

**A PRELIMINARY GENETIC STUDY OF THE WILD HORSE
(*Equus caballus*) IN THE BRITTANY TRIANGLE
(TACHELACH'ED) REGION OF THE ?ELEGESI QAYUS
(NEMIAH) WILD HORSE PRESERVE OF BRITISH COLUMBIA**



November 2014

Dr. E. Gus Cothran

Department of Veterinary Integrative Bioscience
Texas A & M University, College Station, TX 77843-4458
email: GCothran@cvm.tamu.edu

Wayne P. McCrory, RPBio

McCrory Wildlife Services
208 Laktin Road, New Denver, British Columbia V0G 1S1
Phone 250-358-7796, email: waynem@vws.org

[Study for Valhalla Wilderness Society (VWS), Friends of Nemaiah Valley (FONV), & Xení Gwet'in First Nation]



LEGAL COVENANT FROM THE XENI GWET'IN GOVERNMENT



TSILHQOT'IN
PEOPLE OF XENI

Tsilhqot'in Rights & Title

The Supreme Court of Canada recognized the Tsilhqot'in Nation's Aboriginal title in the lands described in *Tsilhqot'in Nation v. British Columbia*, 2013 SCC 44, and their Aboriginal rights to hunt and trap throughout the area claimed in *Tsilhqot'in Nation v. British Columbia*, 2007 BCSC 1700 (the "Claim Area"). This includes the rights to hunt and trap birds and animals for the purposes of securing animals for work and transportation, food, clothing, shelter, mats, blankets, and crafts, as well as for spiritual, ceremonial, and cultural uses throughout the Brittany Triangle (Tachelach'ed) and the Xeni Gwet'in Trapline. This right is inclusive of a right to capture and use horses for transportation and work. The Claim Area is within the Tsilhqot'in traditional territory and the Xeni Gwet'in First Nation's caretaking area. The Tsilhqot'in Nation also enjoys Aboriginal rights and title outside the Claim Area. Nothing said in our meetings or documents shall abrogate or derogate from any Aboriginal title or rights of the Tsilhqot'in, the Xeni Gwet'in First Nation or any Tsilhqot'in or Xeni Gwet'in members. The Xeni Gwet'in First Nation is one of six Tsilhqot'in communities which include: Yunesit'in, TI'etincox, Tsi Del Del, ?Esdilagh, and TI'esqox.



Characteristic wild horse pocket/wetland sedge/grassland habitat of the Brittany Triangle Plateau. This is one of the most remote and harsh wild horse areas left in Canada. This is an unusually large group of wild horses, as bands in the Brittany Triangle usually number 10-14 horses. Chris Harris photo.

Wayne McCrory ACKNOWLEDGEMENTS

Thanks are expressed for financial support from The Vancouver Foundation, Friends of Nemaiah Valley (FONV), Valhalla Wilderness Society (VWS), anonymous donors, and others. Thanks are also extended to the genetics lab at the Department of Veterinary Integrative Bioscience, Texas A&M University, for doing the genetic analysis at nominal costs.

The Xeni Gwet'in (Nemiah) First Nations Government is thanked for allowing us to carry out this research in their Caretaker and Rights Area. Special thanks to Chief Roger William and former Chief Marilyn Baptiste for their ongoing advice and support. BC Parks is thanked for providing research permits for our main sample area, Nunsti Provincial Park.

David Williams and Pat Swift of FONV are particularly thanked for their tireless support, enduring faith in the Nemiah People and their horse culture, and for holding so many things together that make things work, as well as for their generosity in providing a comfortable and always interesting research station at Far Meadow.

The following are thanked for allowing us to take blood samples from their domestic horses previously captured from wild stock in the Brittany Triangle:

- ❖ Raphael Williams
- ❖ George Colgate
- ❖ Ian Bridge
- ❖ Terry Lulua
- ❖ Bob Blatchford
- ❖ Rosie Pierce

Special thanks to Veterinarian Dr. Corrine Long for her volunteer collection of blood samples. Thanks also to Dr. Dag Goering, who trained David Williams in the art of collecting blood samples from horses.

Many volunteers helped with our collection of wild horse hair samples from the Brittany Triangle, including David Williams, Pat Swift, Jessica Setah, the late Harry Setah, Jon Huizinga, Katherine Card, Dr. Jonaki Bhattacharyya, and others.

Biologist and editor Maggie Paquet is thanked for her editing and formatting of the final report.

E. Gus Cothran ACKNOWLEDGEMENTS

I would like to thank all those people who were involved in the permissions for the sample collection and study, and those people who provided or collected samples. In addition, I must thank Dr. Rytis Jursas, who performed the laboratory work that the genetic part of this study is based upon. He also did much of the statistical analysis. Dr. Eleanore Hempsey also helped with the lab work and analysis, and Dr. Anas Khashour helped with data analysis and figure preparation.

TABLE OF CONTENTS

LEGAL COVENANT FROM THE XENI GWET'IN GOVERNMENT

ACKNOWLEDGMENTS W. MCCRORY	3
ACKNOWLEDGMENTS G. COTHRAN	4
EXECUTIVE SUMMARY	7
1.0 INTRODUCTION	9
1.1 Study Background.....	9
2.0 STUDY AREA	11
2.1 Wild Horse Sub-populations in the Study Area.....	12
2.2 Estimated Wild Horse Distribution in the BC Chilcotin and Estimated Numbers in the North Brittany Triangle Plateau Study Area.....	14
3.0 METHODS AND APPROACH	16
3.1 Blood Sample Approach.....	16
3.2 Hair Sample Approach.....	16
3.3 Laboratory Analysis.....	18
3.3.1 Statistical Analysis.....	19
4.0 RESULTS	22
4.1 Field Sampling Constraints.....	22
4.2 Genetic Results.....	22
5.0 DISCUSSION	25
5.1 Genetic Interpretation.....	25
5.2 Implications of Genetic Results; Historic and Present Context.....	27
6.0 LITERATURE CITED	30
LIST OF MAPS	
Map 1. Shows the spread of the Spanish horse in North America.....	10
Map 2. Xení Gwet'in First Nations traditional lands showing the Brittany Triangle within the greater ?Elegesí Qiyus (Nemiah) Wild Horse Preserve or Xení Gwet'in Caretaker Area.....	11
Map 3. Distribution of Chilcotin feral horses today, Brittany genetic study area and interpolated horse distribution by subarea based on winter horse counts.....	15
LIST OF FIGURES	
Figure 1. Dr. Colleen Long preparing to take a blood sample from a captured wild horse.....	16
Figure 2. Field researchers D. Williams and W. McCrory collecting wild horse hair samples.....	17
Figure 3. 2010 expedition to collect wild horse hair at the north end of the Brittany Triangle.....	18
Figure 4. Likelihood tree showing Brittany Triangle Feral Herd in relation to 68 domestic breeds and with the Przewalski Horse used as an outgroup.....	20
Figure 5. Likelihood tree showing the same dataset as in Figure 4, plus data from 43 feral horse populations from the northwestern US, plus one herd from Alberta.....	21
Figure 6. Medium-sized adult black stallion in spring habitat in the Brittany Triangle, 2002.....	28
Figure 7. Wild horses in spring time to the east of the Brittany Triangle.....	29
LIST OF TABLES	
Table 1. Breed list, Nei data.....	23



Free-ranging horses in the Brittany Triangle have survived harsh winter conditions since apparently being introduced to the area by First Nations in about 1750. W. McCrory, photo.

EXECUTIVE SUMMARY

There is widespread historic documentation that the common ancestral lineage of the wild horse (*Equus caballus*) found in the Americas came from Spanish horses brought over from Spain in the early 1500s. A small number of New World wild horse populations still maintain characteristics ascribed to their Spanish heritage, despite horses from other parts of Europe subsequently being introduced to North America.

It is assumed that the horses observed by European fur traders in the early 1800s in association with Tsilhqot'in First Nations in the Chilcotin area of south central British Columbia were descended from Spanish-derived horses brought in about 1740 along native trade routes from plateau grasslands in what is now Washington State. Today, an estimated 1,000 feral horses still survive in remote areas of the Chilcotin Plateau, including an estimated 150-215 in the Brittany Triangle sup-population. The semi-isolated Brittany sup-population is likely the most remote herd left in mainland Canada and has survived for several centuries alongside major predators, including grizzly bears (*Ursus arctos*), mountain lions (*Felis concolor*), and grey wolves (*Canis lupus*). In 2002, to protect and recover these horses, the Xenigwet'in, a horse culture and member group of the Tsilhqot'in Nation, established a large wild horse preserve, an area that includes the Brittany Triangle (?Elegesi Qayus [Nemiah] Wild Horse Preserve). The Xenigwet'in have won recognition through the BC Supreme Court of their aboriginal right to capture wild horses for domestic and work purposes.

Based on a preliminary review of First Nations oral history and written historical documents regarding the origin of wild horses in the Chilcotin, we decided to test the hypothesis that Chilcotin-Brittany Triangle wild horses came from Spanish bloodlines, and to explore other ancestral genetic lineages. Our genetic testing of the Brittany sup-population meets the requirements of the 2005 Convention of Biological Diversity for livestock.

An iterative approach was used with respect to testing methods, first with eight blood samples collected in 2003 and 2004 from domestic ranch horses that had been captured wild in the Brittany Triangle. A qualitative assessment of the variation seen in the Brittany sup-population indicated at least some probability of Iberian influence and possibly old Spanish ancestry, but this was considered preliminary because of the very small sample size. Following this, 99 viable hair samples were collected from the wilds between 2006-2010. The hair was collected over a broad sample area from trees where horses traveled or sheltered. These were submitted to Texas A & M University for a more in-depth and reliable genetic analysis.

As the Brittany sup-population has been semi-isolated for several centuries by major river valleys, tests were also done for other genetic uniqueness. Genetic background of hair samples was analysed using a set of 15 microsatellite markers and mitochondrial DNA (mtDNA) sequence variation. MtDNA sequencing (n=32 random samples) revealed three unique haplotypes. Microsatellite data was used to assess levels of genetic diversity within the population and for phylogenetic analysis to compare with data from 85 other domestic and feral horse breeds.

