Winter 2017

Effects of Wildfires on Rattlesnake (Crotalus Oreganus) Growth and Movement in Washington State

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EFFECTS OF WILDFIRES ON RATTLESNAKE (*CROTALUS OREGANUS*) GROWTH AND MOVEMENT IN WASHINGTON STATE

A Thesis

Presented to

The Graduate Faculty

Central Washington University

In Partial Fulfillment

of the Requirements for the Degree

Master of Science

Biology

Joseph Chase

November 2017

CENTRAL WASHINGTON UNIVERSITY

Graduate Studies

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ABSTRACT

EFFECTS OF WILDFIRES ON RATTLESNAKE (*CROTALUS OREGANUS*)

GROWTH AND MOVEMENT IN WASHINGTON STATE

by

Joseph Chase

November 2017

Fire is a dominant force in the Pacific Northwest that shapes ecosystems and influences wildlife, yet little is known of its effects on local predators. Northern Pacific rattlesnakes (*Crotalus oreganus*) comprise an excellent model to investigate how fire may influence wildlife because they are important predators that contribute to controlling prey populations, but are also unable to readily escape from wildfires. We developed a novel technique to assess growth rates of rattlesnakes by using digital photography to analyze differences in widths of their rattle segments laid down over time. We compared growth rates of rattlesnakes in habitats that were affected by recent fires with those inhabiting areas unaffected by recent fires. The snakes from the Methow Valley region, in dens affected by the Carlton Complex wildfire of 2014, showed no difference in growth rates before as compared to after the fire, which may be because those snakes have not had sufficient time to respond to potential changes in their prey populations brought about by fire. Methow snake populations from dens affected by fire, however, showed a size structure that was significantly skewed toward smaller individuals than those in dens outside the wildfire area. Snakes that were tracked using radio telemetry in different burned areas did not show any avoidance of burned habitat during the tracking

period.

ACKNOWLEDGMENTS

There are many people who I would like to thank for making this experience and time in Central Washington a true success. Firstly, big thanks to my advisor, Dr. Dan Beck, whose constant curiosity and devotion to the field has instilled me with a pride for the field, and his high standards make me strive to write better, think deeply, and always, always carry a field journal. Other thanks to my other committee members, Dr. Bob Hickey and Mr. John Rohrer, whose expertise in mapping and local wildlife gave me an invaluable amount of knowledge and advice. To John Rohrer, and the staff of the Methow USFS, I am extremely grateful for the housing provided during the field season and the eager volunteers. To the owners of the Swauk Ranch, thanks for the permission to study at their incredible location, as well as a big thanks to the caretakers, Alex and Eric, for their great support of my research while I was there. While collecting snakes, my field assistants made it possible to do anything and everything, big thanks to Carmen, Bryce, Naomi, Alex, John, Scott and many others. Thanks to all my friends and family for the unwavering support throughout this process, even if I was across the country from most of them. Lastly, a big thanks to the School of Graduate Studies and Research for providing funding that made all of this possible.

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CHAPTER I

LITERATURE REVIEW

The Northern Pacific rattlesnake (*Crotalus oreganus*) has an extensive range from Mexico into British Columbia and is one of the most abundant and important predators in dry forest and shrub steppe ecosystems of Central Washington (Fitch, 1949; Klauber, 1972). Molecular systematics has assigned what was once *Crotalus viridis oreganus* to the separate species of *Crotalus oreganus* (Ashton and de Queiroz, 2011; Davis et al., 2016).

Snakes occupying temperate habitats, such as the Northern Pacific rattlesnake, must be able to cope with the regular periods of freezing temperatures. As reptiles, they are ectotherms, so they need heat gathered from the environment rather than internal metabolism, as occurs in mammals. Washington State rattlesnakes communally den underground in hibernacula and may remain there for six months, using particular characteristics of slope, aspect, and distance from other hibernacula for selecting these overwintering den sites (Gienger and Beck, 2011). In British Columbia, rattlesnakes move into hibernacula (ingress) from September to October, and egress (move away from dens) from late March to April (Hobbs, 2007). During the remaining months of the year, the “active season”, snakes forage and reproduce. The earlier part of the season is used for reproduction, as it was observed that ovulation occurred from mid-May to mid-June in Northern Idaho populations of rattlesnakes (Diller and Wallace, 1984). During the reproductive season males move more in search of females, but during the feeding season both sexes demonstrate similar movement (Putman et al., 2013), the exception being that gravid females tend to remain closer to the den site and move less during the active season (Gomez et al., 2015). Site fidelity is important for these snakes, as they return to the same den year after year. This was shown through attempted translocations of rattlesnakes in British Columbia, where 12 out of 14 snakes that were translocated returned to the area they were captured and traveled further on average (Brown et al., 2009).

Since Northern Pacific rattlesnakes in Washington occupy dens, there may be patterns of habitat and resource use that are unique to each den. In British Columbia, the movement patterns were specific to each den, showing different habitat use during the active season for each den (Gomez, 2007). Northern Pacific rattlesnakes had a diet consisting of about 90% rodents and fed predominantly from June to August in British Columbia (Macartney, 1989). Northern Pacific rattlesnakes in California given supplemental hydration had boosted both body condition and reproductive success (Capehart et al., 2016). Also, in diamond-backed rattlesnakes (*Crotalus atrox*), the amount of food consumed by the snakes positively correlated to growth, and it was the size of the animal, rather than the age, that determined reproductive maturity (Taylor and Denardo, 2005). For *C. oreganus* body condition (length and weight) was more similar within dens, and animals that were in closer proximity to humans and associated disturbance tended to be smaller and had lower body condition (Jenkins et al., 2009; Lomas et al., 2015). These lines of evidence suggest that rattlesnakes, including the Northern Pacific rattlesnake, are driven by resource limitations, habitat suitability, and human disturbance that influence individual and den-level success.

For Northern Pacific rattlesnakes and their relatives, the distinguishing feature is the rattle. These keratin structures are segmented, and a new segment is added to the rattle every time the rattlesnake sheds its skin. The exposed segments are composed of lobes that, powered by the shaker muscle during rattling, strike against each other rapidly, to create a buzzing noise (Klauber, 1972). Young and Brown (1995) described the rattle as a multi-dimensional oscillator, where the frequency of sound produced is derived by the proximal (basal) rattle segment.

Shedding is an important, regular occurrence in all snakes. Snakes tend to be more vulnerable to predation when they are shedding, partly because clouding of the ocular scales which cover the snakes' eyes. In Washington, *C. oreganus* are social during these times and used aggregated shed sites near/in basalt outcrops, different from the den, as a conspicuous and safe place to shed (Loughran et al., 2015). When rattlesnakes are born, they have a pre-button segment that is shed days after birth and replaced with the first permanent segment called the button. As they shed, they add another segment to the proximal end of the rattle, forming a string of rattles (Klauber, 1972). Early observations of rattlesnakes in Kansas gave insight into these features, including that strings of rattles tended to get longer with bigger snakes and that correlated strongly with the age of the animal (Fitch, 1985). Research has also shown a strong correlation between the width of the basal rattle segment and the snout to vent length (SVL) of the snake (Beaupre et al., 1998; Wittenberg and Beaupre, 2014; Beck et al., 2014; Geroso, 2014). Thus, the rattle is an important, quantitative record of how the individual snake has changed in size through time. Large-scale natural events like wildfires may cause population level changes in growth that can be tracked by looking at individual rattles.

In Washington, the increase in number and size of wildfires has been of growing concern (Littell et al., 2010; Rogers et al., 2011). Current studies into the direct, immediate effects of wildfire in similar communities have been conducted in the deserts of the southwestern United States. These studies found that animal mortality and alteration of vegetation cover were the largest effects of the wildfires on the community (Esque et al., 2003; Rochester et al., 2010). Since rattlesnakes are predators, the cascading effects of vegetation and small mammal prey populations would be important in understanding how fires influence their environment and food base. In Washington, the Northern Pacific rattlesnake predominantly occupies shrub steppe habitat, which has an entangled history of fire and fire suppression.

Cycles of fire suppression have resulted in more shade-tolerant species occurring in the northwestern US, as well as homogenization of the landscape (Hessburg and Agee, 2003). However, fire also had an effect of homogenizing the soil surface texture for years following fire in New Mexico (White, 2011). Homogenization could influence the habitat suitability for small mammals, which in turn may influence suitability for snakes.

