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A Characterization of West Virginia Coyotes (Canis Latrans) Utilizing Skull Morphology

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A CHARACTERIZATION OF WEST VIRGINIA COYOTES (*CANIS LATRANS*) UTILIZING SKULL MORPHOLOGY

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In partial fulfillment of
the requirements for the degree of
Master of Science
In
Biological Sciences: Organismal, Evolutionary, and Ecological Biology
by
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Approved by
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ABSTRACT

Coyotes (*Canis latrans*) are now found throughout North and Central America, but before European colonization were restricted to west of the Mississippi. Migration occurred in two major paths to the East; north over the Great Lakes (through Canada) and south below the Great Lakes. The location of these routes is significant because those migrating north interbred with the wolves that reside there. These hybrid animals are larger and behaviorally different from their western counterparts. It is possible to differentiate these hybrids morphologically and genetically. Hybrids are known to be located in Maine, New York, and Pennsylvania, but the interest of this study was to determine if their range has spread to include West Virginia. Fourteen measurements were taken by hand using digital calipers on 126 skulls from West Virginia and Ohio and 25 domestic dog skulls. Utilizing PCA, ANOVAs, and multivariate allometry, these data were compared to data collected on coyote populations from western and northeastern North America. Results conclude that while West Virginia coyotes show some similarities to both comparative populations, they are a distinct population with unique morphological variation, and additionally show no similarities to dogs. The distinct morphology of West Virginia coyotes may be due to ecological pressure to adapt that varies from the West and is influenced to lesser degree by admixture with other species than the Northeast.

CHAPTER ONE

INTRODUCTION

The coyote, *Canis latrans* Say (1823), evolved in North America between 500,000 and one million years ago (Nowak, 1978; Tedford, Wang, & Taylor, 2009). While coyotes of the Pleistocene were larger than those in the current day, the extinction of the North American megafauna pushed coyote evolution quickly toward their current size and morphology (Meachen & Samuels, 2012). A wealth of studies have focused on varied aspects of coyotes, including genetics, morphology, habitat selection, diet, and impacts of urbanization, among others (Crête, Ouellet, Tremblay, & Arsenault, 2001; Gese & Grothe, 1995; Hill, Sumner, & Wooding, 1987; Lehman & Wayne, 1991; Poessel, Breck, Teel, Shwif, & Crooks, 2012; Thurber & Peterson, 1991; Timm & Baker, 2007; Way, 2007; Wykle, 1999). Because coyotes have been hunted consistently across North America for decades, it is often relatively easy to access coyote remains (such as skulls) for research.

The Marshall University collection possesses 125 coyote skulls collected in or near West Virginia, collected between 1989 and 2000. A former graduate student at Marshall University, Jennifer Wykle, was responsible for the collection of these specimens via the USDA, WVDNR, trappers, hunters, and taxidermists. Her study was broad, examining distribution, abundance, hybridization with domestic dogs, taxonomy, ecology, and behavior. She performed a few morphological measurements and utilized ratios to compare similarities between West Virginia coyotes and those in the neighboring states of Ohio, Kentucky, and Pennsylvania (Wykle, 1999). Wykle's findings were somewhat inconclusive, however, and she did not explore hybridization with wolves, which is one major implication of coyote migration from the West. Kays, Curtis, and Kirchman (2010) examined via morphology and genetics the adaptive evolution of coyotes in the Northeast due to introgression with wolves, which sparked an interest to perform a more extensive morphological study of the skulls in the Marshall University museum collection (MUMC), using the morphology data collected in Kays et al. (2010) as reference populations of coyotes from across North America to explore similarities and differences among populations.

Fossil Record

The fossil record for canids is extensive, but assigning species and deducing their relatedness to one another has been difficult and hotly contended. The genus *Canis* may have arisen as early as the Turolian, at the end of the Miocene in Europe. *"Canis" cipio* Crusafont-Pairó, 1950, was discovered in Spain and is dated as old as 7-8 million years before present (Bartolini Lucenti, Alba, Rook, Moyà-Solà, & Madurell-Malapeira, 2017). Properly assigning this specimen is difficult, as the entirety of the fossil is only a partial mandible, and given the limited characters it possesses, could instead belong to the extinct genus *Eucyon*. Irrefutable evidence of *Canis* in Europe is not found until the late Pliocene (Bartolini Lucenti et al., 2017).

The canid family (Canidae) has evidence to support its foundation in North America (Pires, Silvestro, & Quental, 2015). The oldest known *Canis* from the North American continent comes from the late Hemphillian (near the Miocene-Pliocene boundary) of Mexico and the west/southwest of the United States. This specimen, *Canis ferox* Miller and Carranza-Castañeda, 1998, persists until the medial Blancan (~3.5 Ma) in the North American fossil record. It possesses characters intermediate between those of *Eucyon davisi* Merriam, 1911, and *Canis lepophagus* Johnston, 1938, but is more similar to the larger *C. lepophagus*. *C. ferox* is thought to be the most primitive form of *Canis,* at least in North America (Tedford et al., 2009). Tedford et. al. 2009 proposed a tree of phyletic relationships for *Canis* based on both stratigraphic and morphologic data (Fig. 1). Branching from the base species, *C. ferox*, gives rise to *C. lepophagus*. At one time, it was proposed that *C. lepophagus* was directly ancestral to the coyote (*Canis latrans*), with one intermediate form between them (Kurtén 1974). Bekoff (2001) suggests that *C. lepophagus* was already too specialized to be a direct ancestor of the coyote, and furthermore that they likely co-existed for some time.

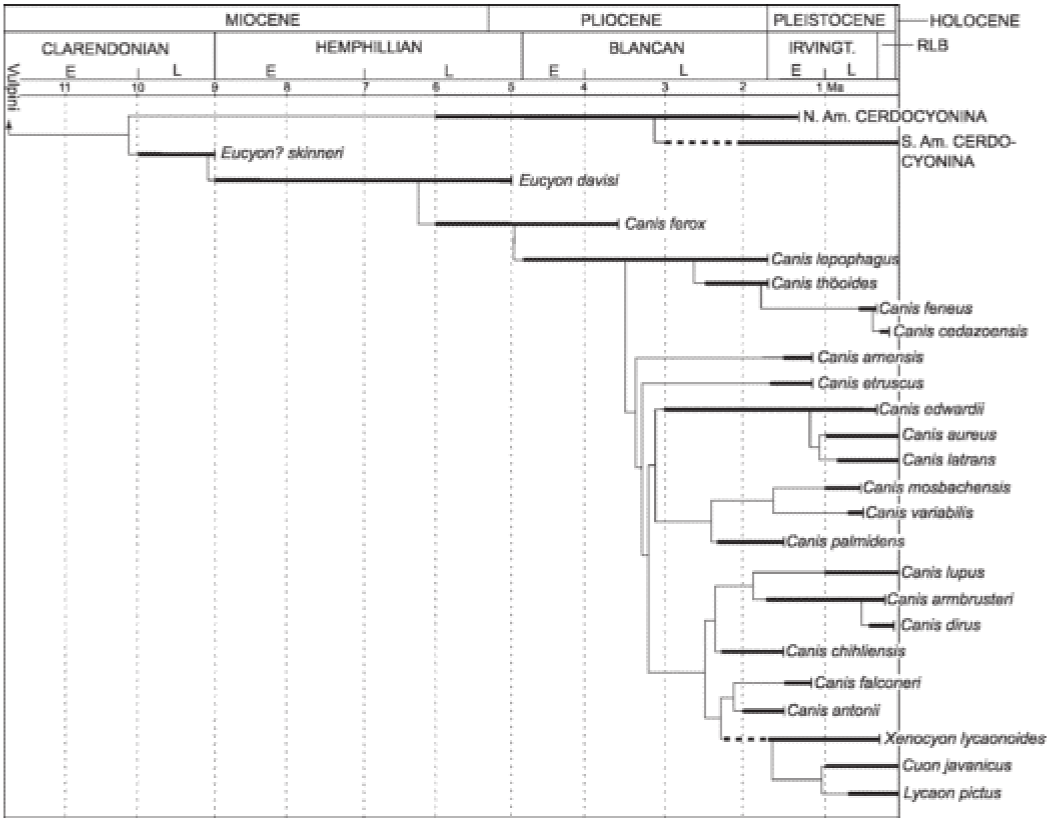


Figure 1. Proposed cladogram with stratigraphic ranges for subfamily Caninae from Tedford et. al. (2009)

The Tedford et al. (2009) tree supports Bekoff (2001), and suggests that a late Blancan branching from *C. lepophagus* initiates the appearance of the jackal-like clade in North America, beginning with *Canis thöoides,* of which only two fossils have been found from the late Blancan of Arizona. The clade continued with *Canis feneus* of the late Irvingtonian in Nebraska, the dentition of which suggests a more mesocarnivorous lifestyle than its predecessors. The clade then terminated with *Canis cedazoensis* Mooser and Dalquest, 1975, from the Rancholabrean of Mexico. *C. cedazoensis* fit well into the jackal-like niche, being an intermediate size between fox and coyote with dentition suggesting a hypercarnivorous lifestyle. Jackals are now limited to southern Eurasia and Africa (Fig. 2).

Figure 2. Current-day range of golden jackal (*Canis aureus*; IUCN, 2008), side-striped jackal (*Canis adustus*; IUCN, 2014a), and black-backed jackal (*Canis mesomelas*; (IUCN, 2014b). Jackals are found throughout areas in orange.

An earlier branching event from *C. lepophagus* began much of the radiation in diversity of *Canis*. Along this path, the European *Canis arnensis* Del Campana, 1913, and *Canis etruscus* Forsyth Major, 1877, share the nearest common ancestors, and arose from branching events that occurred before the mid-late Blancan. Both of these species terminated in the Irvingtonian and neither are directly ancestral to any other *Canis* species. *C. arnensis* has been variably described as either jackal-like or coyote-like since its first characterization, given by Del Campana (1913) (Kurtén, 1974). *Canis etruscus* is considered one of the first true wolf-like canids to appear in Europe. Shortly after the event that gave rise to *C. etruscus*, another branching led to two sets of cascading branches. One set of branches holds the coyote/jackal type canids, while the other leads to the wolf-like canids. The coyote/jackal lineage begins with *Canis edwardii* Gazin, 1942, which dates back further than both *C. arnensis* and *C. etruscus* but is the result of further branching away from each of their lineages, occurring near the mid-late Blancan. The record for *C. edwardii* terminates in the Late Irvingtonian of Texas, Kansas, Nebraska, Oregon, Idaho, Arizona, California, and Florida. Branching from *C. edwardii* in the Early Irvingtonian led to the sister taxa *Canis latrans* (modern-day coyote), and *Canis aureus* Linnaeus, 1758 (Golden/Common Jackal). Both species persist today in geographic isolation from one another. From the same branching event that gave rise to *C. edwardii* came a lineage that branched in the later part of the late Blancan (~2.5 Ma), giving rise to *Canis palmidens* Teilhard de Chardin and Piveteau, 1930 (extinct in Early Irvingtonian). A branch neighboring *C. palmidens* begot *Canis mosbachensi*s Soergel, 1925, and *Canis variabilis* Pei, 1934, around the Pliocene-Pleistocene boundary. *C. palmidens*, *C. mosbachensis*, and *C. variabilis* are present in the Eurasian but not North American fossil record. These three species round out the coyote/jackal type canids (Tedford et al., 2009).

Both *C. mosbachensis* and *C. variabilis* went extinct around the same time in the Late Irvingtonian. The lineage of wolf-like canids began branching around 2.5 Ma. Two separate branches arose after the original branching event. One of these gave rise to *Canis falconeri* Forsythe-Major, 1877, of European origin, and *Canis antonii* Zdansky, 1924, of Asian origin, both of which went extinct in the Early Irvingtonian. To the other belongs *Canis chihliensis* Zdansky, 1924 (extinct around the Pliocene-Pleistocene boundary), and a series of branching from which came *Canis lupus* Linnaeus, 1758, *Canis armbrusteri* Gidley, 1913, and from *C. armbrusteri* comes *Canis dirus* Leidy, 1858. Both *C. armbrusteri* and *C. dirus* became extinct in the Rancholabrean. *Canis dirus* spread throughout North, Central, and South America, while *C. armbrusteri* records in the Rancholabrean are restricted to Florida (Tedford et al., 2009).

Cope's Rule and Hypercarnivory

Of the large diversity that once existed in *Canis*, only three of the species aforementioned are extant: *C. aureus*, *C. latrans,* and *C. lupus*. The decline in diversity of the group can be attributed to a number of factors. According to Cope's rule, there is an evolutionary trend to increasing body size. Cope formulated this rule due to the extreme increase in size of mammals during the Cenozoic (Cope, 1886). Although this trend is not always apparent, Alroy (1998) ran analyses on over 1,500 mammalian species ranging from late in the Cretaceous to the late Pleistocene, comparing relative ages and weight estimates for each species. Over the entire set of data, it was found that novel species were on average 9.1% larger than their assigned predecessor (the more ancestral species). The trend was assessed for change over time, as this increase could have been one anomalous event, but it was discovered that the increase in size actually increased with time, all the way up to a 21% increase in size as the lineages progressed. As size cannot continue to increase to infinity, an upper limit must exist. Once reaching that upper limit, the lineages plateau but are much less likely to regress (Alroy, 1998). A tendency to get larger over time is usually advantageous in that it reduces the chance of being predated upon and can increase the chance of successfully subduing prey (if the species is predatory), as well as increased longevity of individuals of the species.

The size limit of certain lineages depends heavily upon their ecology, and for completely terrestrial species, the ability to form limbs strong enough to constantly support their weight (Stanley, 1973). For canids living in the Cenozoic, growing to larger sizes than seen in present times was possible due to the physically larger prey base. As some canid species progressively grew larger, their adaptations for hypercarnivory grew with them (Van Valkenburgh, Wang, & Damuth, 2004). Hypercarnivores are defined as an intake of greater than 70% of a total diet in meat, while mesocarnivores consume less than 70% but greater than 50%, and hypocarnivores less than 50% meat (Van Valkenburgh, 2007). These adaptations are marked by greatly reduced grinding surfaces of molars, a lengthening of the shearing surfaces of carnassials, jaws that are more deeply set, and lengthening of the canines. Dental characteristics such as these provide species with the ability to take down larger prey more efficiently (deep jaws, large canines) and process more meat (greater shearing carnassials, reduction in grinding surfaces). The move from hypo- or mesocarnivory toward hypercarnivory indicates a move from a more generalized species to one that is specialized (Van Valkenburgh et al., 2004). Because of the abundance of large herbivorous species in North America during the Cenozoic, hypercarnivory was sustainable for co-occurring carnivores. With the drastic decline in herbivore prey base came the decline of the large-bodied, hypercarnivorous predators, such as the dire wolf (Van Valkenburgh, 1988).

Another factor that could contribute to the decline in diversity of canids is that an increase in body size is negatively correlated with population density. Larger species logically require a greater amount of resources relative to smaller species, so populations are likely to be less dense for larger species. This decrease in density will likely equate to a decrease in the probability of finding a mate and additionally a decrease in overall number of species. All these factors combined seem to put larger, more specialized species at a higher risk for extinction as compared to the generalized, smaller species (Cardillo et al., 2005). Thus, the large hypercarnivores of the Pleistocene became extinct along with the other megafauna, whilst the smaller, more generalist canids (coyote, gray wolf, and common jackal) were able to survive into the present day.

Pleistocene Coyotes

Coyotes of the Pleistocene were quite similar to current-day descendants, but less gracile in overall size (Kurtén, 1974). These more ancient coyotes are sometimes referred to as *C. latrans orcutti*, and were larger and more apt for the acquisition of large prey compared to coyotes of the Holocene. The extinction event at the end of the Pleistocene triggered a shift in ecological role for coyotes, and thus a shift in morphology and behavior (Meachen & Samuels, 2012). Meachen, Janowicz, Avery, and Sadleir (2014) found that Pleistocene coyotes possessed a thicker mandible, especially under the carnassials, extending under the molars, suggesting both greater meat and bone processing. Further evidence of bone processing is inferred from the less prominent coronoid process (Meachen et al., 2014). The jaw morphology of Pleistocene coyotes lends support to the idea that carnivores of the Pleistocene utilized carcasses more due to greater interspecific competition.

Van Valkenburgh and Hertel (1993) found that Pleistocene carnivores, including the coyote, had a higher incidence of tooth breakage than extant species do. Pleistocene predators suffered a 5-11% occurrence of any tooth breakage, while their current counterparts suffer only 0.5-2.7% breakage. This increased occurrence of tooth breakage has been attributed to greater contact of teeth to bone, which is the most common mode of tooth breakage. Predators were more diverse in the Pleistocene and it is also possible there was more overlap among predators, making competition more intense and increasing the extent of carcass utilization relative to current times. In the Pleistocene, predators were more likely to suffer a break in any of their teeth relative to extant predators, whereas extant predators suffered from canine breakages disproportionately more than in any other tooth (Van Valkenburgh & Hertel, 1993). This rate of tooth breakage is not a constant, however, as tooth breakage varied over time and was dependent on nutrient stresses that likely caused greater interspecific competition between carnivores and therefore greater levels of breakages at certain periods during the Rancholabrean (O'Keefe, Binder, Frost, Sadlier, & Van Valkenburgh, 2014). This increased rate in breakage of any teeth reinforces the idea that Pleistocene predator teeth came in contact with bone more frequently (greater utilization of the carcass), whereas extant predator teeth tend to break during the process of subduing prey because of the great force exerted on the canine teeth to hold onto large, moving prey. Breakages in the canine teeth of Pleistocene coyotes was more common than observed in Holocene specimens, indicating coyotes of the Pleistocene likely were more active large prey hunters than they are now (Van Valkenburgh & Hertel, 1993). Later Pleistocene coyotes show some characters intermediate between older and recent forms, illustrating change toward a more omnivorous lifestyle. Prior to the end Pleistocene extinction event, coyotes lived in concert with dire wolves. The extinction of dire wolves left an open top canid niche, which gray wolves migrated from Eurasia to fill. Dire wolves were significantly larger than gray wolves and thus overlap between resources for dire wolves and coyotes was lesser. Once gray wolves began to occupy the same space as coyotes, this increased competition, and their greater similarity in body size meant a greater overlap in necessary resources. This overlap is likely the evolutionary stress that pushed coyotes to be smaller and more omnivorous over time (Meachen et al., 2014; Meachen & Samuels, 2012).

