (Pica hudsonia), scorpions (Paruroctonus sp.), and centipedes (Family Scolopendridae). We also found 3 bats using a day roost between sheets of rock at ROC01001, the same site where the milksnake was detected. No evidence of snake hibernacula was found, however, timing of rock outcrop surveys was not ideal for observing snakes recently emerged from or about to enter hibernation. If the ATG or BLM are interested in identifying snake hibernacula in the future, specific surveys for hibernacula in fall or early spring would be optimal. Success, however, varies with weather patterns and size of the survey area.

Roadkill and Basking Surveys

We conducted multiple surveys of 5 one-mile stretches of dirt and paved roads to search for roadkill/basking reptiles and amphibians (Figure 3). We detected 20 individuals, 17 of which were dead, from at least 4 species. As in 2009, Bullsnakes were the most common species found dead on roads (41%). Toads accounted for 29% of roadkill followed by Common Gartersnakes (*Thamnophis sirtalis parietalis*; 18%), Prairie Rattlesnakes (6%), and Northern Leopard Frogs (6%).

Because we targeted key roads and roadkill "hotspots" identified during 2008 and 2009 inventory surveys, the average roadkill rate at monitoring sites in 2010 was higher than in previous years. We determined the average roadkill rate for sites that were visited more than once during the field season. The overall average roadkill rate was 3.2 mortalities/mile, however, 88% of mortalities were found along two, one-mile stretches of SR338 near the Welch Management Area along the Tongue River in Sheridan County (Table 7). The overall roadkill rate for the same 5 sites surveyed in 2009 was 1.4 mortalities/mile. The increase in roadkill rates from 2009 to 2010 is almost entirely due to a large increase in the number of mortalities at site RD0908 on SR338 in 2010. We documented 11 dead reptiles and amphibians at RD0908 in 2010, as well as numerous dead swallows on the bridge section where SR338 crosses the Tongue River. Roadkill rates are likely consistently high along this portion of SR338 due to its proximity to riparian areas along the Tongue River and multiple adjacent rock outcroppings. The potential influence of such high roadkill rates on local herpetofauna is difficult to predict without formal population studies. However, if managers are concerned about reptile and amphibian populations in the Welch Management Area, seasonal speed restrictions or increased signage warning motorists to watch for wildlife on roads may help to decrease the significant number of road mortalities in this area.

In addition to formal roadkill/basking surveys, we recorded all incidental occurrences of dead or basking reptiles and amphibians found while driving between surveys in the PRB. In 2010, 23 reptiles and amphibians were found on roadways, the majority of which (74%) were dead. Bullsnakes were again the most common species found on roadways (48%) followed by Prairie Rattlesnakes (22%). We also documented a Pale Milksnake dead on SR14/16 near the intersection of Arvada-Gillette Road (Figure 12). Incidental occurrences of other species found on the road include Eastern Yellowbelly Racer, Wandering Gartersnake, Tiger Salamander, Woodhouse's Toad, Great Plains Toad, and Greater Short-horned Lizard (*Phrynosoma hernandesi*). Although formal surveys in past years did not detect roadkill along Dead Horse Road south of Hwy 93 on the east side of the Powder River, we documented incidental occurrences of 5 dead snakes (3 Bullsnakes, 2 Prairie Rattlesnakes) along in about a 3.5 mile stretch of this road. Managers may want to monitor roadkill rates along this road in future years.

Species Distributions

Although surveys conducted in 2008 and 2009 primarily focused on inventorying amphibians and reptiles in the PRB and surveys in 2010 focused on collecting baseline data at proposed monitoring sites, we continued to add to our knowledge of the distribution and abundance of species in the area. Inventories at additional areas of interest to the BLM in 2010 further augmented our knowledge. Interpretation of patterns are limited to sites surveyed because large portions of the PRB are private lands or are inaccessible due to lack of roads or to surrounding land ownership. Because water is rare and patchily distributed across the PRB, amphibians (and certain reptiles) are concentrated near water bodies. Occupancy by a given species is not only influenced by presence of water, but could also be influenced by water permanence, proximity to other water bodies, water quality, recent disease outbreaks, and the length of time the water body has been present on the landscape. Thus, understanding patterns of occupancy in such a large, complex, and changing landscape is difficult.

Taking the above limitations into consideration, some patterns are revealed in the data. In general, surveys in the northern portion of the PRB in Wyoming tended to detect more species (Figure 4). Surveys of lentic sites in the northwest, in particular, resulted in a diversity of both amphibians and reptiles. Rock outcrops, used by many reptile species, also generally are more common and larger in the northern portion of the PRB. Thus, the northern PRB may provide more habitat for reptiles than the eroded clay breaks in the southern portion of the PRB.

Based on results from multiple survey methods, Boreal Chorus Frogs, Woodhouse's Toads, and Tiger Salamanders appear to be common throughout the PRB (Figures 5, 6, 7). Boreal Chorus Frogs were primarily detected by their calling during nocturnal call surveys and VES surveys at both lotic and lentic sites. Tiger salamanders, however, are more common in areas with standing water than flowing water and the majority of Tiger Salamanders detected were dead or dying, likely due to ranavirus. Northern Leopard Frogs were also relatively common throughout the PRB and were most easily detected using VES surveys at water bodies (Figure 5). Plains Spadefoot Toads and Great Plains Toads had the lowest occupancy rates for amphibians; however, detectability for these pulse breeders is notoriously low. We only detected Great Plains Toads in the northern half of the PRB (Figure 8), but lack of detections in the south could be due to low detectability and fewer surveys in that region. Plains Spadefoot Toads were found in low numbers throughout much of the PRB (Figure 8).

We also documented most of the reptile species thought to occur in the PRB. Bullsnakes were the most common snake observed in the PRB and were detected throughout the area both near water bodies and in upland sites on roads and at rock outcrops (Figure 9). Prairie Rattlesnakes also were common in both upland and riparian habitats throughout the PRB (Figure 10) though were most often detected at rock outcrops or on roads. Gatersnakes were primarily restricted to areas near water bodies and were most often detected using VES surveys. Of the 3 species detected, Wandering Gartersnakes were the most common and were distributed throughout the PRB at both lentic and lotic sites (Figure 11). Common Gartersnakes were only documented at a couple of sites, but occurred in both the northern and southern portions of the PRB (Figure 11). Plains Gartersnakes, however, were only detected near water bodies in the northern half of the PRB. Eastern Yellowbelly Racers were not common, but could occasionally be found near water bodies and on roads and were distributed throughout the PRB (Figure 12). The two rarest snakes in the PRB were the Western Hog-nosed Snake and the Pale Milksnake. Both had too few occurrences to understand their distributions in this area (Figure 12). Surveys in 2010, however, resulted in the only recorded sightings of Pale Milksnakes in the PRB.

Other reptiles documented include lizards and turtles. Although Northern Sagebrush Lizards were common throughout the PRB both near water bodies and at upland sites, Greater Short-Horned Lizards were rarely detected (Figure 13). Greater Short-horned Lizards, however, prefer sandy upland habitats, areas which were not targeted for surveys. Observations of turtles were rare but could result from low detectability for these species using VES surveys. Of the 3 turtle species, Western Painted Turtles were the most common and likely the most detectable (Figure 14). Spiny Softshell Turtles were only documented in the Welch Management Area (Figure 14), however, this species prefers large rivers and further surveys along the Powder River and its major tributaries could result in more documented occurrences of this species.

Predictive distribution maps for amphibian and reptile species were generated using data from this project, past surveys, and documented sightings/collections in Wyoming. Draft maps were provided in the 2009 annual report (Estes-Zumpf et al. 2010). These distribution models are part of an ongoing project on Assessment of Wildlife Vulnerability to Energy Development (AWVED) being conducted by WYNDD's senior zoologist, Doug Keinath, and GIS specialist, Mark Andersen. Final distribution maps are available online by request from WYNDD. A summary of habitat types, habitat characteristics, and water quality parameters for each amphibian species documented in the PRB was provided in the 2009 annual report (Estes-Zumpf et al. 2010; Table 5).