In Washington State, effects of fire on the vegetation were shown to be inversely related to elevation; as elevation increased there was a loss in vegetative cover, species richness, and seedling density (Dodson and Peterson, 2010). Although higher elevations had lower richness attributed to the fire, they were more resilient to change in composition; however, lower elevations, where rattlesnakes occur, changed from primarily seeding species to weedy, early-successional species two years after a small controlled burn (Davies et al., 2012). In these lower elevation areas, the dominant vegetation is sagebrush (*Artemisia spp*.), a study in Oregon has shown that sagebrush may increase productivity for two years after a fire (Davies et al., 2007) This indicates that for the sagebrush species, there may be increased resources available to them after a fire. The resilience to change in the post-fire landscape for sagebrush in Wyoming was enough that livestock grazing didn't impact herbaceous plant community recovery, it had only resulted in less seed production (Bates et al., 2009). Alternatively, the dominant shrub in the Methow region is bitterbrush (*Purshia tridentata*), which responds a little differently to fire. A study from Central Oregon found that regrowth of bitterbrush occurred quickly after the first fire, however when subject to yearly repeated burns had a lower recovery and recruitment rate (Busse and Riegel, 2009). Cheatgrass (*Bromus tectorum*) is an invasive species that has been increasing in presence in Washington State, and can affect the landscapes more than fire alone. The success of cheatgrass in Nevada was evident two years after a fire; it was also responsible for negatively affecting the soil water content, water potential, and biomass of native species in their proximity (Melgoza et al., 1990). Invasive species have adverse effects for other shrub steppe plants, for example, sagebrush in Wyoming had poor efficiency after a fire in systems dominated by invasive plants (Taylor et al., 2013). Sagebrush steppe habitat is typically able to rebound from a single fire well as these studies have shown, however the introduction of invasive species by human activity is adding another facet that these communities haven't had time to cope with.

Habitat influences the snakes directly, but also indirectly through their prey. The main prey for Northern Pacific rattlesnakes in British Columbia are small mammals including voles (*Microtus spp.*), deer mice (*Peromyscus maniculatus*), northern pocket gophers (*Thomomys talpoides*), and red squirrels (*Tamiasciurus hudsonicus*) (Macartney, 1989). Little research has been done in Washington on the effects of fires on these small mammals; however, research in other areas can provide insight. In California, prescribed burns did not affect small mammal abundance, and some species actually increased after the fire (Amacher et al., 2007). Timing of the prescribed fires in another California study revealed that there was no difference in early and late season fires on both deer mouse and lodgepole chipmunk populations (Monroe and Converse, 2006). Through a meta-analysis of small mammal response to small-scale burns in the southwestern US, similar results were found, i.e. little evidence of negative effects in the short-term after low intensity fires (Kalies et al., 2010). These studies all suggest that mammalian populations have adapted to recent fire conditions, however some of these studies also acknowledge the need for both long-term and larger spatial scale studies to better understand the situation (Amacher et al., 2007; Kalies et al, 2010).

Studies on larger spatial scale effects of fires on Northern Pacific rattlesnakes are also needed, as there are no published studies on the effects of wildfire on *C. oreganus*. The few studies of fire effects on other reptile species give some insight into the potential direct and indirect effects that could be expected. For reptile populations affected by fire in Australia, the biggest change was in the assemblage of reptiles, where some species were only present before the fire (Abom and Schwarzkopf, 2016). Indirect effects of fires can influence habitat use; in Australia, habitat specialist snakes (desert death adder, *Acanthophis pyrrhus,* and monk snake, *Parasuta monachus*) were more likely to avoid burned habitat (McDonald et al., 2012). Yet, in massasauga rattlesnakes (*Sistrurus catenatus*), another habitat specialist, movement, home range size, and habitat use were not influenced by a low intensity prescribed fire (Cross et al., 2015). The Orsini's viper (*Vipera ursinii*) did show a difference in pre- and post-fire survival within the French Alps after a prescribed fire (Lyet et al., 2009). For species already under pressure, like the ridge-nosed rattlesnake (*Crotalus willardi obscurus*), climate change is restricting the upper elevation with increased wildfire risk and lower elevations with higher drought; these combined factors lead to high risk for extinction (Davies et al., 2015). Results from such studies suggest variable snake responses to fire or disturbance. However, when one considers studies similar in scale and intensity to the bigger wildfires in Washington State (~1000km^2), only the specialist snakes studied in Australia (~1400km^2), which showed a preference for unburned areas, is comparable (McDonald et al., 2012). The situation in Washington provides a valuable opportunity to more effectively draw conclusions about potential effects.

The Washington wildfires relevant to this study occurred in mid-summer, during the active season of the snakes, April-October, when they were likely foraging for mammalian prey (Macartney 1989; Wallace and Diller 1990). During these times, the rattlesnakes would mostly be dispersed from the dens and in areas that were exposed to the fire. Unlike other predators in the area, like hawks, coyotes, and cougars, snakes cannot move quickly to escape the fire. Through tracking rattlesnakes, one can assess the habitat they prefer and whether they avoid the burned habitat, which would indicate a behavioral response to these fires. Any of these changes in the predators may have cascading effects on the rest of the ecosystem; such effects have been shown across many ecosystems (Estes and Palmisano, 1974; Ripple and Beschta, 2004). Thus, these rattlesnakes represent a vital indicator of the ecosystem and the factors affecting them may be amplified onto both lower and higher trophic levels, and indicate effects on the broader ecosystem's health.

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CHAPTER II

ARTICLE

ABSTRACT

Fire is a dominant force in the Pacific Northwest that shapes ecosystems and influences wildlife, however little is known of its effects on local predators. Northern Pacific rattlesnakes (*Crotalus oreganus*) are important predators in the area that influence prey populations and are, in turn, prey to some raptors, yet are also unable to readily escape from wildfires. These snakes thus comprise an excellent model to investigate how fire may influence wildlife. To determine whether the snakes were affected by the fire, the growth rates of rattlesnakes were measured in habitats that were affected by recent fires and compared with those in areas unaffected by recent fires. The snakes measured in the Methow region, affected by the Carlton Complex wildfire of 2014, showed no difference in growth rates with nearby populations unaffected by fire, possibly a result of snakes having insufficient time to respond to potential changes in their prey populations brought about by fire. Snake populations from dens affected by fire, however, showed a size structure that was significantly skewed toward smaller individuals than those outside the wildfire area. Snakes that were tracked in different burned areas using radio telemetry did not show any avoidance of burned habitat during the tracking period. Rattlesnakes in habitats affected by less recent fires, like the Taylor Bridge fire of 2012, may be more likely to show a growth response to wildfire. From this experiment a technique was developed to assess growth rates of rattlesnakes by analyzing the widths of their rattle segments over time using digital photography. The technique developed resulted in quicker rattle measurements to reduce handling time of the animal as well as providing a

permanent record to monitor over time.

INTRODUCTION

Understanding how organisms grow through time is a way to observe the effects of a change in the environment. Monitoring growth usually requires immense time commitments. However, there are some cases where growth records are preserved by the organism. Annual growth rings on trees are one of the well-known examples, where seasonal differences in growth are visible in trunk horizontal cross sections (Studhalter, 1956). These features allow for historic size and growth to be immediately available for measurement, and thus can be very useful ecological metrics. For terrestrial animals, there are few examples of these features; however, one comes from an unexpected place, namely in the rattle of the rattlesnake. The rattlesnake makes an ideal study organism to better understand how large events, like wildfires in Washington State or elsewhere, may change trajectories in growth and be an indicator of the surrounding areas' health.

Although rattlesnakes are given a bad reputation, there are many reasons that these animals can provide an abundance of information. For Washington State, the only native rattlesnake species is the Northern Pacific rattlesnake (*C. oreganus*). This snake however, has populations far outside of Washington State, with an extensive range from southern California into British Columbia (Klauber, 1972). As a predator, snakes represent a factor in maintaining small mammal populations, and comprise a component in trophic structure stability (Ripple and Beschta, 2004; Bestion et al., 2015). However, unlike many other predators, they are unable to escape an oncoming fire quickly and may only move approximately 2 km away from their den (Klauber, 1972; Gomez, 2007). The large numbers of snakes congregating in dens during winter time may be a useful mechanism for thermoregulation (Graves and Duvall, 1987) or driven by a lack of adequate refuges in the area (Hobbs, 2007), and ultimately is an effective way for increasing sampling success since they all emerge around the same time in spring.