Holocene Coyotes

The current-day coyote, *Canis latrans,* is a widely ranging species with large size variability depending upon location and subspecies (8 to 16 kg in Bekoff and Gese, 2003; up to 22 kg in Benson, Patterson, and Wheeldon, 2012). Males trend larger in size than females, but do not exhibit as much sexual dimorphism as the gray wolf. Although there are 19 subspecies of coyote (Jackson, 1951), due to their high ability to disperse, identification of individual subspecies is difficult and the utility of doing so is questionable. Coyotes can be found far to the north in northern Alaska and south to Costa Rica and Panama. They are present essentially everywhere within the United States, including major cities (Bekoff & Gese, 2003). Figure 3 is a photograph of a typical coyote.

Figure 3. Photograph of a coyote from Vermont, USA. Copyright Kyle Jones.

Diet

Coyotes exhibit a considerable amount of diet plasticity, varying both seasonally and spatially (Bekoff & Gese, 2003). In South Carolina, coyotes relied upon vegetative matter for at least 50% of their diet during summer and fall. A shift in diet toward mammals occurred in winter and spring, with white-tailed deer (*Odocoileus virginianus* Zimmerman, 1780) occurring in 40% of scats collected throughout December, and 31% of both wild boar (*Sus scrofa* L., 1758) and lagomorphs during February (Schrecengost, Kilgo, Mallard, Ray, & Miller, 2008). In Wyoming, spring and summer diets were dominated by rodents, especially the Uinta ground squirrel (*Urocitellus armatus* Kennicott, 1863), and the occasional cattle carcass. Winter diets were supplemented with small rodents, but primarily consisted of elk carrion (*Cervus canadensis* Erxleben, 1777; Bekoff & Wells, 1981). In Nova Scotia, greatest prey volume consisted of deer fawns in summer (32.5%), followed by snowshoe hares (*Lepus americanus* Erxleben, 1777) (23.5%) and adult deer (19.5%). In late summer to fall, snowshoe hare dominated the diet (36-40%), followed by adult deer (9-24%) and fruits (7-18%). Winter scats exhibited similar percent volumes of adult deer (36.3%) and hares (38.8%). The greatest occurrence of hares was in spring (43.5%), followed by adult deer (28%). It was also observed that predation on hares increased with increasing hare density, but increasing deer density did not impact predation on hares. It is likely coyotes predate more heavily upon lagomorphs when available because they are hunted in a solitary manner, rather than adult deer which must either be scavenged or pack hunted, and require a certain amount of vulnerability (i.e. deep snow) (Patterson, Benjamin, & Messier, 1998). A central West Virginia study found coyotes relied on deer in both winter and summer, but occurrence of deer remains was significantly higher in winter (76% vs 45% of scats). Rodents were also an important prey item, occurring more frequently in summer than in winter (nearly 50% vs ~22%). These findings support the claim of coyotes being opportunistic feeders, depending upon relative densities, availability, and vulnerability of different prey items (Crimmins, Edwards, & Houben, 2012).

Habitat Utilization

Being such a wide ranging species, it follows that coyotes inhabit a broad range of habitats. Not all habitat types are equally productive or suitable for the species, however. Crête et al. (2001) found that between forested and rural landscapes in Québec, individuals inhabiting the forest had home range averages of over double (100 km^2) that of rural coyotes (30-40 km^2). Habitat reflected diet as well, as rural coyotes fed primarily on deer in winter, whereas those in the forest subsisted on hare; thus it follows that they must cover more area in search of enough hare to satisfy their energetic requirements. In northern New York, coyotes were found to be most dense in forested habitat, specifically open canopied (disturbed) forest with natural edges along wetlands or shoreline, which is generally associated with greater prey density and vulnerability. Density was negatively correlated with human-related rural structures (R. W. Kays, Gompper, & Ray, 2008). A study performed in Indiana found that coyote core areas (where they spend the majority of their time) contained forest more than any other habitat. Non-core portions of home ranges contained more open habitat types (grassland, urban, and corridor). In urban areas with greater human infrastructure and activity, coyote home ranges were significantly smaller than in more rural areas, likely in order to limit direct contact with humans (Atwood, Weeks, & Gehring, 2004). Crimmins et al. (2012) observed that coyotes in central West Virginia also prefer disturbed, open forest with ample ground cover over more heavily forested areas, following the premise that these open areas tend to present greater opportunity for hunting.

Urbanization

Coyote utilization of urban and suburban habitats has been steadily increasing, especially in the last two decades, which can be evidenced by increased sightings of coyotes in urban/suburban areas, and the increasing number of antagonistic interactions occurring between coyotes and humans (Gehrt, 2007; Timm & Baker, 2007). Given increased exposure to human infrastructure, especially roads, one might expect urban coyotes to have a greater mortality rate than those inhabiting rural spaces. A study in Chicago, Illinois, found that even though the density of roads for the area was 6.11 km/km^2 and mortality via collisions with vehicles was the greatest risk of mortality for coyotes in the area (62% of all deaths), Chicago coyotes had a mean yearly (averaged over six years) survival rate of 0.62 (Gehrt, 2007). This higher rate of survival is in contrast to findings from Albany, New York, which found an annual survival of just 0.20 for coyotes tracked. In this case, a much higher rate of mortality came from hunting activity (43%) than one would expect from an urban landscape, as hunting is generally prohibited inside city limits. The difference here can be reflected in comparing the density of the human populations for each city. The population density in Chicago at the time was 5,684/mi^2 (Morey, 2004), whereas in Albany it was 563/mi^2 (Bogan, 2004). The population of Albany is more spread out and overall smaller than that of Chicago, so hunting and trapping may be permissible in some suburban areas, or coyotes may include more rural areas in their home ranges than Chicago coyotes do. Another factor to note is the number of coyotes collared in each study. The Chicago study tracked a denser population of 150 coyotes (Morey, 2004), whereas the Albany study only tracked 21 (Bogan, 2004; Gehrt, 2007).

Coyotes inhabiting urban areas are likely to cause the greater human population of those areas some alarm, in some cases for good reason. The relatively smaller home ranges of coyotes is not only indicative of their attempt at avoiding humans, it also suggests that they are residing in urban areas at higher densities than elsewhere (Gehrt, 2007). A dense population of top predators living in close proximity to dense populations of humans is likely to cause trouble for both species. Tending to be a relatively minor part of an urban coyote’s diet, human refuse can still be a motivator for closer interactions between the species (Morey, Gese, & Gehrt, 2007).

Non-livestock domesticated animals (pets) can fall prey to coyotes as well. Grubbs and Krausman (2009) observed interactions between coyotes and domestic cats (*Felis catus* Linnaeus, 1758) in Tucson, Arizona. Of the 36 interactions observed, 19 (53%) resulted in the death of the cat. Poessel et al. (2012) compiled reports about coyotes from in and around Denver, Colorado during the time period 2003 to mid-2010. Of the thousands of reports processed, most were merely observations. Actual conflict reports numbered 510, and of those, 92% were attacks on pets. These reports had a seasonal bias, as it was found to be 1.5 times more likely for a pet to be attacked during the winter months than any other time of year. This increase in winter attacks is likely related to the greater cost of foraging in the winter months and the limited amount of daylight, meaning people and their pets are outside in the dusk/dark more often than at other times of year. Less than 3% of all reports were attacks on humans (Poessel et al., 2012). Lukasik and Alexander (2011) observed similar conflict incidences in Calgary, Alberta (Canada). Of 781 incidents over a three year period, approximately 7% were attributed to human-coyote direct conflict, though they observed an increase in conflicts during the summer months which they attributed to protection of pups.

Coyote attacks on humans occur at relatively low frequencies and can take place due to a number of reasons. White and Gehrt (2009) compiled 142 coyote attacks on humans across North America ranging from 1960 to 2006 and ranked them based on behavioral intent. The greatest proportion of attacks were classified as predatory, meaning the coyote or pack pursued the victim(s) with intent to kill, often causing the most serious of injuries in the process. Most often, these attacks were carried out on children (10 years of age or younger). Attacks classified as investigative made up the next largest proportion. In these cases, coyotes were experimenting with the possibility of predation on humans by sneaking up on them, or stealing things directly from them. Both these types of attacks require some level of habituation to humans, which often involves intentional or unintentional feeding of wildlife. No significance was found between times of day or year.

Range, Colonization, and Introgression with Wolves

Although populations of coyotes exist throughout North America currently, their presence east of the Mississippi River began after the arrival of European settlers. Originating from the West, coyotes began migrating and colonizing new habitats with the human-instigated extirpation of the gray wolf from nearly the entirety of its historic range (R. Kays, Curtis, & Kirchman, 2010; Ripple, Wirsing, Wilmers, & Letnic, 2013). Along with this anthropogenic persecution of the wolf came changes in habitat and relative abundance of prey (Bozarth, Hailer, Rockwood, Edwards, & Maldano, 2011). Because coyotes are a generalist species, colonization across a broad range of habitats was relatively easy (Bozarth et al., 2011; Crête et al., 2001; Crimmins et al., 2012; Schrecengost et al., 2008). Two major migration paths have been proposed for coyote expansion to the East. One route passed north over the Great Lakes and into Canada. The second route took migrants below the Great Lakes, through Ohio (Kays et al., 2010; Figure 4). The coyotes following the southern route did not encounter wolves because they no longer inhabited those areas. Although, Monzõn, Kays, Dykhuizen (2014) found there was admixture of both wolf and dog in Ohio coyotes, it is likely this introgression occurred before coyote colonization of the East.

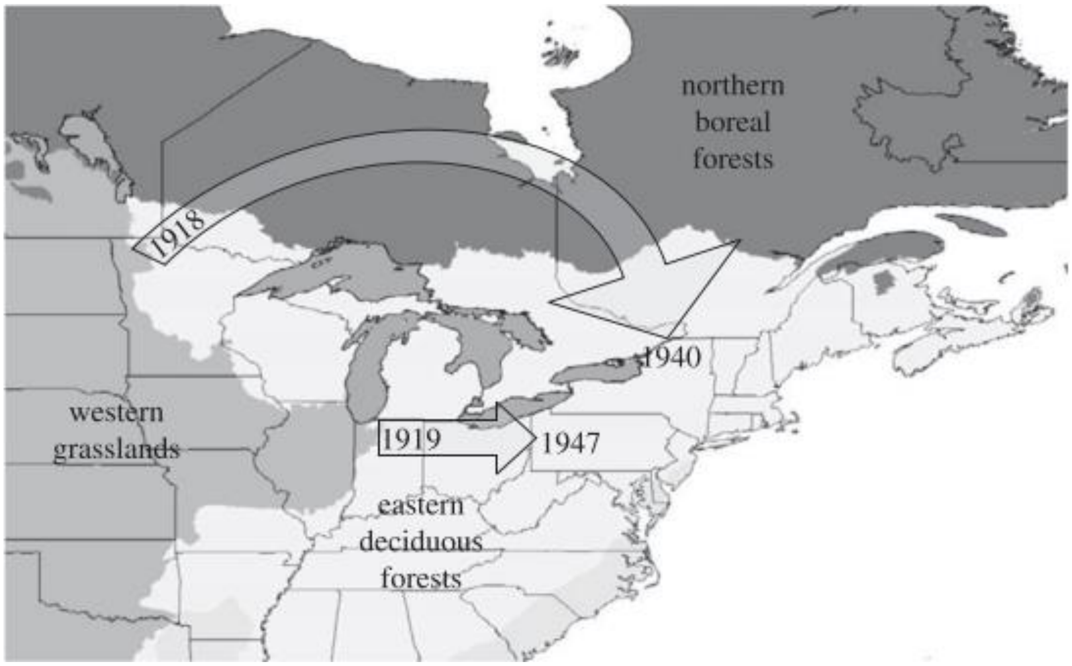


Figure 4. Proposed range expansion of coyotes to the East. Taken from Kays et al. (2010).

While traveling along the northern route, a number of dispersing coyotes interbred with eastern wolves, also referred to as Great Lakes wolves (*C. lycaon* Schreber, 1775). Wolves found refuge in Canada, but population sizes were limited. In addition, coyotes were moving into areas yet uncolonized by the species (Kays et al., 2010). This colonization of new territories led to an Allee effect, where conspecific mates are difficult to find due to low density of individuals (Allee, Park, Emerson, Park, & Schmidt, 1949). These hybridization events were not isolated, leading to introgression and the creation of a hybrid swarm. These hybrid "coywolves," commonly referred to as eastern coyotes, are significantly larger than their western counterparts. It is fairly easy to distinguish the two both genetically and morphologically, as these hybrids tend to be larger overall and possess distinct cranial and dental features (Benson et al., 2012; Kays et al., 2010). Due to the relatively recent nature of this introgression, gene flow between descendants of the two migration paths has not yet been extensive; therefore, the genetic admixtures of populations vary spatially (Kays et al. 2010).

In the United States, the states in the Northeast, such as Maine and New York, are inhabited by a population descended from individuals that migrated through Canada. These individuals are highly admixed with wolf mitochondrial DNA. According to Kays et al. (2010), approximately one-quarter of the population sampled in the northeast exhibited distinct eastern wolf haplotypes. In contrast, coyotes sampled from Ohio, Texas, and Nebraska possessed only coyote haplotypes. It has been hypothesized that wolf haplotypes have not been found in populations of coyotes in the West because the eastern wolf has enforced gene flow between gray wolves and coyotes due to its intermediate size (Wheeldon & White, 2009; Wilson, Grewal, Mallory, & White, 2009). An evolutionary linkage between eastern wolves and coyotes may have better support though, as Mexican gray wolves (*C. l. baileyi*) will not readily hybridize with coyotes, even though they are of a similar size to eastern wolves. This closer relatedness may instead be the source of such extensive hybridization (Wilson et al., 2000). In some areas where gray wolves have been reintroduced, coyotes live in close proximity but will not readily hybridize. However, Lehman and Wayne (1991) found that populations of gray wolves across North America contained varying levels of distinct coyote mtDNA, ranging from 0% to 100% occurrence. Wolf specific mtDNA was not found in any of the coyotes sampled, indicating hybridization occurs mainly between male wolves and female coyotes. These hybridization events most likely occur when young male wolves leave their natal home ranges and disperse into areas with substantial coyote populations and few mature female wolves. Thiel (2006) made observations that seem to support that theory in Wisconsin, despite not finding evidence of any hybridization events during the course of the study. Where species ranges overlap, gray wolves act as a source of interference competition, in most cases acting aggressively towards coyotes at prey kill sites. In some cases, these interactions will lead to the death of a coyote, especially if the coyote is transient (Bekoff & Gese, 2003; Berger & Gese, 2007; Merkle, Stahler, & Smith, 2009). Coyotes will actively avoid areas with increased wolf activity (i.e. den sites), even when prey is readily available in those areas (Miller, Harlow, Harlow, Biggins, & Ripple, 2012).

Coyote vs. Wolf Behavior

Coyotes and wolves have behavioral differences that set them apart, particularly in pack hunting and human interaction. Wolves, due to their larger body size and therefore higher energetic requirements, focus on larger prey such as ungulates. Coyotes often feed on small mammals, especially in summer and rely on ungulate carcasses in winter. Coyotes form packs similar in structure to wolves, with an alpha breeding pair and potentially offspring from previous years. They are less able than wolves to take down larger prey, even while in packs, though alpha coyotes will initiate attacks on vulnerable large prey, such as calves or older individuals. Coyote pack hunting strategy differs from wolf strategy in that two or three (usually the alpha pair, sometimes joined by a beta) members will participate in the attack of prey while other pack members watch. In the case of wolves, most of the pack will participate in the chasing and taking of prey (Gese & Grothe, 1995). Fully cooperative pack hunts displayed in wolves may not have developed in coyotes because their energy requirement is lesser, and can be satisfied by scavenging, smaller prey, and the occasional cooperative hunt (Moehlman, 1989). In addition, wolves tend to be more wary of humans and areas of higher human activity than coyotes. Wolves were found in highest densities when the density of roads in the area was the lowest. A threshold for road densities was posed by Thiel (1985) of 0.58 km/km^2 in Wisconsin, in which wolves would be more common below that threshold and very uncommon above it. This threshold was reinforced by Mech, Fritts, Radde, and Paul (1988) in Minnesota. Coyotes, in contrast, seem to have no issue with being in close proximity to humans and human infrastructure. Being both habitat and diet generalists, they will feed on nearly anything they can scavenge, and human refuse is no exception (Fuller, Destefano, & Warren, 2010). Coyotes in all environments with noticeable human presence have been found to shift to a primarily nocturnal activity pattern, likely in order to avoid direct human contact (Atwood et al., 2004). An increase in diurnal activity in urban/suburban environments suggests a move toward habituation to humans, increasing the likelihood of antagonistic interactions (Gehrt, 2007).

Coyote-Wolf Hybrids

Hybrids, on the other hand, may possess some mixture of both coyote and wolf behavioral characteristics, similar to their intermediate morphology. Changes to teeth, parts of the skull, and overall larger body size is suggested as making them more effective than western coyotes at actively hunting adult ungulates (Kays et al., 2010). Benson and Patterson (2013) found packs led by a hybrid alpha to be effective at preying on healthy adult moose (*Alces alces* Linnaeus, 1758) in Ontario, even when its mate was a coyote. Interestingly, a pack led by two non-hybrid coyotes was also documented as taking a young moose during the course of the study. This predation of coyote on moose could be attributed to the moose still being young and inexperienced, as well as the larger than average body size of coyotes resident in Ontario. Previously, it was believed that among canids, only wolves were effective moose predators. These morphological and behavioral changes brought about by hybridization may make these eastern coyotes a better substitute than their western counterparts for the wolves they replaced.

West Virginia Coyote Characterization

For the purpose of this study, the nature of coyotes inhabiting West Virginia was in question. Where Kays et al. 2010 was primarily a study of genetics, this study focuses on morphology, using measurements from that prior study as populations for comparison. Because *Canis* species are morphologically distinguishable, it is possible to make inferences as to the genetic admixture of the population in question by comparing morphological measurements from populations of known admixture. In addition, combining genetic studies with morphology gives the most complete characterization of a population. Kays' et al. (2010) mitochondrial study found evidence of widespread introgression of Great Lakes wolf haplotypes in coyotes of the Northeast and their "contact zone" (where the two coyote migration routes met) in western New York and Pennsylvania. They found no wolf haplotypes in Ohio or western coyotes. Bozarth et al. (2011) found the contact zone mentioned by Kays extends as far as at least northern Virginia, as a diverse range of coyote mitochondrial haplotypes and a haplotype attributed to Great Lakes wolves were all found in the population of coyotes sampled. In contrast, a microsatellite study in West Virginia and Virginia found low levels of admixture (no greater than 16% of individuals). Microsatellite analysis detects more recent hybridization than the mitochondrial DNA surveys, meaning the wolf haplotypes detected in Bozarth et al. (2011) are likely the result of older hybridization events (Bohling et al., 2017). Bohling et al. (2017) also found evidence of several individuals highly admixed with domestic dogs, meaning some contemporary hybridization between coyotes and dogs is ongoing, at least in the West Virginia-Virginia area. Monzõn et al. (2014) found mitochondrial evidence for admixture with dogs in Ohio, but these were not thought to be contemporary hybridization events.