BLM Special Area Results

We conducted inventories for reptiles and amphibians at 10 areas of special interest to the Buffalo Field Office of the BLM (Table 8; Figure 15). All but one of these areas occurred on BLM lands. The Wild Horse Creek area is privately owned but permission was given to survey a portion of the creek. Wild Horse Creek is of interest to the BLM because considerable amounts of CBNG water have been released into Wild Horse Creek for several years. We were unable to survey Dead Horse Reservoir 2 as requested because it was dry in 2010. The type of survey methods used varied between sites with type and accessibility of habitat (water bodies and rock outcrops).

The Fortification Creek area had the most habitat for reptiles and amphibians and we were able to conduct several VES, nocturnal call surveys, and rock outcrop surveys. We identified 8 species of herps (4 amphibians, 4 reptiles) as well as a gartersnake, toad, and turtle which we were unable to get a sufficient view of to identify to species (Table 8). Chytrid fungus was not documented in any of the amphibians sampled from the Fortification Creek area, however, we did find at least 10 dead tiger salamanders in a pond on state land. Due to access issues, we only surveyed a small portion of this watershed, but were able to target the few areas in the watershed with consistent water. More upland searches could yield detections of additional reptile species.

We first inventoried reptiles and amphibians in the Welch Management Area in 2009 at the request of the BLM. We surveyed the area a second time in 2010 and documented 5 of the 9 species detected in 2010 (Table 8). We also detected several Common Gartersnakes, which were not observed in 2009, and 5 toads that had been killed by vehicles and which we were unable to identify to species. Thus, during surveys in 2009 and 2010, we confirmed at least 11 species of reptiles and amphibians in the Welch Management Area, making this area one of the most herpetologically diverse sites surveyed. Unfortunately, many of the individuals in this area were found during roadkill/basking surveys and had been killed by vehicles on SR338.

The Burnt Hollow Management Area also contained several species of herps (4 amphibians, 1 reptile detected), despite the relative lack of water bodies and rock outcrops (Table 8). We were able to find 3 ponds in the Management Area that contained water and conducted opportunistic searches while looking for suitable rock outcrops. The most unique individual detected in Burnt Hollow was a likely Woodhouse's Toad adult with no cranial crests. Toads in Wyoming can be identified by the presence and shape of their cranial crests. Although cranial crests are sometimes not very obvious on young toads, crests are typically prominent on adults. The only toad species in Wyoming that lacks a cranial crest is the Boreal Toad (*Anaxyrus boreas boreas*) which occurs in some mountain ranges in Wyoming. Because the Burnt Hollow Management Area is well outside the range of the Boreal Toad in Wyoming both WGFD herpetologists and WYNDD experts assume, based on photographic evidence, that the large toad found was an aberrant Woodhouse's Toad that lacked any cranial crests. Further investigations into toads in this area, however, could prove interesting.

Tiger Salamander Mortality

Tiger Salamander mortality was documented at 50% (9 of 18 sites) of lentic sites (standing water bodies) in 2010, including sites in the Fortification Creek area and near Ice Cave west of Kaycee (Figure 7). The proportion of sites with dead or dying salamanders in 2010 was greater than the 25% of lentic sites in 2008 but less than the 63% of sites with dead salamanders in 2009. In total, over 700 dead salamanders have been found throughout the PRB during survey activities from 2008 to 2010 (Figure 7). In 2009, diagnostic tests of two dead salamanders sent the National Wildlife Health Center in Madison, Wisconsin confirmed that both salamanders died of a ranavirus infection. No salamanders were sent in for diagnostic tests in 2010, however, many exhibited hemorrhaging consistent with (but not limited to) a ranavirus infection.

Although testing of dead Tiger Salamanders from multiple sites across the PRB has yet to be done, it is likely that the majority of deaths are the result of ranavirus infection due to presence of the virus in the area and obvious external symptom evident on dead and dying individuals.

Because other amphibians in the area, and even in the same ponds, do not show evidence of a ranavirus infection, it is also likely that the ranavirus is the genus-specific *Ambystoma tigrinum* virus (ATV). However, more widespread diagnostic tests should be conducted to confirm this. Tiger Salamanders are currently common throughout Wyoming, however, the apparent rapid spread of ranavirus through populations in eastern Wyoming warrants monitoring. Ranavirus is a lethal and highly contagious virus that could potentially impact Tiger Salamander populations in the state. At this point, it is unknown if the ranavirus outbreak is natural or if changes in water quality in the PRB due to CBNG development could be increasing susceptibility of Tiger Salamanders to the virus. We recommend monitoring tiger salamander mortalities in the PRB. Research investigating the possible role of CBNG discharge in susceptibility of salamanders to ranavirus could also help resource managers and scientists better understand the dynamics of this virus.

Chytrid Analyses

We sampled 26 individual amphibians (11 Woodhouse's toads, 12 Northern Leopard Frogs, 2 Plains Spadefoot Toads, and 1 Great Plains Toad) for chytrid fungus. Chytrid was only detected in 2 samples, 1 Woodhouse's Toad and 1 Great Plains Toad. The overall chytrid infection rate for individuals sampled in 2010 (7.7%) is a drastic decrease from the 23% documented in 2009 and the lowest it has been since Turner (2007) began documenting chytrid infection rates in the PRB. We also observed a drop in chytrid infection rate when analyzed by site rather than by individual. In 2009, 3 of 11 sites (27%) contained amphibians which tested positive for chytrid. In 2010, 2 of 12 sites (17%) contained individuals with chytrid fungus. Furthermore, in 2010 no Northern Leopard Frogs tested positive for chytrid, while 28% tested positive in 2009. This decrease in chytrid is encouraging, especially given the cold wet spring in 2010, conditions which typically decrease the ability of infected amphibians to fight off the fungus through basking behavior. The perceived decrease in chytrid infection rate could indicate a decrease in the prevalence of chytrid fungus in the PRB. Results from surveys conducted by WYNDD from 2008 to 2010 indicate that chytrid has been documented along the Powder River and several of its tributaries (except in the very south) and from the Tongue River in the West to the just northeast of Gillette (Figure 16). As with the ranavirus infection, sufficient data does not yet exist to determine the impact of chytrid on local amphibian populations, and we recommend continued monitoring of chytrid infection rates in the PRB.

RECOMMENDATIONS

 Proposed monitoring sites and survey protocols should be reviewed by the BLM and the ATG to make sure they meet the needs of the group. Surveys sites focus on areas near water bodies and detections of upland species are expected to be lower. Some species, such as Northern Sagebrush Lizards, Bullsnakes, and Prairie Rattlesnakes are better detected using rock outcrop surveys. If the ATG decides these species are important to monitor, rock outcrop surveys should be incorporated into monitoring efforts. Only five, 1-mile stretches of road are currently proposed for monitoring road mortalities based on concern over the number of dead herps detected during inventories. If the ATG is concerned about road mortality due to increased traffic associated with CBNG, we recommend establishing more monitoring sites throughout the PRB. Unfortunately, although even low road mortality rates can impact local herp population (Rosen and Lowe 1994, Fahrig et al. 1995, Pope et al. 2000, Carr and Fahrig 2001, Row et al. 2007), detection of mortality events using formal surveys is generally low, limiting the power to assess trends in roadkill rates even with a large number of survey sites.

