USING DENTAL METRIC ANALYSIS TO UNDERSTAND PREHISTORIC POPULATION

VARIABILITY ON THE NORTH CAROLINA COASTAL PLAIN

by

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Biodistance studies can quantify intra- and inter- population relatedness through non-metric and metric skeletal variables. In this study, dental metric traits of two linguistically distinct Late Woodland (AD 800-1650) populations, the Algonquian and Tuscarora-speaking groups within the North Carolina coastal regions, were assessed to determine if presumed linguistic barriers led to a reduced gene flow. Previous research by Kakaliouras (2003) and Killgrove (2002) using cranial and dental non-metric traits identified few significant differences in frequencies of these traits between the Algonquian and Tuscarora, suggesting little genetic differentiation between the two groups. This research used the dental metrics of 170 Algonquian and 53 Tuscarora individuals found that the Algonquians had significantly more variation in only the canine buccolingual measurement (CBL) (Levene's F=8.6644; p=0.0049). The Tuscarora had significantly more variation in the first premolar mesiodistal measurement (PM1MD) (Levene's F=65.5607; p<0.0001) but otherwise identified no overall significant differences in variation (Van Valen Z=1.45012, p=0.1470). These results largely agree with other studies that utilized various cranial and dental non-metric traits, and indicate that genetic dissimilarity did not follow that of language variability. Furthermore, one site linguistically categorized as Tuscarora but which shows a mixture of Algonquian and Tuscarora culturally-affiliated artifacts, was

distinguished as Algonquian in all ten measurements, including the PM1MD (t=-1.99254, p=0.0085), first molar buccolingual measurement (M1BL) (t=1.99254, p=0.0124) and first molar mesiodistal measurement (M1MD) (t=1.99354, p=0.0120).

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CHAPTER 1: INTRODUCTION AND BACKGROUND

Bioarchaeology is the study of human remains from archaeological contexts (Martin, Harrod and Pérez 2013:1). Through the use of a biocultural perspective within an evolutionary framework, bioarchaeologists can make inferences about human behavior and health, all of which are culturally influenced (Larsen 2003:3; Martin, Harrod and Pérez 2013:1-2). The role of culture in shaping human interactions, from environmental to mate choice, affects genotypic and therefore phenotypic expression within a population. Human populations vary in norms and customs associated with marriage, and genetic variation, which can be altered by varying social organization (Kumar et al 2006). In other words, genetics is shaped by cultural practices (Kumar et al 2006).

Previous bioarchaeological studies in North Carolina, particularly in biological distance analysis, have addressed potential genetic differences between Algonquian and Tuscarora populations (Kakaliouras 2003; Killgrove 2002). Archaeological differences have been documented between Algonquian and Tuscarora settlement and subsistence patterns and artifact assemblages. These appear to be consistent with the linguistic divisions historically documented between the Algonquian-speaking Algonquians and the Iroquoian-speaking Tuscarora. This study attempts to further examine potential genetic differences between Algonquian and Tuscarora populations through an odontometric analysis.

This chapter presents background information regarding previous biometric studies and the hypothesis addressed in this research. First, however, I present the archaeological context for this study.