If coyotes from West Virginia are morphologically distinguishable from both the West and Northeast, then the novel morphology would not be the result of hybridization, as this would make West Virginia coyotes more similar to the Northeast. Novel morphology would also indicate adaptive pressure from the environment in West Virginia that differs from the West; otherwise they would likely be indistinguishable from each other. The first reports of coyotes in Ohio come from 1919 (Weeks, Gildo, and Shieldcastle, 1990), and from 1947 for western Pennsylvania (Williams, McLaren, & Burgwin, 1985). Given the proximity of West Virginia to both Ohio and Pennsylvania, it was predicted coyotes originating from West Virginia would be more similar to western coyotes morphologically, and therefore descended primarily from coyotes migrating along the southern route through Ohio, as coyotes began migrating east through Ohio before the other route of coyote expansion had reached Pennsylvania.

CHAPTER TWO

INTRODUCTION

Coyotes, while possessing the ability to disperse great distances, have a detectable genetic signature related to their population of origin (vonHoldt et al. 2011). These genetic signatures may be expressed morphologically as well, allowing detection of distinct populations in morphometric space. Morphological differences can be useful in separating out populations of individuals, but can also be used to make inferences about the ecology of the organism, such as dietary habits, which can be explored via skull and tooth morphology (Meloro, Hudson, and Rook 2015; Meloro and Louys 2015). Morphological studies will generally utilize a wide variety of measurements, which when taken on an individual will necessarily covary as they are not independent. This covariation is hard to separate out from overall patterns of variation among individuals, so principle components analysis (PCA) is often used to help make these patterns more evident and easier to detect than in the original variables (Zelditch, Swiderski, Sheets, & Fink, 2004, p. 156).

The way PCA works is by reordinating variables originally measured with a linear combination of all the variables, which removes their dependence. Depending on the variation these variables describe, PCA will generate a number of principle components (PCs), which are different shape and size metrics with associated values that describe a certain percentage of variation in the data set. Most of the variation in the data can be explained in the first few principle components because the first PC will always be the one with the longest axis, or rather that which describes the greatest percentage of variation (Zelditch et al., 2004, p. 156-168). As the number of PCs increases, the amount of variation they describe decreases, so that PC 1 will describe the most variation, followed by PC2 and so forth. When examining PCA for clusters of individuals, detecting distinct groups can be accomplished by combining PCA with multivariate statistics (ANOVA, for example), as appearance of a cluster may not always indicate a distinct group (Zelditch et al., 2004, p. 156).

Allometry

As an organism grows larger its constituent parts must grow as well, but not every part of an organism grows at the same rate or has the same response to an increase in overall body size. Differences in proportions of growth between different segments of an individual is allometry. If the ratio or proportion between two measurements is held constant, this is instead referred to as isometry. Positive allometry occurs when a measurement is increasing in proportion to the reference measure. Negative allometry occurs when a measurement is increasing at a rate lesser than the reference measure. Some components of an individual must get larger in order to accommodate for an overall larger body size, whereas others need not grow at such a pace. Allometry can be studied within a population of the same species (static allometry), phylogenetically (evolutionary allometry), and developmentally (ontogenetic allometry) (Gould 1966; Jolicoeur 1963). Many of the specimens used in this study were of unknown age, although the majority were determined to be adults because adult dentition is achieved by 6-7 months of age (Kreeger, 2003). This study is not concerned with allometry related to ontogeny. When examining differences among populations, the focus is primarily on evolutionary allometry (even though these are the same species, they reside in separate areas and have distinct recent histories), and additionally static allometry when examining variation within each population (sexual dimorphism, for example).

MATERIALS AND METHODS

Experimental design

In this study, we elected to take morphological measurements of the skulls of coyotes harvested in West Virginia, the total number being 123, with an additional 2 in the collection from Ohio. These coyotes were harvested between 1989 and 2000, and no tissue or hair samples were saved. The West Virginia-Ohio skulls were compared to coyote skulls from Kays, Curtis, and Kirchman (2010). That particular study focused on genetics, but skull measurements were also taken. We replicated those measurements in our study to make comparison straightforward. A complete list and description of each measurement is provided in Table 1, and a visual representation is given in Figure 5. Due to skull fragmentation, a full suite of measurements was not possible on the entirety of the collection. Full measurements were collected on 89 out of 125 specimens.

Skulls measured for this study were compared to Kays' et. al. (2010) measurements of specimens from Arizona (n=11), Montana (n=2), Nebraska (n=8), Texas (n=1), Ohio (n=22), Connecticut (n=4), Massachusetts (n=16), New Hampshire (n=20), New York (n=77), Vermont (n=10), Maine (n=42), New Jersey (n=4), Pennsylvania (n=1), Quebec (n=14), and Ontario (n=2). The number of skulls measured in that study is 234. Full measurements were taken on 190 of the 234 specimens. The total number of skulls analyzed in this study was 302. These states were grouped into regions based on proximal locality and how they cluster genetically (vonHoldt et al., 2011). West (W; N=22) constitutes Arizona, Montana, Texas, and Nebraska. This region represents the ancestral western coyote population. Northeast (NE; N=190) includes Connecticut, Massachusetts, New Hampshire, New York, Vermont, Maine, New Jersey, Pennsylvania, Ontario, and Quebec. This region represents the hybrid swarm of eastern coyotes. West Virginia is grouped with Ohio into WV-OH (N=125; Fig. 6).

Table 1. List of the 14 measurements taken and their description

Measurement	Description
Greatest length of skull (GSL)	Length from anterior tip of premaxillae to posterior point of union
Zygomatic width (ZW)	Greatest distance across zygomata
Alveolar length of maxillary toothrow (ALM)	Distance from anterior edge of alveolus of P1 to posterior edge of alveolus of M2
Maximum width across upper cheek teeth (MXP)	Greatest breadth of skull measured between outer sides of crowns of P4
Palatal width at first premolars (MNP)	Minimum width between inner margins of alveoli of P1
Width of frontal shield (WPOP)	Maximum breadth across postorbital process of frontals
Height from toothrow to orbit (M1O)	Minimum distance from outer alveolar margin of M1 to most ventral point of orbit
Depth of jugal (HJ)	Minimum depth of jugal anterior to postorbital process, at right angle to its anteroposterior axis
Crown length of upper carnassial (LP4)	Maximum anteroposterior length of crown of P4 measured on outer side
Crown width of second upper molar (WM2)	Maximum transverse diameter of M2 measured from outermost point to innermost point of crown
Height of mandible at M1 (HMm1)	Dorsoventral distance from alveolus at mid-point of M1 to ventral edge of mandible
Width of mandible at M1 (WMm1)	Thickness of mandible at mid-point of M1
Height of coronoid process (HCP)	Dorsoventral distance from ventral edge of mandible to tallest point of coronoid process
Width of anterior portion of ramus (WAR)	Thickness of ramus posterior to M2

Because the data being analyzed in this study are physical size measurements, which in each individual are correlated to one another by virtue of being a part of the same organism and are thus relative to one another, the data were natural logarithmically transformed (Jolicoeur 1963). Transformation is also helpful in simplifying the scale of measurements, since they vary widely across the data set. Principle components analysis (PCA) was then performed on the natural logarithm-transformed data to examine size and shape. Principle components analysis was chosen to reduce the number of dimensions across the data set, making for simpler detection of trends in the data. Due to the heterogenous nature of variance among measurements, the correlation matrix was used in eigenanalysis instead of the covariance matrix (Reyment & Jvreskog, 1996).

Figure 5. Measurements taken in this study, replicated from Kays et. al. (2010). Measurements are as follows: A) greatest length of skull; B) zygomatic width; C) width of frontal shield; D) maximum width across upper cheek teeth; E) crow length of upper carnassial; F) height from toothrow to orbit; G) depth of jugal; H) alveolar length of maxillary toothrow; I) palatal width at first premolars; J) crown width of second upper molar; K) height of coronoid process; L) height of mandible at M1; M) width of mandible at M1; and N) width of anterior portion of ramus.

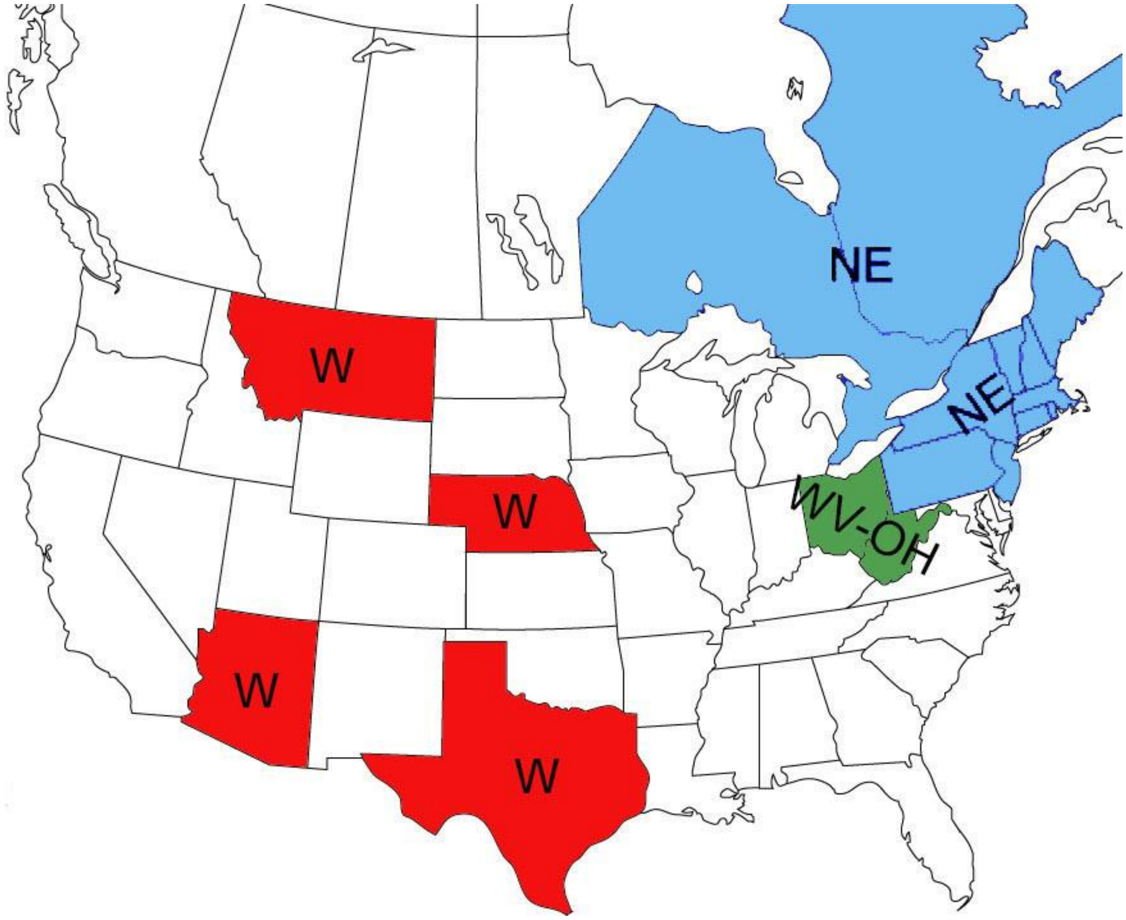


Figure 6. Map of specimen distribution and the regions grouped for analysis. “NE” denotes Northeast (in blue), “WV-OH” denotes West Virginia and Ohio (in green), and “W” denotes West (in red).

While domestic dogs were also measured in this study, the first PCA performed was split by species, so as to exclude domestic dogs from analyses pertaining to only coyotes. Inclusion of domestic dogs in the PCA skews the data in a way that makes for unreliable comparison when attempting to examine differences among coyote populations (see Chapter Three).

PCA generated 14 principle component axes that accounted for 100% of the variability in the data, but only the first two components were used in analysis, as they accounted for approximately 69% of the variation. As a general rule only axes that may be biologically relevant should be interpreted. In this case, only the first three principle components account for greater than 5% variance in the data set (Zelditch et al. 2004); however PC 3 is not featured in any analysis because it possessed no apparent trends.

In order to examine statistical differences among groups, parametric statistics, specifically one-way ANOVAs and Student’s t-tests, were utilized. First, presence of sexual dimorphism was examined both within the entire set of coyotes and additionally by region to examine if dimorphism varies regionally, using one-way ANOVAs. The regional variation in dimorphism also required pairwise comparisons, so the Student’s t-test was also run. Overall sexual dimorphism was examined for PC 2 as well, but pairwise comparison was not made. Overall regional, non-sex related differences were explored for both PC 1 and PC 2, using one-way ANOVAs and Student’s t-test.

Allometric differences among populations were also examined. Eigenvalues taken from the covariance matrix of the first principle component were used to create Figure 7, which represents the multivariate allometry vector. (Jolicoeur, 1963; O’Keefe, Meachen, Fet, & Brannick, 2013).

RESULTS

Principle components analysis

A summary of the eigenanalysis on the correlation matrix for principle components 1-5 is given in Table 2. The first PC accounts for the greatest amount of variation in the data set (60.9%) and is largely a size-descriptive axis because all eigenvalues are positive. The second PC accounts for the second greatest amount of variation (8.2%) and relates to bone and tooth shape variation, as it is largely tooth measurements loading positively while the bone measurements load negatively (Table 2). These two PCs were the two used in analyses because they contain distinct patterns, whereas PCs 3-14 have non-distinct, difficult to interpret patterns.

Table 2. Summary of eigenanalysis on correlation matrix for all coyotes

	PC1	**PC2**	**PC3**	**PC4**	**PC5**
Eigenvalue	8.526	1.155	0.844	0.643	0.542
Percent	60.902	8.247	6.031	4.595	3.869
Cumulative percent	60.902	69.149	75.180	79.776	83.645
LP4	0.253	0.362	-0.117	-0.346	0.459
WM2	0.153	0.630	0.155	0.659	0.076
ALM	0.258	0.211	-0.545	-0.160	0.203
M1O	0.291	-0.126	-0.113	0.065	-0.293
HJ	0.279	-0.116	0.132	-0.220	-0.160
GSL	0.289	-0.039	-0.426	0.089	-0.151
ZW	0.298	-0.257	0.056	0.011	0.199
WPOP	0.228	-0.465	0.130	0.390	0.411
MXP	0.303	0.042	0.141	-0.063	0.243
MNP	0.257	-0.062	0.314	0.115	0.075
HMm1	0.296	-0.147	-0.176	0.076	-0.159
WMm1	0.251	0.292	0.308	-0.160	-0.419
HCP	0.287	-0.046	-0.129	0.180	-0.366
WAR	0.259	0.027	0.416	-0.356	0.018

Figures 7 and 8 show PC 2 plotted against PC 1. In Figure 7, the pattern of differentiation between regions is difficult to discern, but the higher variability in PC 1 is illustrated by the wider range (approx. -9 to 8) when compared to the variation in PC 2 (approx. -4 to 4). In Figure 8 the scatterplot is replaced by 95% density clouds with associated reduced major regression lines. The relative trajectories of PC 2 relative to PC 1 become more apparent in this figure. The slopes for the Northeast and West regression lines are both positive and very similar, in contrast to the regression line for the West Virginia and Ohio region which has a negative slope (Table 3). The West possesses the strongest positive correlation (0.55), the Northeast possesses a weaker positive correlation (0.24), and West Virginia-Ohio possesses a weak negative correlation (-0.12) between PC 2 and PC 1 (Table 3).

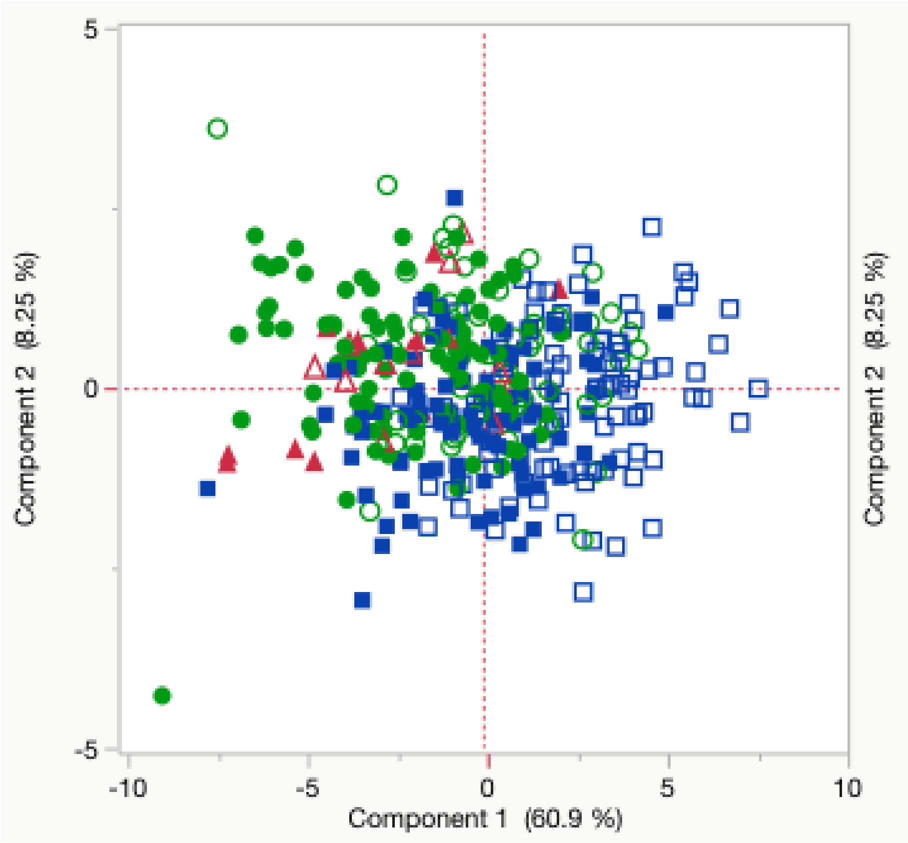


Figure 7. PC 2 plotted against PC 1 for all coyotes. Regions are differentiated by color and shape: Northeast = blue squares; West = red triangles; West Virginia and Ohio = green circles. Sex is differentiated by filled or unfilled shapes: shape filled in = female; shape hollow = male.