- 2. Occupancy modeling is the most efficient method to assess population trends for amphibians and reptiles across such a large landscape (Heyer et al. 1994). This method, however, is dependent on detections at repeat surveys to allow estimation of detection probability. Two visits to nocturnal call routes did not provide sufficient data to estimate detection probabilities for most anurans, and detections decreased from spring to early summer. Therefore, we recommend that future nocturnal call survey protocols require surveys to be conducted in the spring (May mid June) and require a portion of the sites to be visited at least 3 times. Because field time and funding typically are limiting, a more easily applied approach would be to sub-sample existing routes for 1 to 2 stretches composed of 3 stops. Each stretch would cover an area of 0.6 miles, a distance from which most local anuran calls should be audible in that landscape. Stops could then be treated as repeat sampling events (from different angles) and used to assess detectability. Even with this alternative, each stretch should be visited at least twice in the spring.
- 3. Data on reproduction can be easily collected during VES surveys. Presence of egg masses, tadpoles, larvae, and metamorphs can be documented and incorporated in occupancy modeling. Because egg mass and larval life stages are most sensitive to changes in water quality and temperature, we strongly recommend a sampling scheme that, at the very least, incorporates documentation of reproduction for key species of amphibians. This is best accomplished by incorporating use of dip nets at regular intervals during VES surveys at water bodies to detect tadpoles in murky waters.
- 4. New long-term monitoring reaches along the Powder and Tongue Rivers are currently being established by the ATG and the USGS. Because a number of ecological components will be monitored at these sites, WYNDD recommends including amphibian and reptile surveys at set time intervals decided upon by the ATG. Because data will exist on other biotic and abiotic components at these sites, should changes in herp populations occur researches will be able to investigate the cause of these changes.

We recommend that VES surveys for amphibians and reptiles be conducted at these USGS monitoring reaches, targeting areas with backwater and oxbows if available. Protocols should be guided by those used during the 2010 field season of this study (i.e., dual observer method with dipnets) and at least 2 visits should be conducted at a large portion of the sites. These protocols would result in 4 independent surveys in 2 visits and should allow for estimation of detection probabilities and occupancy rates for most species. Results could then be used to examine trends in occupancy rates across time.

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LITERATURE CITED

- Aquatic Task Group, 2006. Aquatic monitoring plan. An online document prepared by a subcommittee of the Coal Bed Natural Gas Interagency Working Group. http://www.wy.blm.gov/prbgroup/04minutes/aquaticbiotaplan06-16.pdf
- BFO BLM, 2003. Powder River Basin Final EIS/ Proposed Resource Management Proposal Amendment. Buffalo Field Office, Bureau of Land Management, Buffalo, Wyoming. http://www.blm.gov/wy/st/en/info/NEPA/bfodocs/prb_eis.html
- Burnham, K. P., and D. R. Anderson. 2002. Model selection and multimodel inference: a practical information-theoretic approach. 2nd ed. Springer, New York, New York.
- Carr, L. W., and L. Fahrig. 2001. Effect of road traffic on two amphibian species of differing vagility. Conservation Biology 15:1071–1078.
- Davis, W.N., R.G. Bramblett, and A.V. Zale. 2006. The effects of coalbed natural gas activities on fish assemblages: a review of the literature. Montana Cooperative Fish Research Unit, Montana State University, Bozeman, Montana.
- Estes-Zumpf, W. H. Griscom, D. Keinath. 2010. Inventory of Amphibians and Reptiles in the Powder River Basin Area of Wyoming: Year Two Progress Report. Report prepared for the Buffalo Field Office of the Bureau of Land Management by the Wyoming Natural Diversity Database-University of Wyoming, Laramie, Wyoming
- Fahrig, L., J. H. Pedlar, S. E. Pope, P. D. Taylor, and J. F. Wegner. 1995. Effect of road traffic on amphibian density. Biological Conservation 73:177–182.
- Griscom, H., W. Estes-Zumpf, D. Keinath. 2009. Pre-drilling surveys of amphibian and reptile habitats in the Powder River Basin of Wyoming. Prepared by the Wyoming Natural Diversity Database, Laramie, Wyoming. January 2009.
- Heyer, W. R., M. A. Donnelly, R. W. McDiarmid, L. C. Hayek, and M. S. Foster. 1994. Measuring and Monitoring Biological Diversity: Standard Methods for Amphibians. Smithsonian Institution Press, Washington D.C.
- Hines, J. E. (2006). PRESENCE2- Software to estimate patch occupancy and related parameters. USGS-PWRC. <u>http://www.mbr-pwrc.usgs.gov/software/presence.html</u>.
- Livo, L.J. 2003. Methods for obtaining *Batrachochytrium dendrobatidis* (Bd) samples for PCR testing. Department of Integrative Physiology, University of Colorado, Boulder, Colorado.
- Mackenzie, D. I., J. D. Nichols, J. A. Royle, K. H. Pollock, L. L. Bailey, J. E. Hines. 2006. Occupancy Estimation and Modeling: Inferring Patterns and Dynamics of Species Occurrence. New York, Academic Press.
- Parker, J. and S. Anderson. 2001. Identification guide to the herptiles of Wyoming. Prepared by the Wyoming Cooperative Fish and Wildlife Research Unit for the Wyoming Game and Fish Department.
- Pope, S. E., L. Fahrig, and N. G. Merriam. 2000. Landscape complementation and metapopulation effects on leopard frog populations. Ecology 81:2498–2508.

- Rosen, P. C., and C. H. Lowe. 1994. Highway mortality of snakes in the Sonoran Desert of southern Arizona. Biological Conservation 68:143–148.
- Row, J. R., G. Blouin-Demers, and P. J. Weatherhead. 2007. Demographic effects of road mortality in black ratsnakes (*Elaphe obsoleta*). Biological Conservation 137:117-124.
- Turner, W. 2007. Survey of herpefauna in the Powder River Basin. Prepared by the Wyoming Game and Fish Department for the US Environmental Protection Agency.

TABLES AND FIGURES

Table 1. Species of amphibians and reptiles thought to potentially occur in the Powder River Basin of Wyoming (Turner, 2007; Parker and Anderson, 2001) and their detection history during past surveys.

Common Name	Scientific Name	WGFD 2004-2006	WYNDD 2008	WYNDD 2009	WYNDD 2010
Bullfrog*	Lithobates catesbeianus	Maybe	N	N	N
Northern Prairie Lizard*	Sceloporus undulatus garmani	N	N	N	N
Pale Milksnake ⁺	Lampropeltis triangulum	Ν	Ν	Ν	Y
Boreal Chorus Frog	Pseudacris maculata	Y	Y	Y	Y
Northern Leopard Frog	Lithobates pipiens	Y	Y	Y	Y
Great Plains Toad	Anaxyrus cognatus	Y	Y	Ν	Y
Woodhouse's Toad	Anaxyrus woodhousii	Y	Y	Y	Y
Plains Spadefoot	Spea bombifrons	Y	Y	Y	Y
Tiger Salamander	Ambystoma mavortium	Y	Y	Y	Y
Greater Short-horned Lizard	Phrynosoma hernandesi	Y	Y	Y	Y
Northern Sagebrush Lizard	Sceloporus graciosus	Y	Y	Y	Y
Eastern Yellowbelly Racer	Coluber constrictor flaviventris	Y	Y	Y	Y
Prairie Rattlesnake	Crotalus viridis	Y	Y	Y	Y
Western Hog-nosed Snake	Heterodon nasicus	Y	Y	Ν	Ν
Bullsnake	Pituophis catenifer sayi	Y	Y	Y	Y
Wandering Garter Snake	Thamnophis elegans vagrans	Y	Y	Y	Y
Plains Garter Snake	Thamnophis radix	Y	Y	Ν	Y
Common Garter Snake	Thamnophis sirtalis parietalis	Y	Y	Y	Y
Snapping Turtle	Chelydra serpentina	Y	Y	Ν	Y
Western Painted Turtle	Chrysemys picta	Y	Y	Y	Y
Spiny Softshell Turtle	Apalone spinifera	Y	Ν	Y	Y

* Indicates questionable occurrence in PRB due to lack of supporting documentation from field survey efforts.

[†] Occurrence in PRB was previously questionable due to lack of sightings

BCF	Boreal Chorus Frog	PR	Prairie Rattlesnake
NLF	Northern Leopard Frog	WHS	Western Hog-nosed Snake
GPT	Great Plains Toad	BS	Bullsnake
WT	Woodhouse's Toad	WGS	Wandering Garter Snake
PSFT	Plains Spadefoot Toad	PGS	Plains Garter Snake
TS	Tiger Salamander	CGS	Common Garter Snake
SHL	Short-horned Lizard	ST	Snapping Turtle
NSL	Northern Sagebrush Lizard	SST	Spiny Softshell Turtle
EYR	Eastern Yellowbelly Racer	WPT	Western Painted Turtle

Table 2. List of acronyms for species names used in tables and figures.

