Elk Hoof Disease in Southwest Washington

WDFW Hoof Disease Public Working Group Meeting 02 December 2015

A.13 650

Agenda

Welcome

- TAHD Diagnostics
- Hoof Disease Euthanasia Protocol
 - **Hoof Disease Prevalence/Distribution**
 - **Hoof Disease Survival Study**
 - Next steps
- Public Testimony

Public Testimony

- Members of the public are requested to fill out a Public Testimony Form
- Members of the public will be requested to provide their public testimony to the HDPWG in the order the Public Testimony Forms were received
- Each member of the public wishing to relay their comments will have 3 minutes each to do so
 - This time frame is provided to allow the opportunity for all members of the public to provide their testimony to the HDPWG

Hoof Disease Public Working Group

- Understanding hoof disease in elk is a priority and WDFW is committed to the sound management of these important resources
- WDFW established the Public Working Group as we believe it is important to work together as we try to better understand and address this issue
- The purpose of this Working Group is to provide the opportunity to:
 - share information about the hoof disease phenomenon and WDFW activities,
 - discuss research and management questions with regard to hoof disease and solicit feedback, and
 - public outreach

Willapa Hills and MSH Elk Herds



Collections

Four collections from affected and unaffected areas:

Adult elk

- March 2009 :
- March 2013:
- August 2013:
- January 2014:

- 9-10 month elk
- 3-4 month calf elk
- 7-8 month calf elk
- Summary: 43 elk examined from March 2009 -Jan 2014
- Extensive analyses with multiple national and international partners including 5 independent labs
- Detected Treponema sp. bacteria in diseased samples from the multiple collections

Findings

 Treponema species are known to be highly associated with serious and widespread hoof disease in domestic livestock throughout the world,

 and more recently have caused very severe disease in sheep and goats, particularly in the UK

PF3

PF1&2

OM

PF4

Diagnosis of TAHD

 Diagnosis of Treponeme Associated Hoof Disease (TAHD) (June 2014)

- Available evidence is most consistent with an infectious bacterial hoof disease
- The disease shares many features and most resembles treponeme-associated contagious ovine digital dermatitis (CODD)
- Environmental factors, including wet conditions, are likely important in disease initiation and propagation

Prioritized Efforts

- Prioritized 4 immediate questions/efforts to address and inform management:
 - Better understand prevalence of hoof disease in elk herds in Southwest Washington,
 - Better understand the distribution of hoof disease in elk herds in Southwest Washington,
 - Understand the impacts of hoof disease on elk survival and productivity, and
 - Remove elk severely affected with hoof disease
 - Euthanasia Protocol

TAHD

 Continue to work with collaborators on additional TAHD-related information needs identified from HDTAG and HDPWG

Identified Information Needs

- Being maintained in elk population?
- Elk movements/habitat use?
- Do elk develop immunity?
- Effects on survival & reproduction?
- Progression of disease over time (individual & herd)?
- How transmitted?
- Presence in environment (fecal & soil sampling)?

Identified Information Needs

- How TAHD affects body condition and other health parameters
- How elk immune system responds to TAHD
- What other bacteria are involved in the development of TAHD
- Are treponemes shed in the feces (we know this is the case in cattle)
- Whether some elk are genetically resistant (or susceptible) to developing TAHD

TAHD Diagnostics

Research Occurring at USDA

- Identifying other bacteria besides treponemes that might be involved in the development of TAHD
 - Evaluating the immune response of elk to these TAHD bacteria
 - inform our knowledge of whether the elk eventually become immune to the disease

Research Occurring at USDA

 Evaluating whether blood can be tested for evidence of previous exposure to the bacteria that cause TAHD

- Provided USDA with archived serum:
 - 2005 Nooksack relocations (before TAHD recognized in area elk),
 - 2010 during WDFW population study (when sporadic cases were being detected),
 - Feb 2015 captures (when apparent prevalence of TAHD had increased)

Prep for Additional Research

 Collecting oral and rectal swabs, as well as feces, for future testing to see if the bacteria are spread via a "fecaloral" route, as is suspected may occur in cattle

 Collecting samples for WDFW genetics lab to see if we can detect evidence of genetic susceptibility/resistance to the disease

Prep for Additional Research

- Recapture animals to:
 - ensure that the negative controls remain truly negative throughout the duration of the study,
 - to follow individual animals over time to better understand the progression of the disease, including whether they recover

Hoof Disease Euthanasia Protocol

SOP

- HDTAG and HDPWG agreed to removal for humane reasons
- Finalized the Standard Operating Procedure for Lethally Removing Elk Severely Affected by Hoof Disease
 - Guideline for euthanasia procedures





WILDCOM: 360-902-2936

SOP

- DNS finding from SEPA review
- WDFW staff implementing
 - Future assistance from Master Hunters trained in criteria



Understanding Hoof Disease Prevalence/Distribution

Distribution Effort



Distribution

Reports of Observations of Limping Elk to Date from Website Reporting Tool:

Report Type	Total # of Reports Since 2012	Total # of Reports After 09/15/2014	
Limping	639	207	
Dead	182	68	66/55 reported as harvested
Total	821	275	

Prevalence

Goal:

- Understand the prevalence of hoof disease and monitor changes throughout a defined area within the range of the Mount St. Helens and Willapa elk herds
- Determine the ratio of limping elk to the total number of elk observed within accessible areas in southwest Washington
- Develop a simplified survey strategy using a consistent, observable indicator of TAHD
- Develop a simplistic sampling strategy at landscape scale
- Implement citizen science effort to accomplish study goals

Survey Strategy

 Boundaries of the study area
 GMUs chosen based on distribution of incidental observations of limping elk reported by the public

> From online reporting website since 2012

Survey Strategy



Survey Strategy

- Boundaries of the study area
 - GMUs chosen based on distribution of incidental observations of limping elk reported by the public (online reporting website since 2012)
 - Acceptable visual indication of TAHD for this study
 - Having confirmed TAHD in the area through diagnostic testing on harvested elk
 - In the absence of a non-intrusive and immediate test for TAHD in live elk that does not require capture
 - In this study:
 - Used limping elk as a surrogate for TAHD infection, reasoning a limping gait as an acceptable visual indication of TAHD

Prevalence

- Pilot Study August 2014
- Citizen Science Project: March May 2015
 - Five training sessions 1st week in March
 - 223 volunteers participated; 218 signed up for a survey
 - Participation from USFS, WADNR, Weyerhaeuser, Sierra Pacific, Green Diamond, Pope
 - 148 survey waypoints across 10 counties reasonably accessible by vehicle
 - Another 20-40 surveys were identified for WDFW staff to fill in coverage gaps over areas that determined inaccessible to volunteers

Prevalence

Survey Start Point Locations



Prevalence Survey Grid



Prevalence Survey

- At survey start point:
 - Volunteer and staff teams drove 50 miles or for 4 hours
- Searching for elk:
 - Focused on agricultural fields, forest openings, and clear cuts (but not limited to these areas)
- Surveys took place in the mornings or evenings when elk are most active
- Collected data (e.g., distance, number of elk/limping, wind, elevation, etc.)

Prevalence Results

- 142 surveys successfully completed by volunteers
 - 33 surveys were completed by WDFW staff
- Over 7,300 miles driven by volunteers
- 96% return on survey data from volunteer teams
- Surveys at mean elevation of 1,127ft.

Prevalence Results

On average teams spent:

- 48% of time in forests; 29% in open habitat;
 16% in shrub, and 7% in other habitat types
- Teams surveyed on public lands 19% of the time
- 283 elk group encounters (~ 2,600 elk observed)
 - 83 groups had one or more limping elk (29%)
 - Elk group sizes were mostly <4 or >11
 - The ratio of limping elk to total elk always declined as group size increased

Prevalence Completed Surveys



Prevalence of Limping Elk






OR

- Rate of detection is not constant across types of encounters with groups (different levels of detection)
- Have to assume that we are not detecting early levels of the disease when using limping as our indicator of a TAHD infection
 - Likely to miss individual limping elk when examining groups under certain situations
- Therefore:
 - Cannot determine the ratio of limping elk to total elk at the individual level

- TAHD is an infectious disease
 - Believed to be contagious among elk
- Assume that if a single elk is detected limping within a group
 - Potential for other elk within group to be susceptible to disease and be infected as well
- At the group level can apply correction factors to our observation methods (visual, scope, distance, etc.)