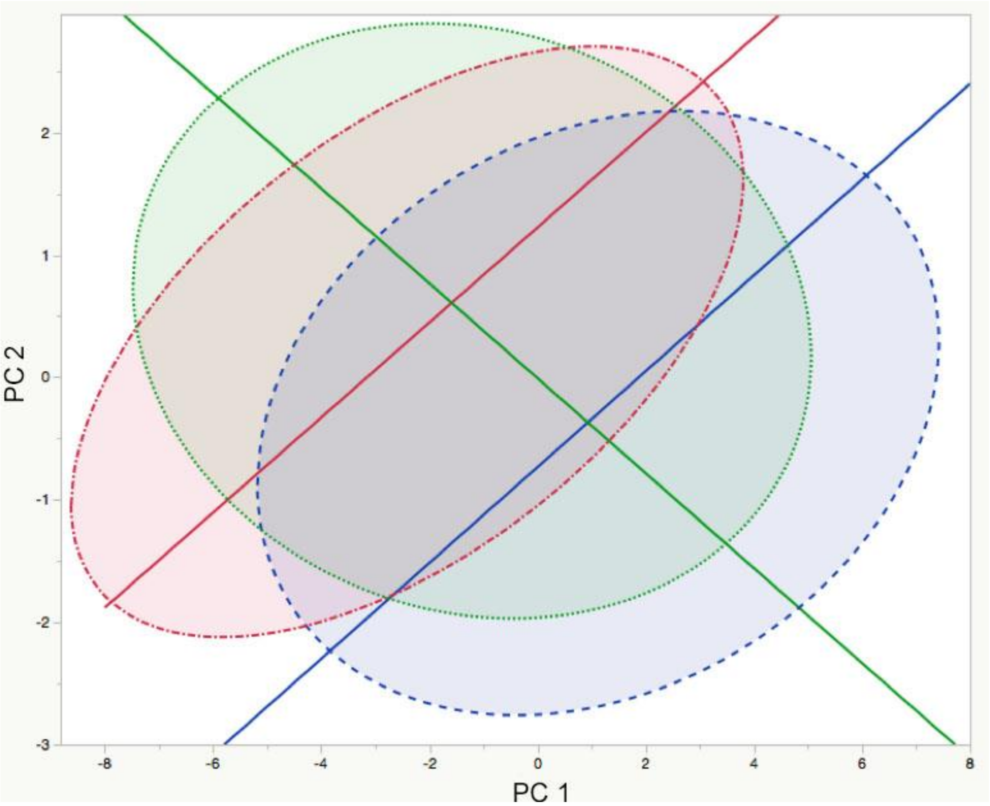


Figure 8. PC 2 plotted against PC 1 with 95% density clouds for regions and reduced major axis regressions. Green is West Virginia and Ohio, blue is Northeast, and red is West.

Table 3. Summary data for the reduced major axis regressions for coyotes by region.

West Virginia-Ohio	**Variable**	**Mean**	**Std. Dev.**	**Variance Ratio**	**Correlation**
	PC 1	-1.20944	2.561813	0.150337	-0.1208
	PC 2	0.461335	0.993299		
	Intercept	**Slope**	**Lower CL**	**Upper** CL	**Alpha**
	-0.0076	-0.38773	-	-	0.05
Northeast	**Variable**	**Mean**	**Std. Dev.**	**Variance Ratio**	**Correlation**
	PC 1	1.121964	2.574686	0.15326	0.2387
	PC 2	-0.29303	1.00795		
	Intercept	**Slope**	**Lower CL**	**Upper CL**	**Alpha**
	-0.73226	0.391485	0.189695	0.807927	0.05
West	**Variable**	**Mean**	**Std. Dev.**	**Variance Ratio**	**Correlation**
	PC 1	-2.4075	2.538434	0.150729	0.5520
	PC 2	0.290436	0.985517		
	Intercept	**Slope**	**Lower CL**	**Upper CL**	**Alpha**
	1.225118	0.388238	0.147415	1.022483	0.05

Allometry

Differences illustrated among the regional groupings reflect differential growth in the bones and teeth. Figure 9 shows differences in allometry between each region. The vector of isometry is included as a vertical line (√1/14=0.267). While some features of the skull seem rather conserved in size or shape changes, such as the length of the upper carnassial (LP4), others are highly variable by region, such as the width of the mandible at the lower carnassial (WMm1). Overall, West Virginia-Ohio seems to be more allometrically similar to the Northeast than to the West (Figure 9).

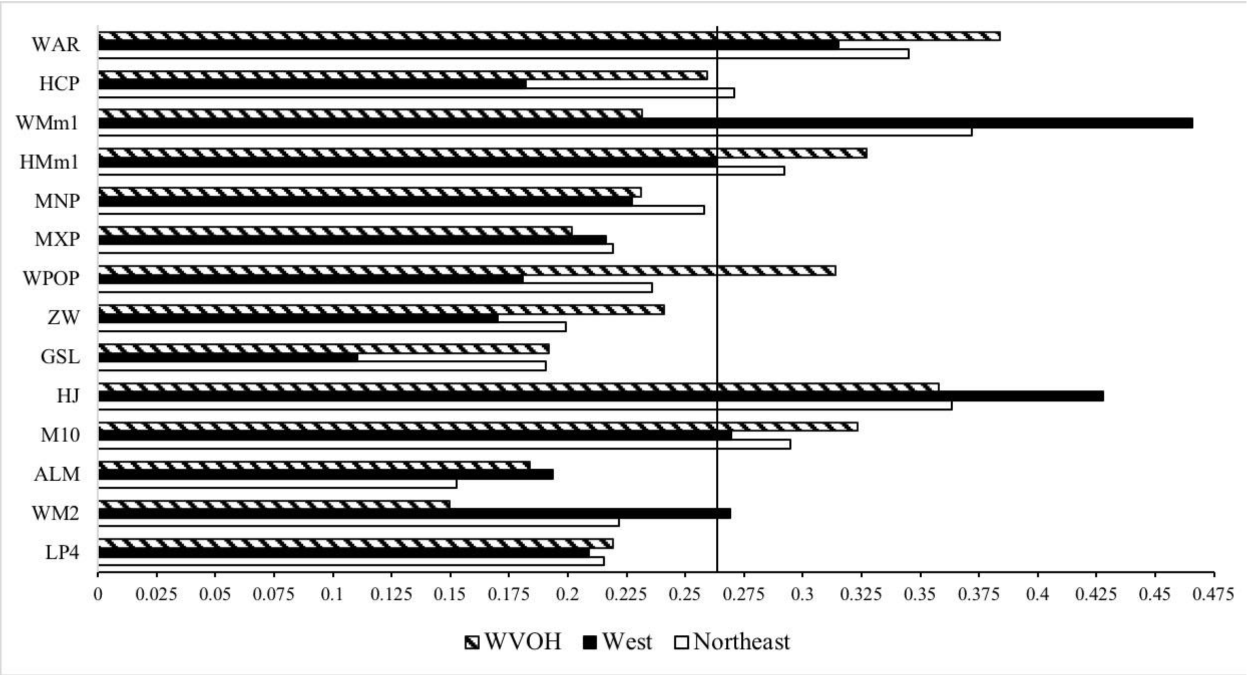


Figure 9. Allometry vectors for each of the 14 measurements, split by region. Values for each bar reflect eigenvalues taken from the covariance matrix of PC 1. The vector of isometry is represented as the vertical line at 0.267.

Sexual dimorphism

While coyotes can be sexually dimorphic, the differences between the sexes tend to be more subtle than those found in other more dimorphic species, such as the gray wolf (Hillis and Mallory, 1996; Kays et al., 2010; Kennedy, Mech, Tran, Grubaugh, and Lance, 2003; O'Keefe et al., 2013). However, statistically significant dimorphism can still occur, as illustrated in the

ANOVA in Figure 10 and Table 4. The difference in the size axis between males and females is highly significant (p<.0001). Figure 11 splits the sexual dimorphism out by region for PC 1 to explore in which region sexual differences are greater or more apparent. An ANOVA was also run in this case, with the summary data in Table 5 and ordered differences report in Table 6. The group in the Northeast appears to be the most highly dimorphic, with the tightest, most distant error bars for each sex. Additionally, Northeastern coyotes of both sexes were significantly different from coyotes from any other region (Table 6). West Virginia and Ohio coyotes also have apparent sexual dimorphism, though not as dramatic as seen in the Northeast, and more similar to that seen in the West, although sexual dimorphism in the Western coyotes is not at all apparent in Figure 11. Male coyotes are significantly different between the West and West Virginia-Ohio (p=0.04), but female coyotes show no significant difference between these two regions (p=0.26; Table 6).

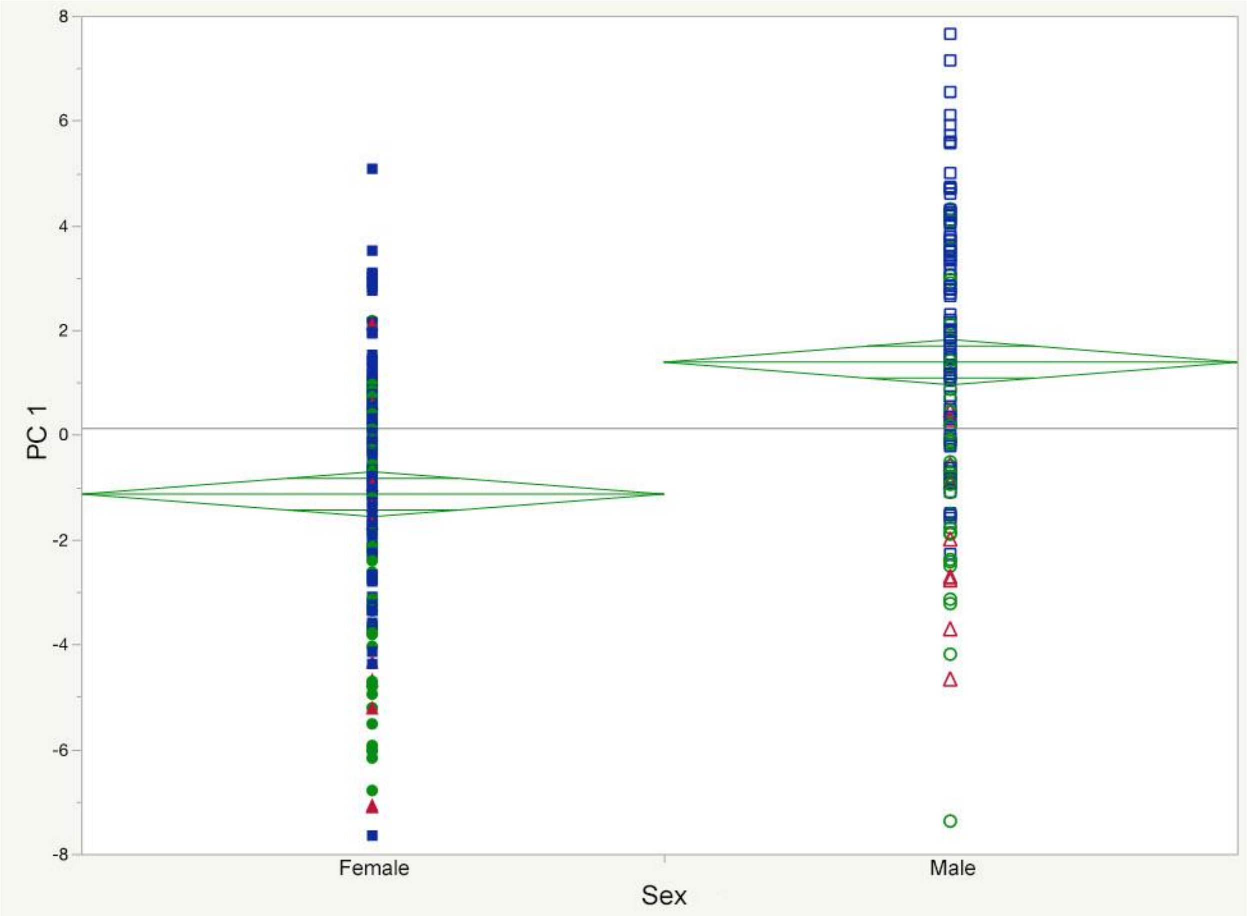


Figure 10. ANOVA on PC 1 grouped by sex for coyotes. Illustrates significant sexual dimorphism across all coyotes analyzed in this study.

Table 4. ANOVA summary for PC 1 by sex for coyotes. Star indicates significance at α = 0.05.

Source	DF	Sum of squares	Mean Square	F Ratio	Prob > F
Sex	1	441.294	441.294	66.526	<.0001*
Error	276	1830.821	6.633		
C. Total	277	2272.115			

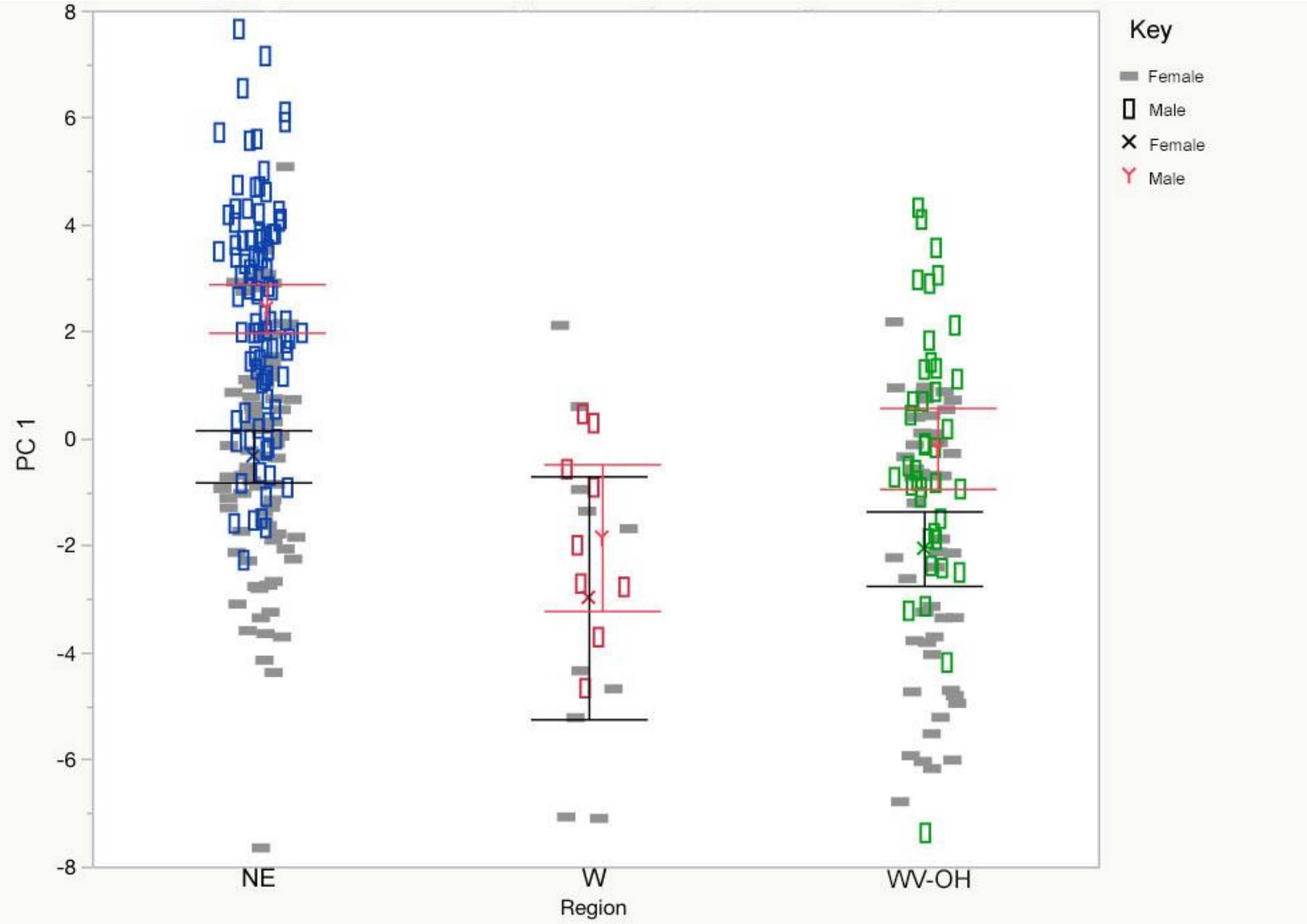


Figure 11. Relative sexual dimorphism in PC 1 by region for coyotes. Hollow, non-gray rectangles represent males; filled in, gray rectangles represent females. The means and standard error bars are included for each sex. "Y" with red bars represents male; "X" with black bars represents female.

Table 5. ANOVA summary for PC 1 by region and sex for coyotes. Star indicates significance at α = 0.05.

Source	DF	Sum of Squares	Mean Square	F Ratio	Prob > F
Model	3	840.307	280.102	53.602	<.0001*
Error	274	1431.807	5.226		
C. Total	277	2272.115			

Table 6. Ordered differences report for PC 1 split by region and sex for coyotes. Star indicates significance at α = 0.05.

Level	- Level	Sex	Difference	Std. Err Dif	Lower CL	Upper CL	p-Value
Northeast	West	Female	2.645	0.790	1.082	4.207	0.0011*
		Male	4.285	0.768	2.765	5.804	<.0001*
Northeast	WV-OH	Female	1.726	0.427	0.882	2.569	<.0001*
		Male	2.617	0.418	1.791	3.444	<.0001*
WV-OH	West	Female	0.919	0.818	-0.698	2.536	0.2632
		Male	1.667	0.810	0.065	3.270	0.0416*

The second PC was also explored to discover if there is any significant sexual dimorphism on this axis. The ANOVA results indicate that significant sexual dimorphism cannot be used to explain variation in PC 2, as males and female overlap heavily on this axis (Fig. 12; Table 7). For this reason, it was decided not to split this PC by species to make pairwise comparisons as was done for PC 1.