Table 3. Locations of proposed monitoring sites for long-term monitoring of amphibian and reptile populations in the Powder River Basin area of Wyoming.

		Start		En	d
Site Name	Survey Type	Easting	Northing	Easting	Northing
Burger	VES	411947	4887314	411688	4887345
Cave Pond	VES	346508	4855404	346508	4855404
Cave Pond 2	VES	350972	4855218	350972	4855218
Dry Creek	VES	402642	4899622	402271	4899482
Fallen Eagle	VES	410576	4889560	410576	4889560
Flowing E Pond 1	VES	395674	4900159	395674	4900159
Flowing E Pond 2	VES	394660	4903281	394660	4903281
LS Bar Pond	VES	429597	4965028	429597	4965028
Powder 0713	VES	405950	4863489	405199	4862817
RVE0901 / Tongue River	VES	346714	4978631	346441	4978532
RVE0902 / Tongue River	VES	346427	4978525	346221	4978481
RVE0903 / Tongue River	VES	345343	4978204	345266	4978175
Spotted Horse	VES	433967	4949937	433967	4949937
Trabing Reservoir	VES	376819	4884368	376819	4884368
Van Houten Draw	VES	414267	4891773	413526	4892372
Van Houten Pond	VES	415143	4891914	415143	4891914
WB0901	VES	414972	4890175	414972	4890175
WB0902	VES	415119	4889986	415119	4889986
WB0908	VES	398457	4885127	398457	4885127
Wild Horse Creek	VES	411021	4944716	410809	4944809

Wild Horse Creek 2	VES	411021	4944716	411239	4945059
BTRD07	Roadkill	408908	4925314	409413	4926690
RD0902	Roadkill	407808	4898063	407771	4898373
RD0903	Roadkill	407671	4895349	408296	4893919
RD0907	Roadkill	347250	4979896	347891	4981343
RD0908	Roadkill	346698	4978801	347349	4977405
BTNOC09	Nocturnal	395987	4923083	394618	4922329
BTNOC11	Nocturnal	396386	4925557	395300	4926657
BTNOC14	Nocturnal	382290	4898499	383150	4897258
BTNOC15	Nocturnal	377246	4882746	376867	4884280
NOC009B	Nocturnal	398748	4838009	395736	4838390
NOC010	Nocturnal	410347	4944639	413280	4943471
NOC011	Nocturnal	424203	4932914	424719	4929900
NOC016	Nocturnal	390402	4917339	386462	4917769
NOC018B	Nocturnal	432976	4899074	432180	4893840
NOC1001	Nocturnal	429086	4925178	430790	4922544
NOC019B	Nocturnal	411690	4896632	410438	4894389
NOCBVER	Nocturnal	345595	4837382	345102	4838857

Table 4. Occupancy estimates (Ψ) for amphibians detected during nocturnal call surveys in 2010. Naïve occupancy rates (number of sites where detected / number of sites surveyed) are provided for each species. For species with sufficient detections, we estimated detection probabilities (p) for surveys and corrected occupancy estimates for bias due to imperfect detection. Standard errors (S.E.) are provided for estimates.

				Nocturnal Call	
Species	Naïve Ψ	Ψ	S.E.	р	S.E.
BCF	0.867	0.867	0.088	varied with survey (1,2) p1 = 1.0 (fixed) p2 = 0.727	p1= 0 (fixed) p2 = 0.134
NLF	0.2	-	-	-	-
WT	0.467	0.606	0.15	varied with day of year (-)	n/a
GPT	0.2				
PSFT	0.6	-	-	-	-

Table 5. Occupancy estimates (Ψ) for amphibians detected during visual encounter (VES) surveys in 2010. Naïve occupancy rates (number of sites where detected / number of sites surveyed) are provided for each species. For species with sufficient detections, we estimated detection probabilities (p) and corrected occupancy estimates for bias due to imperfect detection. Standard errors (S.E.) are provided for estimates.

			VES		
Species	Naïve Ψ	Ψ	S.E.	р	S.E.
NLF	0.379	0.416	0.101	0.634	0.094
BCF	0.414	0.754	0.23	0.286	0.086
WT	0.379	0.492	0.128	0.466	0.096
GPT	0	-	-	-	-
PSFT	0.035	-	-	-	-
TS	0.552	0.307 lotic 0.788 lentic	0.152 lotic 0.117 lentic	0.665	0.071

Table 6. Occupancy estimates (Ψ) for reptiles detected during visual encounter (VES) surveys in 2010. Naïve occupancy rates (number of sites where detected / number of sites surveyed) are provided for each species. For species with sufficient detections, we estimated detection probabilities (p) and corrected occupancy estimates for bias due to imperfect detection. Standard errors (S.E.) are provided for estimates.

Species	Naïve Ψ	Ψ	S.E.	р	S.E.
WGS	0.138	0.18	0.085	varied with cloud cover (-)	variable
PGS	0.069	0.114	0.089	0.305	0.17
EYR	0.069				
BS	0.034				
PR	0.069				
WPT	0.103				
SST	0.034				
ST	0.034				
NSL	0.069				

Route	2009 Date	2010 Date	2009 Mortality Rate	2009 Species	2010 Mortality Rate	2010 Species
BTRD07	5/27	5/25, 6/21	0	-	0	-
RD0902	5/26, 6/27	5/25, 6/14	1	2 Bullsnake	1	2 Bullsnake
RD0903	5/28	5/25, 6/14	0	-	0	-
RD0907	6/5, 6/28	7/19	3	5 Bullsnake 1 Western Painted Turtle	4	1 Prairie Rattlesnake 1 Bullsnake 1 Common Gartersnake 1 Unknown toad
RD0908	6/5, 6/29	7/19	3	3 Bullsnake 1 Eastern Yellowbelly Racer 1 Prairie Rattlesnake 1 Western Painted Turtle	11	4 Bullsnake 2 Common Gartersnake 1 Northern Leopard Frog 4 Unknown toad
Avg. mo	ortalities/mi	ile	1.4		3.2	

Table 7. Comparison between vehicle mortality rates documented for the same 1-mile stretches of road (n = 5) surveyed by WYNDD in 2009 & 2010.

Table 8. Results of amphibian and reptile surveys in key areas of the Powder River Basin identified by the BLM. Type of surveys conducted varied between sites based on habitat present. We recorded species detected, if water was present, if amphibians sampled tested positive for chytrid fungus, and if dead salamanders were found. Notes about amphibian/reptile habitat at each site area also reported. Detailed species information can be found on Figures 5-14 and in corresponding GIS shapefiles.

BLM Special Interest Areas	Surveys Conducted	Species Detected	Water Present?	Chytrid Detected?	Dead Salamanders?	Notes
Burnt Hollow Management Area	3 VES (lentic) opportunistic	Great Plains Toad Boreal Chorus Frog Bullsnake Northern Leopard Frog Woodhouse's Toad?	Yes	Yes	No	Area was mostly eroded clay with no obvious large rock outcrops; creeks all dry. Only a few ponds present. Found large adult toad with NO CRANIAL CRESTS believed by WGFD and WYNDD experts to be aberrant Woodhouse's Toad.
Welch Management Area	2 Roadkill 3 VES (lotic) 4 Rock outcrop	Bullsnake Common Gartersnake Northern Leopard Frog Northern Sagebrush Lizard Prairie Rattlesnake Spiny Softshell Turtle Gartersnake Sp. Toad sp.	Yes	2009 only	n/a	Tongue River flows through area. Rock outcrops present. Highest roadkill rates in 2009 and 2010 along 2 sections of SR338, especially where highway cross the Tongue River.
Van Houten Draw Area	2 VES (1 lotic, 1 lentic)	Tiger Salamander Woodhouse's Toad	Yes	No	Yes	Small creek that was dry for long stretches. Cattails present in small bunches. Also a small CBNG pond with submerged grasses.
Middle Fork Area near Kaycee	3 Rock outcrop 2 Nocturnal call opportunistic	Boreal Chorus Frog Bullsnake Northern Sagebrush Lizard	Nearby on private land	n/a	n/a	No water except on private land along Barnum Road. Rocky slopes and rock outcrops present near canyon. Sagebrush, juniper, and cacti present near rock outcrops.
Ice Cave	2 VES (lentic) 2 Rock outcrop	Tiger Salamander Toad sp.	Yes	n/a	Yes	Small stock ponds present with heavy cattle disturbance. Ponds had reeds and submerged vegetation. Rock outcrops with sagebrush present.