As the Pacific Northwest is seeing more frequent and intense wildfires during the summer months (Littell et al., 2010; Rogers et al., 2011), we need to understand these effects on local fauna. The persistence of increasing regional droughts, further adds to the possibility that these fires will be a recurring issue (Perry et al., 2011). These recent, large-scale events may bring dramatic change to the landscape but they also provide an opportunity to address key questions regarding the long term effects of the fires on wildlife. Obvious direct effects are still visible as blackened scars on the landscape, but how the wildlife rebounded after the incident is largely unknown. Using the rattlesnake as the focal species, and the rattle segment width as a growth indicator, information can be extracted about possible long- and short-term consequences of these fires on snake growth.

Wildfire's influence on rattlesnake growth is most likely mediated through its effect on their prey populations. The main food sources for *C. oreganus* are small mammals, lizards and birds (Macartney, 1989). Other studies have provided insight into how prey populations may be affected by wildfire. In Victoria, Australia, where 3500 km^2 burned, two small mammal species in the area of a fire occurred at one third the density of nearby unburned sites one year after the fire (Banks et al., 2011). However, in a smaller Chihuahuan Desert grassland fire, where 0.2 km^2 burned, canopy cover was markedly reduced, yet neither rodent abundance nor species richness was altered

(Killgore et al., 2009). These contrasting results were apparent through a meta-analysis of response to fire in United States fire-prone forests, where there were mixed rodent response to fire (Fontaine and Kennedy, 2012). When smaller, less intense fires were studied there was often a benefit to biodiversity in the burned area following the fire. However, in the larger, high-severity fires the rodents had a negative response and experienced a loss in biodiversity. The avian responses to the prescribed fires and wildfires in the United States forests were predominantly positive with both small and large scale fires (Fontaine and Kennedy, 2012). Given the size and severity of the Carlton Complex fire, the Australia study (Banks et al., 2011) is most comparable, and would indicate there might be a loss of rodents in the area. Since rodents accounted for around 90% of the Northern Pacific rattlesnake diet (Macartney, 1989), a severe reduction in food would likely cause growth in the snakes to be negatively affected after the fire. Western yellow-bellied racer (*Coluber constrictor mormon*) abundance was not different in burned and unburned sites in California (Thompson et al., 2013). This would suggest snakes may be more tolerant to the direct effects of the fire. If the rattlesnakes are influenced less by the fire than are the rodent populations, intraspecific competition for resources among snakes would be stronger and snakes could thereby experience negative growth.

Each string of rattles contains a record of growth and thus provides a unique opportunity to examine how wildfire may influence growth rates in an important predator. Every time these animals shed they create a new rattle segment, the basal (most proximal) segment of a string of rattle segments. As adults, Northern Pacific rattlesnakes shed once per year, thus adding one new rattle segment per year (young may do so two or more times a year in their first two years, Fitch, 1949; Diller and Wallace 2002). The width of that basal rattle segment is strongly correlated with the snout-to-vent length of the snake (Beaupre et al., 1998; Beck et al., 2014; Geroso, 2014; Wittenberg and Beaupre, 2014). Each year when they shed, individuals add one new segment to their string of rattles. Therefore, an adult string of rattles reflects changes in growth at yearly intervals, and thus contains a record of the snakes' size through time (Beck et al., 2014). In this study, digital photography was used to capture a record of the strings of rattles and associated growth rates of rattlesnakes.

A second, complementary approach to understanding potential effects of wildfire on snakes is to observe behavior. Previous research has shown that Australian desert death adders (*Acanthophis pyrrhus*) and monk snakes (*Parasuta monashus*) avoided habitats that were previously burned (McDonald et al., 2012), whereas massasasaugas (*Sistrurus catenatus*) in Michigan, U.S. showed no avoidance of burned habitats in a prescribed fire (Cross et al., 2015). As rattlesnakes, *Crotalus oreganus* and massasaugas (*S. catenatus*) may show similar behaviors and exhibit no preferential avoidance of burned habitat. However, to date, no study has explored such effects on Northern Pacific rattlesnakes, or snakes in general in the Pacific Northwest. Two locations with known rattlesnake dens, the Swauk and Frenchman Coulee, in Central Washington were burned in recent years. Both of these areas, even several years after the fire, still have charred remnants of trees and vegetation blackened from the fire, which present opportunities to explore whether burned habitat features are avoided by wildlife.

The following study was conducted to investigate growth and movement responses of rattlesnakes to wildfire. For growth, rattles provided the key information that was used to test whether wildfires influence growth rates of snakes within and outside the wildfire area, using the Carlton Complex as the focal area. The driving hypothesis being that there is an effect on individual rattlesnake growth in response to wildfires. Because growth is mediated through prey populations, and wildfire may reduce prey abundance, rattlesnakes might experience reduced growth after the fire. For movement, snakes were radio-tracked to determine whether they avoid habitat that was previously burned, using both the Taylor Bridge and George fires as focal areas. The main hypothesis is that snakes will avoid habitat that was previously burned. As these snakes are likely to return to the den it would be expected that a noticeable change in their previous foraging habitat may cause the snakes to search nearby for more suitable unburned habitat.

STUDY SITES

There were three study sites used for this investigation, the Methow, Frenchman Coulee, and Swauk areas of Washington State (Figure 1), each of which have experienced recent fires and have Northern Pacific rattlesnake dens in the vicinity. In order to answer the two research questions, the sites were used to collect two different sets of data for the different analyses. The Methow area was utilized to measure and compare growth rates of snakes from dens within fires to those outside the fire area, this round of data collection occurred first. The Frenchman Coulee and Swauk areas then were used for tracking individual snakes to observe habitat selection in a burned landscape.

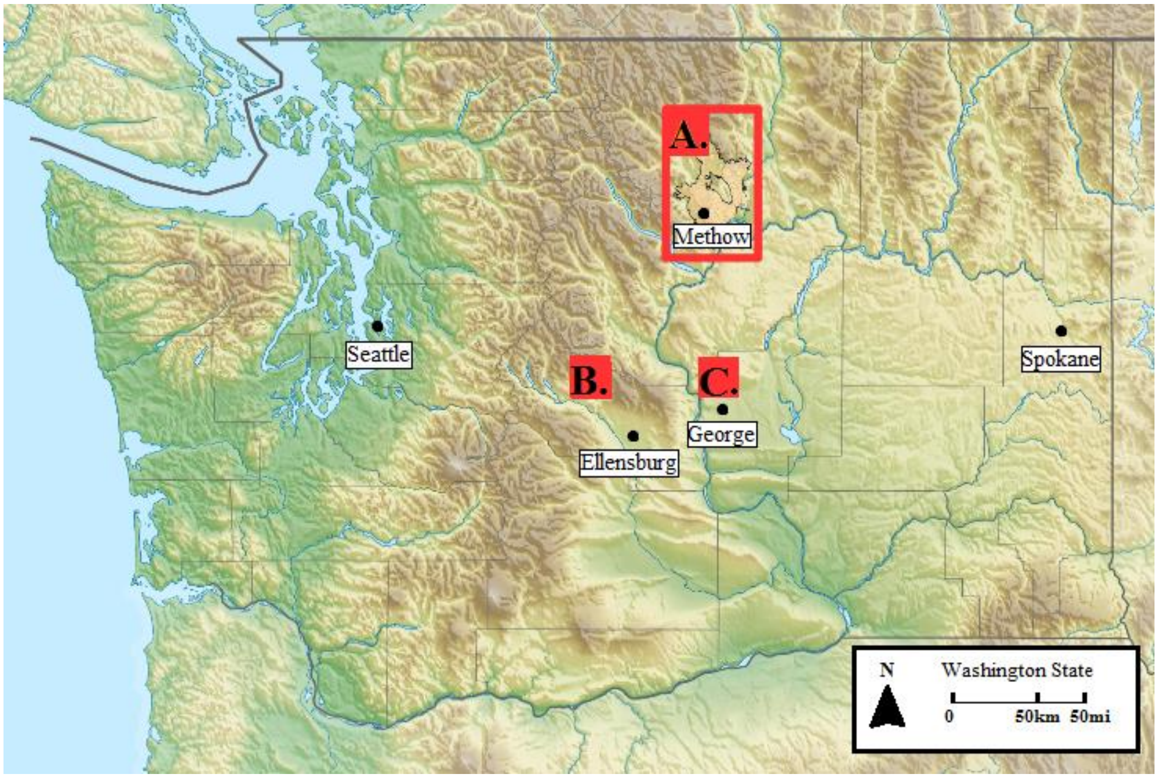

Figure 1: Washington state physical map (Carport, 2012) with the study sites indicated; A. Carlton Complex fire in the Methow Valley, B. Taylor Bridge fire in the Swauk, C. George fire in the Frenchman Coulee.