Therefore:

 Use limping as a group level indicator of TAHD

Thereby, determine the proportion of groups affected by TAHD across the survey area

Prevalence of Groups

• 48% of observed groups contain at least one limping elk across the study area

Range from 40% to 57%

Region 5:

 51% of observed groups contain at least one limping elk (Range 41%-61%)

Region 6:

- 42% of observed groups contain at least one limping elk (Range 28%-59%)
 - Fewer observations in region 6 resulted in a greater range





Prevalence – What We Learned

- Individual Level
 - Current sampling design:
 - We are unable to detect the early stages of the disease
 - We are unable to estimate prevalence at the individual elk level
- Group Level
 - Determine the proportion of groups affected to total number of groups observed
 - More thoroughly define the perimeter of TAHD

Prevalence – What We Learned Group Level (cont.)

- Determine the distribution at finer scales, such as GMU or County, within survey area
- Group level may be a more feasible measure for monitoring changes in TAHD distribution over time
- Achieve with a smaller, more focused volunteer effort
- More is known about the disease:
 - Have more options available for determining prevalence at the individual elk level

Prevalence

- **2015 Hunter Harvested Hoof Collection Effort**
- Pilot Effort:
 - Contacted 483 modern firearm special elk permit holders in Game Management Units 520 (Winston), 522 (Loo-wit), 524 (Margaret), 550 (Coweeman), 556 (Toutle), and 673 (Williams Creek) to assist
- Created a simple scoring system for hunters to determine the grade of hoof disease in harvested elk

2015 Hunter Harvested Hoof Collection Effort



Prevalence

2015 Hunter Harvested Hoof Collection Effort

- Contacted 483 modern firearm special elk permit holders in Game Management Units 520, 522,524, 556, 550, and 673 to assist in the pilot effort
- Created a simple scoring system for hunters to determine the grade of hoof disease in harvested elk
- Hooves are being collected and scored again by WDFW staff to test for consistencies among hunters
- If successful the effort will be expanded to include a larger study area

Prevalence 2015 Hunter Harvested Hoof Collection Effort

- To date collected:
 - Region 5:56 samples
 - Region 6:22 samples
- Still collecting samples in both Regions

Prevalence

- To inform our understanding of prevalence and distribution of TAHD and guide our management direction:
 - Continue to collect data from both the citizen science survey and hunter assessment
 - Adapt and refine these or any additional methodologies as needed
 - Especially as we learn more from the continued diagnostic work and the ongoing study looking at the effects of TAHD on elk population dynamics
- Thank all of the volunteers, volunteer leads, landowners, WDFW staff

Hoof Disease Survival Study

Hoof Disease Study

- Potential Effects of TAHD
- May reduce survival of affected elk
- Secondary effect on nutritional condition
 - Reduced probability of conception
 - Limit the ability of a cow to support a calf
- Alter the way affected elk use the landscape





Study Objectives

Objective 1: Estimate the effects of TAHD on survival of adult (>2 years old) female elk

- Objective 2: Determine cause-specific mortality rates for adult female elk that have TAHD
- Objective 3: Estimate the effects of TAHD on the pregnancy rates of adult female elk
- Objective 4: Estimate the effects of TAHD on elk productivity (i.e., survivorship of calves)
- Objective 5: Estimate the effects of TAHD on the level of condition that hunter-harvested female elk are able to achieve in autumn

Study Area



Capture and Marking Conducted captures in February 2015 81 elk captured within study area GMUs 520 (Winston), 522 (Loo-wit), 524 (Margaret), 550 (Coweeman), and 556 (Toutle) 78 elk marked and fitted with GPS-equipped 524 Margaret radio-collars 58 had TAHD 550 Coweeman 556 Toutle 20 had no visible signs of being affected 2015 Capture Locations Capture Mortality FISH and

TLDLIFI

Capture and Marking

- Estimated age at time of capture via tooth wear and replacement Removed upper canine to estimate age via cementum annuli
- **Assessed body condition**
 - Measured rump fat thickness
 - Determined a rump body condition score (rBCS)
- Measured chest girth to estimate body mass



TAHD Observations

Observed wide variation in the condition of hooves of elk affected by TAHD
Involved rear hooves = 98%
Involved front hooves = 10%
Involved >1 hoof = 27%
Involved just 1 rear hoof = 72%



Disease Severity

0





Control Grade 1 Grade 2 Grade 3 Grade 4 Grade 5 Multiple TAHD Severity

Body Condition and Age

		Age	<u>IFBF</u>		Body Mass	
Score	n =	\overline{x}	\overline{x}	95% CI	\overline{x}	95% CI
Control	19	7.6	5.13	3.95-6.31	205.2	197.0-213.3
Early	7	7.3	5.19	3.42-6.95	208.5	194.9-222.2
Late	28	6.5	4.26	3.45-5.08	194.4	188.3-200.4
Multiple	21	5.6	3.86	2.80-4.91	188.3	178.7–198.0
Early Late Multiple	7 28 21	7.3 6.5 5.6	5.19 4.26 3.86	3.42–6.95 3.45–5.08 2.80–4.91	208.5 194.4 188.3	194.9–222.2 188.3–200.4 178.7–198.0





BM x Severity



Control 6 4 3 2 1 0 1 2 3 4 5 6 7 8 9 10111213141516 Age





TAHD Severity



Survival and Cause of Mortality

Survival March 1-December 1



Survival May 1-December 1







Pregnancy

- Assessed pregnancy for 80 elk
- 76 Adults

- 4 Yearlings
- Overall pregnancy rate for adults was 0.64
 - 0.84 for controls
 - 0.58 for TAHD
 - Pregnancy rate varied with disease severity
 0.67 for McCorquodale et al. (2014)

Score	n	Number Pregnant	Proportion Pregnant
Control	19	16	0.84
Early	7	7	1.00
Late	28	18	0.64
Multiple	21	7	0.33

Productivity

Defined as early survivorship of calves Assess via calf-at-heel and/or lactation rates of hunter harvested elk Calf-at-heel not effective strategy Will use lactation rates of captured elk moving forward 4 of 6 cows with **TAHD** have been lactating at time of death

Changes Moving Forward

- Will conduct captures in December
 - Allows assessment of lactation status and autumn body condition
 - Will not collect samples from hunterharvested elk
- Will capture elk to replace study animals that have died,
- but will also attempt to recapture all control elk and subset of affected elk
 Include new research objective related to increasing our understanding of disease progression

Next Steps

Next Steps

- Alternative design on prevalence survey
 - Evaluate hoof assessment methodology
- Second year of survival study
- Continue diagnostic research to address identified management and research questions
- Adaptive as we learn from these efforts with respect to management and research
- Assess/prioritize/address remaining information needs
- Continue working with HDPWG and HDTAG as moving forward

Thank youany questions....

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Public Testimony

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Using Citizen Scientists to Aid in Estimating the Prevalence and Distribution of Treponeme-associated Hoof Disease in Southwest Washington



December 1, 2015

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Background

Southwest Washington is generally defined by the Cascade Mountains to the east, the Pacific coast to the west, the southern end of Puget Sound to the north and the Columbia River to the south. Two elk herds inhabit the area: the Mount St. Helens and the Willapa Hills elk herds. The Mount St. Helens herd is recognized as one of the largest herds in the state and predominately occupies the mountainous terrain of the western Cascades but extends west beyond the Cascades to the Interstate Highway 5 corridor through mid to low level forests, floodplains and valley bottoms (McCorquodale et al. 2014). The Willapa Hills elk herd area is located in the southwest corner of Washington with boundaries roughly defined by State Highways 8 and 12 to the north, the Interstate Highway 5 corridor to the east, the Columbia River to the south, and the Pacific Ocean to the west (Washington Department of Fish and Wildlife, WDFW 2014). Topography of the Willapa Hills herd area consists of tidal flats along the coastal Long Beach Peninsula, level to rolling terrain to the North, and mountainous terrain in the core of the Willapa Hills (WDFW 2014). Both herds are considered an important resource to the state of Washington, providing ecological, recreational, aesthetic, cultural, and economic benefits (WDFW 2014).

WDFW first received sporadic reports of lame elk or elk with deformed hooves during the 1990s in areas associated with the Cowlitz River Basin in Cowlitz and Lewis counties. In 2008, the number and geographic area of these reports increased significantly, and reports have continued to increase since that time (Mansfield et al. 2011, WDFW unpublished data). Reports are still concentrated in the Cowlitz River Basin; however, suspected incidences of hoof disease occur in 10 southwest Washington counties and the disease is known to affect both the Mount St. Helens and Willapa Hills elk herds (Han and Mansfield 2014).