Mosier Gulch	opportunistic	Bullsnake	No	n/a	n/a	Conducted multiple opportunistic surveys throughout the survey season in Mosier Gulch. Total search time 9-11 hours.
Fortification Creek Area	4 VES (2 lentic, 2 lotic) 2 Nocturnal Call 3 Rock outcrop opportunistic	Boreal Chorus Frog Northern Leopard Frog Woodhouse's Toad Tiger Salamander Bullsnake Eastern Yellowbelly Racer Plains Gartersnake Northern Sagebrush Lizard Gartersnake sp. Toad sp. Turtle sp.	Yes	No	Yes	Most surveys right along Fortification Creek and Fortification Road. Creek varied from dry spots to deep pools. Lots of invertebrates and submerged vegetation present. Ducks also seen. Local landowner reported seeing turtles in the area. Small, often sandy rock outcrops present.
Wild Horse Creek	2 VES (lotic) 1 Nocturnal call	Northern Leopard Frog Boreal Chorus Frog Eastern Yellowbelly Racer Bullsnake	Yes	n/a	n/a	Wild Horse Creek had lots of fish. Tall grass along shoreline was filled with grasshoppers making VES surveys difficult. Lots of dragonflies also present. Nocturnal call route had Wild Horse Creek on the north and the Powder River on the south.
Dead Horse Reservoir 2	none	n/a	No	n/a	n/a	Did not survey due to lack of water.
Dry Creek Area	3 VES (1 lotic, 2 lentic)	Boreal Chorus Frog Woodhouse's Toad Tiger Salamander Bullsnake Wandering Gartersnake	Yes	Yes	No	Dry Creek itself had some stretches with water and some stretches with just patchy pools. Also surveyed 2 ponds in the Flowing E area. Both had lots of pondweed and algae present.
Petrified Tree Area	opportunistic	Eastern Yellowbelly Racer	No	n/a	n/a	Conducted opportunistic searches for ~2 hours but no water present.

Figure 1. Map of the Powder River Basin and Welch Management Area where amphibian and reptile surveys were conducted by WYNDD from 2008 to2010.

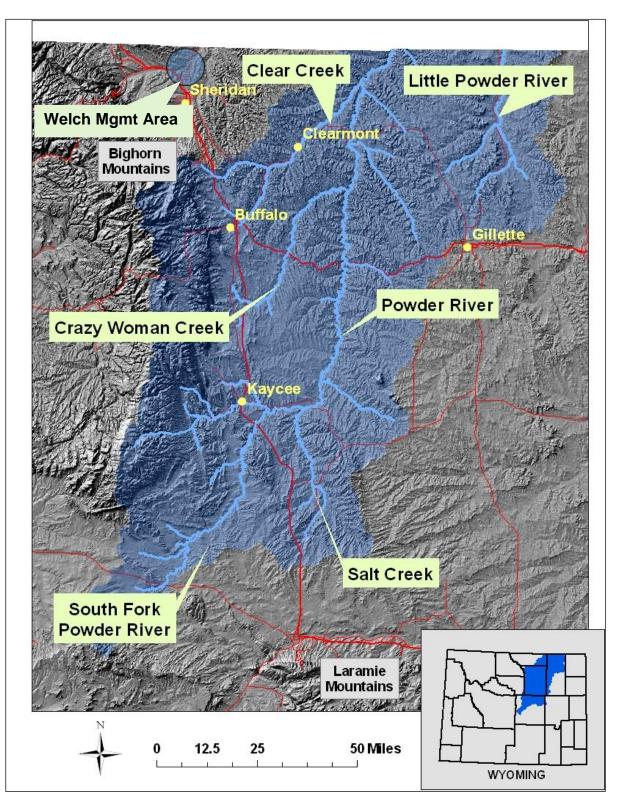


Figure 2. Location of proposed monitoring sites for amphibians and reptiles in the Powder River Basin and surrounding area. Surveys include nocturnal call surveys, Visual Encounter Surveys (VES) at water bodies, and roadkill/basking surveys along roads. Additional longterm monitoring sites are being established by the USGS along the Powder River and are not included on this map. See Table 3 for specific location information.

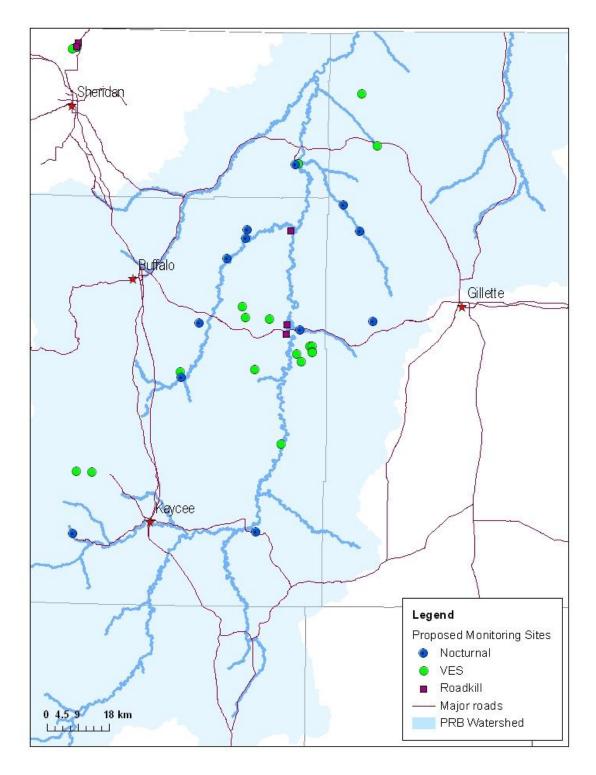


Figure 3. Location of amphibian and reptile surveys conducted in the Powder River Basin in 2010. Surveys included nocturnal call surveys, Visual Encounter Surveys (VES) at water bodies, roadkill/basking surveys along roads, and rock outcrop VES surveys for reptiles.