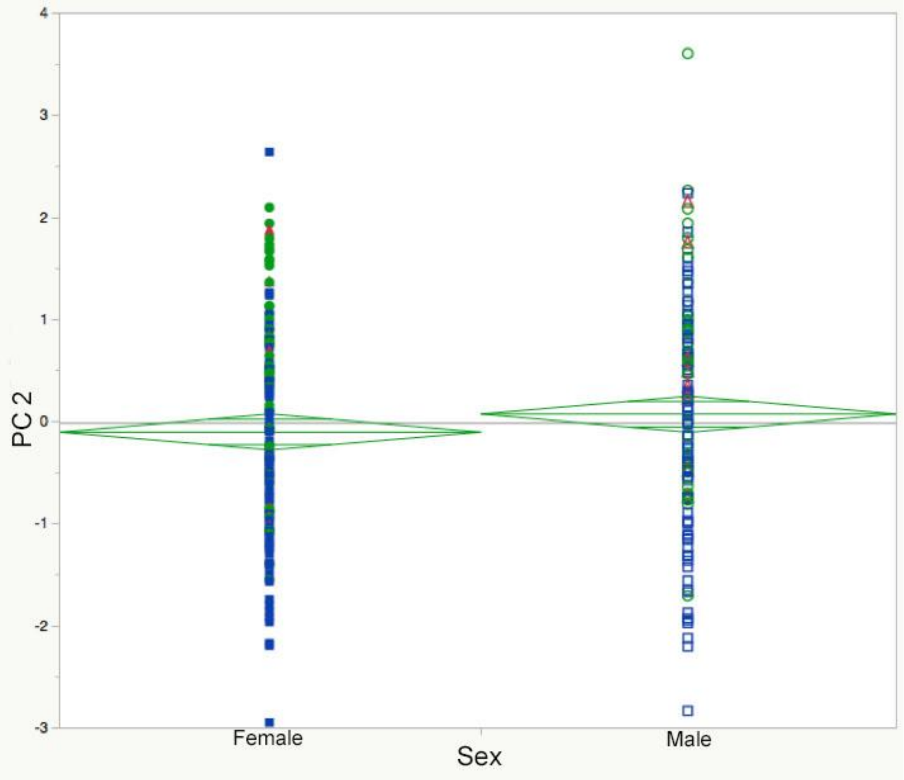


Figure 12. ANOVA on PC 2 grouped by sex for coyotes. Sexes heavily overlap, indicating little to no sexual dimorphism in this principle component.

Table 7. ANOVA summary for PC 2 by sex for coyotes.

Source	**DF**	**Sum of Squares**	**Mean Square**	**F Ratio**	**Prob > F**
Sex	1	2.14914	2.14914	1.9160	0.1674
Error	276	309.58152	1.12167		
C. Total	277	311.73065			

Regional differences

A one-way ANOVA was run on both the first and second PCs split by region to examine the overall differences among the regions for both the “size” and “shape” axes. Figure 13 shows the Northeast as the most positively scoring region for PC 1, while both West Virginia-Ohio and the West score negatively on PC 1. Table 8 gives the ANOVA summary data describing the overall difference between these populations for PC 1 as very significant (p<.0001). Table 9 splits out the differences using the Student's t-test, giving pairwise comparisons. The Northeast is shown to be the most different from either of the other two regions, with both pairwise values for the Northeast as p<.0001. As suggested by Figure 13, the difference between West Virginia-Ohio and the West is not as dramatic, as there is some overlap between their confidence intervals. The pairwise comparison between the West and West Virginia-Ohio gives a marginal significance of p=0.06.

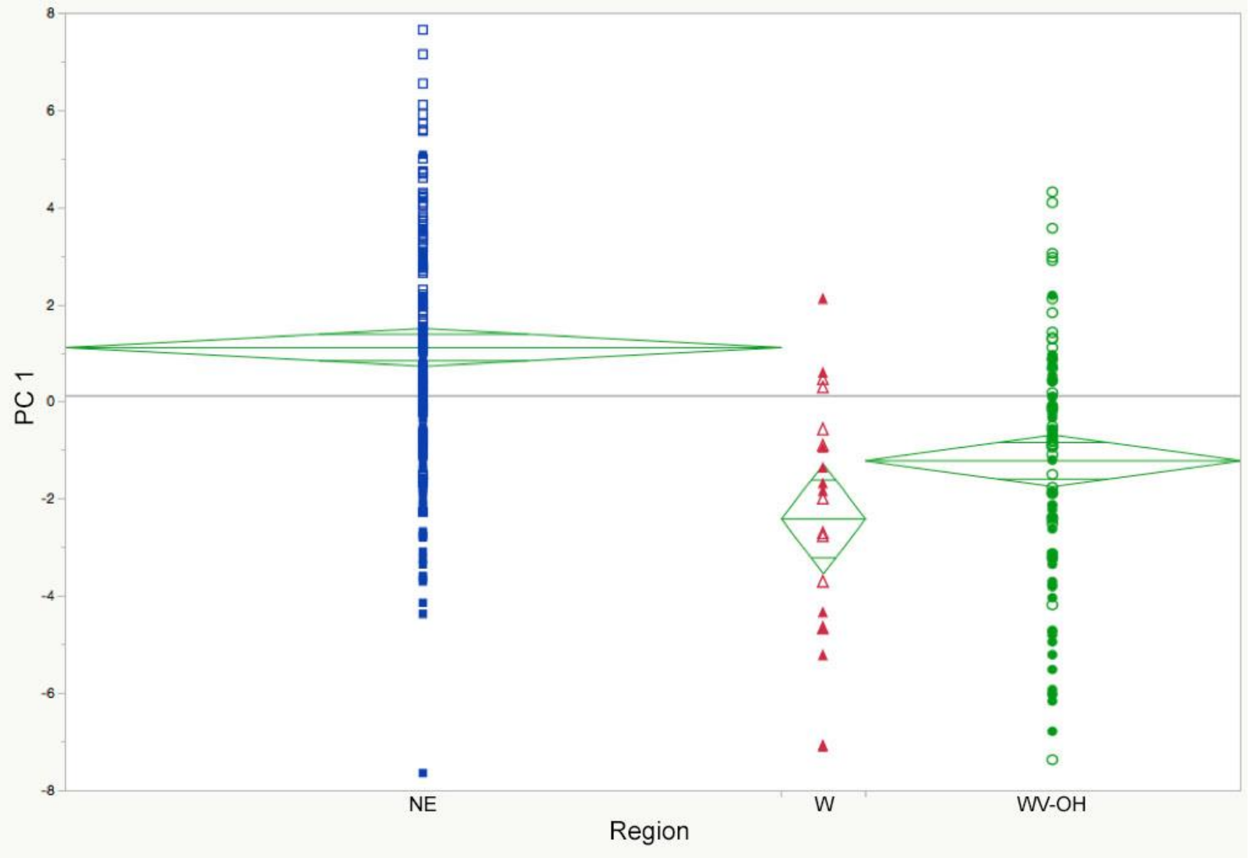


Figure 13. ANOVA on PC 1 split by region for coyotes.

Table 8. ANOVA summary for PC 1 split by region for coyotes.

Source	**DF**	**Sum of Squares**	**Mean Square**	**F Ratio**	**Prob > F**
Region	2	455.7253	227.863	34.5500	<.0001*
Error	276	1820.2655	6.595		
C. Total	278	2275.9908			

Table 9. Ordered differences report for ANOVA on PC 1 split by region for coyotes. Star indicates significance at α = 0.05.

Level	**- Level**	**Difference**	**Std Err Dif**	**Lower CL**	**Upper CL**	**p-Value**
Northeast	West	3.529460	0.6070859	2.33435	4.724567	<.0001*
Northeast	WV-OH	2.331405	0.3360031	1.66995	2.992860	<.0001*
WV-OH	West	1.198054	0.6355008	-0.05299	2.449099	0.0605

When examining PC 2 for regional differences, Figure 14 gives an essentially inverse trend as shown in Figure 13. West Virginia-Ohio ranks most positive on PC 2, followed closely by the West, while the Northeast ranks just below zero with a negative score. Table 10 shows that the overall difference for this PC is significant across the regions, but the pairwise comparison shows it is the Northeast again that is significantly different from the other two regions (p<.0001; p=0.01). In this case, West Virginia-Ohio and the West are much more similar (p=0.49) and overlap greatly.

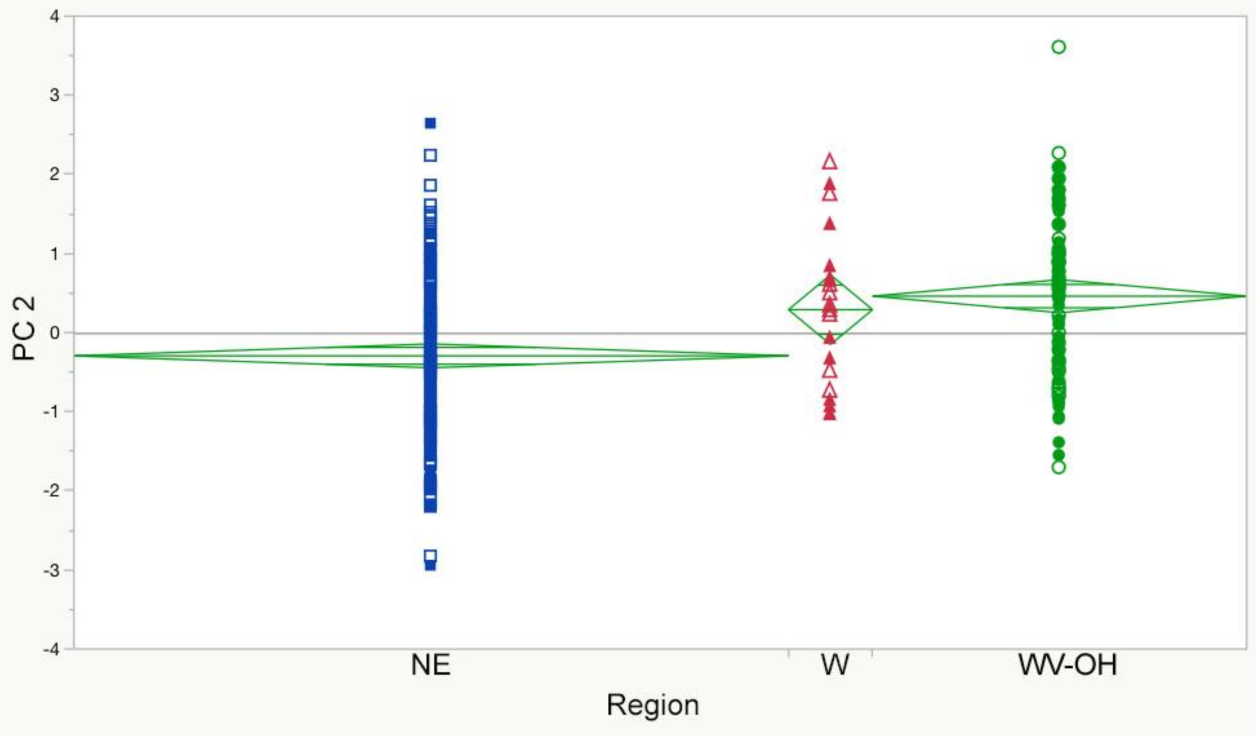


Figure 14. ANOVA on PC 2 split by region for coyotes.

Table 10. ANOVA summary for PC 2 split by region for coyotes. Star indicates significance at α = 0.05.

Source	DF	Sum of Squares	Mean Square	F Ratio	Prob > F
Model	2	35.195	17.598	17.536	<.0001*
Error	276	276.976	1.004		
C. Total	278	312.171			

Table 11. Ordered differences report for ANOVA on PC 2 split by region for coyotes. Star indicates significance at α = 0.05.

Level	- Level	Difference	Std Err Dif	Lower CL	Upper CL	p-Value
WV-OH	Northeast	0.754	0.131	0.496	1.012	<.0001*
West	Northeast	0.583	0.237	0.117	1.050	0.0144*
WV-OH	West	0.171	0.248	-0.317	0.659	0.4912

DISCUSSION

Sexual Dimorphism

Coyotes appear not to follow Bergmann's rule, which states body size increases with latitude (Meachen and Samuels 2012), and sexual dimorphism can vary by latitude, in that it is more likely when body size is more variable and resources are more limited by the seasons (Isaac 2005). This non-adherence to Bergmann's rule would suggest coyotes are likely not very sexually dimorphic; however, the results presented in this study give evidence to support sexual dimorphism in coyotes that is highly variable by region. Sexual dimorphism was first explored across regions, to discover if any significant dimorphism could be detected in coyotes as a whole. The first principle component shows a highly significant difference between males and females (Fig. 10). Because PC 1 is a size axis, this suggests males are overall larger in skull measures than females (which in turn suggests larger body size). No sexual dimorphism is apparent in PC 2 (Fig. 12). Because PC 1 illustrates such clear sexual dimorphism, it was of interest to explore how that varies by region. When split apart by region, it is evident that the Northeast is the most sexually dimorphic, followed by West Virginia and Ohio, and the West shows no evidence of sexual dimorphism (Fig. 11). The evidence of such strong sexual dimorphism could be attributed to Bergmann's rule; however it is more likely that the dramatic increase in size in this region is more attributable to introgression with wolves, though likely not the only push toward larger body size, as large prey is more readily available in the Northeast than elsewhere. Hybridization as the primary cause for rapid increase in size is also supported by the difference in size between the Northeast and West Virginia-Ohio, as both these regions are eastern forest and possess white-tailed deer populations. If the selection for larger size was primarily driven in the Northeast by predation on white-tailed deer, they would likely be much more similar in size to coyotes in West Virginia and Ohio, rather than being so significantly larger (Kays et al., 2010). The West represents the ancestral population, and is overall smaller in size, which is likely the reason sexual dimorphism is not present/apparent in the region. Smaller sample size (n<30) may also contribute to lack of evidence for sexual dimorphism, but based on previous data for western coyotes, it is more likely they are simply too small to show significant sexual dimorphism (Way, 2007). For example, sexual dimorphism in wolves increases with increasing body size (O'Keefe et al., 2013).

Scatterplots

Coyotes found in West Virginia and Ohio appear to have a unique trajectory pertaining to PC 2's response to PC 1. Figure 8 shows the density plots by region for PC 2 against PC 1, with reduced major axis regressions for each region. Both the Northeast and the West have regression lines with positive slopes of similar magnitude. The West has a stronger positive correlation between variation in PC 2 in response to PC 1 than the Northeast; however both correlations are still positive, whereas West Virginia-Ohio possesses a regression line with a negative correlation and negative slope (Table 3). Therefore, the size and shape trajectories of the Northeast and the West appear to be more similar to each other than either is to the West Virginia-Ohio.

Allometry

In terms of allometry, the regions both vary widely from one another in some measurements and maintain close similarities in others. Because the allometry vector was created based on PC 1, the eigenvalues for each measurement are all growth relative to overall size. All values are positive, meaning the features are all growing as the individual as a whole grows, but some grow more quickly than others. The largest region-based difference is the width of the mandible at the lower carnassial (WMm1). This measure, above any other, is the greatest positively allometric in coyotes of the West. While at a lesser value for the Northeast, this measure is also its most positively allometric value. For coyotes of West Virginia and Ohio, however, this particular measurement is one of their lesser values, and lies below the line of isometry. A wider mandibular area underneath the carnassial would suggest a higher propensity for the processing of meat, as this is the main shearing tooth. In the case of similar measures across all regions, the length of the upper carnassial (LP4) seems to be the most highly conserved. All three regions have eigenvalues for this measure that lie tightly together between 0.2 and 0.225. Again, carnassial size is related to meat processing, and in this case it seems the allometry of the upper carnassial is similar among all coyote populations (Van Valkenburgh, 2007).

In most measures, West Virginia-Ohio has PC scores more similar to the Northeast than to the West. Width of the anterior portion of the ramus, height of the coronoid process, height of the mandible at the lower carnassial, width of the post orbital processes, zygomatic width, greatest skull length, height of the jugal, and height from toothrow to orbit all show West Virginia-Ohio more similar to the Northeast, whereas West Virginia-Ohio are more similar to the West in only minimum width of the palate, maximum width of the palate, and length of the maxillary toothrow (Fig. 9). Most of the similarities in measurement to the Northeast are not directly tooth related, but seem to relate more to overall skull size (greatest skull length, for example). The similarities between West Virginia-Ohio and the West are all directly tooth related measures. However, not all tooth measurements show similarities between West Virginia-Ohio and the West, as the width of the upper second molar is positively allometric for the West, while it is quite negatively allometric for West Virginia-Ohio. A larger molar would be more indicative of a diet that required greater grinding of food and utilization of more of the carcass, including bones, which is a common occurrence in scavengers (Van Valkenburgh, 1987).

Regional differences

The main goal of this study was to explore similarities and differences among coyotes in West Virginia and two reference populations: the West (the more ancestral), and the Northeast (the highly admixed). The heart of this comparison was performed examining regional differences in the first two principle components. In the first principle component (the size axis), the Northeast lies in the positive space of the axis, whereas West Virginia-Ohio and the West place in the negative space (Fig. 13). The one-way ANOVA between regions was found to be significant (Table 8), but this must be broken down into pairwise comparisons. The Northeast is significantly different from both the West and West Virginia-Ohio, but the latter two are not significantly different from one another (Table 9). This difference suggests the coyotes of the Northeast are significant larger overall than those of the West and West Virginia and Ohio. In the second principle component (the tooth versus skull axis), West Virginia-Ohio and the West both lie in the positive space of the axis, while the Northeast lies just below zero in the negative space (Fig. 14). Again, the one-way ANOVA between regions was found to be significant (Table 10), but pairwise comparisons show that while the regions load different on this axis, the significance between regions is the same. The Northeast is significantly different from both the West and

West Virginia-Ohio, while the latter two are much more similar (Table 11). Because this axis loads the teeth against the rest of the skull, it appears that coyotes of West Virginia and Ohio have teeth more similar to those of the West, rather than the Northeast.