North Carolina Coastal Plain

The North Carolina Coastal Plain consists of the Outer Coastal Plain and the Inner Coastal Plain (Figure 1). Both the Tidewater and Outer Banks (barrier islands) regions make up the Outer Coastal Plain, extending from the eastern edge of the state to the western edge of the Tidewater, encompassing the length of the coast between Virginia and South Carolina. The Outer Coastal Plain is flat, rising at most six to seven meters above sea level. Rivers, lakes and wetlands with sounds (Albemarle, Pamlico, and Currituck) occupy the coastline. The Inner Coastal Plain is bounded by the Tidewater region to the east and the Fall Line to the west. Compared to the Outer Coastal Plain, the Inner Coastal Plain has better drainage with a higher elevation.
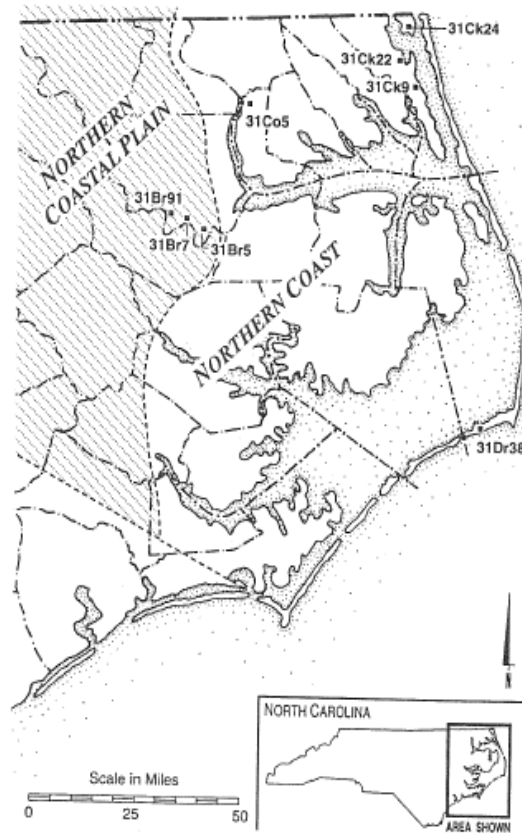
Figure 1: Inner coast and outer coast separation in North Carolina showing the locations of sites included in this study (Hutchinson 2002:20)

North Carolina's climate is humid with short winters. A multitude of plant and animal life thrive in the diverse ecosystems of the Coastal Plain. North Carolina coastal ecosystems have been described as areas containing freshwater, brackish water, saltwater and terrestrial ecosystems (Hutchinson 2002:1).

Late Woodland

Native peoples in the Late Woodland exploited a diverse array of plant and animal resources depending on their location, whether near the ocean or inland. For both the Inner and Outer Coastal regions, resources included hickory nuts (*Caraya* spp.), acorns (*Quercus* spp.), amaranth (*Amaranthus* spp.), sturgeon (*Acipenser* spp.), catfish (*Amiurus* spp., *Ictalurus* spp.), sea trout (*CynoscionI* spp.), yellow perch (*Perca flavescens*), oysters (*Crassostrea virginia*), clams (*Mercenaria mercenaria* and *Chione* spp.), with the inland sites also providing freshwater mussels (*Elliptio* spp., *Fusconaia* spp. and *Ligumia nasuta*) (Hutchinson 2002:27). Other animals recovered at archaeological sites include alligator (*Alligator mississippiensis*), several species of turtles (*Chelydra serpentine*, *Chrysemys* spp. and *Terrapene carolina*), wild turkey (*Meleagris gallopavo),* beaver (*Castor canadensis*), opossum (*Didelphis marsupialis*), white-tailed deer (*Odocoileus virginiams*), black bear (Ur*sus americanus*) and marsh rabbit (*Sylvilagus palustris*) (Hutchinson 2002:27-28).

Algonquians. Ethnohistorical and archaeological evidence identify two geographically separated groups, the Algonquian (Algonquian language) and the Tuscarora (Iroquoian language), within the northern Coastal Plain during the Late Woodland Period (AD 800-AD 1650). By AD 600, Algonquians had migrated to the North Carolina Coastal Plain (Loftfield 1990:121). By about AD 1000, Algonquians exhibited semi-sedentary settlements in the outer coastal region, consisting of permanent villages and summer fishing and shell-fishing camps

dispersed along estuaries and tidal waters in the Tidewater region of Virginia and North Carolina (Hutchinson 2002:19; Loftfield and Jones 1995:122; Phelps 1983:39). Based on material remains examined in the archaeological record such as longhouse construction, shell-tempered ceramics and ossuary burials, it appears that the most southern expansion of the Algonquians was north of the Cape Fear River (Mathis 1997:3). This "Algonquian" style artifact assemblage includes shell-tempered Colington phase ceramics with plain, fabric-impressed, simple-stamped, and incised surface treatments and rims decorated with incised lines, geometric patterns or, to a lesser extent, punctuations (Phelps 1983:36; Horning 2009:137). Lithic artifacts associated with the Colington phase are small triangular projectile points (Roanoke and Clarksville), other bifaces, polished stone celts, grinding stones, gorgets, and sandstone abraders. Ceramic pipes as well as various bone artifacts, including punches, awls, fishhooks, antler flakers and pins are also part of these artifact assemblages (Phelps 1983:39).