Our genetic study raises more questions than it answers, but it does dispel our earlier tentative results from blood samples and the hypothesis that the Brittany horses today still have Spanish ancestry. The more refined analysis and larger sample size using hair samples showed no conclusive Spanish ancestry. These results however do not disprove the considerable historic evidence that the Tsilhqot'in

had horses pre-contact as early as 1740, and the hypothesis that these likely came from Spanish horses to the south. If this hypothesis is true, our study indicates that the genetic make-up of today's wild horses in the Brittany Triangle has undergone considerable transformation since these earlier times, as with many other New World horse sup-populations where few retain any Spanish ancestry. Since European contact in the early 1800s, many different Old World breeds have been brought into the area, including during the Caribou gold rush in the 1860s, with some escaping or being deliberately released into the wilds. Or, alternatively, the foundation herds in the Brittany were actually different from Spanish horses altogether. These will remain unanswered questions.

In a comparison with 69 different horse breeds by phylogenetic analysis, the Brittany Triangle (BT) horse of today paired with the Canadian Horse breed within the cluster that includes the Shire, Clydesdale, Highland Pony, Eriskay Pony, Fell Pony, and Dales Pony. This is a natural cluster of the domestic breeds, which are native British breeds except for the Canadian. The origins of the herd are largely from the heavy horse types and, specifically, it appears that the Canadian Horse breed (or its ancestors) contributed significantly to the ancestry of the Brittany Triangle sup-population. This warrants further investigation.

The most intriguing result of our genetic study was the possibility that Yakut horses, an ancient horse of Russian heritage, also contributed to the origins of the herd. However, this requires more study and probably more baseline samples from Eastern Russia. We found very limited historic documentation to support the most obvious hypothesis that the Yakut horse bloodlines arrived in the remote Brittany Triangle of British Columbia from Russian fur traders along the adjacent Pacific Coast.

The results, including the great degree of similarity among individuals, are also indicative of isolation. There is no indication of specific subgroups within the herd based upon cluster analysis of individuals, with the possible exception of the haplotype 3 horses.

Further research is needed to determine the possible genetic pathways of what appear to be the somewhat unique Canadian and Yakut ancestry in the Brittany Triangle wild horse herds. It also appears important to conserve the gene pool until more is understood.

1.0 INTRODUCTION

1.1 Study background

The forerunners of today's domestic horse evolved in North America but disappeared from the continent about 8,000-10,000 years ago. It was reintroduced to the Americas by Spanish explorers in the early 1500s. Although horses from other parts of Europe were subsequently brought to the Americas, some New World sub-populations still maintain characteristics ascribed to their Spanish ancestry (Conant et al. 2011). A historic review of wild horses in the Brittany Triangle (McCrary 2002a) demonstrated that the Xenigwet'in First Nation had horses prior to European contact and that the original horses most likely came from Spanish horses.

The Xenigwet'in are a member tribe of the Tsilhqot'in (Chilcotin) Nation. They have never signed treaties but through their land claims process have recently won, through both the provincial and federal supreme courts, recognition of aboriginal rights and title for part of their traditional territory. This includes the right to capture wild horses for transportation and work. Some Xenigwet'in elders who were interviewed expressed the viewpoint that they had horses for a "*long time*" or "*since time immemorial*." One elder claimed the Tsilhqot'in had horses before the 1700s. In 1808, Simon Fraser, one of the first European explorers in search of a fur trade route to the Pacific, made numerous observations in his journal of native people with horses in the grasslands and desert-like canyonlands along the great salmon river now bearing his name.

The Tsilhqot'in had horses and lived near the top of the river named after them (which is where the Brittany Triangle is located). In 1845, Father Giovanni Nobili, an Italian Jesuit priest, journeyed on horseback from Fort Alexandria, a Hudson's Bay post situated along the Fraser River, deep into Chilcotin territory. His detailed letters, dated November 30 and December 27, 1845, indicated that the Xenigwet'in had horses but not in great numbers and they did not use them for trade with the Hudson's Bay Company on the coast (Nobili, G. 1845-1846).

In his 1808 journal, Simon Fraser recorded a number of different words that the Tsilhqot'in used in reference to horses (Lamb 1966). Today, the Xenigwet'in call the horse *Našlhiny*. The pre-European linguistic history of unique naming of the species suggests that the Tsilhqot'in had horses for quite some time prior to contact when compared to the moose, which did not arrive in the region until about 1920 as a result of a natural post-glacial re-colonization. The moose has no First Nation linguistically distinct name other than *Mus*, which reflects the European name.

The combined evidence of First Nations oral history and the Simon Fraser-Nobili journals and early Hudson's Bay records strongly indicate that the early occurrence of horses in what is now the Cariboo-Chilcotin region was the result of native people bringing Spanish-derived horses along their well-established trade routes from the Columbia Plateau grassland areas to the south from what is now Washington State. This is consistent with the widespread historic documentation that free-roaming and domestic First Nations horses noted by early European fur traders, explorers, and settlers in association with many North American native cultures in the vast prairie grasslands and canyonlands of the continent originated from Iberian horses brought from Spain in the early 1500s (Cowdrey et al. 2012). Using early historic documents to trace the spread of the Spanish horse northward and westward across the continent, Cowdrey et al. (2012) assume horses probably arrived in the Cariboo-Chilcotin around 1740 (see Map 1).

In a recent background study of wild horses in the Brittany Triangle of BC's Chilcotin, McCrory (2002a) concluded, from field research and information provided by the Xeni Gwet'in, that free-ranging horses likely existed in the remote Triangle for several centuries or more. Geographic isolation created by mountain ranges and deeply incised river valleys suggest the horse bands in the Triangle are likely the most isolated surviving in western Canada. These factors, plus anecdotal observations by the Xeni Gwet'in and at least one Spanish horse expert (Ginger Kathrens pers. comm.) suggested the possibility that some ancestral Spanish bloodlines still exist within the Brittany wild horse sub-population. Subsequently, a research proposal was prepared in February 2002 to test this hypothesis (McCrory 2002b). After consultations with Dr. G. Cothran, a world expert on horse lineage and genetics at Texas A & M University, we decided to begin testing. Funding was obtained by Friends of Nemaiah Valley (FONV) to do a pilot study using a small number of blood samples from domesticated horses originally captured in the Brittany Triangle. Dr. Cothran's initial results, based on eight blood samples, were somewhat positive, but not conclusive, for Spanish ancestry, and he recommended that we switch to hair samples collected from the wilds not to increase the sample size but to obtain more reliable results. Subsequently, with further funding from the Vancouver Foundation, FONV, and the Valhalla Wilderness Society (VWS), more intensive field collection of hair was done. The sampling period for both blood and hair samples ran from 2003 to 2011.

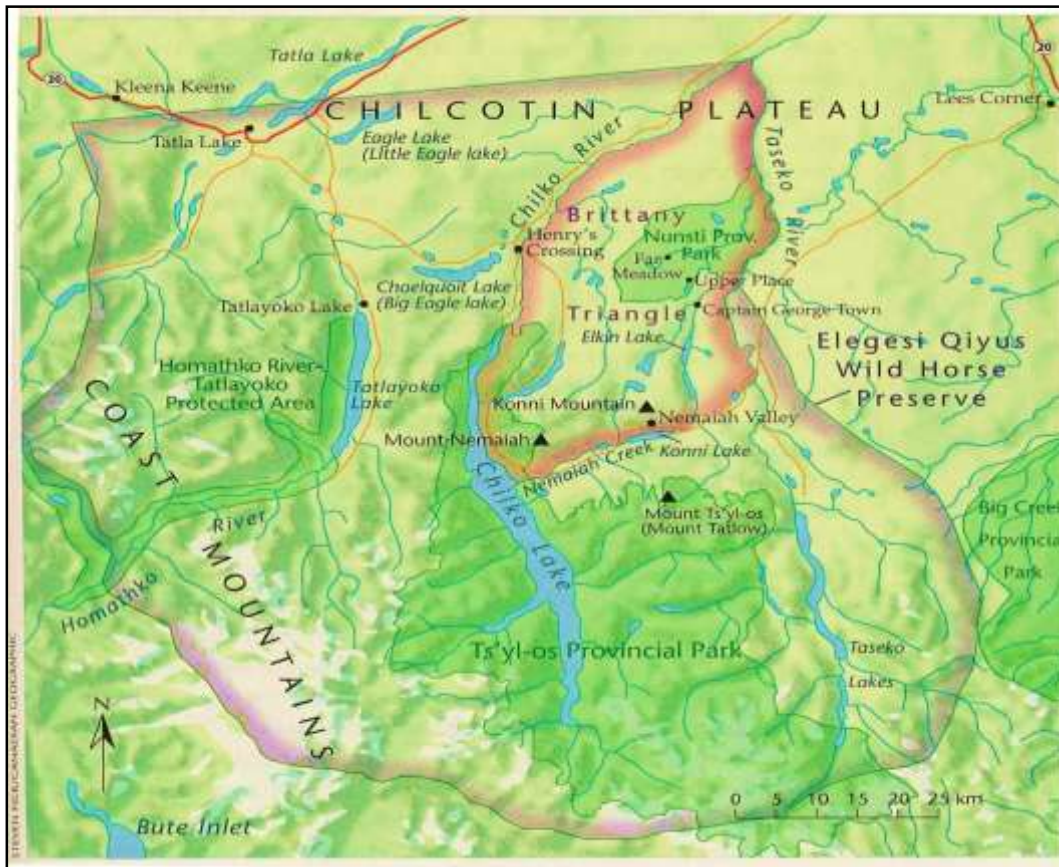


Map 1. Showing the spread of the Spanish horse in North America including (top left) when they were thought to have arrived in the Brittany Triangle and the Nemaiah. Map compliments of Mike Cowdrey, and Ned and Jody Martin, Horses and Bridles of the American Indians. Hawkhill Press, Nicasio, Cal., 2012. www.hawkhillpress.com.