In 2009, WDFW initiated diagnostic testing of elk in southwest Washington, and of control elk believed to be in non-affected areas immediately east of affected areas (Han and Mansfield 2014). Results from this initial study revealed that the hoof disease in elk was chronic and limited to the hooves; however, due to the advanced stages of the disease in collected samples, a causal factor was not determined (Han and Mansfield 2014). Further testing on elk hooves in the Mount St. Helens and Willapa Hills herds in 2013, 2014, and 2015 resulted in the detection of bacteria in the genus Treponema associated with hoof lesions (Clegg et al. 2015; Wilson-Welder et al. 2015; WDFW unpublished data). Treponemes are described as causing severe foot diseases known as bovine digital dermatitis (BDD) in cattle and contagious ovine digital dermatitis (CODD) in sheep (Clegg et al. 2015; Wilson-Welder et al. 2015). The hoof disease in elk in Southwest Washington, referred to as treponeme-associated hoof disease (TAHD), most closely resembles CODD in sheep and typically results in abnormal hoof growth, cavitating sole ulcers, chronic laminitis and, in severe cases, eventual sloughing of the hoof capsule. It is likely that environmental factors are important in disease initiation and propagation as the bacteria are suspected to persist in wet soil conditions and are spread to new areas by elk with infected hooves.

TAHD is currently thought to occur among elk across 5,343 mi² (3.4 million acres) in southwest Washington (Mansfield et al. 2011). Determining a reliable estimate of the prevalence and geographic distribution of TAHD at this scale presents many challenges. The leading limitations are the absence of complete elk density estimates for the geographic region in question and an inability to adequately sample the area where elk occur due to factors such as land accessibility and staffing constraints Anecdotal evidence from surveys of elk groups and interviews with landowners conducted in 2008-2009 suggest that up to 80% of elk groups in the affected geographical area contain lame elk and that 30-90% of individuals in an affected group are lame (Han and Mansfield 2011). These original estimates do not occur systematically across the affected area and range widely among the individual elk affected within groups.

WDFW initiated two studies in 2015 to learn more about TAHD in southwest Washington. The first is designed to assess the potential effects of TAHD on elk population dynamics in southwest Washington with a focus on cow elk survival, reproduction, and body condition (Hoenes et al. 2014). The second is to understand more about the distribution and prevalence of TAHD using citizen science to systematically survey for elk within accessible landscapes in Southwest Washington. The two studies together will provide critical information regarding elk herds exposed to TAHD, and therefore inform management decisions within the affected geographic region (Hoenes et al. 2014). This report summarizes the distribution and prevalence survey and analyses.

Study Goal and objectives

To increase our understanding of TAHD distribution across the southwest Washington landscape and to reasonably determine the prevalence of the disease in elk groups, we developed the following objectives:

- 1. To develop a simplified survey strategy using a consistent, observable indicator of TAHD across the apparent hoof disease area
- 2. Determine the apparent prevalence (ratio of affected elk to total number of elk observed) of *TAHD* among elk within accessible areas in southwest Washington.
- 3. To utilize and assess the effectiveness of citizen science to accomplish Objectives 1 and 2

Study Area

A pilot study, conducted in August 2014 tested different methodologies to inform the early development of sampling strategies for the current survey effort. In this effort, twenty-nine Game Management Units (GMUs) were used across 10 counties, including both the Mount Saint Helens and the Willapa Hills elk herd areas. The area is a mix of mountainous, rolling, and level terrain interspersed with naturally occurring mid to low elevation conifer and deciduous forests, industrial timber plantations with a mosaic of clear cuts in various successional stages, agricultural farm lands, natural meadows, and coastal tidal flats.

GMUs used to define the boundaries of the study area were chosen based on the distribution of incidental observations of limping elk reported by the public through an online reporting website in place since 2012 (Fig. 1). To systematically distribute surveys across the study area we used watershed features defined by the U.S. Geological Survey hydrologic unit code (HUC12). In ArcGIS 10.2, we established a GPS waypoint in the center of each HUC12 and then moved it to the nearest accessible road, and in many cases, onto public land. GPS coordinates originally consisted of 320 centroid points. We prioritized these points based on road accessibility and private timber ownership within the survey area. Prioritizing the points resulted in 148 survey starting waypoints that we believed were suitable for a volunteer survey designation. We also identified approximately 84 additional, less accessible, waypoints that were suitable to be completed by staff. Based on the distribution of volunteer starting waypoints, we determined that 20-40 designated staff starting waypoints could be conducted in certain locations to fill in coverage gaps across the survey area (Fig. 2).



Figure 1. Map depicts incidental observations reported on the WDFW hoof disease website since 2012. Grey triangles indicate observations of limping elk, and black circles represent reports of dead or harvested elk. The hash-lined polygon represents GMUs chosen as the area for this study.


Figure 2. Map depicts the 148 volunteer and 46 potential staff starting locations. Blue circles represent volunteer designated surveys and black squares represent surveys designated as staff surveys.

Survey Procedures

Over 300 individuals, generally from western Washington and northwest Oregon, voluntarily registered with WDFW to participate in the survey effort. Each volunteer was required to attend a training session covering requirements for participation, the characteristics of TAHD, survey procedures, and GPS navigation. Two hundred and twenty-three volunteers participated in 5 trainings held across Southwest Washington and 218 actually signed up for a survey point.

Ten WDFW staff members and 112 volunteer survey teams, consisting of two observers, conducted vehicle based surveys from March 6–May 1, 2015. Each team made one attempt to reach each designated survey waypoint on one occasion. In some cases, teams were unable to reach waypoints due to locked gates or decommissioned roads. Surveys were conducted when teams were able to get within 5 miles of a starting waypoint, when visibility was high, and when winds were below 40 miles an hour. The time and duration of a survey were not restricted,

however; observers attempted to survey for at least 50 miles or four hours in the early mornings or evenings when elk were most likely to be active. To ensure adequate visibility for examining elk, observers began morning surveys no earlier than 20 minutes after sunrise and ended evening surveys one half hour before sunset.

Once a starting waypoint was reached, observers utilized aerial maps and GPS units to drive 50 miles around the survey start point, whenever possible. An emphasis was placed on utilizing secondary roads while attempting to locate forest clearings, clear cuts, and habitat where elk were more likely to be observed. Observers maintained speeds of 25-35 mph on improved, paved or gravel roads, 10-20 mph on unimproved or dirt roads, and 5 miles below the speed limit if greater than 35 mph. Track logs were recorded during 30 surveys to obtain trends in spatial information such as elevation and survey habitat types.

When elk were encountered we made every attempt to minimize disturbance that would alter their behavior. We classified observer distance to a group of elk as being < 50 yards or being > 50 yards. Observers used binoculars and/or spotting scopes to examine elk when groups were estimated to be >50 yards away. Observers counted the total number of visible elk in a group and counted the total number of elk observed limping (Appendix A1).

In the absence of a non-intrusive and immediate test for TAHD in live elk that does not require capture; for this study limping elk was used as a surrogate for a TAHD infection, reasoning that a limping gait is an acceptable visual indication of TAHD within the defined study area where TAHD has previously been confirmed diagnostically.

Observers attempted to identify subtle limping behaviors by looking for characteristics such as a shortened gait and uneven weight bearing on limbs. The counts of limping elk were considered approximate or exact depending on whether a group could be examined adequately and/or in its entirety. The length of time spent observing elk varied, but were classified as fleeting (<1 minute), 1-10 minutes, or >10 minutes. We did not observe any groups of elk for longer than 15 minutes. An individual elk was considered a group and groups larger than 10 elk were placed into categories of 11-20, 21-40, and >40.

Analysis

Observations at the group level were treated as a logistic (0 or 1) response, where all members of groups with a limping elk are potentially diseased. While in reality some members of infected groups may not be diseased at the time of observation, we can assume they will contract the disease at a higher probability than members of a group with no limping elk. We chose this approach because it fit naturally with our solution to the problem of incomplete detection of limping elk, which preliminary analysis indicated was a significant source of variance in the data. To account for this uncertainty in classification of individual elk as limping, we assumed every individual limps to a degree (including zero). There exists some uncertainty when

classifying elk this way based on factors such as observation methods, time of observation, etc.; however, the uncertainty in classification is reduced at the group scale as more individuals in the group are scored.

We then applied a correction factor based on observation attributes such as length of time, distance to elk, and use of visual aid to every encounter to get an unbiased estimate of relative frequency of limping elk groups across the geographical region. Moreover, this approach accounts for group size in the variance estimate because confidence in presence/absence of the disease in a given location is allowed to be higher for an observation of multiple individuals than for a single elk.