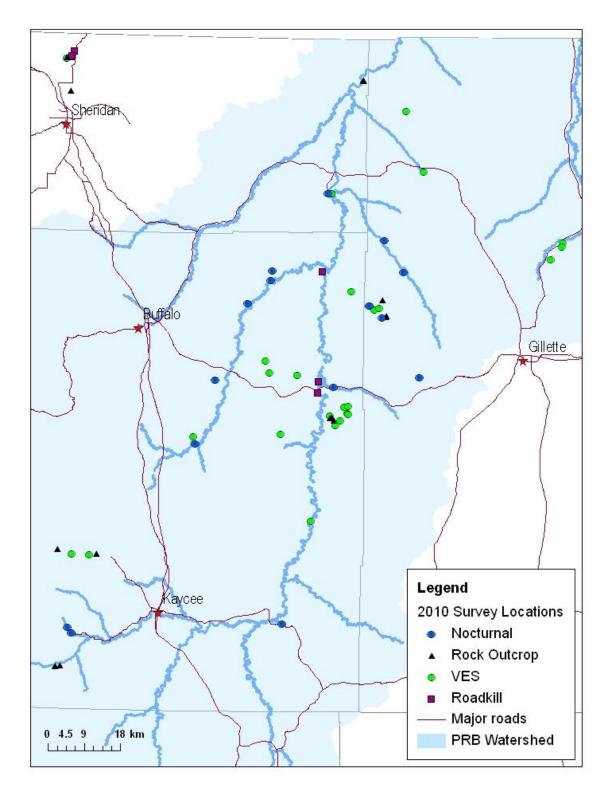
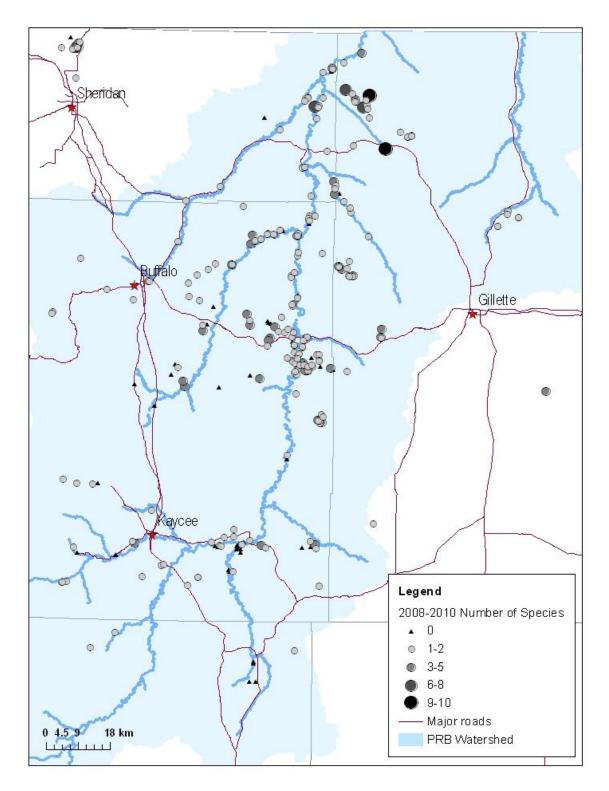


Figure 4. Relative diversity of species detected at survey locations from 2008 to 2010 during WYNDD field work in the Powder River Basin. Surveys included nocturnal call surveys, Visual Encounter Surveys (VES) at water bodies, roadkill/basking surveys along roads, and VES surveys at rock outcrops for reptiles.



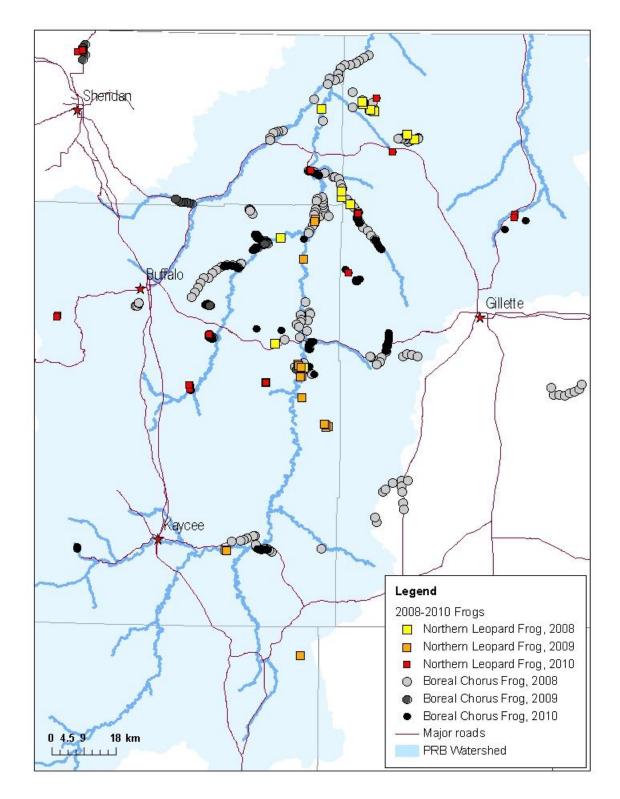


Figure 5. Locations of occurrences for Northern Leopard Frogs and Boreal Chorus Frogs detected 2008-2010 during WYNDD field surveys in the Powder River Basin.

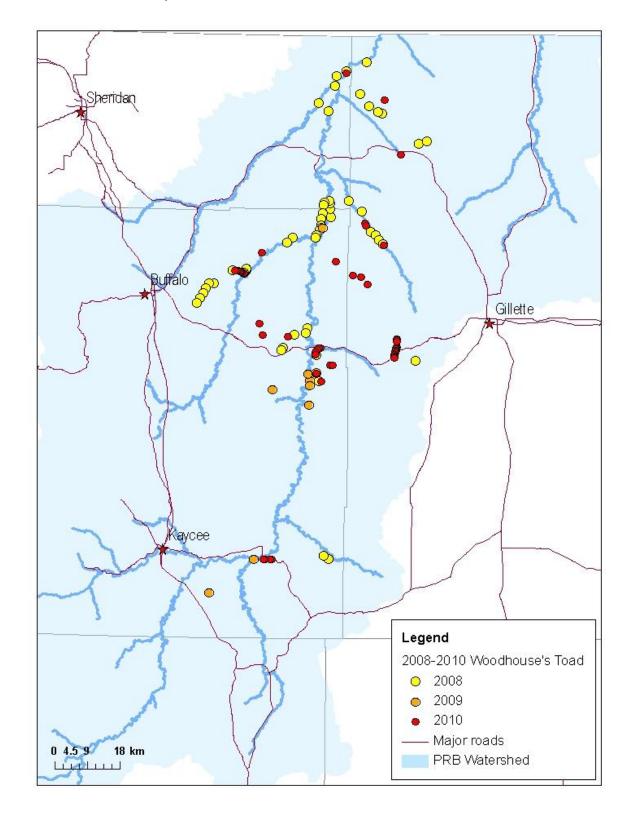
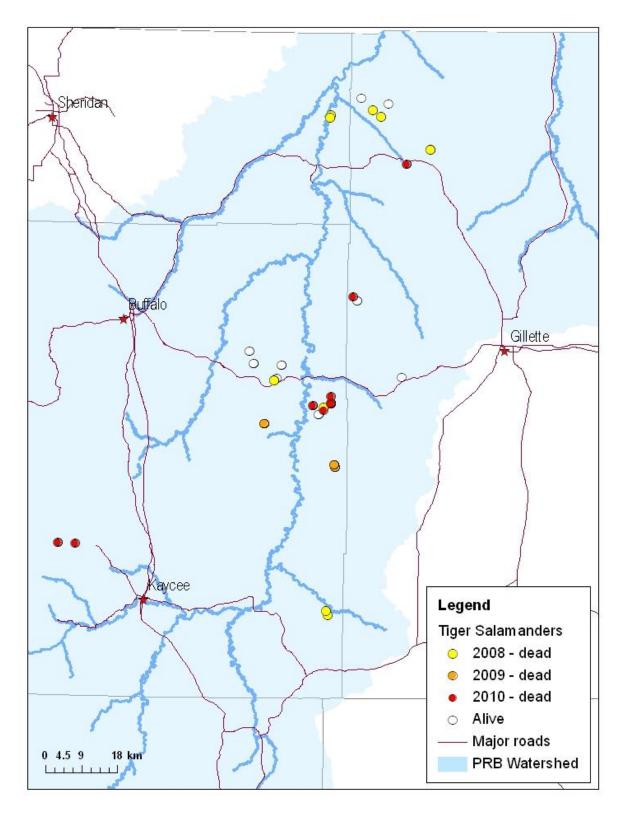


Figure 6. Locations of occurrences for Woodhouse's Toads detected 2008-2010 during WYNDD field surveys in the Powder River Basin.

Figure 7. Locations of occurrences for Tiger Salamanders detected 2008-2010 during WYNDD field surveys in the Powder River Basin. Locations where dead or dying salamanders were documented are noted.