The three study sites have several ecological properties that are shared. All of these areas are predominantly sagebrush steppe habitats with hills covered by big sagebrush (*Artemisia tridentata*), wild buckwheat (*Eriogonum spp*), bitterbrush (*Purshia tridentata*) and grasses, the most common of which is bluebunch wheatgrass (*Pseudoroegneria spicata*). The Northern Pacific rattlesnake dens were usually found on talus slopes with predominantly southern and eastern facing exposures (Gienger and Beck, 2011). Site differences and properties are discussed below.

The Methow Valley in late July of 2014 was the location of the largest wildfire to occur in Washington State recently, the Carlton Complex fire, which burned an area of 1000 km^2 (250,000 acres) in northern Washington (USFS, 2014). Previous work in the area had been conducted on rattlesnakes and many den locations are known (Figure 2; J. Rohrer, pers.comm.; Geroso, 2014). Five dens were sampled, three from within the wildfire area and two outside its perimeter. The dens occupied by rattlesnakes were found from 760-910m elevation (2500-3000ft). For 2016, the total liquid content (TLC) of precipitation was 40.5cm (15.9in; NOAA, 2016). The Methow River and Chewuch River run north to south in the valley. The area is a shrub-steppe habitat containing both local and invasive ground cover, as well as forests dominated by ponderosa pine (*Pinus ponderosa*).

Since this fire was the most recent to occur and covered the largest area, there were several observations about how the wildfire had influenced the landscape, both for individual snakes and the population at large. Individual snakes provided evidence for the effects of fire on the animals' physical exterior. Burns, which did not noticeably affect the snakes' behavior, were found on multiple snakes, both on the bodies and rattles of the animals (Figure 3). The landscape level changes from the fire are clearly visible from these dens, as trees, shrubs, and grass alike were charred and destroyed (Figure 4). Recent rains at the dens had left some snakes in the fire dens coated in dirt, from their head to the rattle, snakes' behavior was unnoticeably different as well (Figure 5). None of these lines of evidence were found on snakes or at dens outside of the wildfire area, and indicate potential changes that were not directly measured but should be noted.

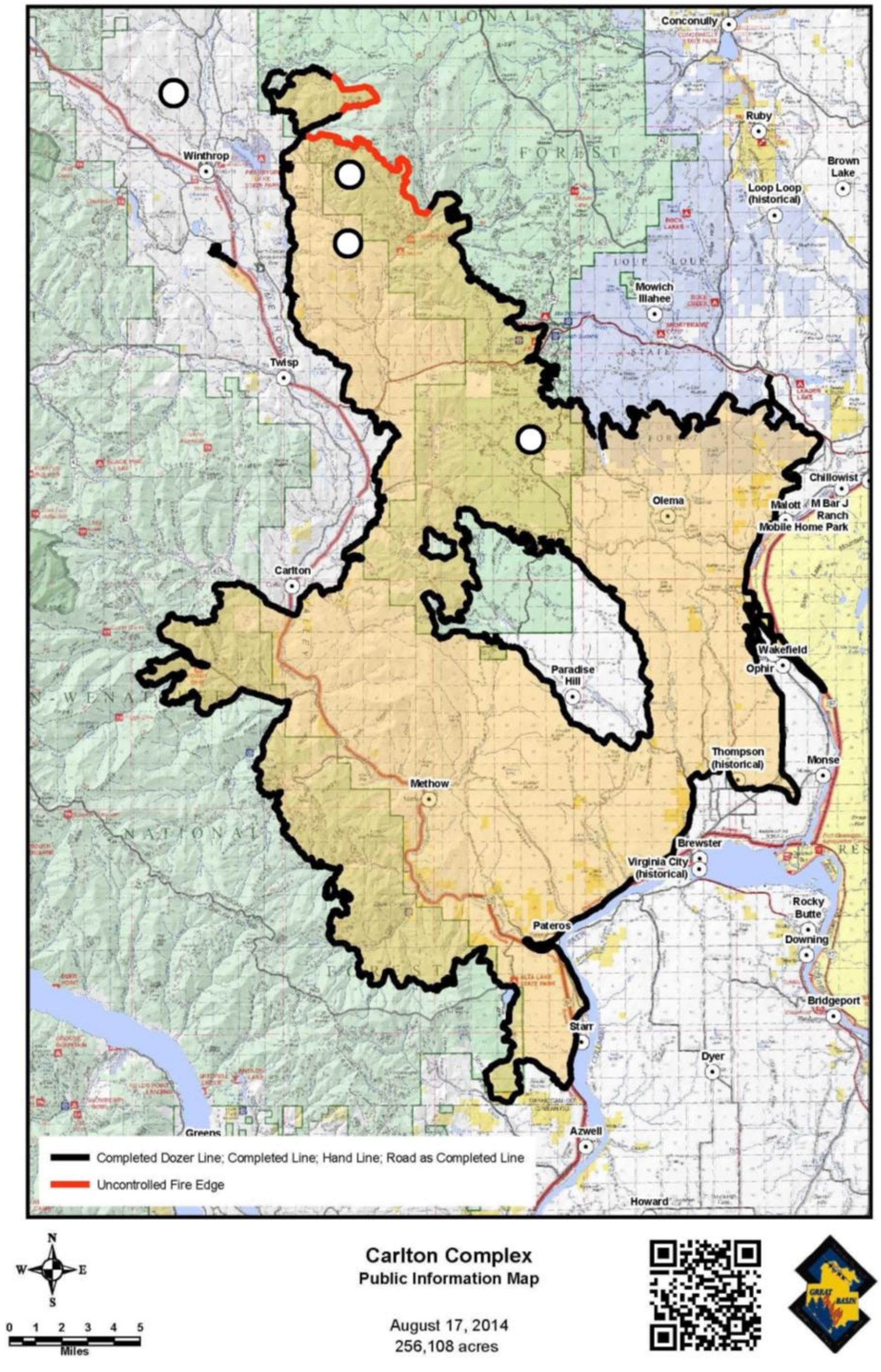

Figure 2: Carlton Complex fire situation map (USFS, 2014) outlining the extent of the 2014 fires in the Methow Valley. The four dots indicate all but one of the dens sampled in the area. They were each given a name to identify them, from top to bottom: Gunn Ranch, Cougar, Pipestone, and Finley. Uphill is the only den not visible, and was north of the Gunn Ranch den.

Figure 3: Presence of external scarring on individuals, both on the body and the rattle.

Figure 4: View of the scorched landscape from the den rock in Finley Canyon.

Figure 5: Several snakes from the Pipestone den, located within the fire area, showed evidence of having been coated in mud.

In August of 2012, the Taylor Bridge fire burned a total of 95 km^2 or 23,500 acres (USFS, 2012), including Swauk Creek canyon, which harbors several known rattlesnake dens. The Swauk den, affected by the fire (Figure 6), was at 580m elevation (1900ft) and was situated at the mouth of a canyon with southern exposure. The den was a talus slope with scattered large rocks (1m or larger), and surrounding shrub steppe habitat. TLC of precipitation in 2016 was 25cm (10in; NOAA, 2016). Swauk Creek, a perennial stream approximately 50m south of the den, is the main water source.