All analysis was done in R 3.2.2 (2015) where a Bayesian hierarchical model (Appendix A2) was estimated with Markov Chain Monte Carlo simulations in JAGS 3.4 (2014). While the analysis is Bayesian in nature, allowing a probabilistic interpretation of the results, we utilized completely diffuse (non-informative) priors for all hyper-parameters such that our posterior estimates are effectively identical to the "objective" maximized likelihood solution.

The base model described in A2 that jointly estimates "limping detection" θ and rate of limping groups ϕ , was fit to three location functions for ϕ : first, a single rate for Southwest Washington, second, broken down by game management units where each unit is independent but the estimate is "shrunk" towards the global mean (units with less observations borrow or share information from all other GMU's, and finally, we predicted the relative rate where the spatial dynamics of TAHD are explicitly accounted for via Ordinary Kriging. In Kriging, each location on a surface is predicted and weighted by the distance to all other neighboring observations (Cressie 1988).

Results

Designing this effort, training participants, and coordinating volunteers required one staff member working full time, and more than 16 staff contributing partial time from January through May. Additionally, five volunteer leads contributed 110 volunteer hours in March and April. Volunteer observers were 96% successful in completing a survey and returning data results to WDFW. Each volunteer spent an average of six hours conducting a survey for a total of 1,308 hours during the project period. One hundred seventy five surveys were conducted (142 by volunteers and 33 by WDFW staff) and 7,300 miles were driven by volunteer observers within the study area. According to track log data, observers surveyed at a mean elevation of 1,127 feet and spent an average of 29% of their time surveying open habitats, 16% in shrub, 48% in forests, and 7% in other habitat. Overall, 19% of observer's time was spent surveying public lands.

Teams encountered a total of 283 elk groups during surveys (Fig. 3). Of the total encounters, 83 (29%) groups had at least one elk detected as limping within the group (6-8% of all individuals seen). Groups were more commonly observed as less than 4 individual elk or more than 11 elk.

Limping counts were not a constant rate within groups, and the proportion of elk within a group that were limping declined sharply as group size increased (Fig. 4). These rates applied to both Regions 5 and 6 and over the entire survey area (Fig. 5).



Figure 3. Map depicts completed surveys and observations of groups of elk across the survey area. Symbols are graduated based on the elk group size.



Figure 4. Histogram representing encounters for group size (total count) and the limping count. Colors blue to red denote an increasing ratio of limping elk within a group.



Figure 5. Map depicts GMU boundaries and proportion of limping elk within groups. Graduated point sizes represent the group count, and green to red colors represent an increasing proportion of limping elk to total elk within the group.

We considered observation time, distance to group, and type of visual observation (i.e., ocular, binoculars, or scope) as covariates that related to detecting limping elk in an effort to determine if we were accurately quantifying the number of limping elk within a group, i.e., detection, in all combinations of observation methods. This analysis revealed statistically significant trends related to the covariates suggesting that detecting limping elk is a function of the observer or survey conditions (e.g., habitat type, group size, etc.) and not just the presence of the disease.

When examining the odds of classifying an elk as limping (detection rate) we found that observers were most confident in classifying elk when they were able to observe groups for 1-10 minutes, at a distance of less than 50 yards, and while using binoculars. The likelihood of classifying an elk confidently varied with any deviation from this method. When we consider this method (1-10 minutes, <50 yards, with binoculars), where we were most confident in our classifications, as a baseline; the odds of classifying an elk as limping increased when observation distance was >50 yards (P = 0.023, 97.7%), when the observation was fleeting (i.e.,

< 1 minute) (P = 0.054, 94.6%), or when the observation time was > 10 minutes (P = 0.016, 98.4%). The odds of classifying an elk as limping decreased when the observer used a scope (P = 0.04, 96%) or when the elk were observed without the aid of binoculars or a spotting scope (P= 0.001, 99.9%) (Fig. 6).



Figure 6. Graphs representing the estimated odds ratio for "detection" of limping elk (or % of limping elk within a group) for deviations in time, distance, and observation type from the baseline (bottom left corner) of 1-10 minutes, <50 yards, and binoculars. P is a Bayesian one sided p-value, 1 minus the probability of the null. TimeDistMethod is the baseline when we were most confident in our classifications. With all other factors remaining at the baseline level, d50 is observations at >50 yards; mScope is the use of a scope; mVis is visual only; t1 represent fleeting observations, and t10 is observations lasting longer than 10 minutes. The black line in all other graphs represents the TimeDistMethod baseline.

When we applied this correction factor across all observations in the entire study area, we estimated that 48% (95% CI = 40–57%) of groups encountered contained at least one limping elk. At the regional level, we estimated the proportion of groups encountered that contained at least one limping elk ranged 41–61% (median = 51%) in Region 5 and ranged 28–59% (median = 42%) in Region 6. The wider interval in region 6 is attributed to the lower sample size in that area.

We used a partial pooling model, allowing data to be borrowed from neighboring units, in an attempt to estimate the rate of limping elk at the GMU level. This model was utilized to balance units with a low sample size (few observations of elk); however, in some cases when observations of groups of elk were low within a GMU, the estimates of the rate of limping groups within these GMU's still ranged considerably (Fig. 7).



Figure 7. A representation of the rates of limping groups by GMU. Numbers represent the number of elk group encounters in each GMU; Triangles represent the estimate of the rate of limping groups; the bars represent the range of the rate of limping groups at a 95% confidence interval; the grey bars span the range estimate at a distribution of 50%; and the vertical line and shaded bar represent the global estimate of the rate of limping groups within the entire study area.

To spatially analyze the proportion of limping groups across the study area, we used an ordinary Kriging prediction. We used the proportion of groups observed limping to produce a reasonable interpolation of occurrence of TAHD across the study area (Fig. 8). The map loosely depicts areas where the likelihood of encountering groups with TAHD may be higher and the proportion of elk affected by TAHD may be greater. When only considering the results from this study, there are issues with scale and over smoothing using this spatial prediction. Further studies with more robust sample sizes may help to refine this prediction.



Figure 8. Ordinary Kriging prediction overlaid with GMU boundaries and proportion of limping elk within groups. Point size represents group count and green to red colors represent an increasing proportion of limping elk to total elk within the group. The prediction surface is not smooth as the model accounts for other factors such as group size, neighboring observations and detection.

Discussion:

With the current sampling design we used a limping gait as a surrogate for an elk being infected with TAHD. While we are sampling in an area with confirmed cases of TAHD, we have assumed that all of the elk that were observed limping during the study were not affected by any other type of deformity, injury, or illness. Furthermore, by using this method it is likely that we

are underestimating TAHD by not detecting limping that is subtle, or in other words, we are not detecting the early stages of the disease. Additionally, we have observed anecdotally that limping behavior can be masked by elk, especially during a disturbance event.

When attempting to classify elk as limping we found that detection rates varied based on the method of observation. Therefore, if we examined a group of elk using any method other than observing through binoculars for 1-10 minutes at less than 50 yards, our odds of detecting limping deviated. All of these scenarios have the potential to result in artificially adjusting our estimate of the proportion of individual elk affected by TAHD. Because of these factors, we suspect under this study design, that using limping elk as a surrogate for TAHD is not an appropriate indicator when trying to estimate prevalence at an individual level.

Since TAHD is an infectious disease and is believed to be contagious among elk, it is reasonable to assume that if a single elk is detected limping within a group there is the potential for other elk within a group to be or become infected as well. Considering infection rates at the group scale allows us to apply correction factors to our observation methods, and therefore determine the proportion of groups affected by TAHD across the survey area; i.e., allows an understanding of apparent prevalence at the group level.

The global estimate of 48% of groups affected by TAHD implies that of the groups we observed, 48% had at least one elk that was limping and detected by our observers. At the regional level we observed a lower estimate of 42% of groups affected for Region 6 as opposed to 51% for Region 5. Both the global estimate and the regional estimates are reasonable given what we know from incidental observations reported since 2012. Previous surveys, while anecdotal, had estimates as high as 80% of groups containing limping elk in the affected geographic region. After a more systematic survey, we now see that the estimate of groups containing limping elk is lower across the study area.

As we attempt to estimate the group infection rate at smaller scales, i.e., at the GMU level, we find that our estimates become unstable due to low sample sizes (too few observations of groups) in some GMUs. We can borrow information from neighboring GMUs to estimate in areas where observations were low, but estimates in this circumstance range widely and should only be considered with caveats in mind. We can see, however, a general concept of the distribution and prevalence of the disease across the Southwest Washington landscape even when considering the existing caveats. Future surveys designed to sample at the GMU level that also target GMUs with low elk group encounters would allow for a more accurate analysis and smoother prediction surfaces of models at a finer scale.