2.0 STUDY AREA

The Brittany Triangle (Tachelach'ed), a large plateau and mountainous area of approximately 155,000 hectares (381,300 acres), is located about 120 kms southwest of the city of Williams Lake in the Chilcotin Plateau of the interior of British Columbia (Map 2). The study area is in the traditional territory of the Xeni Gwet'in First Nation. Currently, part of the Brittany Triangle is now a recognized Aboriginal Rights area (Vickers 2007) and Title area (SCC 2014). Part of it is protected by Nunsti Provincial Park. The remainder is being considered as a Xeni Gwet'in Tribal Park (D. Williams pers. comm. 2014).

The study area has a rich ethnography and numerous First Nation cultural/heritage sites. The Xeni Gwet'in First Nation is a horse culture whose ancestry in the area goes back thousands of years. Provoked by threats of extensive roading, clearcut logging, and open-pit mining, the Xeni Gwet'in declared protection of their traditional territory in 1989. They decreed that no industrial mining, logging, and hydroelectric development would be allowed. In 2002, as a result of a biological assessment of wild horses and wildlife of the Brittany Triangle (McCrorry 2002a), the Xeni Gwet'in established the ?Elegesi Qayus Wild Horse Preserve (Eagle Lake Henry Cayuse Wild Horse Preserve), which overlays the same area as their 1989 Aboriginal Preserve. The area is approximately the same size as Yellowstone National Park. The objective of the wild horse preserve is to maintain and restore the threatened population of wild horses to be used as part of the Xeni Gwet'in cultural and wilderness tourism program and for capture for domestic uses.



Map 2: Xeni Gwet'in First Nations traditional lands showing the Brittany Triangle (pink boundary) within the greater ?Elegesi Qiyus Wild Horse Preserve (purple boundary) or Xeni Gwet'in Caretaker Area. Nunsti Provincial Park (green) and the adjacent Brittany Plateau was the centre of our wild horse genetics study (Map courtesy of www.canadiangeographic.ca/Magazine/ma05/indepth/maps.asp?from=maps).

Unlike the U.S., which federally protects wild horses and has established many preserves, wild horses in Canada are not protected by any statutes; only in 2013 was the Sable Island wild horse area off the coast of Nova Scotia protected as a national park. The Xeni Gwet'in Aboriginal/Wild Horse Preserve decreed protection area is the first wild horse preserve established in western Canada and was designed by the Xeni Gwet'in Elders to provide full protection from industrial forestry, mining, and hydroelectric development in a manner similar to a fully protected provincial or national park. The preserve meets the International Union for the Conservation of Nature's (IUCN) definition of a protected area (Dudley 2008) and the 2003 World Congress' definition of an Indigenous and Community Conserved Area (ICCA) (World Parks Congress 2003). However, because the Xeni Gwet'in Aboriginal/Wild Horse Preserve areas have not been officially recognized by the provincial or federal governments, industrial-scale logging has already degraded some 16% of it. A large proposed open pit mine, recently turned down a second time by the federal government, has generated significant local, provincial, and national controversy (McCrary 2014). Other than several large wildfires over the past decade, the Brittany wild horse study area remains largely a pristine wilderness.

Geographically, the "Triangle" is defined by the natural boundaries of two large sister river valleys, the Taseko and Chilko, that flow northward from large glacial massifs and lakes where they are widely separated at the outset as they cut through moderately incised canyons over the plateau, then eventually join together at the "confluence" that forms the distinct apex of the triangle. The watersheds are also significant migratory and spawning grounds for Pacific wild salmon.

Elevations in the Triangle range from about 1,000 metres along the river bottoms to 1,200–1,400 m on the plateau and up to mountain peaks over 2000 m that form the south boundary of the Triangle.

The Nemiah Valley, a large and spectacular pocket grassland, nestles to the south of this mountain range, and is the current homeland for the Xeni Gwet'in and a small number of non-native residents, including ranchers and tourism operators.

2.1 Wild horse sub-populations in the study area

The general area has a number of apparently separate sub-populations of free-ranging horses; the two main ones of interest being the Nemiah Valley sub-population on the south and the main Brittany Triangle sub-population on the north. Other horse bands exist to the north and east of these Triangle horses (Map 3), such as on the east side of the Taseko River. Today, the Nemiah Valley free-ranging herds are a mix of domestic (branded and/or halter-broke) and wild horses due to the strong, historic settlement pattern of private ranch lands and Indian Reserves. The generally isolated Brittany Triangle appears to have a predominantly wild horse sub-population of 150-215 animals with little current domestic influence, which is why we chose it as our genetics study area. These two sub-populations are connected by a small number of valley bottom and mountain pass wildlife corridors, but ten years of intermittent observations by Wayne McCrary and others, including a remote camera set up for a season in a mountain pass between the two horse sub-population areas, strongly suggests that these sub-populations remain largely genetically isolated from one another.

The main study area was the central portion of the Brittany Triangle. The more remote areas to the north towards the apex of the Triangle proved logistically difficult to sample and one wild horse hair sampling attempt there showed little presence of wild horses. The main study area was comprised of Nunsti Provincial Park and adjacent areas of the plateau. Nunsti Provincial Park is 20,898 ha (51,409

acres) and was established by the province of British Columbia in 1994. A research permit was obtained from BC Parks to carry out the study.

The study area is in the lee of the Coast Ranges. The mountains surrounding the high plateau lands cause a “rainshadow effect” that results in most of the precipitation from Pacific coastal storms falling on the temperate rainforests on the west side of the coastal mountains, thus allowing for much drier conditions on the east side that are favourable to grassland ecosystems. Winter snow depths are generally light with periodic extreme subzero temperatures. Warming winds called “Chinooks” are common, causing snowmelt and then icing and crusting conditions. Summers can be hot with periods of drought. All of these variable weather conditions create periods of harsh survival for the hardy, wild horses in the Brittany. For example, Chinooks followed by freezing temperatures can cause icing of feeding areas in meadows as well as difficult travel conditions in deep snow, while allowing large predators, such as the mountain lion and grey wolf, to hunt wild horses by ease of travel on top of the crusted snow.

The study area encompasses two provincial biogeoclimatic zones. The majority of the area is in the Sub-boreal Pine-Spruce (SBPS) biogeoclimatic zone (SBPSxc subvariant), while some is in the Interior Douglas-fir (IDF) biogeoclimatic zone (IDFdk4 subvariant) (BC Commission on Resources and Environment, 1994). Lodgepole pine (*Pinus contorta*) is the dominant forest cover on the plateau, interspersed with hundreds of small and large native meadows, wetlands, ponds, and lakes. The river breaks are more characterized by old-growth Interior Douglas-fir (*Pseudotsuga menziesii* ssp. *glauca*) and bluebunch wheatgrass (*Agropyron spicatum*). The large size of the study area and the various geological, soil, topographic, and diverse vegetation associations combine to provide considerable habitat diversity both for wildlife and wild horses.

In the Brittany study area, hundreds of natural meadows and wetlands of varying sizes and shapes break up the vast expanses of lodgepole pine forest, creating a quite diverse landscape. The natural meadows and riparian areas comprise the main seasonal habitats for a significant wild horse population. However, the horses also make substantial use of the forested areas, including feeding on pinegrass (*Calamagrostis* spp). There have been three large wildfires in the study area in 2003, 2009, and 2010, that have greatly altered the wild horse feeding ecology of our study area with the herds spending much more time foraging in the recovering burned areas.

There are four native ungulates in the Brittany Triangle: the mule deer (*Odocoileus hemionus*), moose (*Alces alces*), mountain goat (*Oreamnos americanus*), and California bighorn sheep (*Ovis canadensis californica*). Although wild horse habitat overlaps with some of the seasonal ranges of moose and mule deer, there appears to be little or no overlap with habitats of mountain goats and bighorn sheep. The mountains at the south end of the Brittany plateau represent the northernmost distribution of California bighorn sheep in North America. Some of these ungulate species are important subsistence food for the Xeni Gwet'in. The moose did not arrive in the Chilcotin until the 1920s, a good 140 years or more after the horses appeared to have been brought into the area by the Tsilhqot'in (McCrary 2002).

The study area appears to be unique in North America in that it may be one of the last enclaves of remote wild horse bands in a vast wilderness setting where a complete natural guild of large predators still roams, including the grey wolf (*Canis lupus*), mountain lion (*Felis concolor*), coyote (*Canis latrans*), Canada lynx (*Lynx rufus*), wolverine (*Gulo luscus*), grizzly bear (*Ursus horribilis*), and black bear (*Ursus americanus*). The wolverine and grizzly bear are on the federal endangered species list.

Currently a wolf diet study is underway within the Brittany Triangle to help us understand the feeding ecology of wolf packs, including the role of wild horses in their seasonal diet (Parr 2014).

There are very few roads in the Brittany Triangle; the northern half of the plateau is virtually inaccessible. On the west side, there is a tourism lodge and several small cattle-ranching operations. There are also a number of small private holdings, including “Far Meadow,” which served as a research centre for our work, as well as a 98-hectare (240-acre) nature preserve in Elkin Creek owned by the Valhalla Foundation for Ecology. The nature preserve provides late winter-early spring range for a small number of Brittany plateau wild horses.