On average, villages are estimated to have contained 120-200 individuals (Ward and Davis 1999:211) with approximately 12-18 longhouses (Ward and Davis 1999:216). Watercolor drawings produced in 1585 by John White, an English colonist, portrayed the Algonquian village of Pomeiock as a cluster of longhouses surrounded by a palisade, while the village of Secotan was depicted without a palisade and longhouses scattered along a central walkway with nearby cornfields (Harriot 1590:89, 93; Ward and Davis 1999:213). Algonquians relied on fishing, shell-fishing, hunting, gathering, and farming with hunting and gathering being of lesser importance to the diet (Loftfield and Jones 1995:122-3). Shellfish was collected year round, with farming occurring for half the year (Loftfield and Jones 1995:123). Subsistence resources included sea trout, oysters, clams, freshwater mussels, turkey, beaver, opossum, walnuts, acorns, grapes and corn (Loftfield and Jones 1995:123; Ward and Davis 1999:212).

Socio-politically, Algonquian society is characterized as a chiefdom (Loftfield and Jones 1995:134) with evidence of trade with neighboring Tuscarora groups (Health and Swindell 2011:25), which includes the presence of Cashie ceramics in Colington sites (Health and Swindell 2011:25).

The primary data source for Colington phase Algonquian burial practices comes from archaeological evidence and ethnohistorical accounts (Mook 1944:181-182). The majority of individuals are interred in large, usually circular pits containing between 20 and 60 individuals, including males and females of all ages, located close to the associated village (Phelps 1983:42). The disposition of the skeletons in these ossuaries ranges from bundle burials and fully articulated individuals to disarticulated, scattered bones (Phelps 1983:40). Primary burials are either fully or partially articulated skeletons, while secondary burials contain disarticulated and scattered elements (Hutchinson 2002:49; Phelps 1983:40). Males and females of all ages were incorporated within an ossuary (Phelps 1983:42). Grave goods are uncommon during the Late Woodland (Loftfield 1990:118; Phelps 1983:42), and inclusion of antlers and panther muzzles were suspected to mark those of supposed elite status and possibly shamans (Phelps 1983:42; Horning 2009:135).

Interment was often a lengthy ritual, sometimes lasting a decade or more, with temporary storage or burial occurring from death to ossuary burial (Hutchinson and Aragon 2002:31, 35). Temporary storage and processing of the body was done in a charnel house, or mortuary facility (Ward and David 1999:216; Hutchinson 2002:50). Reports indicate that the remains of supposed elites and priests were defleshed and stored in painted containers as depicted by White's watercolors of a charnel house (Horning 2009:135; Hutchinson and Aragon 2002:39). Based upon White's descriptions, Horning (2009) outlines the following burial procedures. Processing

of the body involved the removal of hair and skin. After removal, the skin was then dried and wrapped in a mat, which was laid at the feet of the deceased. Temporary burial has been inferred from empty burial pits, possibly used as primary storage of skeletal remains before the remains were placed in an ossuary (Mathis 1993:46-47). Either cremation or burial of the remains within an ossuary was part of the final interment process (Hutchinson 2002:51).

Tuscarora. The Tuscarora were an Iroquoian linguistic group living to the west of Algonquian territory in small, dispersed villages, camps, and farmsteads that included seasonal sites for the fall and hunting areas for the winter (Phelps 1983:43; Ward and Davis 1999:224). Sites show evidence of mixed subsistence strategy, including agriculture, fishing, and hunting and gathering (Ward and Davis 1999:224). Each village had an autonomous sociopolitical organization, their own chief and a council (Phelps 1983:43; Pritzker 1998:569). The Tuscarora practiced matrilineal descent, with women nominating the clan chiefs (Heath and Swindell 2011:9; Perdue 2003:21; Pritzker 1998:569).