Conclusions

Overall regional differences in the first two principle components show size and shape similarities between the West and West Virginia-Ohio. These regions are not significantly different from each other, while they both are from the Northeast. This similarity would suggest West Virginia and Ohio coyotes are more similar to the more ancestral, "pure" coyote population in the West. However, coyotes of West Virginia and Ohio share more similarities with the Northeast than the region-only comparisons suggest. Taking into account the sexual dimorphic variation by region, West Virginia-Ohio is more similar to the Northeast in that they both show significant sexual dimorphism while the West does not. While the sexual dimorphism is not as significant in West Virginia-Ohio, it could be presented as an intermediate between the insignificant West and the highly significant Northeast. This regional difference follows with Way (2007), who found that coyote body size varies highly with longitude, in a west-to-east increasing gradient. While sexual dimorphism has been found to vary with latitude due to increasing body size (Isaac, 2005), adapting this to longitude is not a far stretch as this seems to be the axis of body size increase for coyotes. Moving on to allometry, West Virginia-Ohio is more similar to the Northeast, especially in cranial (non-tooth) measurements. Where similarities between the West and West Virginia-Ohio exist, they are directly tooth related. The allometry suggests the coyotes of West Virginia and Ohio are under pressure to possess larger, more quickly growing skulls relative to their overall size, while still possessing a tooth structure similar to the western "ancestral" population of coyotes. A larger skull may suggest a move toward more active hunting of larger prey rather than scavenging or hunting rodents. Coyotes of

West Virginia were found to rely heavily on white-tailed deer despite season (Crimmins et al., 2012). Scavenging of carcasses certainly plays a part, but this move to larger skull size rather than larger molar size (which would indicate fuller processing of a carcass, including the bones), suggests a move toward active predation. Crimmins et al. (2012) stated the high survivability of deer in the summer and active hunting of deer by humans in the late fall likely meant coyotes were primarily scavenging gut piles left over from human hunters in the fall and predating upon exclusively fawns in the summer. However, the results presented in this study combined with Crimmins' et al. (2012) habitat use findings, illustrating coyotes of the region prefer recently disturbed (logged) forest, which tend to have both a greater abundance of deer and more ground cover than heavily forested areas, suggest that some active predation of ungulates is occurring in West Virginia. Active predation on deer may be influenced by epigenetics, as vonHoldt, Heppenheimer, Petrenko, Croonquist, and Rutledge (2017) found epigenetic influences may pressure phenotypic changes more quickly than other factors or may actually inhibit the fitness of admixed individuals. Greater understanding of the role of epigenetics in the adaptation of coyotes to their various habitats may help to unravel the underlying mechanisms at play in West Virginia.

Introgression with either wolves or dogs could influence the size of coyote skulls in the West Virginia-Ohio region; however, it has been found that relatively low levels (≤16%) of (recent) hybridization have been detected in the region. Additionally, genetic variation was found in the region not attributable to a western reference group (Bohling et al., 2017). The distinct skull morphology detected in this study may be attributable to the unique genetic variation found by Bohling et al. (2017), suggesting the move toward a regional "ecomorph" of coyote, adapted more to forested regions rather than open grasslands, and hunting larger prey at a higher rate than that found in the West. Larger body size aids greatly in the taking of large prey such as white-tailed deer, as the greater the size of the predator, the more it weighs down its prey during an attack, tiring the prey out more quickly. This unique genetic variation would also suggest that the uniqueness of size and shape found in coyotes of the Northeast cannot entirely be attributed to introgression with wolves, as non-hybrid coyotes in the region are larger than those found in the West (Way, 2007). This increase in body size is likely also adaptive, not merely genetic. Comparing canine tooth size across regions would provide better resolution, as the canine teeth are primarily responsible for anchoring the predator to the prey during a hunt. Unfortunately, canine teeth were not measured in Kays et al. (2010), so no comparison could be made on this metric. Canine teeth would be an interesting morphological aspect to explore in future studies. The theories presented here would hold best if canine size were also shown to increase longitudinally from west to east, with northeastern coyotes who seem to actively hunt the largest prey possessing the largest canines, western coyotes possessing the smallest canines as they predate more frequently on small prey, and coyotes of West Virginia and Ohio having canines intermediate in size between the West and Northeast.

CHAPTER THREE

INTRODUCTION

Hybridization between coyotes and domestic dogs has been discovered to varying degrees in populations of coyotes across North America (Adams, Leonard, and Waits, 2003; Monzõn et al., 2014; vonHoldt et al., 2011). Across the regions examined, Ohio included, about 10% of coyote ancestry can be explained by domestic dog introgression (Monzõn et al., 2014; vonHoldt et al., 2011). Additionally, Bohling et al. (2017) found in a study performed in Virginia and West Virginia that 6-16% of individuals possessed admixture from either wolves or domestic dogs. Because some level of hybridization has been recorded for West Virginia (the area in question in this study), it was of interest to examine if the presence of domestic dog ancestry influenced coyote skull morphology between regions.

MATERIALS AND METHODS

The measurements taken in this study are listed in detail in Chapter Two (Table 1; Fig. 5). Those measurements were collected for the domestic dog skulls measured in this study as well. A set of 25 dog skulls were measured to be used as an additional comparison group. Twenty of the skulls are from the Marshall University teaching collection and the other five were purchased from Skulls Unlimited. Of the 25, two did not have a complete set of all 14 measurements, so analyses include 23 domestic dog skulls. The pattern of statistical analyses performed follows Chapter Two. First, the domestic dog measurement values were transformed to natural logarithm. Once these data were transformed, PCA was performed including coyotes, which were divided by region. Two scatterplots were generated, both of PC 2 dependent on PC 1. The first is a general scatterplot (Fig. 15), while the second groups all coyotes together with 95% density clouds and includes reduced major axis regressions for both the dog and coyote groups (Fig. 16; Table 12). The purpose of these figures is to compare how dogs and coyotes differ in relation to how the shape axis (PC 2) varies with the size axis (PC 1). One-way ANOVAs and Student's t-tests were performed on both PC 1 and PC 2 to examine how the various regional populations of coyotes compare to domestic dogs (Figs. 17 & 18).

RESULTS

Principle components analysis was run on the data, and the summary data is included in Table 12. Only the first two PCs, which account for 79.9% of the variation in the data, were used in analyses. These were the only components deemed biologically relevant and easy to interpret. As in Chapter Two, the first principle component is deemed at the "size" axis, as all the eigenvalues are positive. The second principle component is also designated as the "teeth relative to skull" shape axis because it is largely teeth measurements, such as LP4, WM2, and ALM which possess negative eigenvalues relative to the non-tooth bony parts of the skull.

Table 12. Summary of PCA on correlation matrix for coyotes and domestic dogs.

	PC1	PC2	PC3	PC4	PC5
Eigenvalue	9.717	1.468	0.620	0.426	0.353
Percent	69.409	10.489	4.427	3.040	2.525
Cumulative percent	69.409	79.898	84.324	87.364	89.888
LP4	0.268	-0.282	0.159	-0.094	0.005
WM2	0.231	-0.376	0.238	0.400	0.537
ALM	0.267	-0.391	-0.137	0.066	-0.111
M1O	0.291	0.026	-0.189	0.047	-0.314
HJ	0.273	0.123	0.017	-0.411	-0.302
GSL	0.291	-0.226	-0.205	0.150	-0.161
ZW	0.295	0.093	-0.233	-0.174	0.159
WPOP	0.221	0.387	-0.501	-0.014	0.565
MXP	0.270	0.305	0.197	0.125	0.024
MNP	0.192	0.541	0.241	0.495	-0.197
HMm1	0.296	-0.046	-0.186	0.059	-0.154
WMm1	0.263	0.046	0.527	-0.032	0.028
HCP	0.294	-0.079	-0.120	0.127	-0.138
WAR	0.266	0.066	0.304	-0.568	0.238

Figure 15 shows PC 2 plotted against PC 1. Domestic dogs lie more positively on the "shape" axis (PC 2), but span the entire range of "size" (PC 1) values. Coyotes, on the other hand, even when split by region as they are, clump tightly together in approximately the same region of the plot: more positively on the size axis and more negatively on the shape axis (as compared to dogs). The wide span of dogs on PC 1 supports the high size variability seen across the different breeds of domestic dogs (breeds of dogs measured in this case were unknown, but size was highly variable). Coyotes grouping together away from dogs is also to be expected, despite the regional differences explored in Chapter Two, as they are still members of the same species, and domestic dogs are *C. lupus*.

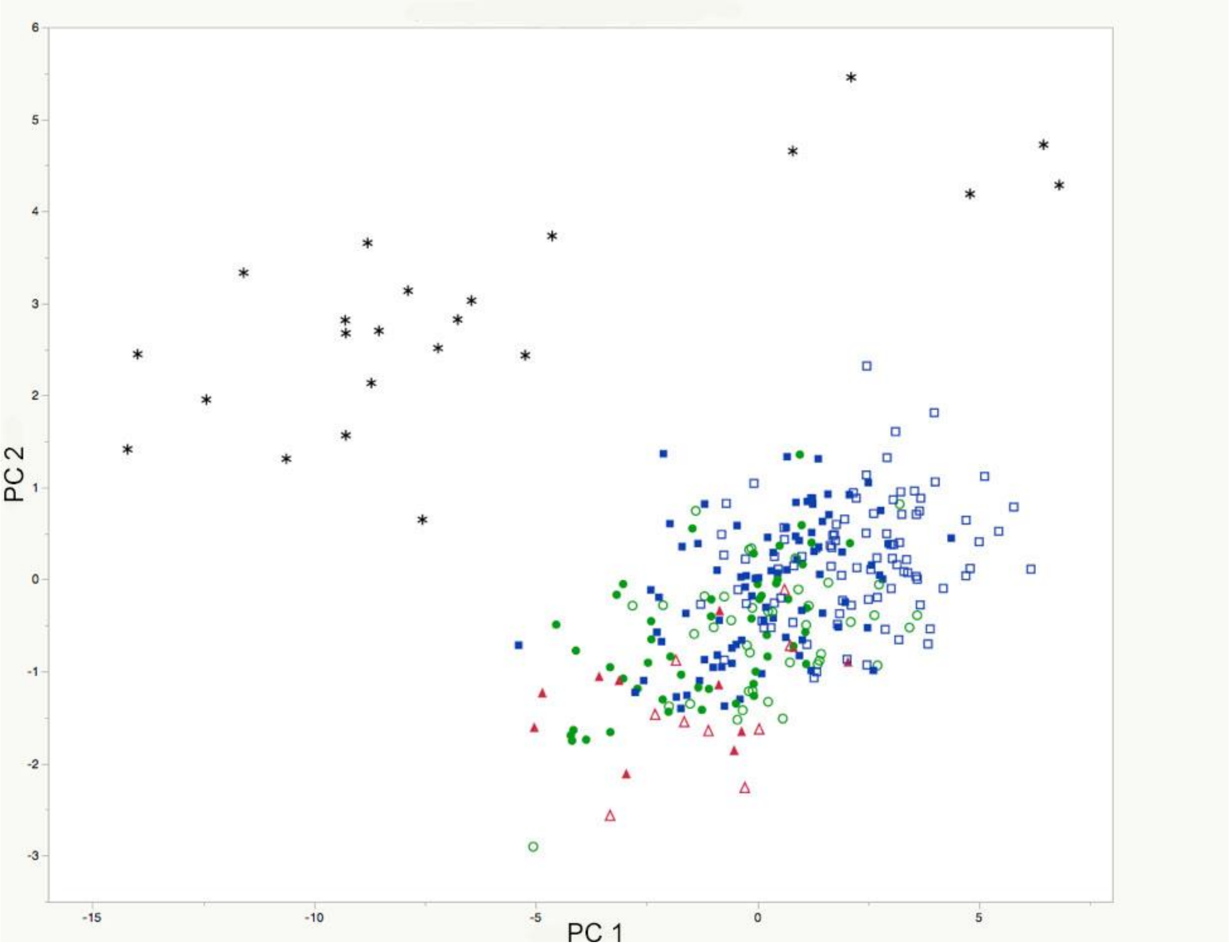


Figure 15. Scatterplot of PC 2 plotted against PC 1 for coyotes and domestic dogs. Stars represent domestic dogs, blue squares represent Northeast coyotes, red triangles represent West coyotes, and green circles represent West Virginia and Ohio coyotes.

Figure 16 is another plot of PC 2 varying with PC 1, but in this case 95% density clouds and reduced major axis regression lines have been added. In this figure, coyotes are regressed as one group rather than split by region. The relative trajectories of both groups are fairly different, with coyotes having a much steeper trajectory than dogs. While both possess regression lines with positive slopes, the slopes are significantly different. The slope of the regression line for dogs is 0.19, while it is 0.38 for coyotes. The strength of the correlation for dogs is greater than it is for coyotes (0.76); however, the correlation for coyotes is still strong (0.55). Both species support a positive relationship between PC 1 and PC 2, though their trajectories lie in different spaces in the plot, clearly showing two distinct groups.

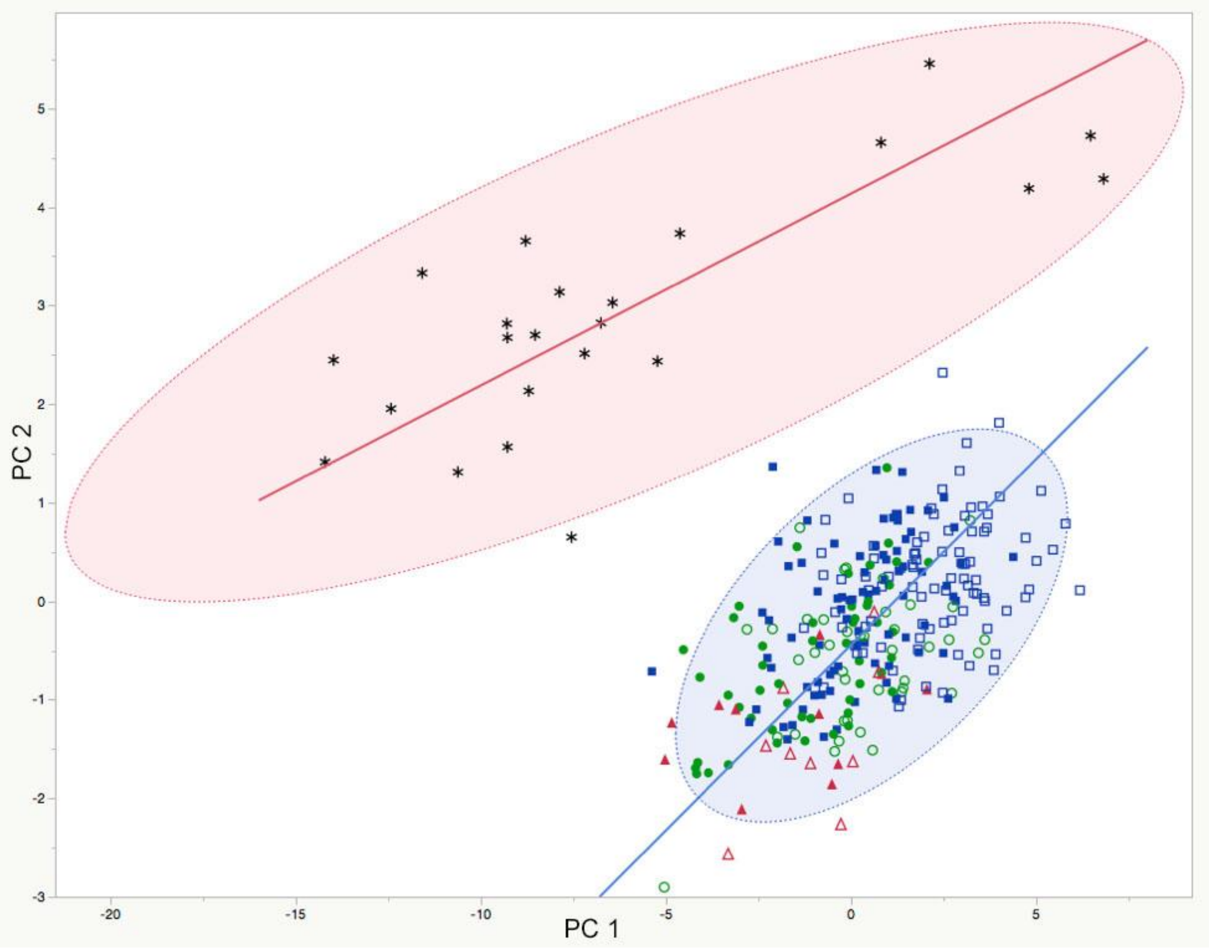


Figure 16. Scatterplot of PC 2 plotted against PC 1 with 95% density clouds and reduced major axis regressions for coyotes and domestic dogs. Blue represents domestic dogs and red represents coyotes.

Table 13. Summary data for the reduced major axis regressions for domestic dogs and coyotes.

	Variable	**Mean**	**Std. Dev.**	**Variance Ratio**	**Correlation**
Domestic dogs **Fit Ratio =** 0.038	PC 1	-6.15156	6.165551	0.037939	0.7619
	PC 2	2.94001	1.200922		
	Intercept	**Slope**	**Lower CL**	**Upper** CL	**Alpha**
	4.138206	0.194779	0.129675	0.29257	0.05
	Variable	**Mean**	**Std. Dev.**	**Variance Ratio**	**Correlation**
Coyotes **Fit Ratio** = 0.142	PC 1	0.523986	2.159906	0.142109	0.5520
	PC 2	-0.23986	0.814227		
	Intercept	**Slope**	**Lower CL**	**Upper CL**	**Alpha**
	-0.43739	0.376973	0.314677	0.451602	0.05

Separating coyotes into regional groups again, a one-way ANOVA was performed on PC 1 (Fig. 17). Compared to the widespread group of dogs, coyotes cluster much more tightly together and in a similar space to one another. The ANOVA is highly significant (p<.0001; Table 14), and the pairwise comparisons (Table 15) show that in fact, all groups are significantly different from one another (all p<.0001) except West and West Virginia-Ohio (p=0.13). The mean for dogs lies in the negative space of the size axis, making them the group with the overall smallest skulls, followed by West and West Virginia-Ohio, and finally the Northeast, which is the largest.