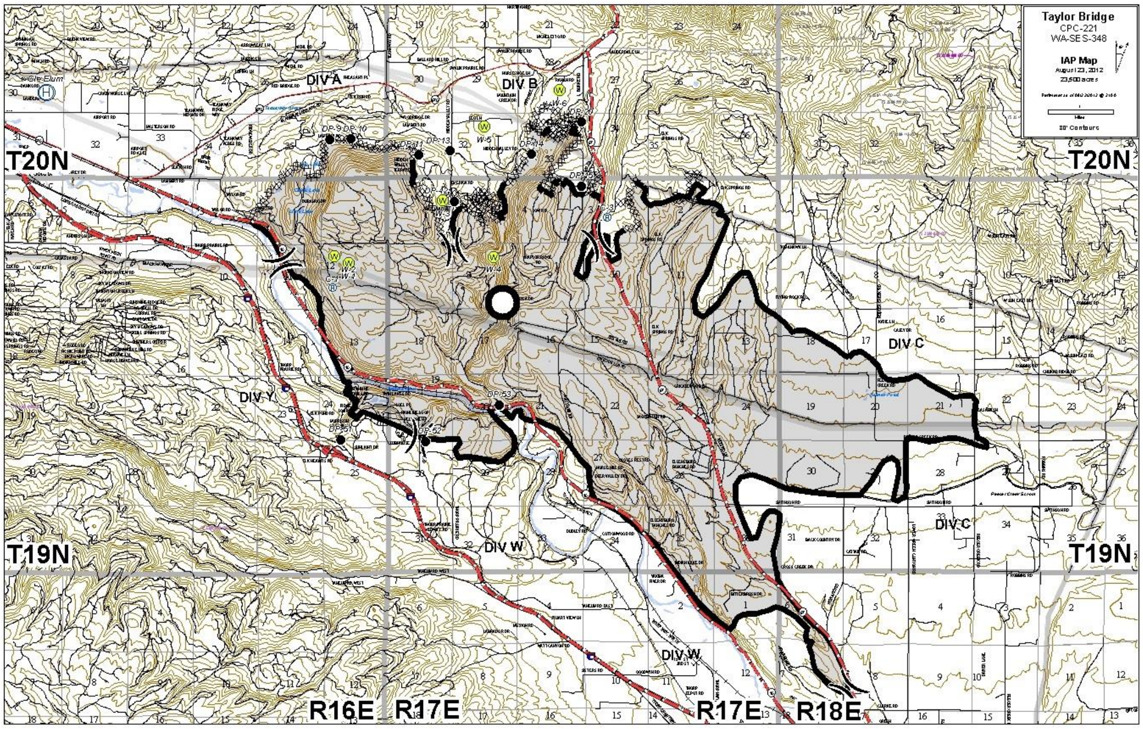

Figure 6: Taylor Bridge fire map (USFS, 2012), indicating the extent of the 2012 fire. The dot indicates the den used in this study.

In 2015, upper Frenchman's Coulee was burned during the George fire on July 20^{th}, burning approximately 2 km^2 (500 acres). The area contains two known dens in an area of shrub steppe habitat bordered by basalt outcrops and cliffs that surround most of the site, adjacent to an interstate highway (Figure 7). Dens were at approximately 350m elevation (1150ft), and the TLC of precipitation was 27cm (10.5in; NOAA, 2016). The foraging area is mostly a flat steppe with an arcing network of wetlands extending along the southern and eastern borders of the expanse. Prior to the fire, sagebrush (big sage, *A. tridentata* and stiff sage *A. rigida*) and wild buckwheat (*Eriogonum spp*.) comprised the dominant vegetation. The open areas were carpeted with invasive cheatgrass (*Bromus* *tectorum*), with trees, mostly Russian olive (*Elaeagnus angustifolia*), occurring along the wetlands.

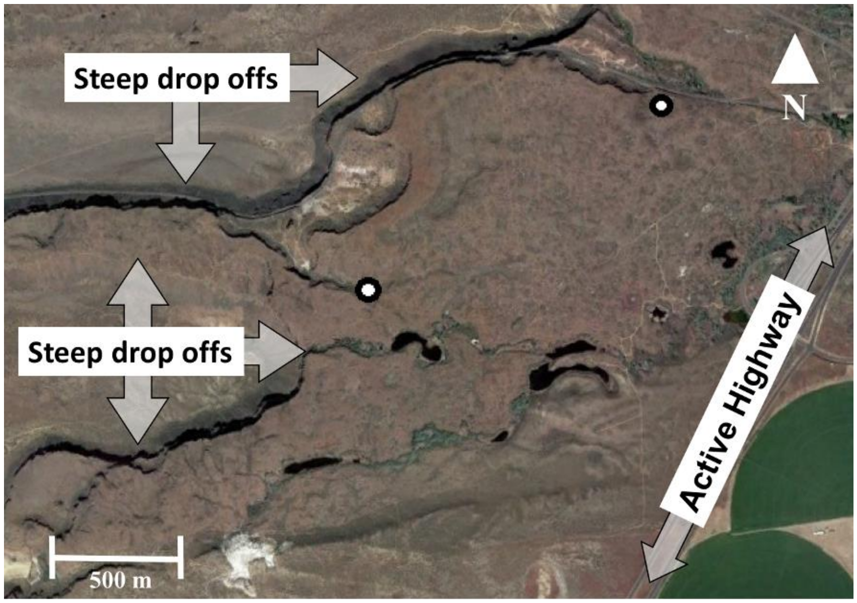

Figure 7: Frenchman Coulee area terrain map (Google Maps, 2017) showing the areas that were likely barriers to the snakes as they were not utilized by the animal in the study. The two dots indicate known den locations in the area, the lower-left den was the focus of this study. The George fire burned the shrub steppe area between the two dens.

METHODS

Growth Measurements

The growth hypothesis data was collected entirely from the Methow area. We sampled rattlesnakes in the Methow Valley from late April until early June, 2016 as the rattlesnakes dispersed from their winter dens to the summer foraging areas. By sampling before dispersal, the width of the basal rattle would more likely represent the previous year's growth, showing potential changes that occurred as a result of the wildfire in 2014. However, the exact foraging patterns were unknown for this population, but this is the most likely scenario given previous research (Wallace and Diller, 2001; Gomez et al., 2015). Three dens were sampled within the wildfire area, and two dens were sampled that were not affected by the wildfire for use as a base of comparison. Dens were frequently sampled during the morning as snakes would all emerge around the same time.

From within wildfire areas 37 snakes were measured and 48 snakes were measured from outside the wildfire area. Snakes were coaxed into clear plastic restraining tubes for safe handling (Antonio, 2014), and then SVL (snout-to-vent length, to the nearest centimeter), tail length, and sex were recorded for each snake. The strong correlation between the width of the basal rattle segment and the SVL of the snake (Figure 8; Beaupre et al., 1998; Wittenberg and Beaupre, 2014; Beck et al., 2014) makes the width of the basal rattle an important measure for looking at growth in the study. The basal rattle segment width was then carefully measured in millimeters using digital calipers. The remaining rattle segment sizes were measured from a standardized digital photograph taken of the rattle. This was accomplished by creating a stand to hold the camera connected to a base for the rattle to be placed upon (Figure 9). With this design, the rattle of each tubed snake was placed flat onto the center of the base beneath the camera, where a high contrasting background was used to preserve the outline of the rattle. The photograph taken of each rattle was stored and recorded for future analysis (Figure 10). The photography method used here was an effort to reduce handling time for the snakes and increase the precision in measuring the remaining rattle segments. The alternative to measuring the segments would rely upon caliper measurements, which are done using human vision and touch to precisely take the width of each segment, the human error associated with this can be high and can be avoided using this digital approach. Additionally, the older rattle segments become worn and fragile over time which may result in segments breaking off or cracking, making them difficult to accurately or precisely measure. To ensure no individuals were sampled twice, a reference photograph of each snake was also taken of the dorsal patterns, which are unique to individuals (Klauber, 1972). Also, ventral scales were clipped in a unique sequence to assign an identification number to each snake for future recognition of marked individuals (Spellerberg, 1977). After processing, usually within one hour, each rattlesnake was released at its capture location.

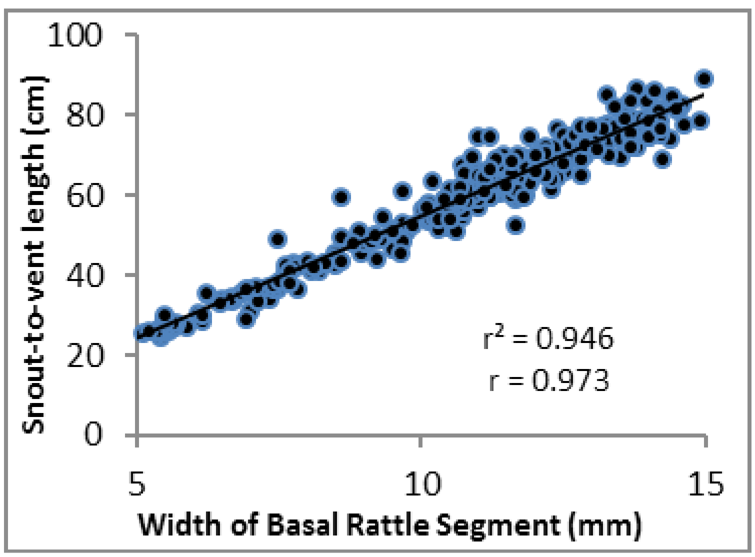

Figure 8: Studies have shown a strong correlation between the basal rattle segment width and body length, which holds true for all sizes of snakes (Beck et al., 2014).