Using citizen science was imperative when attempting to systematically survey for elk over a landscape scale. Due to a successful training effort as well as enthusiastic and interested volunteers, compliance rates among citizen scientists for correctly completing a survey were high and biases between observers when classifying elk were minimal. The logistics, however, of

coordinating 218 volunteers (112 volunteer teams) independently conducting surveys from March through May at a landscape scale presents many constraints on a staffing level. Given that we are unable, under the current sampling design, to determine the apparent prevalence of TAHD at the individual elk level; we need to adjust our original objective to determine the ratio of affected elk groups to the total number of groups observed. This information is critical when monitoring changes in the core area of the Cowlitz River Basin, for further understanding TAHD at smaller scales, and in defining the perimeter of the disease. Therefore, we will pursue an adjusted, smaller scale citizen science survey effort with the understanding that the goal will focus on obtaining group level information across the landscape.

Based on the results of this survey effort, we have launched a second pilot study to explore the possibility that hunter evaluations of hoof conditions for harvested elk could be a means to understand prevalence of TAHD at the individual level. Four hundred and eighty-three modern firearm special elk permit holders in Game Management Units 520 (Winston), 522 (Loo-wit), 524 (Margaret), 550 (Coweeman), 556 (Toutle), and 673 (Williams Creek) are voluntarily being asked to score the hooves of their harvested elk and submit the hooves to WDFW. Hooves will be scored again by WDFW staff to evaluate consistency. If successful, this pilot effort will be expanded to include a larger study area. We are hopeful this method will provide a more complete prevalence estimate of TAHD at the individual level.

In order to inform our understanding of prevalence and distribution of TAHD and guide our management direction, we will continue to collect data from both the citizen science survey and hunter assessment. We will adapt and refine these or any additional methodologies as needed, especially as we learn more from the continued diagnostic work and the ongoing study looking at the effects of TAHD on elk population dynamics.

Acknowledgements:

We would like to thank all of the volunteers who dedicated their time ensuring that this survey effort was possible as well as the volunteer leads: Richard Barlin, Lee Blankenship, Dan Howell, Patrick Miller, and Jim Sevier who spent many hours assisting volunteers; Landowners: Weyerhaeuser, Pope, Sierra Pacific, and Green Diamond; the US Forest Service and the WA Department of Natural Resources; WDFW regional staff as well as Christine Redmond, Hoa Lai, and Anna Marie Sample.

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Appendix

A1 – Elk Hoof Disease – 2015 Protocol Survey Form

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A2 - Bayesian Hierarchical Model

The joint likelihood can be written as

$$l(y,\theta,\phi) = \prod_{j} 1 - \phi (1 - (1 - \theta)^{n_j})$$
(1)

where θ is "detection" or within group limping frequency, and ϕ is the rate of limping groups developed in a hierarchical fashion where if every group encounter is considered a binomial trial

$$y_{ij} \sim Bin(n_j, \theta) \tag{2}$$

where y_{ij} are the limpers out of n_j individuals in group j, then the probability of encountering a group with no limpers is

$$P_{i}(\sum_{i} y_{ij} = 0) = (1 - \phi) + \phi(1 - \theta)^{n_{j}}$$
(3)

 $1 - \phi$ (group is not diseased) *and* the probability the group is diseased (ϕ), but no limpers were detected $(1 - \theta)$ over the n_j "trials". Equation 1 then directly follows Equation 3 after some algebraic manipulation.

ASSESSING THE POTENTIAL EFFECTS OF TREPONEME ASSOCIATED HOOF DISEASE (TAHD) ON ELK POPULATION DYNAMICS IN SOUTHWEST WASHINGTON

PROJECT UPDATE DECEMBER 2015

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METHODS AND RESULTS

Capture and Marking

The Washington Department of Fish and Wildlife (WDFW) conducted captures February 17-27, 2015. We captured female elk via aerial darting from a Bell 206B Jet Ranger helicopter and immobilized elk using carfentanil (3mg) and xylazine (50 mg). We blindfold elk to minimize stress during handling, administered clostridium vaccine and analgesic (flunixin meglumine) injections, and treated the dart wound. We marked each elk using a colored and numbered ear-tag and a mortality-sensitive, GPS (Global Positioning System)-equipped radio-collar. Finally, we antagonized immobilants by administering naltrexone (300 mg) and tolazoline (700 mg).

We captured 81 female elk within Game Management Units (GMUs) 520 (Winston), 522 (Loo-wit), 524 (Margaret), 550 (Coweeman), and 556 (Toutle) (Table 1 and Figure 1). We did not mark three of the elk we captured because two died during the capture process [1 yearling, 1 adult; both had treponeme-associated hoof disease (TAHD)] and the other elk had a broken right rear leg, which was an old injury that was unrelated to us capturing her or TAHD. We chose not to collect samples from the elk with the broken leg and immediately reversed her so this elk is not included in any of the preliminary data that follows.

Of the 78 female elk we marked and fitted with GPS-equipped radio-collars, 58 had visible signs of TAHD. We estimated age at time of capture via tooth wear and replacement and classified three of the 78 elk we marked as yearlings. We radio-collared elk we estimated to be yearlings at time of capture because our official survival analysis period did not begin until May 1 (i.e., start of a new biological year) and any yearling we captured in February would be an adult (\geq 2 years old) at that time. We also removed an upper canine tooth to determine age using microhistological analysis of cementum annuli (Hamlin et al. 2000; Matson's Laboratory, Milltown, MT). The ages of female elk we captured that were affected by TAHD ranged 1-16 years and averaged 6 years old, while the ages of female elk with no visible signs of being affected by TAHD ranged 1-13 years and averaged 7 years old (Figure 2).

that appeared to be unaffected by TAHD (Control).					
GMU	TAHD	Control	Total [*]		
520	24	6	30		
522	11	3	14		
524	1	3	4		
550	15	5	20		
556	9	5	14		
Total	60	20	80		

Table 1. The number of female elk WDFW captured in each GMU, the number of those elk that had visible signs of TAHD and the number of elk that appeared to be unaffected by TAHD (Control).

* Totals do not include the elk that WDFW chose not to mark because it had a severely broken back leg.



Figure 1. Map depicting the location where WDFW captured and radio-collared 80 female elk in February 2015 that were either visibly affected by TAHD (red circles) or appeared to be unaffected by TAHD (blue circles).



Figure 2. Distribution of ages for female elk WDFW captured and radio-collared in February 2015 that were affected by treponeme-associated hoof disease (TAHD) (a) or had no visible signs of being affected by TAHD (b).

Radio-collars

The radio-collars we are using have been functioning properly since we deployed them in February. Since that time, we have documented 13 mortality events (see Survival below) and received prompt notification from the radio-collar in all but one case. In the instance where there was a delayed message from the radio-collar, the elk had died in heavy timber with the radio-collar positioned under her neck, which prevented it from communicating with the satellite. Fix rates during March–August averaged 0.71 (range = 0.50-0.90), which is more than adequate for the purpose of our study. To date, we have collected 23,285 locations from the 78 elk we radio-collared in February 2015 (Figure 3).



Figure 3. Map depicting the > 23,000 GPS locations that WDFW has collected March 2015–November 2015 from the 78 elk they radio-collared in February 2015.

TAHD Observations

We observed a great deal of variation in the condition of hooves of elk that we captured and had visible signs of TAHD (Figure 4). Interesting observations with regard to visible deformities and lesions include:

- ▶ Involved the rear hooves in 98% of affected elk
- Involved just 1 rear hoof in 72% of affected elk
- > Involved the front hooves in 10% of elk
- ▶ Involved more than 1 hoof in 27% of elk
- Involved both back hooves in 21% of elk
- > Involved the rear left hoof in 62% of elk
- ▶ Involved the rear right hoof in 57% of elk

Of the elk we captured that were visibly limping, only two were unaffected by TAHD. One was the elk with the severely broken leg and we believe the other elk had an unknown problem with her hip.

Because we observed such wide variability in the condition of hooves of elk affected by TAHD, we have also begun developing grades of the disease, which relate to disease severity and we anticipate will assist with improving our understanding of how this disease affects elk survival and reproduction (Figure 5). The majority (50/58 = 86%) of elk we radio-collared that were affected by TAHD either had TAHD on multiple hooves (n = 21) or had TAHD on a single hoof with characteristics we have preliminarily associated with a hoof condition score of Grade 3 (n = 8) or Grade 4 (n = 21) (Figure 6). However, we continue to collaborate with other specialists to define criteria we will use to score affected hooves so these frequencies could change.