2.2 Estimated wild horse distribution in the BC Chilcotin and estimated numbers in the North Brittany Triangle Plateau Study Area

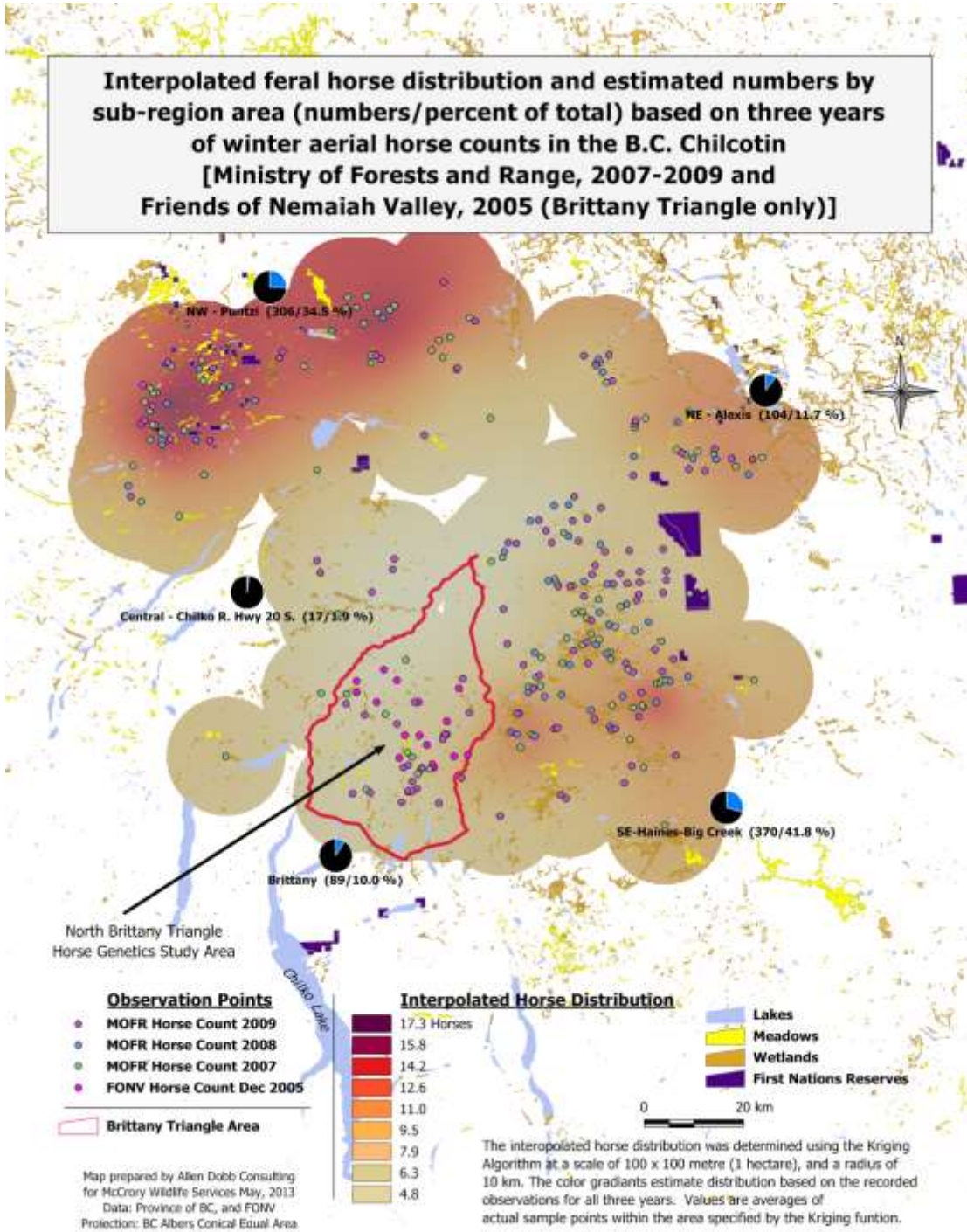
In recent times, attempts have been made to estimate the number of free-ranging horses in the BC Chilcotin. Map 3 shows their approximate range of distribution here based on winter free-ranging horse counts. To create the map, professional agrologist Allen Dobb (pers. comm.) interpolated average winter feral horse numbers for Chilcotin subareas from annual aerial surveys by the provincial Ministry of Forests and Range (MOFR) over three years (2007-2009). For the Brittany Triangle, he also used a winter helicopter count in 2005, done jointly by the Friends of Nemaiah Valley (FONV) and the Xení Gwet'in First Nation (XGFN). The total average number counted by MOFR over the three years for the entire Chilcotin was 886 horses, with about 10% (N=89) from the Brittany sub-population. Given that the counts were done in winter, when any horse movements between survey lines would be constrained, I am assuming these numbers represent “minimal count numbers.”

The Chilcotin minimal number (N = 886) represents the largest population of historically present wild horse bands in British Columbia, if not western Canada, and a small fraction of the numbers that used to exist in the southern grassland ecotones of BC prior to population extirpations sponsored by the provincial government using the bounty system. The last bounty hunt sponsored by the province was on the southeast side of the Brittany Triangle in 1988. Wild horses have avoided this control area since then (McCrary 2002).

For the Brittany Triangle sup-population, minimal counts by the joint FONV-Xeni Gwet'in winter helicopter surveys were 90 in 2001, 80 in 2003 (just after the large wildfire), 121 in 2005, 127 in 2011, and 48 in 2013. As Bhattacharyya (2012) pointed out, the MOFR 2007-2009 winter counts were over an area of about half of the North Brittany Triangle covered by the joint FONV-Xeni Gwet'in helicopter surveys.

What do these minimal aerial counts during the winter period represent in terms of actual population numbers? First of all, there are many potential sampling errors inherent in the aerial survey methods, including weather, lack of visibility of animals in dense forest, whether line transects or random patterns were used, differences between observers, and so on. In addition, without reliable on-the-ground control data to compare more accurate ground counts to aerial-derived numbers, it is difficult to extract from the current database what might be the actual size of the Brittany sup-population. Any “guestimate” should include a wide margin of error. For example, using the joint FONV-XGFN winter counts for 2005 and 2011, Bhattacharyya (2012) subjectively assumed the minimal counts represented approximately 60%-70% of the total population present. She estimated there were 203-215 horses in the Brittany Triangle. However, in February 2013, the joint FONV-XGFN helicopter

count dropped to 48, although less air time was spent compared to previous years. All things considered, in my opinion, a more reliable average range of population estimate that reflects a consistent wide margin of survey error than used by Bhattacharyya, as well as annual fluctuations in winter horse survival and other mortality factors, is between 150 and 215 animals.



Map 3. Distribution of Chilcotin feral horses today, Brittany genetic study area (red), and interpolated horse distribution by sub-area based on winter horse counts (Allen Dobb, pers. comm.).

3.0 METHODS AND APPROACH

Two different types of sampling approaches were used during the duration of the study: blood and hair.

3.1 Blood sample approach

Blood sampling started in February 2003. Detailed instructions were provided by Dr. Gus Cothran, including directions for collection, storage, and shipping. Sampling was done by obtaining permission from local ranchers to take blood samples from horses known to have been captured from wild stock in the Brittany study area. Veterinarian Dr. Colleen Long collected the first five samples from a captive band of horses that were caught from Upper Place in the Brittany Triangle in March 2002. Following this, Dr. Long was not available, so Dr. Dag Goering taught David Williams to obtain blood samples from four other horses. Each horse's age, sex, colouration, capture date and location, and other information was recorded. The total of eight blood samples was considered a very small sample size. The blood samples were analyzed in a preliminary report (Cothran 2006).



Figure 1. Dr. Colleen Long with Ian Bridge preparing to take a blood sample from a captured wild horse from the Brittany Triangle. 2003.

3.2 Hair sample approach

In 2006, on the advice of Dr. Cothran, we decided to switch from collecting blood samples to hair samples because this would assist in obtaining a much larger sample size necessary to draw tentative conclusions about the ancestral bloodlines of the Brittany herds. According to Cothran (2010), using DNA microsatellites from hair samples provides for higher variation levels than blood typing.



Figure 2. Field researchers David Williams and Wayne McCrory collecting wild horse hair samples along horse trail in Nunsti Park in Brittany Triangle.

Dr. Cothran directed our hair collection techniques. This was carried out in the field by biologist Wayne McCrory, graduate student Jonaki Bhattacharyya, and Friends of Nemaiah Valley researcher David Williams. We first explored hair collection in the field by determining if we could actually locate horse hair caught on branches, shed on the ground from molt, and so on. It took some time before we actually found this would work, primarily by very carefully searching on branches under trees where horses sought shelter from winter storms or summer heat. We were careful to not include hair from grizzly bears or range cows and not to duplicate sampling areas from year to year. Field sampling over a number of years ended up being done by collecting one or two stranded hair samples (usually mane hair) from tree branches or tree trunks at each different forested resting site, usually at the edges of feeding meadows. Stranded hair was often located on tree branches near fecal deposits, such as stallion piles, and/or horse trails. Each hair sample of one or more strands was placed in a zip-lock plastic bag and labeled according to site, date, and colour. A total of 62 hair samples were collected in June and September 2006 from Nunsti Provincial Park, and 62 hair samples were sent to Dr. Cothran's lab at Texas A & M University. Following a preliminary analysis (Cothran 2007), we were encouraged to obtain a larger sample size. As additional funding was not obtained until late in 2008, we commenced more sampling in 2009-2010, with a focus on more remote horse bands north and west of Nunsti park. In fall 2010, 53 additional hair samples were submitted for analysis, for a total of 115 samples. After some sorting in the lab, a final number of 99 samples was considered acceptable for the analysis.

3.3 Laboratory analysis

Of the 99 hair samples submitted, only 83 could be used for the estimation of variability. A total of 12 autosomal microsatellite loci (AHT4, AHT5, ASB2, ASB17, ASB23, HMS3, HMS6, HMS7, HTG4, HTG10, LEX33, and VHL20) were examined using the methods described in Juras et al. (2003) and Vega-Pla et al. (2006). The X-linked LEX33 also was typed but not used in the analyses. This specific set of highly polymorphic loci maximizes the number of breeds available to be used for comparison in this study. All 12 loci are included in the Food and Agriculture Organization/International Society of Animal Genetics (FAO/ISAG) Measurement of Domestic Animal Diversity (MoDAD) panel (Hoffmann et al. 2004). Forty random samples were chosen for mitochondrial DNA (MtDNA) sequencing using procedures described in Cothran et al. (2005) and, of these, we were able to obtain reliable sequences from 32.



Figure 3. 2010 expedition to collect wild horse hair at the remote north end of the Brittany Triangle with research assistant Jon Huiziga. To gain access, the river at the confluence of the Dasiqox-Taseko River (milky) and Chilko River (dark blue) had to be crossed with an inflatable raft and motor. There was little evidence of horses and no hair was collected. The more remote apex of the Triangle has yet to be sampled.