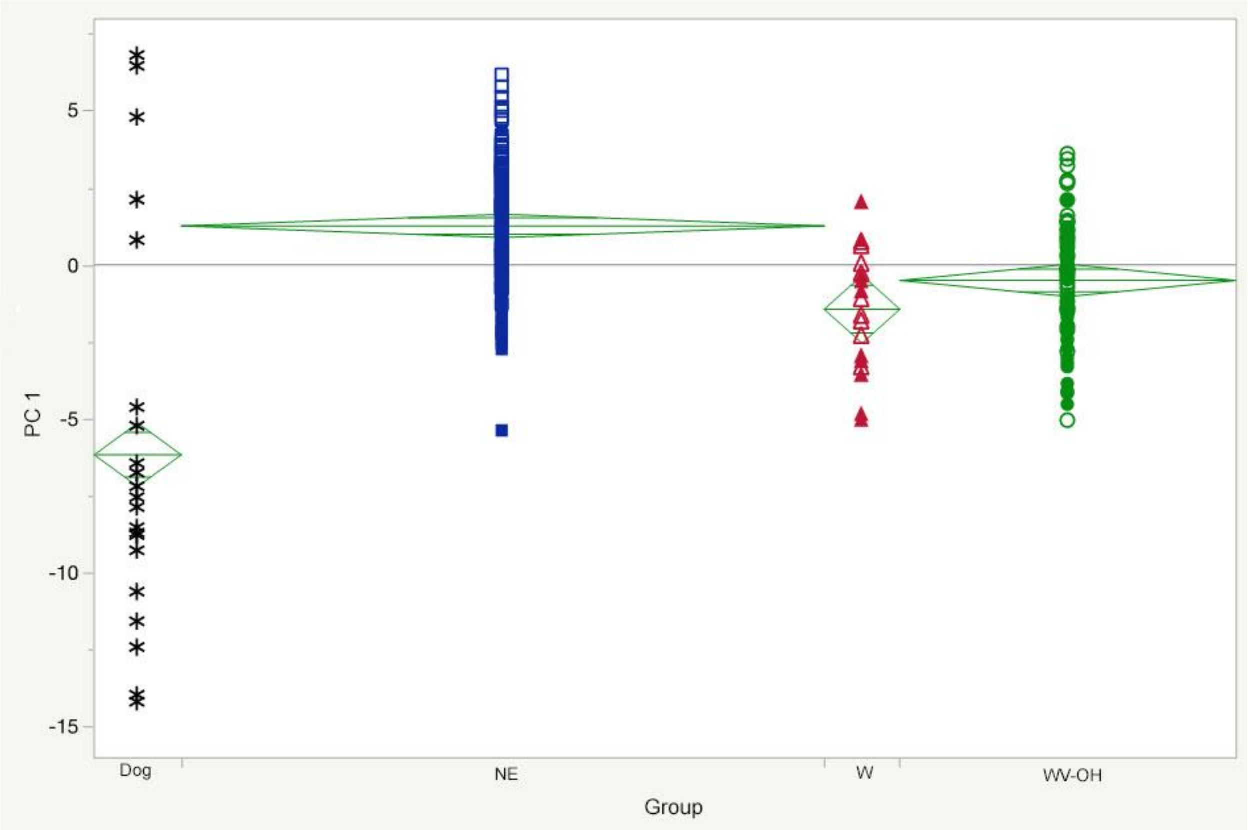


Figure 17. ANOVA on PC 1 split by region including domestic dogs.

Table 14. ANOVA summary for PC 1 split by region including domestic dogs. A star indicates significant at α = 0.05.

Source	**DF**	**Sum of Squares**	**Mean Square**	**F Ratio**	**Prob > F**
Region	3	1211.393	403.798	64.392	<.0001*
Error	298	1868.726	6.271		
C. Total	301	3080.119			

Table 15. Ordered differences report for ANOVA on PC 1 split by region including domestic dogs. A star indicates significant at α = 0.05.

Level	**- Level**	**Difference**	**Std Err Dif**	**Lower CL**	**Upper CL**	**p-Value**
Northeast	Dog	7.434	0.556	6.339	8.529	<.0001*
Northeast	West	2.704	0.592	1.539	3.869	<.0001*
Northeast	WV-OH	1.769	0.328	1.125	2.414	<.0001*
West	Dog	4.729	0.766	3.223	6.236	<.0001*
WV-OH	Dog	5.664	0.586	4.512	6.817	<.0001*
WV-OH	West	0.935	0.620	-0.285	2.154	0.1324

A second one-way ANOVA was performed on PC 2 (Fig. 18). This shape axis has more variability across all the groups than PC 1. The ANOVA is highly significant for this PC as well (p<.0001; Table 16), and the pairwise comparison shows a highly significant difference between every region (Table 17). If the interpretation of this axis as the teeth against the rest of skull holds as it did for coyotes alone, this means the domestic dogs have proportionally small teeth compared to the rest of their skull, while coyotes either have teeth in relative proportion to their overall skull size, or teeth proportionally larger.

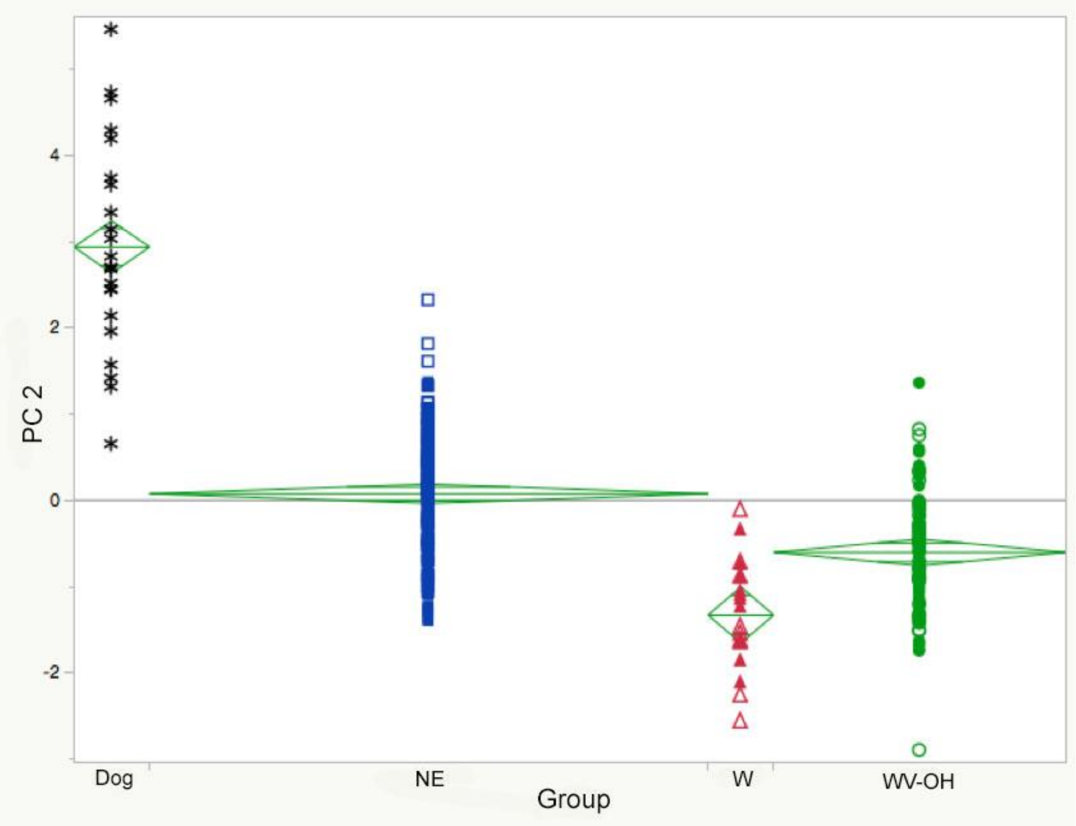


Figure 18. ANOVA on PC 2 split by region including domestic dogs.

Table 16. ANOVA summary for PC 2 split by region including domestic dogs. A star indicates significant at α = 0.05.

Source	DF	Sum of Squares	Mean Square	F Ratio	Prob > F
Region	3	267.385	89.128	162.446	<.0001*
Error	298	163.502	0.549		
C. Total	301	430.887			

Table 17. Ordered differences report for PC 2 split by region including domestic dogs. A star indicates significant at $\alpha = 0.05$.

Level	- Level	Difference	Std Err Dif	Lower CL	Upper CL	p-Value
Dog	West	4.269	0.226	3.823	4.715	<.0001*
Dog	WV-OH	3.542	0.173	3.201	3.883	<.0001*
Dog	Northeast	2.862	0.165	2.538	3.186	<.0001*
Northeast	West	1.407	0.175	1.062	1.751	<.0001*
Northeast	WV-OH	0.680	0.097	0.489	0.870	<.0001*
WV-OH	West	0.727	0.183	0.366	1.088	<.0001*

DISCUSSION

When comparing domestic dogs to coyotes, no true similarities can be gleaned from these results. The domestic dogs measured in this study are widely different in both shape and size from any of the regional populations of coyotes. They were the most distant group among the four groups examined in each of the tests performed. The slopes of the trajectories are fairly different, the dog slope being 0.19 relative to the 0.38 slope for coyotes. Because greatest skull length and most of the teeth measurements score negatively for PC 2, the pressure for dogs to elongate their skull as they get larger is not as strong in domestic dogs as it is for coyotes. Coyotes additionally have larger teeth than dogs, as plotting closer to the negative space of the PC axis for values that score negatively means those measures are greater in size (Fig. 16; Table 13). It follows that if dogs have relatively shorter skulls and therefore shorter muzzles, their teeth must be smaller in order to properly fit in their mouths. As most of the width measurements score positively, this indicates domestic dogs also have wider skulls relative to size as compared to coyotes, which have narrower skulls (Fig. 16; Table 11).

For the one-way ANOVA on PC 1, all groups were significantly different from one another, except for West and West Virginia-Ohio. This being the size axis, the Northeast appears the largest, followed by West and West Virginia-Ohio, and domestic dogs being the smallest (Fig. 17). In this case, domestic dog skulls appear to be more similar in overall size to the West and West Virginia-Ohio, which follows with the body sizes reported in Thurber and Peterson

(1991), indicating coyotes in the western United States are smaller than those in the Northeast. This greater similarity in size between coyotes from the West and West Virginia-Ohio may have nothing to do with domestic dog introgression, as coyotes in the Northeast have been found to have only slightly less ancestry attributed to domestic dogs than Ohio (~9% versus ~10%), and it seems unlikely a one percent increase in domestic dog ancestry would create such a difference in size between the regional populations of coyotes (Monzõn et al., 2014; vonHoldt et al., 2011). More than half of the domestic dog skulls measured in this study were quite small, which is likely a contributing factor.

For the one-way ANOVA on PC 2, all groups were significantly different from each other; however, the Northeastern coyotes lie closer to domestic dogs than the other two regions do (Fig. 18). This component is the teeth relative to the rest of the skull. Domestic dogs appear to have a very distinct shape about them on this axis, likely as a consequence of the artificial selection humans placed (and continue to place) on the species. The wide variance witnessed in the dog group is also attributable to this artificial selection, as there are many different “breeds” of dogs with huge variations in size, shape, and color. The closer placement of Northeastern coyotes to domestic dogs on this PC may be related to their relatedness as species. More ancestral-type dogs, such as fossil specimen, pointers, and dingoes, show greater skull similarities to wolves than other more derived variants of dogs do (Geiger et al., 2017). Geiger et al. (2017) suggested differences between domestic dogs and wolves are the result of some mix between neomorphosis (resulting in novel morphology) and paedomorphosis (a developmental shift in the ancestral morphology).

CONCLUSIONS

Combining the varied genetic studies examining domestic dog introgression with this morphological study suggests the low level of dog ancestry in coyotes is having little to no impact on their skull morphology. The closer similarity on PC 1 of West and West Virginia-Ohio coyotes to dogs has likely little to nothing to do with introgression and more to do with the naturally smaller size of these populations relative to the wolf-introgressed Northeastern coyotes, which is supported by the similar levels of domestic dog introgression found between the West and West Virginia-Ohio. It is possible any morphological impact the dog ancestry might have in the Northeastern population is being masked by the wolf ancestry. The high statistical significance separating West and West Virginia-Ohio from dogs, however, reinforces the idea that dogs are having little if any impact on these coyotes, as you might expect to see much greater similarity if they were. As stated previously, the Northeastern coyotes were more similar to domestic dogs on PC 2, the tooth axis. Domestic dogs are thought to be descended from wolves, and therefore these two species are more closely related than coyotes are to either of the other two (Vilà et al., 1997). Teeth size and structure obviously play a part in diet, and though the majority of dogs are not pack hunters of ungulates, they did descend from this type of diet. Tooth structure would not change much unless there was pressure to do so, and though artificial selection by humans has done much to change dogs morphologically, pressure to change the teeth may have resulted in changes that still bear some resemblance to wolves greater than that of their resemblance to coyotes. It is important to note, however, that domestic dogs are highly significantly different from both coyotes and wolves, and are distinguishable as their own, distinct group.

Because the domestic dog skulls measured for this study were mostly smaller overall and of unknown breeds, they are not a reliable representation of domestic dogs as a whole. To more reliably explore similarities and differences in morphology between domestic dogs and coyotes, it would be pertinent to measure dogs of known breeds and to include a wide range of breeds, spanning a wide range of sizes. A sample inclusive of many different breeds and sizes of domestic dog will still likely show a distinct morphology, but comparison will be more reliable than included in this study.

Any differences illustrated between regional populations of coyotes seem much more likely to be either adaptive to their respective habitats or due to the Northeastern population’s introgression with wolves, rather than any previous introgression that occurred with domestic dogs. The highly significant differences between domestic dogs and coyotes reduces dog introgression as a major morphological influence. Adaptation to habitat occurring in West Virginia, as well as in the Northeast, coupled with the widespread integration of wolf genes into the coyotes in the Northeast, seem to be the most important factors at play when it comes to regional morphological differences.

REFERENCES

Adams, J. R., Leonard, J. A., Waits, L. P. (2003). Widespread occurrence of a domestic dog mitochondrial DNA haplotype in southeastern US coyotes. *Molecular Ecology*, *12*(2), 541–546. https://doi.org/10.1046/j.1365-294X.2003.01708.x

Allee, W. C., Park, O., Emerson, A. E., Park, T., & Schmidt, K. P. (1949). *Principles of animal ecology*. W.B. Saundere Co. Ltd.

Alroy, J. (1998). Cope's Rule and the Dynamics of Body Mass Evolution in North American Fossil Mammals. *Science*, *280*, 731–734. https://doi.org/10.1126/science.280.5364.731

Atwood, T. C., Weeks, H. P., & Gehring, T. M. (2004). Spatial ecology of coyotes along a suburban-to-rural gradient. *Journal of Wildlife Management*, *68*(4), 1000–1009. https://doi.org/10.2193/0022-541X(2004)068

Bartolini Lucenti, S., Alba, D. M., Rook, L., Moyà-Solà, S., & Madurell-Malapeira, J. (2017). Latest Early Pleistocene wolf-like canids from the Iberian Peninsula. *Quaternary Science Reviews*, *162*, 12–25. https://doi.org/10.1016/j.quascirev.2017.02.023

Bekoff, M. (2001). *Coyotes: Biology, Behavior, and Management*. New York: Academic Press.

Bekoff, M., & Gese, E. M. (2003). Coyote (Canis latrans). *Wild Mammals of North America: Biology, Management, and Conservation*, 467–481.

Bekoff, M., & Wells, M. C. (1981). Behavioral budgeting by wild coyotes: The influence of food resources and social organization. *Animal Behaviour*, *29*, 794–801.

Benson, J. F., & Patterson, B. R. (2013). Moose (Alces alces) predation by eastern coyotes (Canis latrans) and eastern coyote × eastern wolf (Canis latrans × Canis lycaon) hybrids. *Canadian Journal of Zoology*, *91*(October), 837–841. https://doi.org/10.1139/cjz-2013-0160

Benson, J. F., Patterson, B. R., & Wheeldon, T. J. (2012). Spatial genetic and morphologic structure of wolves and coyotes in relation to environmental heterogeneity in a Canis hybrid zone. *Molecular Ecology*, *21*(24), 5934–5954. https://doi.org/10.1111/mec.12045

Berger, K. M., & Gese, E. M. (2007). Does interference competition with wolves limit the distribution and abundance of coyotes? *Journal of Animal Ecology*, *76*(6), 1075–1085. https://doi.org/10.1111/j.1365-2656.2007.01287.x

Bogan, D. A. (2004). *Eastern coyote home range, habitat selection and survival in the Albany Pine Bush landscape.* State University of New York, Albany.

Bohling, J. H., Mastro, L. L., Adams, J. R., Gese, E. M., Owen, S. F., & Waits, L. P. (2017). Panmixia and limited interspecific introgression in coyotes (Canis latrans) from West Virginia and Virginia, USA. *Journal of Heredity*, *108*(6), 608–617.

Bozarth, C. A., Hailer, F., Rockwood, L. L., Edwards, C. W., & Maldano, J. E. (2011). Coyote colonization of northern Virginia and admixture with Great Lakes wolves. *Journal of Mammalogy*, *92*(5), 1070–1080. https://doi.org/10.1644/10-MAMM-A-223.1

Cardillo, M., Mace, G. M., Jones, K. E., Bielby, J., Bininda-Emonds, O. R. P., Sechrest, W., … Purvis, A. (2005). Multiple Causes of High Extinction Risk in Large Mammal Species. *Science*, *309*(5738), 1239–1241. https://doi.org/10.1126/science.1116030

Cope, E. D. (1886). *The Origin of the Fittest: Essays on Evolution*. New York: D. Appleton and Company.

Crête, M., Ouellet, J.-P., Tremblay, J.-P., & Arsenault, R. (2001). Suitability of the forest landscape for coyotes in norteastern North America and its implications for coexistence with other carnivores. *Ecoscience*, *8*(3), 311–319. https://doi.org/10.1080/11956860.2001.11682658

Crimmins, S. M., Edwards, J. W., & Houben, J. M. (2012). Canis latrans (Coyote) Habitat Use and Feeding Habits in Central West Virginia. *Northeastern Naturalist*, *19*(3), 411–420. https://doi.org/10.2307/41684250

Fuller, T. K., Destefano, S., & Warren, P. (2010). Carnivore Behavior and Ecology, and Relationship to Urbanization. *Urban Carnivores: Ecology, Conflict, and Conservation*.

Gehrt, S. D. (2007). Ecology of coyotes in urban landscapes. In *12th Wildlife Damage Management Conference* (pp. 303–311).

Geiger, M., Evin, A., Sánchez-Villagra, M. R., Gascho, D., Mainini, C., & Zollikofer, C. P. E. (2017). Neomorphosis and heterochrony of skull shape in dog domestication. *Scientific Reports*, *7*(1), 13443. https://doi.org/10.1038/s41598-017-12582-2

Gese, E. M., & Grothe, S. (1995). Analysis of Coyote Predation on Deer and Elk during Winter in Yellowstone National Park , Wyoming. *The American Midland Naturalist*, *133*(1), 36–43. https://doi.org/10.2307/2426345

Gould, S. J. (1966). Allometry and Size in Ontogeny and Phylogeny. *Biological Reviews*, *41*(4), 587–638. https://doi.org/10.1111/j.1469-185X.1966.tb01624.x

Grubbs, S. E., & Krausman, P. R. (2009). Observations of Coyote-Cat Interactions. *Journal of Wildlife Management*, *73*(5), 683–685.