Body Condition

We determined late-winter body condition [i.e., % ingesta-free body fat (IFBF)] during captures by having an experienced observer use a portable ultrasound to measure maximum subcutaneous rump fat thickness (MAXFAT) and determine a rump body condition score (rBCS) following the procedures of Cook et al. (2001*a*). We then used estimates of MAXFAT and rBCS to estimate IFBF at time of capture following the procedures of Cook et al. (201*a*). We also measured each elk's chest girth to estimate body mass following the procedures of Cook et al. (2003).



Figure 4. Series of photos showing wide variation in the condition of hooves of elk that WDFW radiocollared and were affected by treponeme-associated hoof disease in February 2015.



Figure 5. Diagram depicting characteristics WDFW has associated with the five grades of treponemeassociated hoof disease (TAHD) they defined after capturing 60 female elk in February 2015 that showed wide-variation of the disease.



Figure 6. Distribution of hoof condition scores for 78 female elk WDFW captured and radio-collared in February 2015.

We have not conducted any formal analysis of the body condition data we collected in February 2015. However, we have generated general descriptive statistics (e.g., mean, standard deviation, etc.) based on the preliminary hoof grade scores we assigned to each affected hoof. Although we developed five different grades of TAHD, we provide preliminary descriptive statistics using more generalized classifications. We took that approach at this point in the study for two primary reasons. First, we are still developing the criteria we are going to use to assign hoof condition scores, which means the score we assigned now could change. Second, we have not determined how we will treat elk in our analyses if they have multiple hooves affected. The generalized classifications we used for the statistics we provide below were control, early, late, and multiple. The early group included elk that had TAHD on a single hoof and we assigned a hoof condition score of Grade 1 or Grade 2, the late group included elk that had TAHD on a single hoof and we assigned a hoof condition score of Grade 1 or Grade 2, the late group included elk that had TAHD on a single hoof and we assigned a hoof condition score of Grade 4, and the multiple group included elk that had TAHD on more than one hoof.

Estimates of IFBF and body mass in late winter declined with an increase in TAHD severity, with elk having TAHD on multiple hooves being the most severely affected (Table 2 and Figure 7). However, mean age of radio-collared elk also declined as TAHD severity increased, which is important because there is a strong correlation between body mass and age for elk (Table 2 and Figure 8).

Survival

As of November 25, 2015, 13 (3 controls, 10 affected) of the 78 elk we radio-collared in February have died (Table 3). The first elk died within a day or two of us capturing and marking her, so we will censor that elk from any future survival analyses because she died within 30 days of capture and we could not rule out capture-related stress as a factor that contributed to mortality (Beringer et al. 1996).

We have not conducted a formal analysis of survival at this time. However, naïve survival rates (i.e., $n_{\text{dying}}/n_{\text{at-risk}}$) since March 1 have been 0.84 for elk affected by TAHD and 0.85 for our controls, while naïve survival rates within the biological year (i.e., since May 1) have been 0.87 for elk affected by TAHD and 0.85 for our controls (Figure 9). Seven of 9 (78%) elk affected by TAHD that have died, and we did not censor, were pregnant at time of capture (Table 3).

		Age	IFBF		B	ody Mass
Score	<i>n</i> =	\overline{x}	x 95% CI		\overline{x}	95% CI
Control	19	7.6	5.13	3.95-6.31	205.2	197.0-213.3
Early	7	7.3	5.19	3.42-6.95	208.5	194.9-222.2
Late	28	6.5	4.26	3.45-5.08	194.4	188.3-200.4
Multiple	21	5.6	3.86	2.80-4.91	188.3	178.7-198.0

Table 2. Mean age, percent ingesta-free body fat (IFBF), and body mass (kg) by generalized hoof condition scores (Score) for the 75 adult female elk WDFW captured and radio-collared in 2015.



Figure 7. Boxplots of percent ingesta-free body fat (%IFBF; a) and body mass (kg; b) by TAHD severity for 75 adult female elk WDFW captured in February 2015. Control = elk with no visible signs of TAHD; Early = Elk with TAHD on a single hoof and hoof condition score of Grade 1 or Grade 2; Late = Elk with TAHD on a single hoof and hoof condition scores of Grade 3 or Grade 4; and Multiple = Elk that had TAHD on more than one hoof.



Figure 8. Boxplots of age by TAHD severity for 75 adult female elk WDFW captured in February 2015 (a) and boxplots of age-specific mass for female elk captured by McCorquodale et al. (2014) within our core 5-GMU study area, 2009–2012 (b). Control = elk with no visible signs of TAHD; Early = Elk with TAHD on a single hoof and hoof condition score of Grade 1 or Grade 2; Late = Elk with TAHD on a single hoof and hoof condition scores of Grade 3 or Grade 4; and Multiple = Elk that had TAHD on more than one hoof.

Elk ID	Age	Status	Severity	Mortality	Pregnant	Lactating	Cause of Mortality
111	6	TAHD	Multiple	2/27/15	No	na	Censored
104	9	TAHD	Grade 4	3/22/15	No	na	Malnutrition/Disease
127	11	TAHD	Grade 4	4/3/15	Yes	na	Unknown-natural*
129	11	Control	na	5/22/15	Yes	na	Accident
172	5	TAHD	Grade 4	7/7/15	No	No	Starvation
124	5	TAHD	Multiple	8/15/15	Yes	No	Starvation
107	13	TAHD	Grade 1	8/19/15	Yes	No	Malnutrition/Disease
110	5	TAHD	Grade 4	8/20/15	Yes	Yes	Malnutrition/Disease
159	4	Control	na	9/17/15	Yes	Yes	Human-caused
149	2	TAHD	Grade 2	9/22/15	Yes	Yes	Human-caused
177	8	TAHD	Multiple	11/10/15	Yes	Yes	Unknown-natural*
119	11	Control	na	11/11/15	Yes	Yes	Human-caused
156	9	TAHD	Grade 2	11/16/15	Yes	Yes Unknown-natur	

Table 3. Summary of biological information associated with the 13 mortality events WDFW has documented since February 2015.

*Mortalities we classified as Unknown-natural are mortalities where the carcass was intact, there was no evidence of predation or human-causes, and we collected biological samples to assist with cause-of-mortality determination, but are still waiting for results from the Colorado State University Veterinary Diagnostics Laboratory before assigning a cause of mortality.



Figure 9. Cumulative naïve survival curves (i.e., n_{dying}/n_{total}) for radio-collared elk affected by treponemeassociated hoof disease (TAHD) and for elk with no visible signs of being affected by TAHD (Control). The two survival curves are for cumulative survival () since project initiation (a; TAHD n = 57; Control n = 20) and cumulative survival since the start of the biological year (i.e., May 1) (b; TAHD n = 55; Control n = 20). Numbers on the x-axis correspond to a calendar month (i.e., 3 = March, 4 = April, 5 = May, etc.).

Cause-specific Mortality

We have attempted to investigate all deaths of radio-collared elk within 24 hours of receiving a message that a mortality event occurred. The carcass was intact in all but one instance (hunter harvest) so we collected a variety of samples and submitted them to the Colorado State University (CSU) Veterinary Diagnostic Laboratory to assist with determining cause of death. Samples we collected and shipped to CSU included tissue samples from the heart, lungs, liver, kidney, spleen, pancreas, mammary gland, and brain. We also collected samples of rumen content, a femur, and all four hooves.

To date, we have classified causes of mortality as starvation, malnutrition/disease (aside from TAHD), human-caused (legal harvest, wounding loss, poaching), unknown-natural, and accident. Mortalities we classified as malnutrition/disease included mortalities where it was obvious the elk was severely malnourished but evidence of a disease, other than TAHD, was also present. Mortalities we classified as unknown-natural are mortalities that were not human-caused or related to predation and we are waiting for diagnostic results from CSU before making a final determination.



Figure 10. Number of deaths by cause for elk WDFW radio-collared in February 2015 that were affected by treponeme-associated hoof disease (TAHD) or had no visible signs of being affected by TAHD (Control).

Pregnancy

We determined pregnancy status at time of capture via ultrasonography and analysis of Pregnancy–Specific Protein B (PSPB) in serum samples collected during capture (Noyes et al. 1997). Of the 76 elk we classified as adults, 0.64 (49/76) were pregnant. None of the four elk we classified as yearlings was pregnant. Of the 19 adult female elk that had no visible signs of being affected by TAHD, 0.84 (16/19) were pregnant, and of the 57 adult female elk that had TAHD, 0.58 (33/57) were pregnant. Using the generalized classifications of TAHD severity we described above, pregnancy rates were the lowest for elk that had TAHD on multiple hooves (Table 4). For comparison, McCorquodale et al. (2014) reported an overall pregnancy rate of 67% for the 109 adult female elk they captured 2009-2012.