Figure 9: Camera stand setup made to ensure all images were taken at the same distance and angle to allow for accurate measurements, using a black and white target for contrast to maintain the outline of the rattle.

Figure 10: The raw image of the rattle, used for analysis. The file can be kept as a permanent record.

Radio Telemetry

Radio telemetry was used to track 3 adult male snakes from June 20 to October 10. This time frame captured much of the animals' active season which is typically April to October for other *C. oreganus* (Wallace and Diller, 2001). Adult males were chosen since they have larger ranges and more consistent movement (Gomez et al., 2015). Two of the snakes were gathered from the Swauk den affected by the Taylor Bridge fire; the other was from Frenchman Coulee area affected by the George fire. The tracked snakes were surgically implanted with radio transmitters (Holohil Systems SI-2T 11g); one snake from each site also had an i-button datalogger implanted which recorded body temperature at hourly intervals following methodology of Taylor et al (2004). Animals were anesthetized with isoflurane and had radio transmitters surgically implanted following the techniques of Beck (1995, 1996) and our IACUC protocol no. A011405. Snakes were monitored for a 24 hour period after the surgery to ensure proper recovery, and then released at their collection sites. The animals were each tracked a dozen times and GPS location was recorded for each new observed location, and the presence or absence of burned vegetation was noted.

Data Processing and Analyses

The program ImageJ was used to extract rattle segment sizes from the digital photos, and measure the width of each rattle segment to the nearest pixel. The measured basal rattle segment of each snake, in mm, was divided by the pixel value from ImageJ to generate the scaling factor for that snake. This factor was then multiplied by the pixel size for each of the remaining rattle segments to convert them from pixels to millimeters. The process was repeated for every snake to determine the remaining unknown rattle segment widths. This technique also allowed for rattle segments that were slightly broken to be more accurately measured since they retain the outline in the photo; broken rattle segments tend to flex when being measured with calipers, thus giving unreliable estimates.

Since the growth of the snake was the measurement of interest, the rattle segments within each rattle were compared to adjacent segments to assess annual growth rate. This was done by finding the difference in size between each pair of consecutive segments to identify differences in growth (Figure 11). With this, if two rattle segments next to each other were the same size, then it would be assumed that the snake had experienced no growth. To go even further, if the segment that was formed the year before was larger than the current this would indicate that the snake had experienced a net loss of biomass during that year or negative growth. Positive yearly growth would be expected for an animal unless there is resource scarcity, resource allocation to reproduction, or other issues afflicting the individual. For rattlesnakes, this would appear as a rattle that tapers from the basal rattle towards the button (Figure 11, right). For this investigation, the difference between the basal rattle and adjacent rattle was used for indicating growth rates during 2015, which coincided with the year following the Carlton Complex wildfire. The growth rate in the year following the fire was compared between those snakes in dens that were burned during the fire and those that were spared from it.

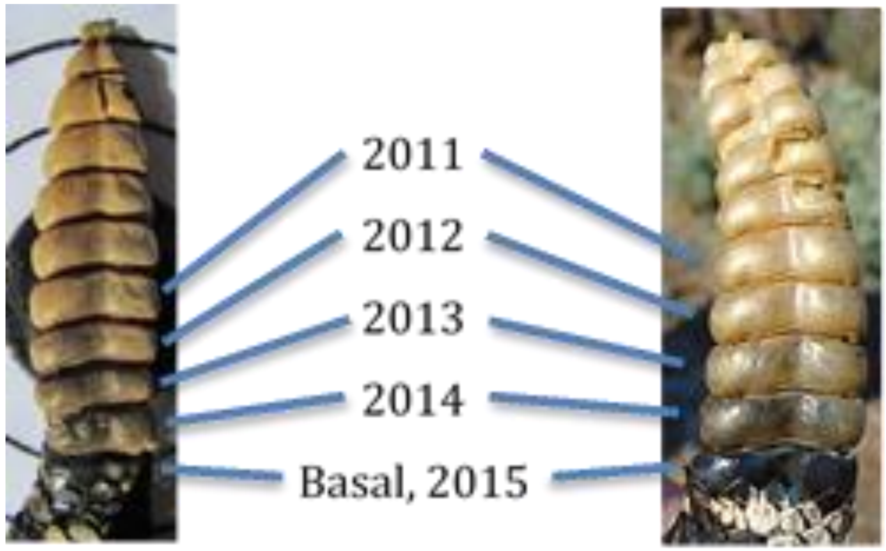

Figure 11: These two animals show different patterns in growth. The right shows a regular taper indicating consistent yearly growth. The image on the left, measured in April 2016, is from a snake located in the area of the Taylor Bridge Fire of 2012, and the rattle segments following those years were reduced in size and misshapen, a potential indicator that the wildfire negatively affected this animal. The difference in widths between two adjacent rattle segments represents the growth in the year leading to the rattle being formed, for this study the 2015 growth was the year of interest.

After all the rattle segment images were converted to mm, I used RStudio for the quantitative analyses conducted on snake size and growth data. To determine whether growth rates differed significantly between the years when rattlesnakes were subjected to wildfire in comparison with the years where wildfires did not occur, two tests were conducted. First a Generalized Linear Mixed Model (GLMM) was fitted to the data in order to test the strength of the association between growth and wildfire presence. The final model was Growth ~ SVL + Fire + Sex + (1|Den), with Growth being the 2015 growth, SVL to take into account the animals' size, Fire was the category of interest and was either fire affected or not, Sex was included to see if sexual dimorphism was a factor, and Den was used as a random effect. Second, an Analysis of Covariance (ANCOVA) was conducted to see if there was a significant relationship between wildfire and growth, with body size as a covariate. The final models were Growth ~ SVL + Fire and Growth ~ SVL * Fire, which looked at the interaction between the snake's size and fire presence on the growth of the individuals.

To explore potential differences in the size structure (SVL) of groups of snakes inhabiting dens affected by wildfire as opposed to those that were not affected, a Kolmogorov-Smirnov "goodness of fit" test was used (Massey, 1951). Lastly, growth trajectories of all snakes that had complete rattles (i.e. having an intact terminal button), were plotted to explore any differences in growth curves over several years between fire (n=24) and non-fire (n=15) snakes. Potential differences were tested with a repeated measures Analysis of Variance (ANOVA).

RESULTS

Growth

A total of 85 adult snakes were measured in the Methow area from 20 April to 21 July 2016, 58 (68%) of which were male and 27 (32%) female. 37 snakes (44% of the sample) came from within the wildfire area and 48 snakes (56%) came from outside the habitat affected by fire. Growth after the wildfire appeared to be similar between fire and non-fire sites for the snakes in each group, confirmed using an ANCOVA (F=0.58, P=0.45; Figure 12). A generalized linear mixed model also demonstrated that there was no significant association between growth and wildfire presence (t=-1.70, critical=12.7).

Growth curves of snakes in wildfire areas did not differ significantly from those inhabiting dens unaffected by the fire (Figure 13). However, the size structures between the groups were different, with a mean SVL of 56 cm for fire-affected snakes and 66 cm for non-fire snakes (Figure 14). This difference was significant following the Kolmogorov-Smirnov test (D= 0.355, P=0.01).