Archaeologically, Tuscarora populations are associated with the Cashie phase. Cashie phase ceramics have fabric-impressed, simple-stamped, plain, and incised surface treatments. Rims could have punctuations, and to a lesser degree, finger pinching and incisions. Surface treatments are similar to that of the Colington ceramics but are tempered with small pebbles and sand (Phelps 1983:43). Along the lower Roanoke River, terrapin turtle shells, marginella shell beads, and conch shells, all native to the coast and Algonquians, have been excavated in Tuscarora sites, hinting at trade between the two linguistic groups (Phelps 1983:44). Other artifacts in Cashie assemblages include ceramic pipes akin to the Colington pipes, Roanoke and Clarksville points, bifaces, unifacial scraping tools, drills and grinding stones. Bone implements include awls, pins and perforators (Phelps 1983:44).

The Tuscarora, like the Algonquian, practiced ossuary burial. Tuscarora ossuary burials frequently include secondary bundles burials containing two to five individuals each (Phelps 1983:46) within a single pit compared to approximately 20 to 60 individuals for Algonquian ossuaries. In general, Tuscarora ossuaries included a greater number of grave goods, especially marginella shell beads, thought to be a status indicator when compared to their Algonquian counterparts (Phelps 1983:46).

Archaeological sites

Six archaeological sites are utilized for this study, three identified archaeologically as Algonquian, two as Tuscarora, and one, the Hollowell site, tentatively identified as Tuscarora, based on nonmetric cranial biodistance analysis by Killgrove (2002). The Algonquian sites include Baum (31CK9), West Village #2 (31CK22), and Hatteras Village (31DR38), all associated with the Colington phase (AD 800-1600). The Tuscarora sites consist of Jordan's Landing (31BR7) and Sans Souci (31BR5), which are associated with the Cashie phase (AD 673-1444). Hollowell has both Colington and Cashie phase materials. Killgrove (2002) discerned Hollowell as belonging to the Inner Coast population, so for this study, Hollowell will be classified as Tuscarora. All material remains are curated at the Phelps Archaeological Laboratory in the Department of Anthropology at East Carolina University.

Algonquian Sites

Baum (31CK9). Baum is an Outer Coastal Plain site located on five acres on Currituck Sound within Currituck County. Investigations began in 1972 by David S. Phelps of East Carolina University after human remains were reported eroding along the beach. Phelps carried out test excavations at the site and identified Algonquian ossuaries (Phelps 1980:1). A total of 8 ossuaries and several single and multiple burials comprise the inhumations at Baum. About 205

individuals were interred in the ossuaries (Hutchinson 2002:64). Ossuaries 1, 5, 6 and 7 date to the Late Woodland period burial, while ossuaries 2, 3, 4 and 8 date from the Middle Woodland period. A radiocarbon date for the site is AD 1315 ± 70 with a calibrated date of AD 1450 (Hutchinson 2002:33)

Ossuary 1 (Figure 2) contained eight articulated burials, scattered bones and a small number of bundle burials. Wave action appeared to have eroded one-quarter to one-third of ossuary 5 by 1980 when it was excavated. Bone awls, bone pins and a portion of a panther muzzle were also found. It was hypothesized that inclusion of the panther muzzle shows the associated human remains were wrapped or laid on a panther skin while in a charnel house, possibly signifying the status of the deceased (Horning 2009:35).