3.3.1 Statistical analysis

Gene diversity was calculated using ARLEQUIN 3.1 (Excoffier et al. 2005). An AMOVA (analysis of molecular variance) was also calculated with ARLEQUIN 3.1. F-statistics, deviation from Hardy-Weinberg equilibrium, and allele frequencies were calculated with FSTAT 2.9.3.2 (Goudet 1995). There was no evidence of linkage disequilibrium, so loci were treated individually. The presence of genetic bottleneck in the population was tested for under the Luikart model using BOTTLENECK 1.2.02 (Cornuet and Luikart 1986). Restricted maximum likelihood (RML) trees based on chord distance were generated from 1000 bootstrapped allele frequency datasets using the CONTML procedure of PHYLIP 3.69 (Felsenstein 2005). Restricted maximum likelihood trees were chosen because they have been shown to generate clades consistent with historical knowledge of horse breeds (Cothran and Luis 2005). Majority-rule consensus trees were created with the CONSENSE procedure PHYLIP 3.69. Trees were visualized with Archaeopteryx version 0.957 beta (Han & Zmasek 2009). Two different trees were generated. The first (Figure 4) had 68 domestic breeds in addition to the Brittany Triangle (BT) population and with the Przewalski Horse used as an outgroup. The second tree (Figure 5) included this same dataset, plus data from 43 feral horse populations from the northwestern US, plus one herd from Alberta, Canada.

The data were also examined using STRUCTURE 2.1 (Pritchard et al. 2000), which assigns samples to clusters using a Bayesian method. Burn-in was set to 20,000, and 100,000 MCMC repetitions were used as recommended by the software developer (Falush et al. 2007). Runs for each different selected value of K (the number of clusters) were repeated ten times. CLUMPP (Jakobsson and Rosenberg 2007) was used to concatenate the data from the multiple runs for each K, and DISTRUCT (Rosenberg 2004) was used to graphically display the results. A factorial correspondence analysis was carried out using GENETIX version 4.05 to further examine the relationship of the five populations studied here to other breeds with which they may share ancestry (Belkhir et al. 1996-2004). GENECLASS 2.0 (Piry et al. 2004) was used for individual assignment to predefined populations using a Bayesian algorithm developed by Rannala and Mountain (1997). This model primarily examines the effects of admixture in a population due to immigration rather than genetic drift or mutation.

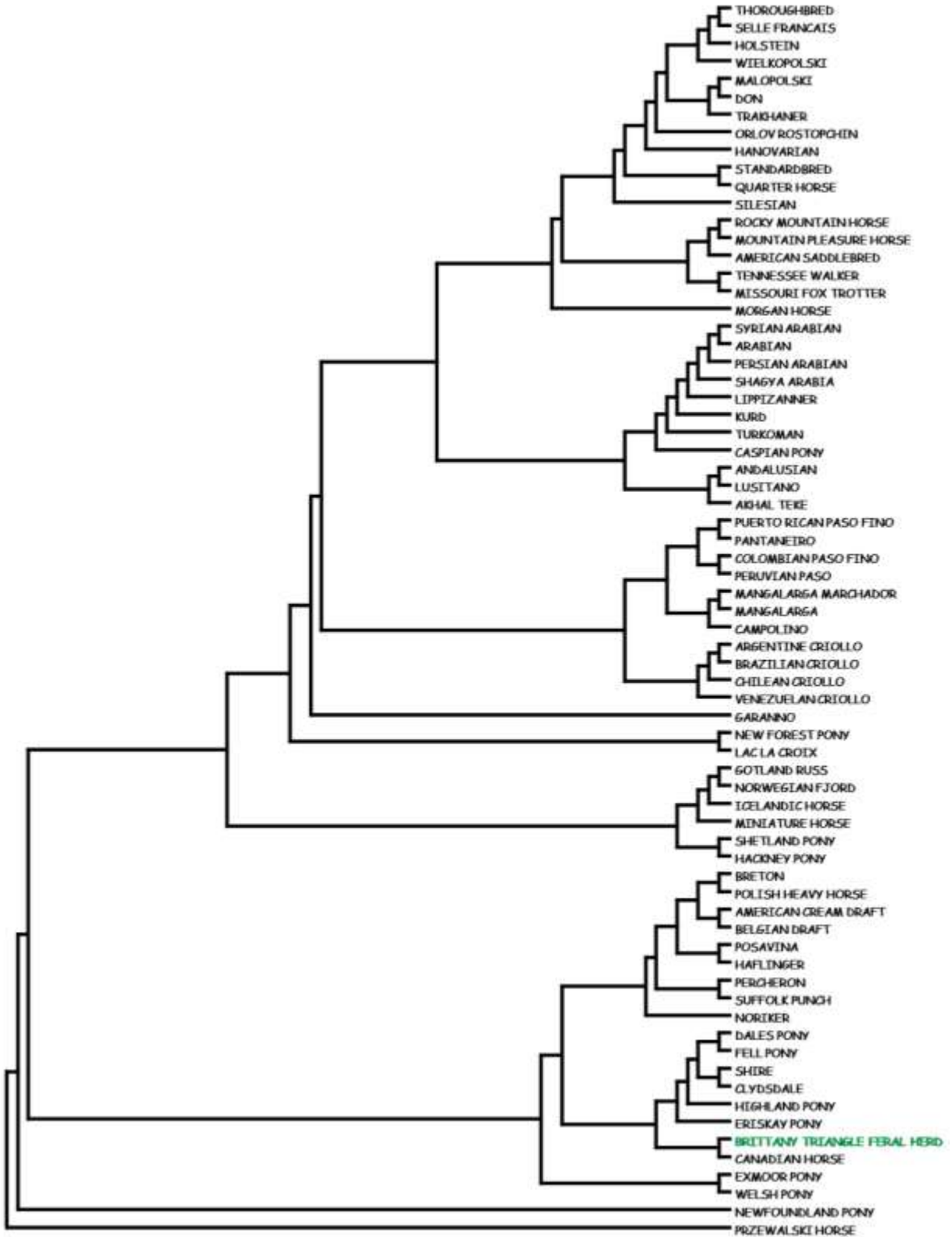


Figure 4. Likelihood tree showing Brittany Triangle Feral Herd (green) in relation to 68 domestic breeds and with the Przewalski Horse used as an outgroup.

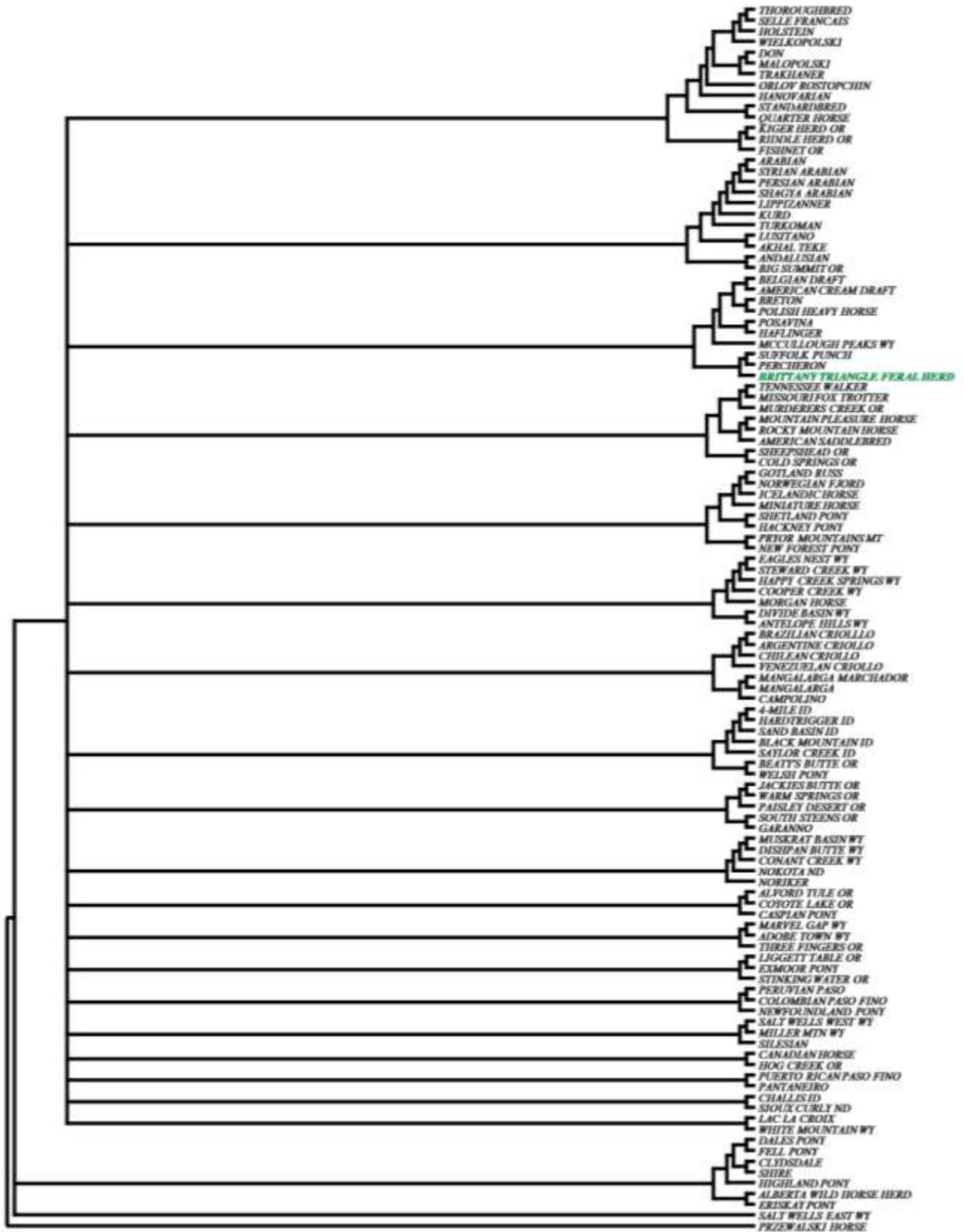


Figure 5. Likelihood tree showing the same dataset as in Figure 4, plus data from 43 feral horse populations from the northwestern US, plus one herd from Alberta, Canada.