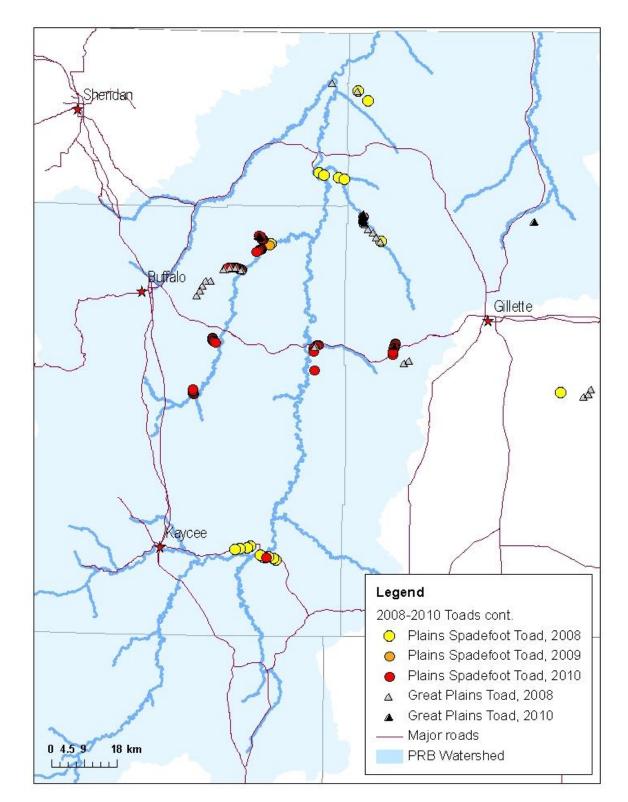


Figure 8. Locations of occurrences for Plains Spadefoot Toads and Great Plains Toads detected 2008-2010 during WYNDD field surveys in the Powder River Basin.

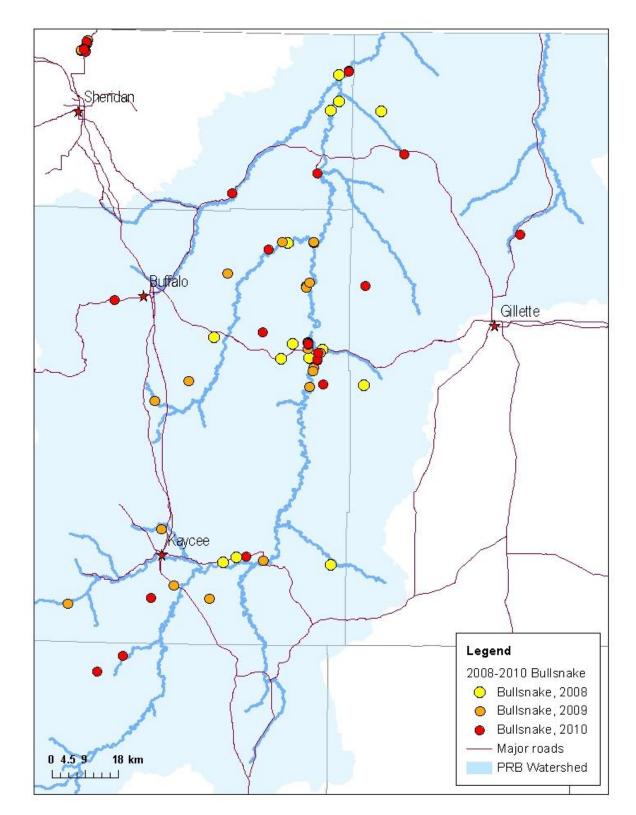


Figure 9. Locations of occurrences for Bullsnakes detected 2008-2010 during WYNDD field surveys in the Powder River Basin.

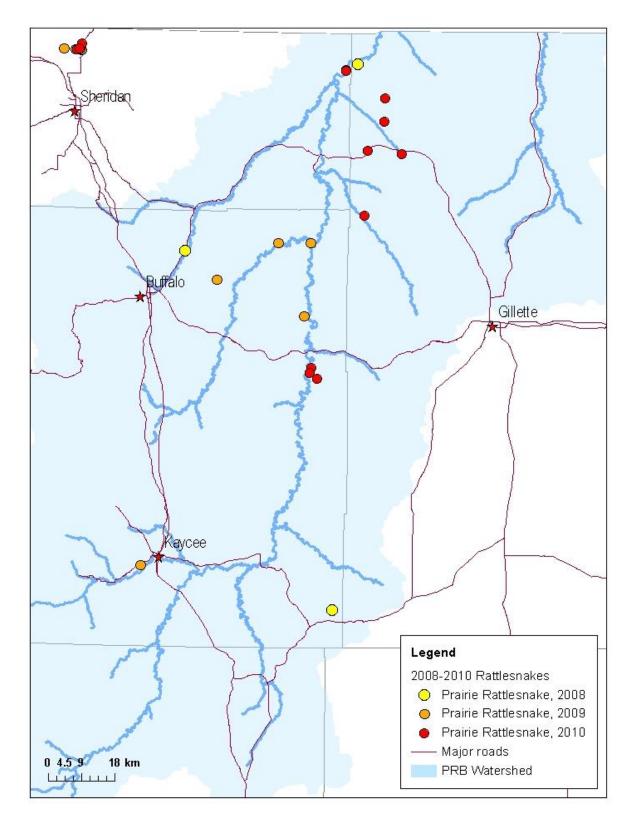


Figure 10. Locations of occurrences for Prairie Rattlenakes detected 2008-2010 during WYNDD field surveys in the Powder River Basin.

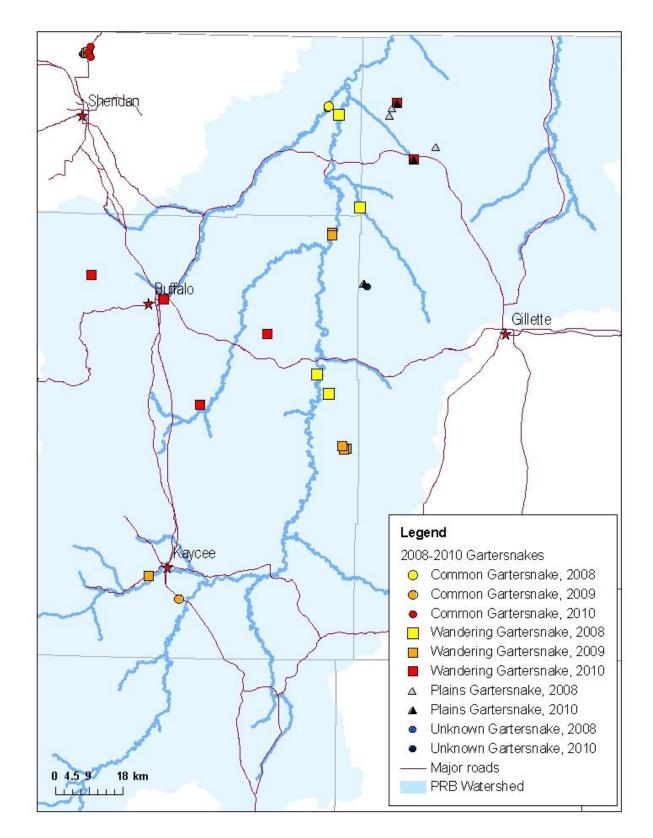
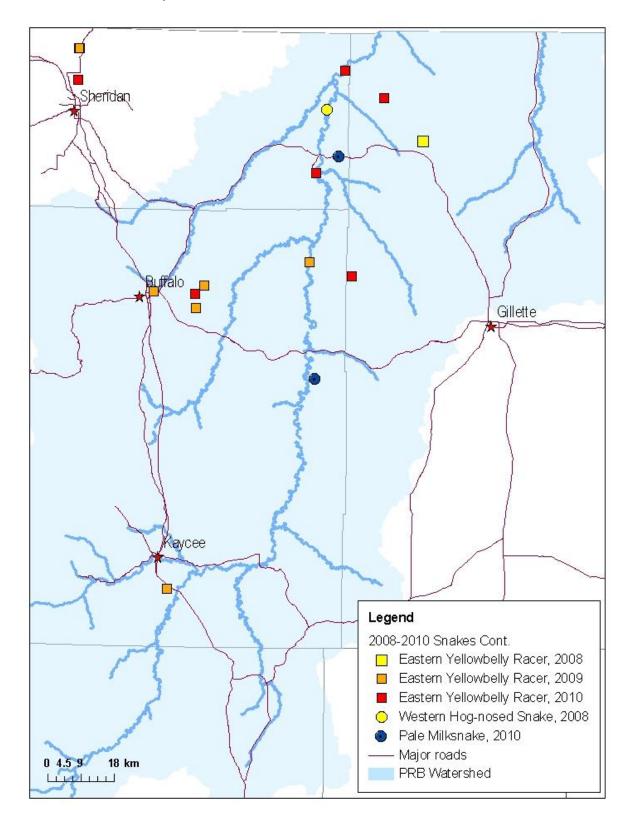
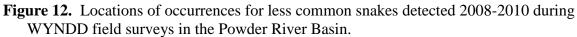


Figure 11. Locations of occurrences for gartersnakes detected 2008-2010 during WYNDD field surveys in the Powder River Basin.