Ossuary 5 (Figure 2) contained scattered bones, two or three bundle burials and three articulated burials (Phelps 1980:10). One fully articulated individual was placed above the main burial within the fill. Phelps (1980:9) hypothesized that either the individual's family arrived too late for the burial ritual but before the ossuary had been completely filled, or the individual died during the ritual, and without the ritual flesh removal, had been
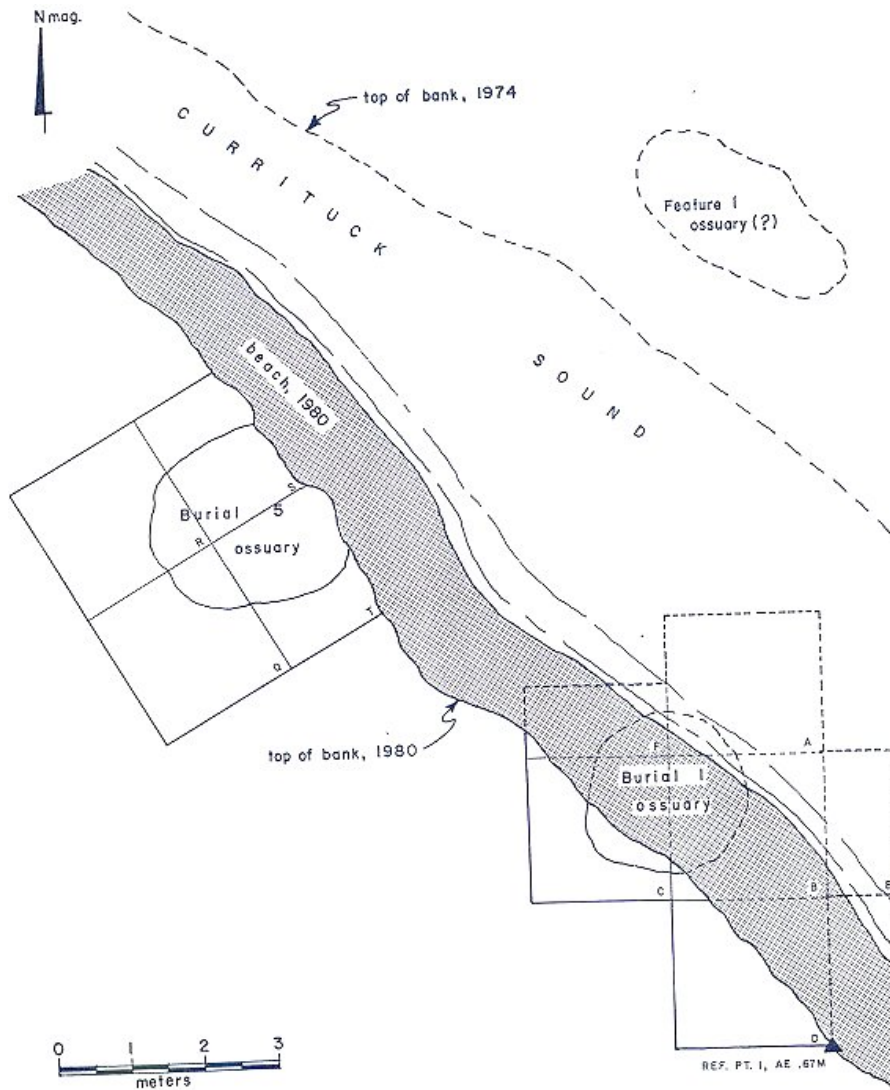
Figure 2: Baum (31CK9) burials 1 and 5 with feature 1 (Phelps, 1980:10)

interred (Phelps 1980:9). Evidence of red staining was apparent on one cranium. Burial goods for

ossuary 5 included a necklace of 15 marginella shells and a disc-shaped copper bead, retrieved

from the southwest corner near crania of infants and children. Besides the marginella shell

necklace and copper bead, the remaining artifacts from the ossuary appear to be unintentional

inclusions (Phelps 1980:11).

Ossuaries 6 and 7 were salvaged in 1983 due to erosion. Ossuary 7 also had evidence of red staining on four crania (Hutchinson 2002:36), much like Ossuary 5. It has been hypothesized the human remains may have been stored in reed containers located at the back of charnel houses, which contributed to the red staining on some crania (Horning 2009:135).

Table 1: Baum archaeological site burial dates

Burial Number	Radiocarbon Date	Calibrated Date	Material Used for Dating
1	AD 1400 ± 60	AD 1410	Human Bone
5	AD 1390 ± 80	AD 1410	Human Bone
6	AD 1310 ± 40	AD 1300	Human Bone
7	No data	No data	No data