4.0 RESULTS

4.1 Field sampling constraints

Eight blood samples and 99 successful hair samples were collected for this study. The small number of captive Brittany horses that were available in the local ranching community limited our 2003-2004 blood-sampling program. As a result, we entertained a proposal by a graduate student to round up wild horses in the Brittany with the help of the Xeni Gwet'in in order to obtain a suitable number of blood samples and complete the genetics study as a master's thesis. However, we felt that a round-up for this purpose would be too expensive and controversial.

When we switched to stranded hair collection, we attempted to spread our sampling over a large portion of a study area in order to collect hair from as many different individual horses as possible. However, some duplication could not be avoided. Sampling was also constrained by limited funds and three large wildfires between 2003 and 2009 that burned much of the study area. In fall 2010, we attempted to sample the very remote north end of the burnt Brittany Triangle by crossing the Chilko River in a motorized inflatable, but found no evidence of wild horse, likely due to the 2009 fire that swept the area.

4.2 Genetic results

Although it is still being strongly debated whether these horses should be managed as livestock, wildlife, or just as wild/feral horses, our genetic testing of the Brittany sup-population meets the requirements of the 2005 Convention of Biological Diversity for livestock (Secretariat of the Convention on Biological Diversity 2005).

Of the 32 mtDNA sequences obtained, only three haplotypes were found. Twelve of the haplotypes belonged to Group A as defined by Jansen et al. (2002). The second haplotype was seen in 17 samples (53.1%) and this represented Haplogroup D. The third haplotype (also Haplogroup A) was only seen in three samples and all of these were from the Brittany Creek area on the west side of the study area. A total of 13 samples were collected from this area, but only four had testable DNA. The 4th animal had haplotype 1. Haplotypes 1 and 2 were found in all other areas sampled to date.

Levels of genetic heterozygosity within the Brittany Triangle population were below mean values for both domestic horse breeds and other feral horse populations but well within the range of variation that has been seen within each group (Table 1). Expected heterozygosity is slightly above observed level but not at a significant level. The number of alleles (*TNV*) and mean number of alleles (*MNA*) are between the feral and domestic horse means. However, effective number of alleles (*A_e*) is low and the number of alleles with a frequency below 5% (*R_a*) is high. The allelic variation numbers are partly a result of a relatively large sample size and possible diversity of origins of the horses (see Discussion).

In a comparison with 69 different horse breeds by phylogenetic analysis (Figure 4), the Brittany Triangle horse paired with the Canadian Horse breed within the cluster that includes the Shire, Clydesdale, Highland Pony, Eriskay Pony, Fell Pony, and Dales Pony. This is a natural cluster of the domestic breeds that are native British breeds, except for the Canadian. The preliminary analysis of the blood typing results indicated some link of the Brittany horses with Spanish horses. There is no indication of relationship with Spanish breeds with the RML analysis of microsatellite data.

Table 1. Breed list Nei data

1	HL	HIGHLAND PONY	9HL
2	AT	AKHAL TEKE	65AT
3	AC	ARGENTINE CRIOLLO	25AC
4	WP	WELSH PONY	35WP
5	SH	SHIRE	41SH
6	RP	PUERTO RICAN PASO FINO	53RP
7	AN	ANDALUSIAN	17AN
8	AR	ARABIAN	5AR
9	CS	CASPIAN PONY	1CS
10	FL	FELL PONY	16FL
11	HF	HAFLINGER	24HF
12	HA	HANOVERIAN	13HA
13	HO	HOLSTEIN	12HO
14	LU	LUSITANO	71LU
15	QH	QUARTER HORSE	27QH
16	PP	PERUVIAN PASO	25PP
17	ST	STANDARD BRED	9ST
18	CF	COLOMBIAN PASO FINO	40CF
19	CP	CAMPOLINA	59CP
20	CC	CHILEAN CRIOLLO	42CC
21	PN	PANTANEIRO	43PN
22	RM	ROCKY MOUNTAIN HORSE	6RM
23	DL	DALES PONY	4DL
24	EX	EXMOOR PONY	44EX
25	GR	GARRANO	45GR
26	SP	SHETLAND PONY	8SP
27	SB	AMERICAN SADDLEBRED	7SB
28	MM	MANGALARGA MARCHADORE	59MM
29	SF	SELLE FRANCAIS	14SF
30	SU	SUFFOLK PUNCH	15SU
31	BZ	BRAZILIAN CRIOLLO	54BZ
32	CA	CANADIAN HORSE	14CA
33	TK	TRAKHNER	76TK
34	VC	VENEZUELAN CRIOLLO	57VC
35	SI	SILESIAN	46SI
36	OR	ORLOV ROSTOPCHIN	20OR
37	KU	KURD	68KU
38	DN	DON	39DN
39	GT	GOTLAND PONY	5GT
40	MK	MALOPOLSKI	46MK
41	PH	POLISH HEAVY HORSE	47PH
42	WI	WEILKOPOLSKI	48WI
43	IC	ICELANDIC HORSE	49IC
44	NK	NORIKER	10NK
45	TU	TURKOMAN	52TU
46	FJ	NORWEGIAN FJORD	67FJ
47	NW	NEWFOUNDLAND PONY	34NW
48	ER	ERISKAY PONY	77ER
49	KA	PERSIAN ARABIAN	3PE
50	BR	BRETON	17BR
51	CD	AMERICAN CREAM DRAFT	21CD

52	FT	MISSOURI FOX TROTTER	22FT
53	PV	POSAVINA	23PV
54	PC	PERCHERON	5PC
55	MP	MOUNTAIN PLEASURE HORSE	30MP
56	CL	CLYDESDALE	30CL
57	HP	HACKNEY PONY	31HP
58	MA	MANGALARGA	33MA
59	LL	LAC LA CROIX	33LL
60	PZ	PRZEWALSKI	
61	MH	MORGAN HORSE	12MH
62	MN	MINIATURE HORSE	38MN
63	HH	HACKNEY HORSE	40HH
64	TB	THOROUGHBRED	10TB
65	BE	BELGIAN DRAFT	50BE
66	TW	TENNESSEE WALKER	59TW
67	LI	LIPPIZANNER	5LI
68	SY	ARABIAN	
69	NP	NEW FOREST PONY	11NP
70	SA	SHAGYA ARABIAN	24SA
71	nv	BRITTANY TRIANGLE	

The breeds are classified into major groups based upon known relationships and phylogenetic analyses done previously. The lowest mean Da distance to group was 0.231 to group one, which is the Thoroughbred and other breeds closely related to the Thoroughbred. The next lowest mean Da was group 2 (0.233), which is the Oriental (Arabian) breeds. The Brittany herd had paired with the Turkoman breed of this group in the blood typing tree. The Spanish horses (group 3) and the North American breeds (group 4) were next with mean Da of 0.248 and 0.280, respectively. The cold blood horses (draft and pony breeds, group 5) had the greatest mean Da , 0.285. Thus the group the Brittany herd clusters with is the group they show the least relationship with based upon Da distance. The RML analysis uses the *Chord* distance measure, but this should have little impact on the structure of the tree. It is not clear why the Brittany herd clusters where it does but it may be due to reduced variability and low allelic diversity. The fit within the cluster was not particularly good with a bootstrap value of 20 out of 100. However, due to the close relationship among horse breeds, bootstrap values tend to be low (Cothran and Luis 2005), even though the trees make sense based upon known relationships and groups are consistent even though the arrangement of populations within groups changes.

In comparison to feral populations and domestic breeds combined, the position of the Brittany Triangle population changes but it still clusters within a group of Draft horses (Figure 5), just not the same ones. This cluster includes the Suffolk Punch, Percheron (these two are closest to the Brittany population), and the Belgian Draft. The heavy horses always tend to form two groups, one with the above breeds and the other with the Shire and Clydesdale as in Figure 4 (see Figure 3 of Bomcke et al. 2011). The Canadian Horse no longer clusters near the BT population and does not fit close to any domestic breeds.

An UPGMA tree using Da distance also was drawn (results not shown). In this tree, the Brittany herd clustered with the Yakut (a breed from Siberia, Russia) in a position that is between the Polish Primitive Horse and all other breeds. This indicates no close relationship to any of the breeds in this

study. This type of result is frequently seen when looking at feral horse herds, especially those with low diversity. It should be noted that the Yakut also has low genetic diversity. The qualitative assessment of the alleles present in the small sample of Brittany horses that were blood typed does suggest some Spanish ancestry as the *Trf-A* allele is most strongly associated with Spanish-derived breeds and is extremely rare in domestic breeds in North America. The presence of *Est-R* is also suggestive of Spanish heritage; however, this allele is found in some North American saddle breeds as well as cold blood horses. With such a small sample size it is not possible to make any strong conclusions.

5.0 DISCUSSION

5.1 Genetic Interpretation

The Brittany Triangle feral horse herd shows low genetic diversity at both the mtDNA and nuclear microsatellite level. The presence of only three mtDNA haplotypes is indicative of low maternal diversity and likely represents a limited number of founders of the population. The rarity and limited geographic distribution of haplotype 3 suggest that this may represent a more recent addition to the herd that was not part of the original founding herd. The three individuals that have this haplotype are somewhat distinct from the rest of the herd at the microsatellite level, while the fourth individual from this area which had haplotype 1 is more like the other horses of the herd, although it does show closest association with the rest of the population based upon assignment using the program WHICHRUN (Banks and Eichert 2000); this analysis will be discussed in more detail below.

Genetic variation based upon microsatellites also is low. Both H_o and H_e values are below the mean values of other feral horse herds and domestic breeds (Table 1). The variability measures do suggest that the population has existed in isolation, at least in the recent past. Because loss of variation over time is highly correlated with population size, it is difficult to estimate the time period of isolation without a good estimate of population size. The key measures that indicate loss of variation are those associated with allelic variation. The number of alleles is a better indicator of genetic diversity for microsatellite loci than is heterozygosity because heterozygosity of microsatellites is highly dependent upon number of alleles and does not decrease unless allele number decreases. However, allelic variation also could be related to founder population size. If the number of founders was small, then the expected number of alleles would also be small. In either case, the results are indicative of isolation. Also indicative of isolation is the great degree of similarity among individuals. There is no indication of specific subgroups within the herd based upon cluster analysis of individuals, with the possible exception of the haplotype 3 horses. Horses such as these, plus a very small number of other horses that do not fit the BT group well based upon WHICHRUN, may be recent immigrants and could have inflated the TVN and MNA values and may be why H_e is higher than H_o .