Hill, E. P., Sumner, P. W., & Wooding, J. B. (1987). Human Influences on Range Expansion of Coyotes in the Southeast. *Wildlife Society Bulletin*, *15*(4), 521–524.

Hillis, T. L., & Mallory, F. F. (1996). Sexual dimorphism in wolves (Canis lupus) of the Keewatin District, Northwest Territories, Canada. *Canadian Journal of Zoology-Revue Canadienne de Zoologie*, *74*(4), 721–725. https://doi.org/10.1139/z96-081

Isaac, J. L. (2005). Potential causes and life-history consequences of sexual size dimorphism in mammals. *Mammal Review*, *35*(1), 101–115. https://doi.org/10.1111/j.1365-2907.2005.00045.x

IUCN. (2008). Canis aureus. Retrieved from http://maps.iucnredlist.org/map.html?id=3744

IUCN. (2014a). Canis adustus. Retrieved from http://maps.iucnredlist.org/map.html?id=3753

IUCN. (2014b). Canis mesomelas. Retrieved from http://maps.iucnredlist.org/map.html?id=3755

Jackson, H. H. T. (1951). *The Clever Coyote*. (S. P. Young & H. H. T. Jackson, Eds.). Washington, D.C.: Wildlife Management Institute.

Jolicoeur, P. (1963). The multivariate generalization of the allometry equation. *Biometrics*, *19*, 497–499. https://doi.org/10.2307/2527939

Kays, R., Curtis, A., & Kirchman, J. J. (2010). Rapid adaptive evolution of northeastern coyotes via hybridization with wolves. *Biology Letters*, *6*(1), 89–93. https://doi.org/10.1098/rsbl.2009.0575

Kays, R. W., Gompper, M. E., & Ray, J. C. (2008). Landscape ecology of eastern coyotes based on large-scale estimates of abundance. *Ecological Applications*, *18*(4), 1014–1027. https://doi.org/10.1890/07-0298.1

Kennedy, M. L., Mech, S. G., Tran, B., Grubaugh, J. W., & Lance, R. F. (2003). An assessment of geographic variation in sexual size dimorphism in the coyote (Canis latrans). *Mammalia*, *67*, 411–417. https://doi.org/10.1515/mamm.2003.67.3.411

Kreeger, T. J. (2003). The internal wolf: physiology, pathology, and pharamcology. In *Wolves: Behavior, Ecology, and Conservation*. Chicago and London: The University of Chicago Press.

Kurtén, B. (1974). A history of coyote-like dogs (Canidae, Mammalia). *Acta Zoologica Fennica*, *140*, 42. https://doi.org/10.1093/brain/122.11.2197-a

Lehman, N., Eisenhawer, A., Hansen, K., Mech, L. D., Peterson, R. O., Gogan, P. J. P., & Wayne, R. K. (1991). Introgression of Coyote Mitochondrial DNA Into Sympatric North American Gray Wolf Populations. *Evolution*, *45*(1), 104–119.

Lehman, N., & Wayne, R. K. (1991). Analysis of coyote mitochondrial DNA genotype frequencies: Estimation of the effective number of alleles. *Genetics*, *128*(2), 405–416.

Lukasik, V. M., & Alexander, S. M. (2011). Human-Coyote interactions in Calgary, Alberta. *Human Dimensions of Wildlife*, *16*(2), 114–127. https://doi.org/10.1080/10871209.2011.544014

Meachen, J. A., Janowicz, A. C., Avery, J. E., & Sadleir, R. W. (2014). Ecological changes in coyotes (canis latrans) in response to the ice age megafaunal extinctions. *PLoS ONE*, *9*(12), 1–15. https://doi.org/10.1371/journal.pone.0116041

Meachen, J. a., & Samuels, J. X. (2012). Evolution in coyotes (Canis latrans) in response to the megafaunal extinctions. *Proceedings of the National Academy of Sciences*, *109*(11), 11–16. https://doi.org/10.1073/pnas.1113788109

Mech, L. D., Fritts, S. H., Radde, G. L., & Paul, W. J. (1988). Wolf distribution and road density in Minnesota. *Wildlife Society Bulletin*, *16*(1), 85–87. https://doi.org/3

Meloro, C., Hudson, A., & Rook, L. (2015). Feeding habits of extant and fossil canids as determined by their skull geometry. *Journal of Zoology*, *295*(3), 178–188. https://doi.org/10.1111/jzo.12196

Meloro, C., & Louys, J. (2015). Ecomorphology of radii in Canidae: Application to fragmentary fossils from Plio-Pleistocene hominin assemblages. *Acta Palaeontol. Pol*, *60*(4), 795–806. https://doi.org/10.4202/app.00080.2014

Merkle, J. A., Stahler, D. R., & Smith, D. W. (2009). Interference competition between gray wolves and coyotes in Yellowstone National Park. *Canadian Journal of Zoology*, *87*(1), 56–63. https://doi.org/10.1139/Z08-136

Miller, B. J., Harlow, H. J., Harlow, T. S., Biggins, D., & Ripple, W. J. (2012). Trophic cascades linking wolves (Canis lupus), coyotes (Canis latrans), and small mammals. *Canadian Journal Of Zoology-Revue Canadienne De Zoologie*, *90*(1), 70–78. https://doi.org/10.1139/z11-115

Moehlman, P. D. (1989). Intraspecific Variation in Canid Social Systems. In J. L. Gittleman (Ed.), *Carnivore Behavior, Ecology, and Evolution* (pp. 143–163). Boston, MA.

Monzõn, J., Kays, R., & Dykhuizen, D. E. (2014). Assessment of coyote-wolf-dog admixture using ancestry-informative diagnostic SNPs. *Molecular Ecology*, *23*(1), 182–197. https://doi.org/10.1111/mec.12570

Morey, P. S. (2004). *Landscape use and diet of coyotes, Canis latrans, in the Chicago Metropolitan Area*. Utah State University, Logan.

Morey, P. S., Gese, E. M., & Gehrt, S. D. (2007). Spatial and Temporal Variation in the Diet of Coyotes in the Chicago Metropolitan Area. *The American Midland Naturalist*, *158*(1), 147–161. https://doi.org/10.1674/0003-0031(2007)158[147:SATVIT]2.0.CO;2

Nowak, R. M. (1978). Evolution and Taxonomy of Coyotes and Related Canis. In M. Bekoff (Ed.), *Coyotes: Biology, Behavior, and Management* (pp. 3–16). New York: Academic Press.

O’Keefe, F. R., Binder, W. J., Frost, S. R., Sadlier, R. W., & Van Valkenburgh, B. (2014). Cranial morphometrics of the dire wolf, Canis dirus, at Rancho La Brea: temporal variability and its links to nutrient stress and climate. *Palaeontologia Electronica*, *17*(1), 1–23.

O’Keefe, F. R., Meachen, J., Fet, E. V., & Brannick, A. (2013). Ecological determinants of clinal morphological variation in the cranium of the North American gray wolf. *Journal of Mammalogy*, *94*(6), 1223–1236. https://doi.org/10.1644/13-MAMM-A-069

Patterson, B. R., Benjamin, L. K., & Messier, F. (1998). Prey switching and feeding habits of eastern coyotes in relation to snowshoe hare and white-tailed deer densities. *Canadian Journal of Zoology*, *76*, 1885–1897. https://doi.org/10.1139/z98-135

Pires, M. M., Silvestro, D., & Quental, T. B. (2015). Continental faunal exchange and the asymmetrical radiation of carnivores. *Proceedings of the Royal Society B: Biological Sciences*, *282*(1817), 20151952. https://doi.org/10.1098/rspb.2015.1952

Poessel, S. A., Breck, S., Teel, T. L., Shwif, S., & Crooks, K. R. (2012). Patterns of Human – Coyote Conflicts in the Denver Metropolitan Area. *The Journal of Wildlife Management*, *77*(2), 297–305. https://doi.org/10.1002/jwmg.454

Reyment, R. A., & Jvreskog, K. G. (1996). *Applied factor analysis in the natural sciences*. Cambridge University Press.

Ripple, W. J., Wirsing, A. J., Wilmers, C. C., & Letnic, M. (2013). Widespread mesopredator effects after wolf extirpation. *Biological Conservation*, *160*, 70–79. https://doi.org/10.1016/j.biocon.2012.12.033

Schrecengost, J. D., Kilgo, J. C., Mallard, D., Ray, H. S., & Miller, K. V. (2008). Seasonal Food Habits of the Coyote in the South Carolina Coastal Plain. *Southeastern Naturalist*, *7*(1), 135–144. https://doi.org/10.1656/1528-7092(2008)7[135:SFHOTC]2.0.CO;2

Stanley, S. M. (1973). An explanation for Cope’s Rule. *Evolution*, *27*(March), 1–26.

Tedford, R. H., Wang, X., & Taylor, B. E. (2009). Phylogenetic Systematics of the North American Fossil Caninae (Carnivora: Canidae). *Bulletin of the American Museum of Natural History*, *325*(325), 1–218. https://doi.org/10.1206/574.1

Thiel, R. P. (1985). Relationship between Road Densities and Wolf Habitat Suitability in Wisconsin. *American Midland Naturalist*, *113*(2), 404–407.

Thiel, R. P. (2006). Conditions for sexual interactions between wild Grey Wolves, Canis lupus, and Coyotes, Canis latrans. *Canadian Field-Naturalist*, *120*(1), 27–30. https://doi.org/Article

Thurber, J. M., & Peterson, R. O. (1991). Changes in Body Size Associated with Range Expansion in the Coyote (Canis latrans). *Journal of Mammalogy*, *72*(4), 750–755.

Timm, R. M., & Baker, R. O. (2007). A history of urban coyote problems. In *12th Wildlife Damage Management Conference* (pp. 272–286).

Van Valkenburgh, B. (1987). Canine tooth strength and large carnivores, *212*, 379_397.

Van Valkenburgh, B. (1988). Trophic diversity in past and present guilds of large predatory mammals. *Paleobiology*, *2*(January 1985), 155–173. https://doi.org/10.1017/S0094837300011891

Van Valkenburgh, B. (2007). Déjà vu: The evolution of feeding morphologies in the Carnivora. *Integrative and Comparative Biology*, *47*(1), 147–163. https://doi.org/10.1093/icb/icm016

Van Valkenburgh, B., & Hertel, F. (1993). Tough Times at La Brea: Tooth Breakage in Large Carnivores of the Late Pleistocene. *Science*, *261*, 456–459. https://doi.org/10.1126/science.261.5120.456

Van Valkenburgh, B., Wang, X., & Damuth, J. (2004). Cope's Rule, Hypercarnivory, and Extinction in North American Canids. *Science*, *306*(5693), 101–104.

Vilà, C., Savolainen, P., Maldonado, J. E., Amorim, I. R., Rice, J. E., Honeycutt, R. L., … Wayne, R. K. (1997). Multiple and Ancient Origins of the Domestic Dog. *Science*, *2761687*(1687), 1687–1689. https://doi.org/10.1126/science.276.5319.1687

VonHoldt, B., Heppenheimer, E., Petrenko, V., Croonquist, P., & Rutledge, L. Y. (2017). Ancestry-specific methylation patterns in admixed offspring from an experimental coyote and gray wolf cross. *Journal of Heredity*, *108*(4), 341–348. https://doi.org/10.1093/jhered/esx004

VonHoldt, B. M., Pollinger, J. P., Earl, D. A., Knowles, J. C., Boyko, A. R., Parker, H., … Wayne, R. K. (2011). A genome-wide perspective on the evolutionary history of enigmatic wolf-like canids. *Genome Research*, *21*(8), 1294–1305. https://doi.org/10.1101/gr.116301.110

Way, J. G. (2007). A comparison of body mass of Canis latrans (coyotes) between eastern and western North America. *Northeastern Naturalist*, *14*(1), 111–124.

Weeks, John L.; Tori, Gildo M.; Shieldcastle, M. C. T. (1990). Coyotes (Canis latrans) in Ohio. *Ohio Journal of Science*, *90*(5), 142–145.

Wheeldon, T., & White, B. N. (2009). Genetic analysis of historic western Great Lakes region wolf samples reveals early *Canis lupus/lycaon* hybridization. *Biology Letters*, *5*(1), 101–104. https://doi.org/10.1098/rsbl.2008.0516

White, L. A., & Gehrt, S. D. (2009). Coyote attacks on humans in the United States and Canada. *Human Dimensions of Wildlife*, *14*(6), 419–432. https://doi.org/10.1080/10871200903055326

Williams, S. L., McLaren, S. B., & Burgwin, M. A. (1985). Palaeo-archaeological and historical records of selected Pennsylvania mammals. *Animals of Carnegie Museum*, *54*, 77–188.

Wilson, P. J., Grewal, S. K., Mallory, F. F., & White, B. N. (2009). Genetic characterization of hybrid wolves across ontario. *Journal of Heredity*, *100*(Supplement 1), S80–S89. https://doi.org/10.1093/jhered/esp034

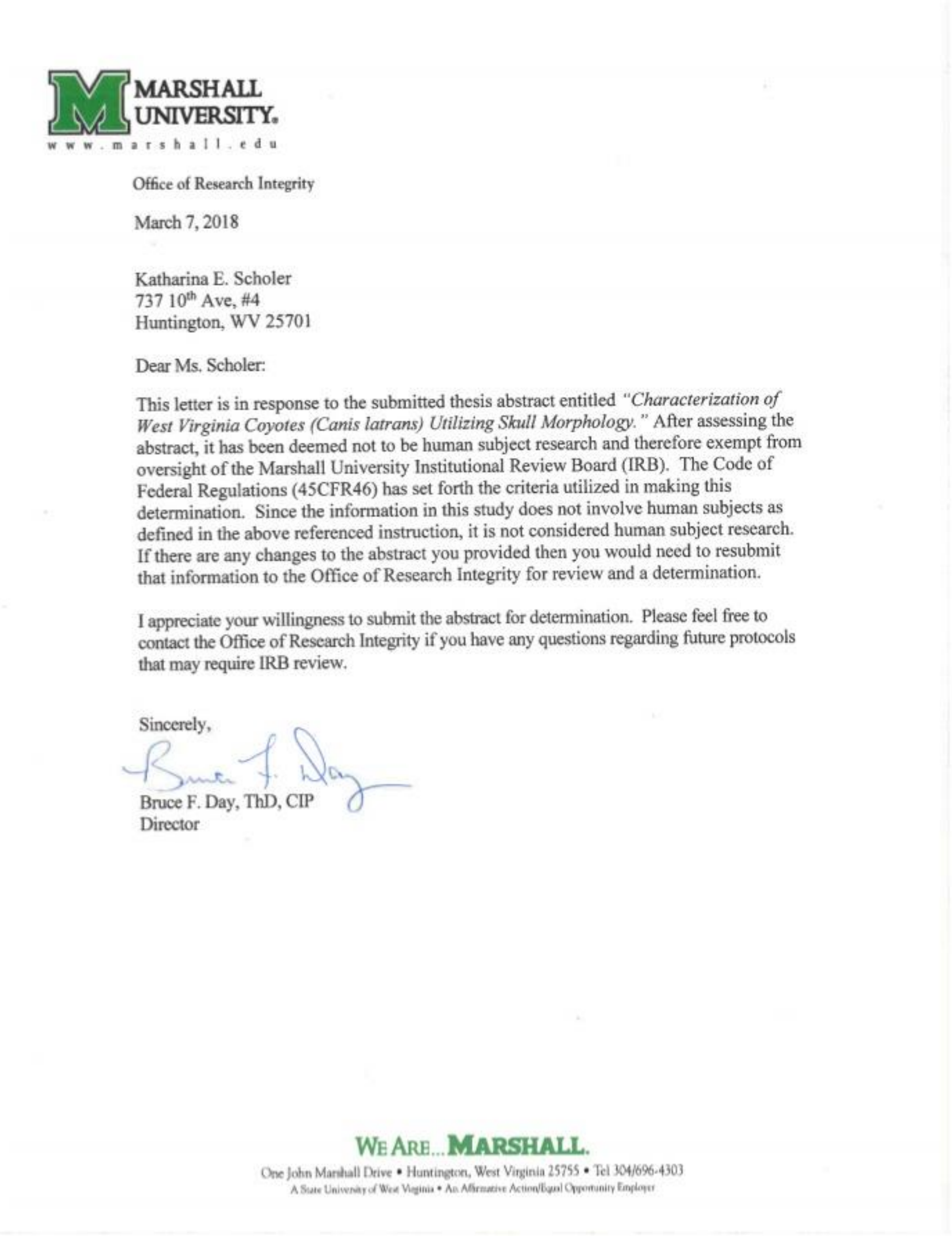
Wilson, P. J., Grewal, S., Lawford, I. D., Heal, J. N., Granacki, A. G., Pennock, D., … White, B. N. (2000). DNA profiles of the eastern Canadian wolf and the red wolf provide evidence for a common evolutionary history independent of the gray wolf. *Canadian Journal of Zoology*, *78*(12), 2156–2166. https://doi.org/10.1139/z00-158

Wykle, J. (1999). *The Status of the Coyote, Canis latrans, in West Virginia*. Marshall University.

Zelditch, M. L., Swiderski, D. L., Sheets, H. D., & Fink, W. L. (2004). *Geometric Morphometrics for Biologists*. Elsevier Inc.

APPENDIX A

Letter from Institutional Research Board



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Office of Research Integrity

March 7, 2018

Katharina E. Scholer
737 10th Ave, #4
Huntington, WV 25701

Dear Ms. Scholer:

This letter is in response to the submitted thesis abstract entitled *"Characterization of West Virginia Coyotes (Canis latrans) Utilizing Skull Morphology."* After assessing the abstract, it has been deemed not to be human subject research and therefore exempt from oversight of the Marshall University Institutional Review Board (IRB). The Code of Federal Regulations (45CFR46) has set forth the criteria utilized in making this determination. Since the information in this study does not involve human subjects as defined in the above referenced instruction, it is not considered human subject research. If there are any changes to the abstract you provided then you would need to resubmit that information to the Office of Research Integrity for review and a determination.

I appreciate your willingness to submit the abstract for determination. Please feel free to contact the Office of Research Integrity if you have any questions regarding future protocols that may require IRB review.

Sincerely,

Bruce F. Day

Bruce F. Day, ThD, CIP
Director

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