West Village #2 (31CK22). West Village #2 is in Currituck County in the Outer Coastal Plain near Currituck. West Village #2 is the second ossuary at this site, with 5 ossuaries in total recovered. The Office of State Archaeology recovered West #1 in 1984 with help from East Carolina University. This ossuary contained at least eight individuals within discrete bundle burials from the Late Woodland based on Colington phase artifacts found within the ossuary (Souther 2010:50). Mark Mathis from the North Carolina Office of State Archaeology excavated West #2 in 1994. By the time the excavation took place, approximately half of the ossuary had eroded. Scattered, disarticulated bones and thirteen bundle burials were uncovered, several of which had red cordage staining on crania. Bone pins, similar to those found at Baum, were discovered in the ossuary. West #3, initially thought to be a separate ossuary, was later confirmed to have belonged to West #2 and was therefore combined. West #4 contained a primary inhumation with two individuals (Souther 2013:38). West #5 contained the remains of one individual (Souther 2013:46). Both West #4 and West #5 were excavated by East Carolina University in 2010. No radiocarbon dates are available (Hutchinson 2002:38).

Hatteras Village (31DR38). Hatteras Village is located on Hatteras Island in the Outer

Banks and this eroding ossuary was excavated in 1974. An estimated 38 individuals were

removed from the ossuary (Hutchinson 2002:36) and a minimum number of individuals was

estimated at 110 (Hutchinson 2002:64). A radiocarbon date of AD 1350 ± 70 was obtained and

calibrated to AD 1395 (Hutchinson 2002:36).

Tuscarora Sites

Jordan's Landing (31BR7). Jordan's Landing covers three acres that are located below

the town of Williamston on the Roanoke River. Phelps excavated the site in 1971 (Hutchinson

2002:42) and discovered two post molds in the east within the village, interpreted as a cooking

pit as well as palisade post molds (Byrd 1997:23, 25). Pits and hearths were found along the west

and north sides of the village and burials were uncovered in the southeast section of the village

(Phelps 1983:46).

While a few burials recovered from Jordan's Landing date to the Middle Woodland

period, the majority (MNI=43) of the burials date to the Late Woodland (Hutchinson 2002:64).

Archaeologists uncovered several clustered small ossuaries and multiple burials on the east side

of the site that likely had been placed either below house floors or in between house structures

(Heath 2003:5). Artifacts associated with burials at Jordan's Landing included bone awls, bone

pins, bone perforators, marginella shell beads and ceramics (Phelps 1983:46). One female

ossuary bundle included a fabric-impressed vessel. A different ossuary contained a worked split

bone pin, which was discovered in association with an adult male and a newborn (Heath 2003:6).

Only one secondary burial containing one individual, Burial #9, was found at Jordan's

Landing (Heath 2003:6), an adult male who had kypho-scoliosis, commonly known as

"hunchback" syndrome (Heath 2003:6). Interred with the male were marginella shell beads and

various modified and unmodified faunal bones, including deer antlers, bear femora and turtle carapaces (Heath 2003:6-7). Associated tools, such as split bone pins and scapula tools, were likely that of a ritual specialist, such as a healer or shaman (Heath 2003:7). One nearly sterile pit feature was located between two primary burials in the northwest section of Jordan's Landing and interpreted as a burial-processing pit for defleshing (Heath 2003:8).

Two samples of charcoal have been radiocarbon dated for the site (Hutchinson 2002:43). One sample has been dated to AD 1280 ± 60, calibrated to AD 1290, and the second charcoal sample has been dated to AD 1425 ± 70, calibrated to AD 1418 (Hutchinson 2002:43).