Phylogenetic analysis clearly places the BT population with the heavy horse breeds group, but specific relationships are not clear. There is no evidence of any relationship to the feral horse populations that we have data for that are geographically closest to the area (Figure 5). In Figure 4, the BT population is closest to the Canadian Horse breed, which appears to be a reasonable candidate for the type of horse that could have founded the population. The Canadian Horse was an important means of transportation in the late 19th and early 20th centuries in Canada (Hendricks 1995), and numbers of

this now rare breed were much greater at that time. We are examining the genetic origins of this breed in ongoing work, but preliminary analyses do show association with the heavy horse breeds. This is consistent with previous but more limited work (Behara et al. 1998; Plante et al. 2007; Prystupa et al. 2011).

One interesting observation that must be examined further was an observed association of the Brittany Triangle (BT) with the Yakut horse, which is a breed from Siberia, Russia. An early analysis of breed relationships of the BT population to a group of about 50 breeds showed the BT paired in a cluster with the Yakut (data not shown but available upon request). We also analyzed the BT population using WHICHRUN in order to see if there were any individuals that did not fit well with the other individuals of the population. WHICHRUN works by using the allele frequency distributions of the multilocus genotypic data and implementing a maximum likelihood approach, breed allocation of individuals, and the certainty of these allocations were estimated using WHICHRUN 4.1 (Banks and Eichert 2000). Incorporating jackknife iterations, this procedure samples individuals one at a time and recalculates the allele frequency in the absence of each genotype before determining the most likely source population of the particular individual. To resolve the stringency of an allocation, WHICHRUN utilizes the log of the odds (LOD) ratio for the two most likely source populations. By restricting breed allocation to apply only for assignments that have a LOD ratio of at least two, a particular assignment will have a 1/100 chance of error, or less (Bjornstad and Roed 2001). The results showed that the Yakut was the breed that the BT horses were most often assigned to after the Pony and Draft breeds, which is the group the BT population clusters with in Figure 4. This raises the possibility that horses from Russia may have contributed to the feral herds at some time in the past, which is not outside the range of possibilities. More analyses need to be done.

Based upon the phylogenetic results, 17 breeds, plus the outgroup Przewalski Horse, were compared to the BT population in the STRUCTURE analysis. Out of the 19 populations, the analysis revealed that the optimum number of clusters was 15. The BT did form a distinct cluster at $k=15$, but the Standardbred and Morgan Horse, the Arabian and Akhal Teke, and the Percheron and Suffolk were not well differentiated. The Yakut showed substructure but also showed some association with the BT population.

The results of this study show that overall diversity within the Brittany Triangle feral horse population is on the low side for horses in general. There are likely a few individuals in this herd that have entered into this population in recent times and have inflated the diversity levels, and if these individuals were not considered, variability would be even lower. The population does appear to have been isolated, although not completely. More work will be needed to fully understand the distribution of variation and how this relates to the demographic history of the population.

The origins of the herd are largely from the heavy horse types and specifically it appears that the Canadian Horse breed (or its ancestors) contributed significantly to the ancestry of the BT population.

There also is the intriguing possibility that Yakut horses of Russian heritage contributed to the origins of the herd but this requires more study and probably more samples from Eastern Russia.

5.2 Implications of genetic results, historic and present context

Our genetic study raises more questions than it answers, but it does dispel our earlier tentative results from blood samples and the hypothesis that the Brittany horses today still have Spanish ancestry. The results, however, do not disprove the considerable historic evidence that the Tsilhqot'in had horses pre-contact as early as 1740, and the hypothesis that these likely came from Spanish horses to the south. If this hypothesis is true, our study indicates that the genetic make-up of today's wild horses in the Brittany Triangle has undergone considerable transformation since these earlier times, unlike some New World sub-populations that still maintain characteristics ascribed to their Spanish ancestry (Conant et al. 2011). Or, alternatively, the original foundation herds in the Brittany were actually the Canadian and Yakut and not Spanish.

According to Sponenberg (1999), most feral horse herds in the U.S. today, including "mustangs" managed in horse reserves by the Bureau of Land Management (BLM), are cross-breeds with non-Spanish horses. Only four remnant herds have been identified with Spanish bloodlines in the U.S. (Sponenberg 1999), including the Pryor Mountain Wild Horse Range (Cothran 2010).

Since the first European contact in 1808 and subsequent early Hudson's Bay influences, many different Old World horse breeds have been brought into the Chilcotin, including draft horses for agricultural purposes. Over the past hundred years or so, some Xeni Gwet'in have also brought in different breeds to improve their chances of winning the very competitive and popular mountain races that continue to this day (Rocky Quilt, pers. comm.; Harry Setah, pers. comm.). Cattle ranching started in the region in the late 1800s and some ranchers deliberately brought in stud horses of different breeds, turning some loose with wild horses, hypothetically to improve wild stock (Harry Setah, pers. comm.). For a long time and continuing through until today, when they run out of winter hay to maintain their horses, some of them mixed breeds, some Xeni Gwet'in families traditionally turn them loose to free-range in the wilds (Roger Williams, pers. comm.). Some of these are not captured and run with established free-ranging herds. Another possible infusion would have been the thousands of domestic horses of different breeds that escaped or were turned loose to fend for themselves during the Cariboo gold rush in the 1860s. A letter to the *Victoria Times-Colonist* dated May 3, 1863, refers to the countless dead horses in the Cariboo on the gold rush trail to Barkerville as a result of packers leaving them to overwinter on their own. Some of these horses also survived in the wilds (McFadden 1965).

That the current horse sub-population in the Brittany Triangle is of average size is consistent with our genetic findings of the heavy horse type-Canadian Horse breed. In 2002, the Canadian Horse was officially deemed by Parliament to be the National Horse of Canada. The Canadian Horse ancestry in the Brittany is certainly intriguing and requires much more historical research as to genetic pathways into the area. This is beyond the scope of our current review.

As an interesting anecdotal note, Figure 6 shows a black stallion in the Brittany Triangle that a member of the Canadian Horse Heritage and Preservation Society strongly insisted was a Canadian Horse when he saw the same picture in a news article in 2002 (Wayne McCrory, pers. comm.).



Figure 6. Medium-sized adult black stallion in spring habitat in the Brittany Triangle, 2002. Photo: Wayne McCrory

Although further research is needed regarding the Yakut influence, we found very limited evidence to support the most obvious hypothesis that the Yakut horse arrived as foundation bloodlines in the remote Brittany Triangle from Russian fur expeditions on the BC coast during trade with interior First Nations that traveled to the coast. Russians began exploring the Pacific coast of North America in the eighteenth century and, after 1799, this was expanded with the chartering of an imperial monopoly, the Russian-American Company, to which was assigned the control of exploitation of what was then considered the Russian possessions in America. This continued until 1867, when Russia surrendered its territory to the USA (Dall et al. 1902).

Since the Brittany study area is within the lee of the Coast Range, the Tsilhqot'in historically traded with First Nations on the adjacent Pacific coast via well-established trade trails over mountain passes in the Coast Range. These traditional interior-coast trails were often known as "grease trails," referring to trade involving the much sought-after oolichan fish oil produced by coastal First Nations. However, although horses may have been present with the Xeni Gwet'in as early as 1740, it was not until more recent times that they were used to travel to the coast. For example, in 1793, when fur trader Alexander McKenzie travelled from the interior Fraser River to coastal Bella Coola, he went on foot along a grease trail and observed no horses (MacKenzie 1801). The Nuxalk-Carrier First Nations grease trail he followed was somewhat to the north of the Brittany Triangle. In 1845, when the Jesuit priest Nobili visited a Xeni Gwet'in winter village near the outlet of Chilko Lake in the Brittany Triangle, he reported that although they had horses, they did not use them to trade at the coast (Nobili, G. 1845-1846). Additionally, research by Thomas (1989) showed that ship logs kept by the Russian fur

traders made no mention of the transport of horses from Russia to the west coast of North America. However, Thomas's research showed that some horses were used by Russians in settlements in present day Alaska on a limited basis during the late 1700s and early 1800s. For example, he notes that in 1817 there were only 16 horses in Russian America and they were more than likely "the hardy Yakut."

The only reference I could find regarding the presence of smaller wild horses extant in the region that could match the diminished size of the Yakut was in a recent interview with Xeni Gwet'in knowledge-keeper and researcher Norman William. William referred to large piles of wild horse bones in the Anvil Mountain area in the upper Dasiqox-Taseko watershed (to the south of the Brittany study area) where the Forest Service had a "bounty" slaughter of the last wild horses there (a wild horse bounty was enacted in 1924 and continued to about 1988). William maintains these horses were small "Shetland"-sized ponies, but in a related interview, his sister Alice William said they were somewhat larger—the size of Arabian horses (McCrary 2014).

Obviously, much more genetic and historic research is needed on the possible origins of the intriguing Canadian and Yakut horse DNA discovered in today's Brittany Triangle wild horses.

Efforts should also be made to maintain this apparent genetic uniqueness.