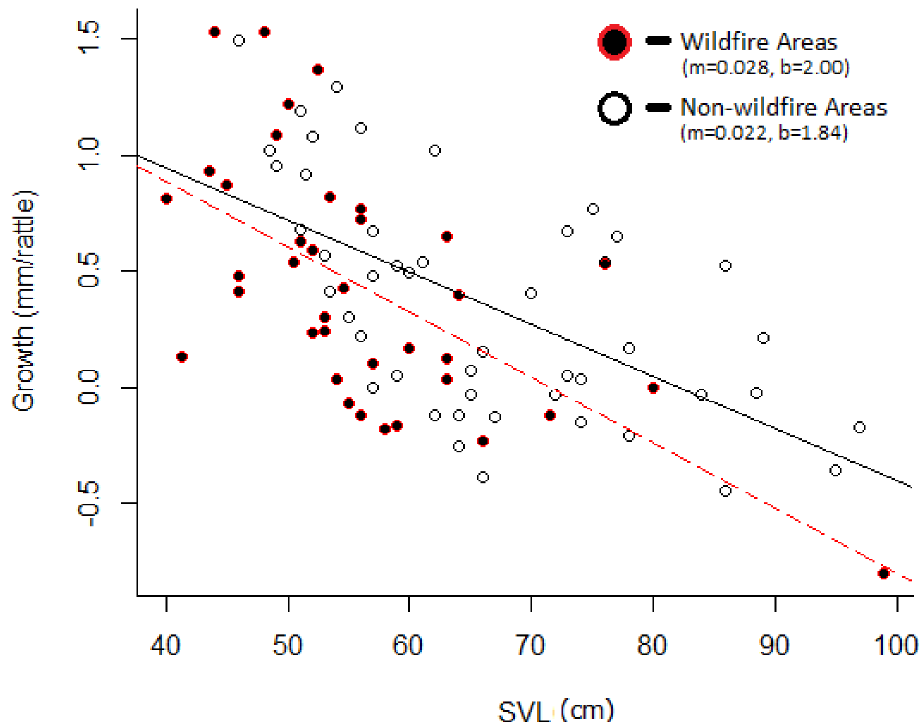

Figure 12: Comparison of rattlesnake growth in fire vs non fire areas.

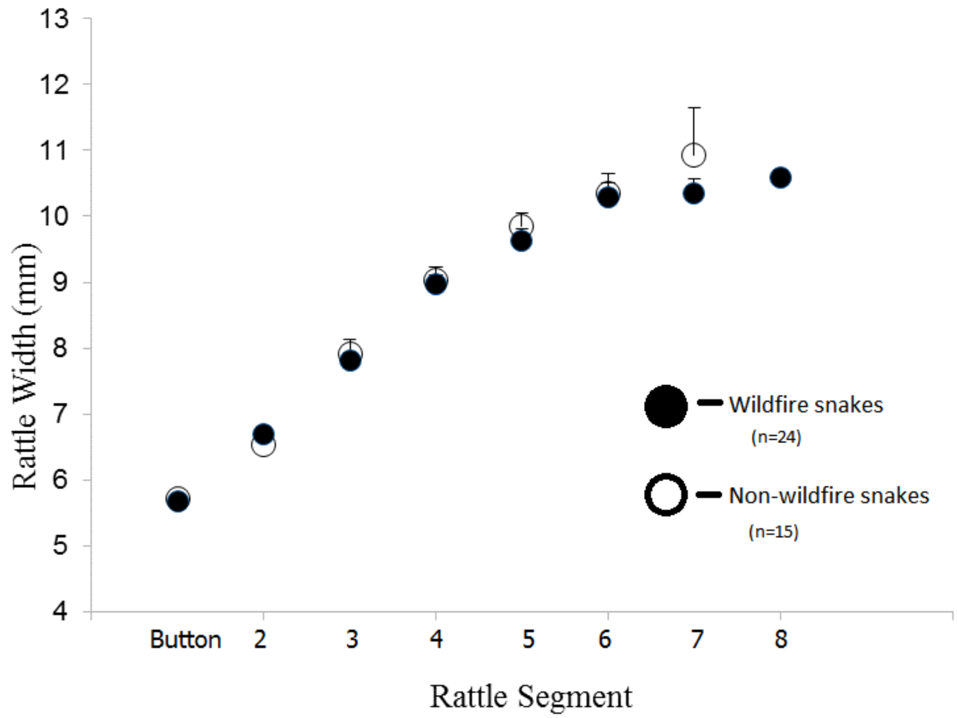

Figure 13: Lifetime growth curves from snakes collected during the 2016 field season with an intact button. Dots are means; horizontal lines depict standard error.

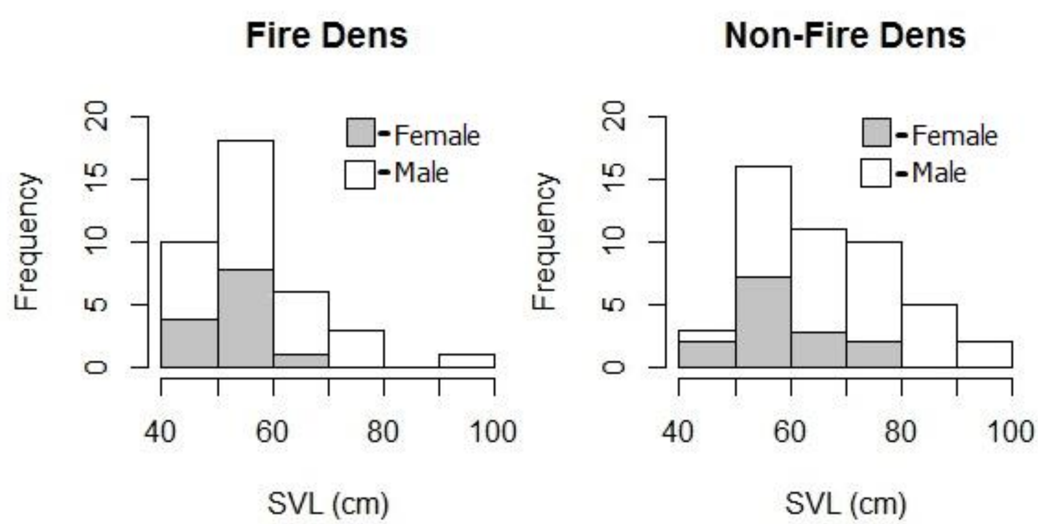

Figure 14: Size distribution of snakes in areas affected by fire and those in areas not affected by fire. The mean SVL for snakes from fire dens was 56 cm and for non-fire dens was 66 cm.

Movement

Three adult male snakes were radio-tracked to observe summer habitat selection in the Frenchman Coulee and Swauk burned areas. Snakes were released for tracking at their original capture sites on June 20 and made their way back to the dens in early October where they remained until spring.

The one snake tracked at Frenchman's Coulee was found under burned vegetation during 8 of 10 relocations and did not demonstrate any avoidance of burned habitat. The snake was found away from wetlands during the summer, in brush covered areas that had strong scorch patterns and trees, predominantly Russian olive (*Elaeagnus angustifolia*), that were likely killed from the fire. Total seasonal movement was 1877m, with ingress back to the den occurring from September 9 to October 10, adding approximately 1240m to the snake's movement with only 2 additional relocations (Figure 15). Total movement during the foraging period (June 20 to September 9) was 638m after 8 relocations, encompassing an area of approximately 200m by 80m, 1.6 ha, (Figure 16).

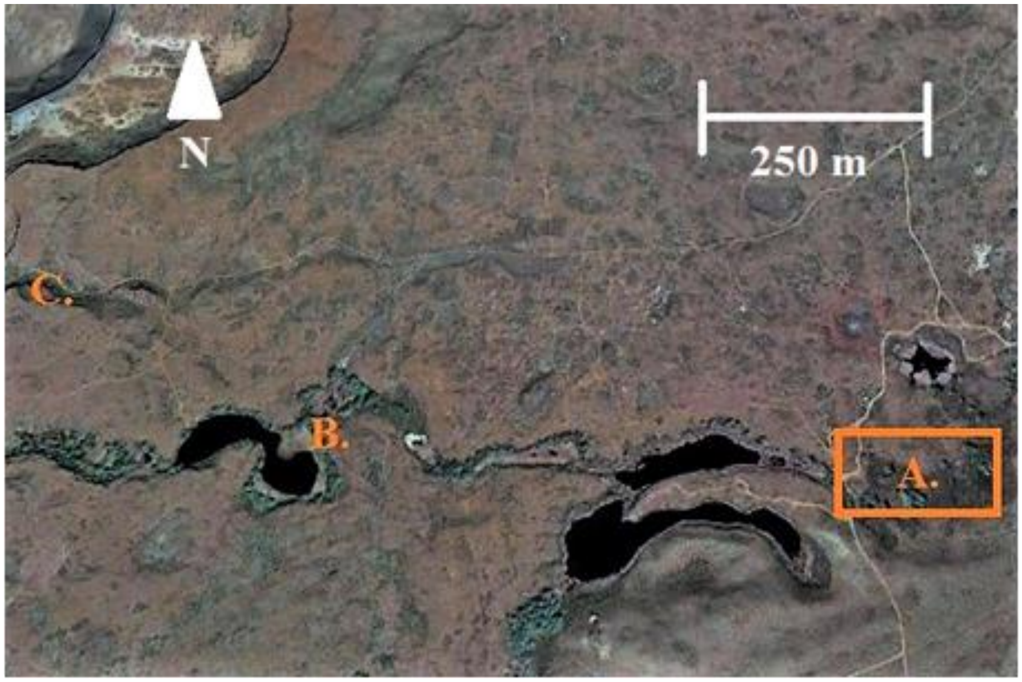

Figure 15: Seasonal movement of the Frenchman Coulee snake from June 20 to October 3, 2016 (Google Maps, 2017). Location A is the foraging area (Figure 16), B is the first movement toward the den on September 24, and C is the den location, which was reached by October 3.