Hollowell (31CO5). Hollowell is located in Chowan County above the Chowan River, near the Algonquian and Tuscarora border in the Inner Coastal Plain. Hollowell was discovered in 1974 when waterline construction cut through the ossuary, and excavations began by Phelps in 1975 (Phelps 1982:30). He discovered nine distinct burial groups within ossuary #1 with an MNI of approximately 90 individuals (Figure 3) (Hutchison 2002:64). Phelps hypothesized the skeletal clusters to be familial and deposited over a 10 year time span (Phelps 1982:31). Contact and trade are inferred through ceramic analysis of the Hollowell site, which contains a combination of Colington and Cashie phase ceramic styles (Phelps 1982:33), and this combination of artifacts makes establishing population and linguistic affiliations difficult (Phelps 1982:33). However, Killgrove (2002), identified through analysis of discrete cranial traits that the Hollowell display closer genetic affiliation to the Tuscarora. One radiocarbon date was obtained from human bone, dated to AD 1460 ± 60 and calibrated at AD 1430 (Hutchinson 2002:40).
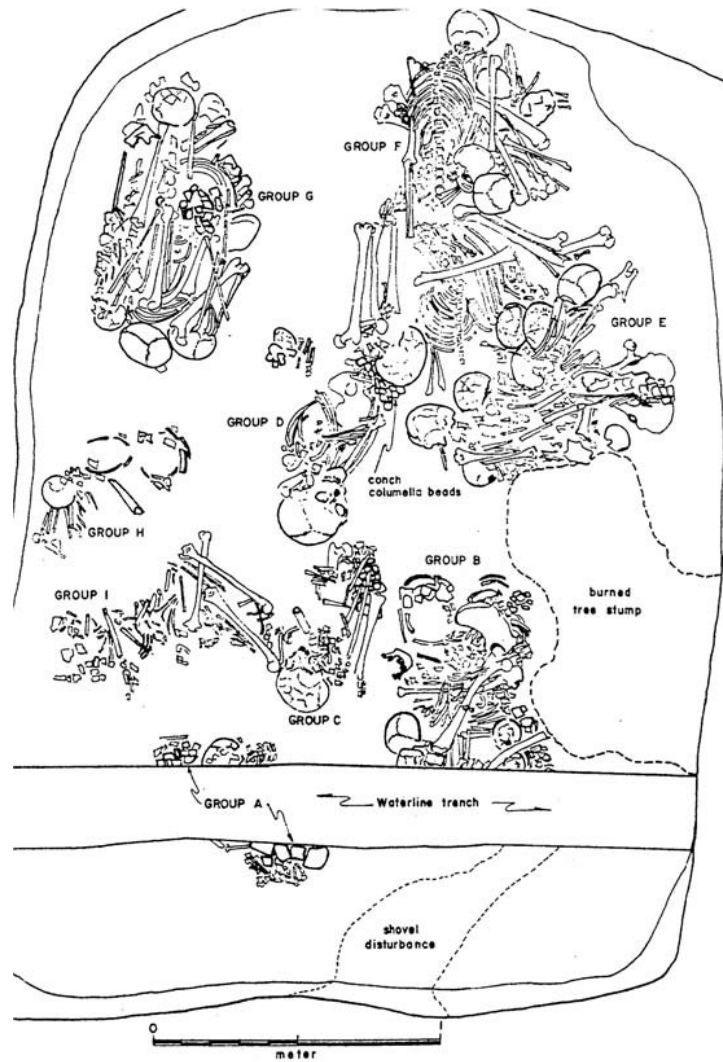
Figure 3: Hollowell (31CO5) nine burials (Phelps 1982:32)

Sans Souci (31BR5). Sans Souci is a Tuscarora Late Woodland site located approximately 10 miles southwest of Jordan's Landing that includes several small ossuaries. A salvage operation by amateurs in 1973 uncovered human remains, which were then donated to East Carolina University. Ossuaries contained discrete bundle burials of two to five individuals (Heath 2003:6), with a total of 33 individuals recovered from the site (Hutchinson 2002:64). Many of the ossuaries contained marginella shell beads, though in lower frequencies than other sites (Heath 2003:6). Additional artifacts included bone and antler awls, bone pins and pendants, a bone

needle, turtle carapaces, freshwater mussel shells, and four deer antlers. The deer antlers were

placed like a headdress around a female cranium (Heath 2003:6-8). Another adult female

interment had a canine skull within the burial fill, which was placed above the pelvis (Heath

2003:4). Burial offerings were associated with either ossuaries containing all female remains, or

ossuaries with at least one reliably sexed female (Heath 2003:6).No radiocarbon dates are

associated with this site (Hutchinson 2002:43).