Table 4. The number and proportion of adult female elk by generalized hoof condition score (Score) that were pregnant at time of capture in February 2015.

Score	п	Number Pregnant	Proportion Pregnant
Control	19	16	0.84
Early	7	7	1.00
Late	28	18	0.64
Multiple	21	7	0.33

Productivity (Early Survivorship of Calves)

In our original proposal, we defined productivity as the early survivorship of calves and stated we would determine the effects of TAHD on elk productivity using two approaches: 1) by estimating calf-at-heel ratios from radio-collared elk and 2) by using lactation rates of hunter harvested elk as surrogates of calf survival. However, we also stated we would employ both approaches in 2015, assess how well each method worked, and then determine whether we would continue to employ these methods during subsequent years of this study.

We conducted calf-at-heel surveys August 10–20, 2015 and spent approximately 174 hours attempting to determine if radio-collared elk had a calf-at-heel. Despite our efforts, we were only able to determine calf-at-heel status for 6/38 radio-collared elk that were still alive and known to be pregnant. Moreover, three of those confirmations occurred because the radio-collared elk died and we were able to assess lactation status post-mortem. The other three confirmations occurred under very fortuitous circumstances—e.g., biologist observed the radio-collared elk briefly in heavy timber, but there were only two cows and two calves, thus the

observer could deduce the radio-collared cow had a calf. Consequently, we have concluded calfat-heel surveys are an ineffective strategy to assess early survivorship of calves and will not employ this strategy during the remainder of this study. We will also not use lactation rates of hunter-harvested elk to assess early survivorship of calves because from this point on, we will conduct captures in December, which should allow us to assess late-autumn condition and lactation status via captured elk (see **Modifications to Study Design** below).

Assuming most calves are not born until June 1 or later, we have had six radio-collared elk that had TAHD and the potential to be caring for a calf at time of death. Of those six elk, four (67%) were lactating at time of death (Table 3) and died late enough into the lactation period that the calf, assuming quantity and quality of milk the cow provided was adequate, had a reasonable chance of survival. Both radio-collared elk that had no signs of being affected by TAHD and died after June 1 were lactating at time of death (Table 3).

DISCUSSION

It is far too soon for us to make any definitive statements that relate to our research objectives or to discuss our results in any detail. Preliminarily, there appears to be a possible relationship between TAHD severity and our late-winter estimates of IFBF and body mass, in addition to pregnancy rates being lowest for the most severely affected elk. However, the mean age of radio-collared elk also tended to have an inverse relationship with TAHD severity, which is a factor that has the potential to interact with TAHD to account for some of the relationships we observed. In addition, we were not able to account for autumn lactation status during captures in February, which is important because estimates of IFBF and body mass in late winter are likely to be significantly related to IFBF and body mass the previous autumn, which have been shown to be significantly affected by lactation status (Cook et al. 2013). We hope to address this limitation in our body condition data by moving our captures to late autumn (i.e. mid-December), which will enable us to determine lactation status (see below).

Survival of radio-collared elk has been similar for both groups up to this point in our study, with estimates of naïve survival rates within the biological year being 0.87 for elk affected by TAHD and 0.85 for our controls. However, radio-collared elk that were affected by TAHD have mostly died of causes related to malnutrition or disease and during times of the year (summer

and autumn) when we would expect survival to be >0.95. In addition, most affected elk that died during summer and autumn were pregnant and/or lactating at time of death (Table 3), which suggests these mortality events may be related to the energetic demands associated with the third trimester of pregnancy and the lactation period.

Although our attempt to assess early survivorship of calves was not successful, we have collected some information via our mortality investigations that indicate some elk affected by TAHD are able to raise a calf to a point when the calf would not be solely reliant on milk from the cow to survive.

MODIFICATIONS TO STUDY DESIGN

Since we initiated our study in February 2015, we have reconsidered some of the methodology we identified in our original study proposal. This reconsideration has occurred for three primary reasons. First, our initial study design did not include recapturing 'control' elk (i.e., no visible signs of TAHD) during subsequent capture events to confirm their control status. We are now proposing to recapture control elk annually so we can ensure we do not bias our survival analysis by having elk in the control group that did not maintain their unaffected status.

Second, although our primary intent during our initial capture effort was to radio-collar elk most severely affected by TAHD, the group of affected elk we captured represented various stages of the disease. Thus, we now believe we have an opportunity to increase our understanding of disease progression by recapturing a subset of affected elk each year to document changes in the condition of their hooves.

Third, some aspects of our original study design would require a great deal of effort, with no way for us to guarantee that the quality and quantity of data we acquired would be sufficient to achieve the associated research objective. For example, our initial study proposal indicated we would estimate autumn body condition of hunter-harvested female elk using modified Kistner scores (Kistner et al. 1980) following the procedures of Cook et al. (2001*b*) and that we would use that data to make inferences about how TAHD affects the nutritional condition of adult female elk in autumn. This approach requires a great deal of staff time and, in terms of usable data, we anticipated would result in very limited return (e.g., McCorquodale et al. 2014), which is why we originally proposed to implement this method across an area much larger than the core 5-GMU area where we radio-collared elk. In addition, accurately assigning lactation status and

identifying the presence of TAHD are requisite for making appropriate inferences about autumn body condition. However, we would have no way of validating the accuracy of this data because we would rely on hunters to collect that information independent of our purview. Although we acknowledged these weaknesses in our original study proposal, we also recognized it was the best we could do without capturing elk in autumn. Now that we are proposing to recapture elk annually, we are also proposing we move our capture efforts to mid-December because that would allow for a study design that is more appropriate for accomplishing our research objectives that relate to assessing the effects of TAHD on body condition of elk in autumn.

Our primary intent below is to identify each aspect of our study design (objectives, methodology, cost, etc.) that will change in response to us recapturing radio-collared elk and conducting captures in December instead of February.

RESEARCH GOALS AND OBJECTIVES

Our initial research goal was to quantify how TAHD affects the survival, pregnancy rates, productivity, and nutritional condition of adult female elk and that will remain unchanged. The only changes we will make to our research objectives are to add a new objective related to disease progression (Objective 6) and reword Objective 5 so it is not restricted to hunter-harvested elk. Our modified study objectives now include:

- 1. Estimate the effects of TAHD on survival of adult (≥ 2 years old) female elk.
- 2. Determine cause-specific mortality rates for adult female elk that have TAHD.
- 3. Estimate the effects of TAHD on the pregnancy rates of adult female elk.
- 4. Estimate the effects of TAHD on elk productivity (i.e., survivorship of calves).
- 5. Estimate the effects of TAHD on the level of condition (i.e., IFBF) adult female elk are able to achieve in autumn.
- 6. Increase our understanding of how TAHD progresses in individual elk, and whether affected elk may recover from the disease.

STUDY AREA

Because we are removing the effort to collect organs from hunter-harvested elk, our study area will no longer include the Willapa Hills elk herd area or areas within the Mount St. Helens elk herd area that are outside our core 5-GMU (520, 522, 524, 550, and 556) study area.

METHODS AND STUDY DESIGN

Capture and Marking

We will employ the same methods we identified in our original proposal to capture and mark elk, but will conduct captures in mid-December instead of late-February. Specifically, we will initiate captures on December 16, which is the first day we can capture elk without conflicting with established black-tailed deer (*Odocoileus hemionus columbianus*) or elk seasons within our study area. In addition, our original proposal indicated we would conduct subsequent captures February 2016–2018 (3 additional capture events), but we are now proposing to conduct subsequent captures December 2015–2018 (4 additional capture events).

We originally indicated we would conduct captures to replace collars that we censor or to replace study animals that have died and anticipated this would result in us capturing 20–25 elk annually. We are now proposing to not only capture elk to maintain our sample size of 80 radio-collared elk, but will also attempt to recapture all radio-collared elk from the control group and a subset of radio-collared elk (10–15) from the affected group, annually. Thus, the number of elk we capture each year will increase from 20–25 to 40–50.

The proportion of elk we capture each year that are new study animals or recaptures will differ for a variety of reasons. First, our primary objective for captures will be to ensure we maintain our sample size of 80 elk, but the number of new study animals we need to capture will vary annually. For example, the proportion of study animals McCorquodale et al. (2014) lost between capture events, ranged from a low of 20% in 2011 to a high of 41% in 2012 (Table 5). If we apply these values to our sample size of 60 affected elk, we would need to capture 12 and 25 new elk, respectively. If we have a year when we need to capture 20–25 new study animals for the affected group, it is unlikely we would have the time to recapture very many radio-collared elk from the affected group.