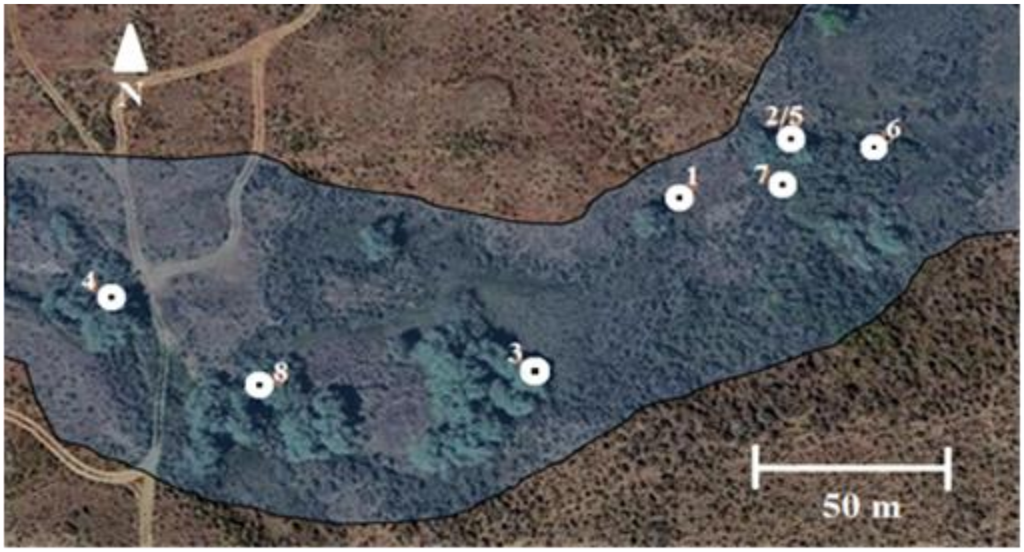

Figure 16: Foraging area of the Frenchman Coulee snake from June 20 to September 9, 2016 (Google Maps, 2017). White dots indicate location and number is the progression of movement, snake was found at 1 and moved towards the den after 8. The blue shaded region is the area that has fire damage still present on the woody vegetation and encompassed the entire foraging area.

Tracking at the Swauk area (n=2) during the active season only resulted in one of the snakes moving (approximately 20m) then returning to the original location, the other snake seemingly remained in the same spot until they made the ingress into the den. Throughout the summer the animals were tracked a dozen times and the habitat that they remained on was the same talus rocky slope in which they were originally captured. The slope had little remaining evidence of the fire that had come through the area, although the nearby riparian vegetation (cottonwood, *Populus spp.*) had been burned by the fire. By the end of the active season both snakes had returned to the hibernacula on the slope. However, neither had left the talus slope area that surrounded the den site in the dozen times that the animals were checked throughout the summer and fall (Figure 17).

Figure 17: Swauk snake summer activity area. White circles represent where snakes were found. The red circle indicates the main talus slope of the den area, the white dot within the den is the initial capture site for both snakes tracked.

DISCUSSION

Since there was no significant difference in growth rates of the rattlesnakes, it may be that these ectothermic animals are able to persist in the post-wildfire landscape with minimal harm. This could mean that even if the prey population had been reduced as result of fire, the rattlesnake population was able to survive long enough for the prey population to rebound to pre-fire levels. In contrast to the rodent prey, the metabolic rate of a snake is much lower, and, as such, could withstand a longer period of unfavorable conditions. Another possibility is that the prey populations may not have been affected by the fire, as other studies have found (Monroe and Converse, 2006; Amacher et al., 2007; Kalies et al., 2010). If there was minimal direct mortality on rodent populations, it would be expected that the snakes that survived would be able to rebound quickly as well.

Alternatively, if these snakes are affected by the wildfire, it may not be evident immediately and there could be a lagging effect. The foraging habits of this population were not directly known, so individuals may have been able to successfully find food before the fire had started on July 14, 2014. With successful foraging they could survive for a longer period without experiencing loss of biomass. However, after the fire, the prey base might have changed, as was discovered in the wildfire study in Victoria, Australia, where two small mammal species within the fire were one-third the density compared to nearby unburned sites a year after the fire (Banks et al., 2011). Since mammals can account for 90% of the snakes' diet (Macartney, 1989), a severe change in prey availability could negatively affect foraging success for the snakes after the fire. These factors would indicate that if more time was given we might see effects of fire on growth. In order to support or refute these conclusions more data gathered from snakes affected by the Taylor Bridge fire of 2012 or future sampling of the Carlton complex snakes could identify if there is a potential delay in wildfire effect on growth.

From the analysis, snakes using dens in wildfires had a different population size structure than the adjacent snake populations in non-fire affected dens. This presents the possibility of a potential direct effect of the wildfire, in that larger individuals are selected out of the population. Since rattlesnakes have limited movement, outrunning the fire would not be plausible, instead finding a refugium would be necessary. Larger snakes may have more difficulty finding a suitable place to safely escape the fire. During the time of the fire, mid-July, these snakes would have been dispersed from the dens and foraging in the adjacent land. In the foraging area some boulder outcrops were available to the snakes for refugia; however the dominant feature was shrub-steppe habitat, low-lying shrubs and grasses. The fire had fully burned the shrubs and grasses, leaving the landscape charred and barren (Figure 4). Further evidence from burn scars found on snakes from within the wildfire area indicates that some individuals were exposed to the flames (Figure 3). These limited places for safety could leave the larger animals more directly exposed to the fire in this region.

Tracking the snakes' use of habitat revealed unexpected results in regard to behavior and possible interactions with the post-wildfire landscape. The Frenchman Coulee tracked individual exhibited relatively normal adult male movement from the den, traveling approximately a kilometer from the den site. The snake was also choosing shelter and hunting locations directly beneath burned vegetation, including during the time of year when wildfires tend to be most prevalent. Similar to other snake studies, this would suggest that there is little avoidance of burned habitat (Cross et al., 2015). Although other studies have found contrasting results (McDonald et al., 2012), this indicates that more research needs to be conducted on the movement behavior of these animals as they interact with these altered landscapes. A larger sample size of radio-tracked individuals from a single den would give more specific insight into how individuals in a similar setting react.

The more unexpected result was in the Swauk snakes which did not leave their den area for the foraging season. In other dens, and from other studies (Wallace and Diller, 2001; Gomez, 2007; Hobbs, 2007; Putman et al., 2014), the normal behavior for full grown adult males, like the animals tracked, is to remain at the den after emerging for about a month and then disperse to their foraging habitat (Fitch, 1949; Klauber, 1972; Macartney, 1989). While the direct reason behind the snakes remaining at the den is unknown, the surrounding habitat gave possible explanations, as populations in British Columbia had similar movement, remaining closer to dens in grassland habitat (Gomez et al., 2015). The most obvious landscape feature in this study site was Swauk Creek, less than 50m from the den, which flows year round. This constant flow is an ideal water resource for the surrounding flora and fauna, including the snakes, and could potentially increase the prey population available to the snakes through the increased primary productivity. With higher food abundance there would be less pressure to travel away from the den and risk unnecessary exposure to predation. This may be plausible as there were several other snakes, up to 20 individuals, also found at the den throughout the season. This would also mean that snakes may have been at the den at the time of the fire, and the rocky slopes would act as good refugia from the fire and a possible way to avoid the surrounding burned habitat.

This study was the first of its kind to look at possible wildfire effects on local predators in the Pacific Northwest. Although there was no indication of reduced growth rates immediately after the fire, more time may be needed to see these effects. However, the use of digital photography, as was performed here, can act as a new tool for future ecological studies using the rattle to explore historic patterns of growth in populations with more precision and efficiency.

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