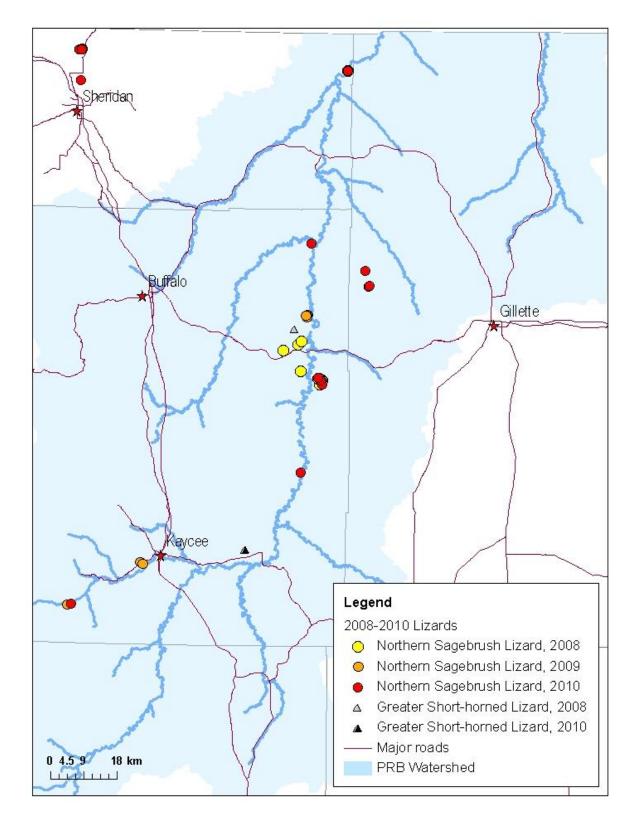


Figure 13. Locations of occurrences for Northern Sagebrush Lizard and Greater Short-horned Lizards detected 2008-2010 during WYNDD field surveys in the Powder River Basin.

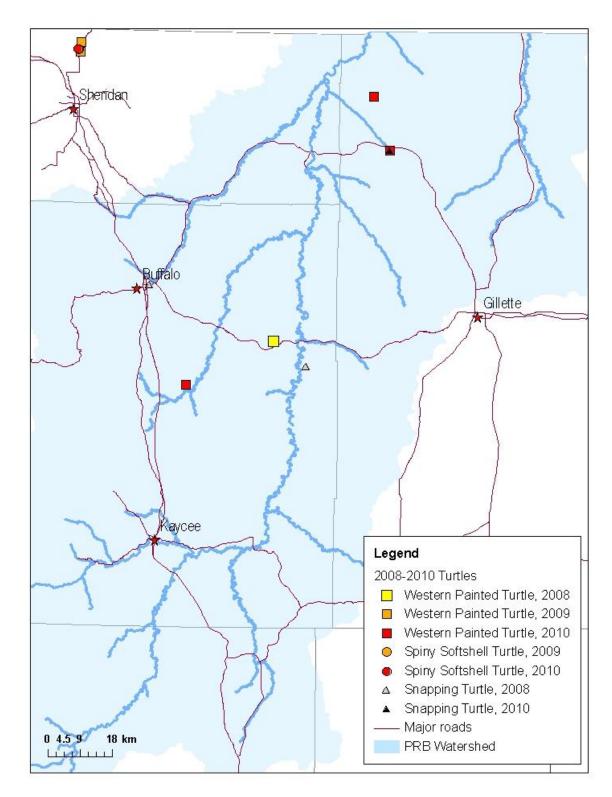
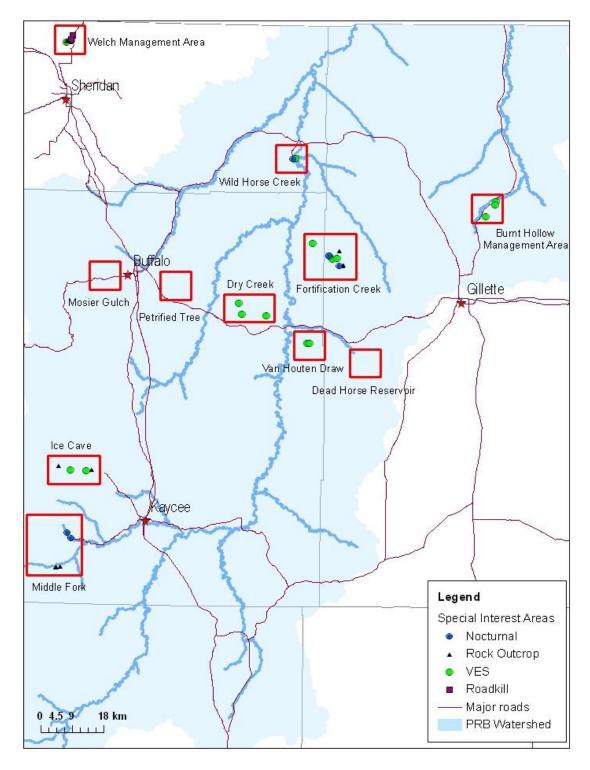


Figure 14. Locations of occurrences for turtles detected 2008-2010 during WYNDD field surveys in the Powder River Basin.

Figure 15. Locations of areas identified by the BLM as areas of interest for amphibian and reptile surveys. The number and type of surveys conducted at each site varied with habitat charicteristics. See Table 8 for habitat descriptions and species results.



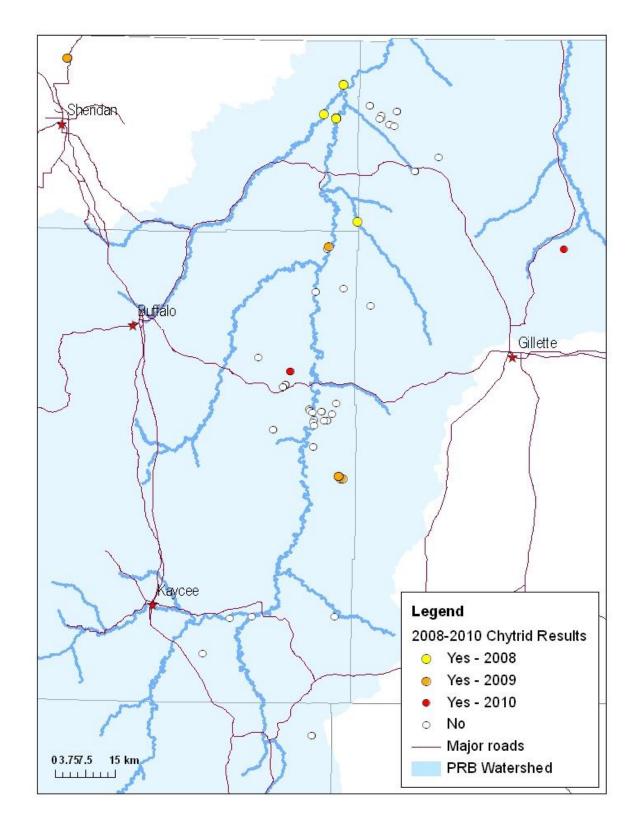


Figure 16. Locations of amphibian in the Powder River Basin area that tested positive for chytrid fungus from 2008 to 2010.