Figure 7. Wild horses in spring time to the east of the Brittany Triangle. Photo: Wayne McCrary

6.0 LITERATURE CITED

- Bhattacharyya, J. 2012. Knowing Našlhiny (Horse), Understanding the Land: Free-Roaming Horses in the Culture and Ecology of the Brittany Triangle and Nemiah Valley. Doctoral Dissertation, School of Planning, University of Waterloo, ON.
- Banks, M.A., and W. Eichert. 2000. WHICHRUN (version 3.2): a computer program for population assignment of individuals based on multilocus genotype data. *J. Hered.* 91:87–89.
- BC Commission on Resources and Environment. 1994. Cariboo-Chilcotin Land Use Plan. 237 pp.
- Behara, A.M.P., D.T. Colling, E.G. Cothran, and J.P. Gibson. 1998. Genetic relationships between horse breeds based on microsatellite data: applications for livestock conservation. *Proceedings of the 6th World Congress on Genetics Applied to Livestock Production* 28:119–22.
- Belkhir, K., P. Borsa, L. Chikhi, N. Raufaste, and F. Bonhomme. 1996-2004 GENETIX 4.05, logiciel sous Windows TM pour la génétique des populations. Laboratoire Génome, Populations, Interactions, CNRS UMR 5000, Université de Montpellier II, Montpellier (France).
- Bjørnstad, G., and K.H. Røed. 2002. Evaluation of factors affecting individual assignment precision using microsatellite data from horse breeds and simulated breed crosses. *Animal Genetics* 33:264–70.
- Bömcke, E., H. Soyeurt, M. Szydlowski, and N. Gengle. 2011. New method to combine molecular and pedigree relationships. *Journal of animal science* 89(4):972–8. doi:10.2527/jas.2010-3135
- Conant, E.K., R. Juras, and E.G. Cothran. 2011. A microsatellite analysis of five Colonial Spanish horse populations of the southeastern United States. *Animal Genetics* 43 (1):53-62.
- Cornuet, J.M., and G. Luikart. 1996. Description and power analysis of two tests for detecting recent population bottlenecks from allele frequency data. *Genetics* 144:2001–2014.
- Cothran, E.G. 2006. Preliminary analysis of the Brittany Triangle feral horse population. 6 pp.
- Cothran, E.G. 2007. Genetic Analysis of feral horses from the Brittany Triangle area (B.C., Canada).
- Cothran, E.G. 2010. Genetic analysis of the Pryor Mountains HMA, WY.
- Cothran, E.G. and C. Luis. 2005. Genetic distance as a tool in the conservation of rare horse breeds. *Eaap Public* 2005(116):55-71.
- Cothran, E.G., R. Juras, and V. Macijauskienė. 2005. Mitochondrial DNA D-loop sequence variation among 5 maternal lines of the Zemaitukai horse breed. *Genet. Mol. Biol.* 28:677–681.
- Cowdrey, M., Martin, J., and N. Martin. 2012. *Horses and Bridles of the American Indians*. Hawk Hill Press. Nicasio, California. 219 pp. Illustr.
- Dall, W.H., C. Keeler, H. Gannett, W.H. Brewer, C. Hart Merriam, G.B. Grinnell, and M.L. Washburn. 1901. *Harriman Alaska Expedition. Alaska Volume II. History, geography, resources*. Doubleday, Page and Company. New York.
- Dudley, N. (ed.). 2008. *Guidelines for Applying Protected Areas Management Categories*. IUCN: Gland, Switzerland. p.8-9. www.unep-wcmc.org/about-protected-areas_163.html. Accessed March 16, 2014.
- Excoffier, L., G. Laval, and S. Schneider. 2005. Arlequin ver. 3.0: An integrated software package for population genetics data analysis. *Evol Bioinform* 1:47–50
- Falush D., M. Stephens, and J.K. Pritchard. 2007. Inference of population structure using multilocus genotype data: dominant markers and null allele. *Molecular Ecology Notes* 7:574-578.
- Felsenstein, J. 2005. PHYLIP (Phylogeny Inference Package) version 3.6. Distributed by the author. Department of Genome Sciences, University of Washington, Seattle. or Felsenstein, J. 1989. PHYLIP - Phylogeny Inference Package (Version 3.2). *Cladistics* 5:164-166.

- Food and Agriculture Organization (FAO) 2007. The State of the World's Animal Genetic Resources for Food and Agriculture (eds. B. Rischkowsky and D. Pilling). FAO, Rome.
[tp://ftp.fao.org/docrep/fao/010/a1250e/a1250e.pdf](http://ftp.fao.org/docrep/fao/010/a1250e/a1250e.pdf). Accessed April 8, 2014.
- Goudet J. 1995. FSTAT Version 1.2: a computer program to calculate F-statistics. *J. Heredity* 86: 485-486.
- Han, M.V., and C.M. Zmasek. 2009. PhyloXML: XML for evolutionary biology and comparative genomics. *BMC Bioinformatics* 10:356.
- Hendricks, B.L. 1995. *International Encyclopedia of Horse Breeds*. Norman, OK, USA: University of Oklahoma Press.
- Hoffmann I., P. Ajmone Marsan, S.F. Barker, E.G. Cothran, O. Hanotte, J.A. Lenstra, D. Milan, S. Weigend, and H. Simianer. 2004. New MoDaD marker sets to be used in diversity studies for the major farm animal species: recommendations of a joint ISAG/FAO working group. *Proceedings of 29th International Conference on Animal Genetics*. Tokyo, Japan. 107pp.
- Jakobsson, M., and N.A. Rosenberg. 2007. CLUMPP: A cluster matching and permutation program for dealing with label switching and multimodality in analysis of population structure. *Bioinformatics* 23: 1801–1806.
- Jansen T., P. Forster, M.A. Levine, H. Oelke, M. Hurles, C. Renfrew, J. Weber, and K. Olek. 2002. Mitochondrial DNA and the origins of the domestic horse. *Natl. Acad. Sci. USA* 99:10905–10910.
- Juras, R., and E.G. Cothran. 2004. Microsatellites in Lithuanian native horse breeds: usefulness for parentage testing. *Biologija* 4:6–9.
- Lamb, W.K. 1966. *Simon Fraser: Letters and Journals, 1806 – 1808*. The Macmillan Company of Canada Ltd., Toronto.
- Mackenzie, A. 1801. *Voyages from Montreal on the River St. Laurence through the Continent of North America to the Frozen and Pacific Oceans in the years 1789 and 1793*. Printed for T. Cadell and Jun. and W. Davies. Strand. Old-Bailey-London. Reprinted as 'Master Works of Canadian Authors'. J. Garvin. 1927.
- McCrary, W. 2002a. Preliminary conservation assessment of the rain shadow wild horse ecosystem, Brittany Triangle, Chilcotin, British Columbia. A review of grizzly and black bears, other wildlife, feral horses, and wild salmon. Report to Friends of Nemaiah Valley.
- McCrary, W. 2002b. Proposal for testing of genetics and historic origins of BC Chilcotin wild horses (Brittany Triangle) - ?Elegesi Qiyus (Nemiah) Wild Horse Preserve: A work in progress. 6 pp.
- McCrary, W.P., A. Williams, L. Smith, B. Cross, and L. Craighead. 2014. Inventory of wildlife, ecological, and landscape connectivity values, Tsilhqot'in First Nations cultural/heritage values & resource conflicts in the Dasiqox-Taseko watershed, BC Chilcotin. Report to Xeni Gwet'in & Yunesit'in First Nations. 158 pp.
- McFadden, L. 1965. Let's stop slaughtering wild horses. *British Columbia Digest*, May-June issue.
- Nobili, Giovanni. 1845-1846. Journals and letters. Vatican archives. Translated into English (with footnotes) by Woodward and Company for *Tsilhqot'in Nation v. British Columbia*. Version *In File*.
- Parr, S. 2014. Pilot study of grey wolf (*Canis lupus*) feeding ecology in the Brittany Triangle and Nemiah Valley Progress Report: May 2013 to February 2014. Report to Valhalla Wilderness Society. 63 pp.
- Piry, S., A. Alapetite, J.M. Cornuet, D. Paetkau, L. Baudouin, and A. Estoup. 2004. GENECLASS2: A software for genetic assignment and first-generation migrant detection. *Journal of Heredity* 95:536–539.
- Plante, Y., J.L. Vega-Pla, Z. Lucas, D. Colling, B. De March, and F. Buchanan. 2007. Genetic diversity in a feral horse population from Sable Island, Canada. *Journal of Heredity* 98:594–602.

- Pritchard, J.K., M. Stephens, and P. Donnelly. 2000. Inference of population structure using multilocus genotype data. *Genetics* 155:945–959.
- Prystupa, J.M., R. Juras, E.G. Cothran, F.C. Buchanan, and Y. Plante. 2012. Genetic diversity and admixture among Canadian, Mountain and Moorland and Nordic pony populations. *Animal* (2012), 6:1, pp 19–30 & The Animal Consortium 2011 doi: 10.1017/S1751731111001212.
- Rannala, B., and J.L. Mountain. 1997. Detecting immigration by using multilocus genotypes. *Proceedings of the National Academy of Sciences, USA* 94:9197–9201.
- Rosenberg, N.A. 2004. DISTRUCT: a program for the graphical display of population structure. *Molecular Ecology Notes* 4:137–138.
- Secretariat of the Convention on Biological Diversity. 2005. Handbook of the Convention on Biological Diversity including its Cartagena Protocol on Biosafety, 3rd edition, (Montreal, Canada). <http://www.cbd.int/doc/handbook/cbd-hb-all-en.pdf>. Accessed August 11, 2014.
- Sponenberg, P. 1999. American Colonial Spanish Horse update. March 1999. Virginia-Maryland Regional College of Veterinary Medicine.
- Supreme Court of Canada (SCC). 2014. Judgments of the Supreme Court of Canada. *Tsilhqot'in Nation v. British Columbia*. Case number 34986. <http://scc-csc.lexum.com/scc-csc/scc-csc/en/item/14246/index.do>. July 12, 2014.
- Thomas, S. 1989. *Myth and Mystery: The Curly Horse in America*. Research project: 1989 Conservancy of Freestone (C.S.) Fund, California.
- Vega-Pla, J.L., J. Calderyn, P.P. Rodriguez-Gallardo, A.M. Martinez, and C. Rico. 2006. Saving feral horse populations: does it really matter? A case study of wild horses from Docana National Park in Southern Spain. *Animal Genetics*, 37, 6:571-578.
- Vickers, J. 2007. *Tsilhqot'in Nation v. British Columbia*. No. BCSC 1700 Registry No. 90-0913 British Columbia Supreme Court 2007. Transcripts: Testimony October 21, 2003; Cross-examination Oct. 18, 2004.
- World Parks Congress. 2003. Recommendation v.26. http://cmsdata.iucn.org/downloads/recommendation_en.pdf. Accessed March 14, 2014.