Second, to make the strongest inferences about nutritional condition of elk in autumn, 12–15 of the adult female elk we capture in each group (control and affected) need to be lactating (R. Cook, NCASI, personal communication), which we anticipate will necessitate that we capture all 20 control elk and 25–30 affected elk before we will meet our required sample size.

Third, the number of new elk we need to capture within each study group may also depend on the ability of our control elk to maintain their unaffected status. For example, if we recapture 15 of our control elk in December 2015 and five of them have developed signs of TAHD, they will automatically transition to our affected group. This would not only require us to capture more new study animals for the control group than we had originally planned, but it would also reduce the number of new elk we need to capture for the affected group.

Lastly, but most importantly, the number of elk we recapture from each group will vary because we already know we will locate some elk in areas that are not conducive to us capturing them. This may include elk that are in heavy timber and will not move out into open terrain or elk that are in locations that jeopardize the safety and well-being of the capture crew and/or elk.

		Harvested		<u>Natur</u>	<u>al Causes</u>	Combined	
Year	At Risk	п	Prop	n	Prop	п	Prop
2009	53	9	0.17	5	0.09	14	0.26
2010	74	11	0.15	7	0.09	18	0.24
2011	80	9	0.11	7	0.09	16	0.20
2012	75	13	0.17	18	0.24	31	0.41
Mean	71	11	0.15	9	0.13	20	0.28

Table 5. The number and proportion (Prop) of radio-collared elk that McCorquodale et al. (2014) reported as being harvested or dying of natural causes in our core 5-GMU study area, 2009-2012.

Confirming the Status of Controls

As discussed previously, we will attempt to recapture all radio-collared elk in the control group annually to confirm they have maintained their control status, but there may be factors that preclude us from doing so. However, we may be comfortable with confirming an elk's control status even if we are not able to recapture them. For example, we may locate an elk in an area that is not conducive to capture, but from our observation, we feel confident the elk has not developed symptoms associated with TAHD, at least not to the extent that would reduce its mobility or cause it to limp. We recognize the potential to misclassify control elk from the helicopter that have contracted TAHD, but only have symptoms associated with early stages of

the disease (e.g., lesions). To address this potential bias, we plan to assign disease status from the helicopter for every control elk we attempt to capture, which, for elk we successfully recapture, will allow us to compare the assessment we made from the helicopter to the assessment we made by visually inspecting each hoof. This in turn will allow us to determine how confident we are in any assessment we made for elk we were not able to recapture.

We will also confirm disease status for any control elk that dies. In instances when the carcass is located with the collar, we will collect the hooves and submit them to CSU for analysis to confirm the elk is unaffected by TAHD. For some elk that hunters harvest (e.g., during general archery seasons) we anticipate we will have to coordinate with the hunter to identify the location where they left the hooves and return to the site as soon as possible to increase our probability of collecting the hooves before they are scavenged. Thus, we recognize we may not always be able to confirm the disease status of our control elk and will deal with that accordingly.

Body Condition

We will only measure body condition at time of capture and will not attempt to estimate autumn body condition from hunter-harvested elk using modified Kistner scores. Instead of estimating late-winter body condition at time of capture, we will be estimating late-autumn body condition. The methodology we will use to evaluate body condition and determine lactation status will remain the same.

Survival Analysis

No change

Cause Specific Mortality

No change

Pregnancy Rate

No Change

Productivity

In our initial proposal, we defined productivity as the early survivorship of calves and indicated we would estimate the effects of TAHD on productivity using calf-at-heel surveys or lactation data collected from hunter-harvested elk. As discussed earlier, we do not feel that calf-at-heel surveys are an effective strategy for assessing the early survivorship of calves and will not conduct these surveys during subsequent years of this study.

As for using lactation data from hunter-harvested elk, our original study design stated that, we would use that data to make population-level inferences about how TAHD affects productivity. More specifically, we were going to use logistic regression to identify covariates (e.g., TAHD status, age, year, etc.) that influence the probability that a hunter-harvested elk would be lactating in autumn. However, we would be limited in our ability to make inferences about calf survival because the probability of an adult female elk lactating in late-autumn of year_t is a function of not only the probability of her calf surviving during summer and early autumn in year, but also the probability of her conceiving in year, We would address this limitation by using information from our analysis of pregnancy data to strengthen the inferences we might be able to make about calf survival. For example, if TAHD status was not a significant predictor of an elk conceiving in autumn, but was a significant predictor of lactation status for hunterharvested elk, then we might be comfortable assuming that was because elk affected by TAHD had lower rates of calf survival. There would be additional assumptions that we must apply when using this approach, but we believed it was a reasonable method to identify potential differences in calf survival between the two groups without estimating calf survival directly (i.e., radio collaring calves). We will still assess the potential effects of TAHD on calf survival using this approach, but will use lactation rates of captured elk instead of hunter-harvested elk.

With our revised study design now including strategies related to recapturing a portion of our radio-collared elk, we may also have the ability to conduct an individual-level analysis of calf survival to 6 months, which would allow for much stronger inferences. This ability may exist because we will know which elk from previous capture events were pregnant and thus, which elk we recapture that are not lactating because they did not conceive the previous autumn or because their calf died. For example, 17 of the 20 control elk we captured in February 2015 are still alive and we determined 13 were pregnant. If we recaptured all 13 in December 2015 and 8 of them are lactating, we would estimate calf survival to 6 months of age to be 0.62.

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However, the number of elk we recapture will vary because of reasons we cited above so we cannot be certain our sample size will be adequate for any analyses beyond providing descriptive statistics.

Disease Progression

We will increase our understanding of disease, including whether elk recover from TAHD, by re-examining the hooves of elk that have died or that we recapture.

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Treponeme-associated Hoof Disease in Elk in Southwest WA: Timeline of Events, Diagnostics, Research, & Management Efforts

1990's	• Sporadic reports of hoof deformities in the Cowlitz River Basin
2008	• Number of reports of abnormal hooves and lameness in elk as well as geographic scope increased significantly
2009	 WDFW conducted first clinical investigation on affected elk in March WDFW conducted a survey of hunters for an initial understanding of prevalence and distribution of limping elk
2010-2011	 Reviewed findings of 2009 investigation to guide future steps Comparison of copper and selenium levels from affected elk versus non-affected herds Consulted with national and international experts in wildlife disease
• WSU • WDF • Deve	College of Veterinary Medicine faculty convened to advise on diagnostic investigation W Public meeting to share information loped Online Hoof Disease Reporting Tool and informational materials on WDFW website
• WDF • WDF and t • WDF optic • Deve	W diagnostic collections of elk in March and August W formalized Hoof Disease Technical Advisory Group to assess results of diagnostic investigation to advise on further diagnostic approaches. W formalized Hoof Disease Public Working Group to share information and discuss management ons and research questions (meetings in October and December) loped Hoof Disease Health/Safety Fact Sheet in partnership with Department of Health
• WDFW • Hoof D • Hoof D trepon • Fish & • WDFW • WDFW • WDFW 2015 • Outrea deform • WDFW • Develo disease • WDFW • Develo disease • WDFW and in • WAC 2 501-56 minimi • Legisla • \$180,00 • \$8,000 • \$250,00	diagnostic collections of elk in January isease Public Working Group meetings in February and May isease Technical Advisory Group reviewed results and reached consensus statement on eme-associated hoof disease in elk Wildlife Commission adopted new rule to leave hooves on site from harvested elk and citizen hosted public meetings (4) hired Hoof Disease Coordinator implemented a pilot prevalence study with volunteers to inform a larger effort for spring ch for public assistance with an expanded effort to report limping elk or dead elk with hoof nities on-line developing management approach based on input from HDTAG, HDPWG, & WDFW staff loping euthanasia criteria for severely affected elk ping study design to implement long term elk survival study in 2015 to evaluate effect of hoof adjuster articles 32-12-286 (Sept 2014): Hunters required to remove the hooves of any elk taken in GMUs 44 and 642-699 in SW WA and leave them on site as a precautionary measure in an attempt to ze the spread of the TAHD in elk tive approved \$200,000 supplemental budget for hoof disease investigation 00 Pittman-Robertson funds for hoof disease work RMEF funds for sample analyses 00 legislative request for 2015-2017 biennium
• Janu diag • Febr • Mare • April • Nove	ary/May – Peer reviewed scientific publications of Treponeme Associated Hoof Disease (TAHD) nosis in elk uary – Initiated TAHD Survival Study ch – April – Conducted citizen science effort to better understand prevalence of TAHD in elk I – Submitted SEPA checklist for removal of severely affected TAHD elk for humane reasons – DNS ember – Initiated pilot hunter elk harvest hoof assessment