The linkage between linguistic and genetic variation in prehistoric North America

In the 19th and first half of the 20th centuries, studies of prehistoric North America (and

elsewhere) inferred linguistic and cultural attributes from skeletal morphology and metric traits

(Coe et al 1982; Hridlička 1916; Loftfield 1990). Thus, based on this assumption, the size and

robustness of crania and post-cranial remains from North Carolinian Native American ossuaries

were used to categorize the groups linguistically, with "robust" groups linked to the Algonquians

and Iroquoians, and "gracile" to the Sioux (Coe et al 1982; Hridlička 1916; Loftfield 1990). (Coe

et al 1982; Loftfield 1990). As a result, some groups that culturally and linguistically were

actually Algonquian, such as individuals living at the Cold Morning site in North Carolina

(31NH28), were attributed to Siouan groups due to their gracile skeletal structure (Coe et al.

1982).

Cranial dimensions also were used to characterize linguistic/cultural groups in eastern

North America. In "Physical Anthropology of the Lenape or Delwares, and of the Eastern

Indians in General," Hrdlička (1916) presented the three typologies he used to group the Lenape,

Delaware, Algonquians, and Iroquoians based on skull shape and size. He grouped skulls into

one of three "types" for from each linguistic group. Hrdlička characterized bracycephic (short

but wide) skulls as Lenape and the dolichocephalic (long but narrow) and mesocephalic

(moderately long and wide) skulls as Algonquians, or Iroquoian groups admixed with groups to the west or southwest. Any skulls varying from these typologies within a group were considered to be adopted or the result of admixture.

Research Question

The goal of this study is to examine the veracity of linguistically-based typologies and genetic divergence and similarity within and between Algonquian and Tuscarora populations on the North Carolina Coastal Plains during the Late Woodland Period. Previous biodistance work has been conducted on the North Carolina Coastal Plain (Killgrove, 2002; Kakliouras 2003), which focused only on the presence of gene flow between the outer and inner coastal plain groups, and did not address the levels of genetic variation within and between these populations. My dental metric study addresses this question, as well as population affiliation of the ambiguous Hollowell site.

Dental metrics are a good phenotypic measure for identifying genetic variability within and between populations due to high preservation of teeth in archaeological contexts, less bias than nonmetric analysis, increased sample sizes and high heritability (Alvesalo and Tigerstedt 1974; Mower 1999:50; Scott 2013:179). Teeth are generally well preserved in archaeological records, even when associated cranial and postrcranial skeletal remains are damaged, leading to larger dental sample sizes (Rathmann et al. 2017; Lukacs 1995:4). Metric studies also have less bias than nonmetric analyses (ordinal data, presence/absence) (Scott and Irish 2013:37). It has been argued that use of metric measurements leads to less intra- and interobserver error rates compared to subjective trait scoring such as those employed by nonmetric analysis (Pilloud and Hefner 2016:144; Hemphill 2016:334). In addition, dental metric analysis can increase sample sizes over dental nonmetric analysis through inclusion of worn teeth that reduce effective scoring

of observable traits, leading to misinterpretation of nonmetric traits (Scott and Irish 2013:536-537). With an increase in sample sizes, it is possible less bias may be introduced in the sample, thus leading to more faithful assessment of population differences. Furthermore, dentition has a high heritability with an estimated .60-.80 heritability of crown size (Scott 2013:179), with dental size and morphology being genetically linked within family units (Scott 2008:271).

Even with the advantages of dental metric analysis, this technique is not without drawbacks. Teeth can be lost or damaged through multiple avenues, resulting in missing data. Due to the conical shape and lack of distal tilting in the roots of first maxillary incisors, loss of these teeth is increased over other tooth positions and tooth classes (Durić, Rakočević and Tuller 2004:8). If remains are removed from a primary burial for placement into a secondary burial, loose teeth may be overlooked and left in the burial pit. With inhumation excavations, teeth may become damaged or lost during excavation, transportation, cleaning or storage. With the loss or damage of teeth, sample sizes shrink, meaning loss of biological data as well.

Biological Distance

Biodistance studies determine genetic convergence (relatedness) or divergence among and between populations (Buikstra, Frankenberg and Konigsberg 1990:1; Stojanowski 2006:273) and are based on the theory that phenotypic expression, in the form of cranial and dental metric and non-metric traits, is linked to genetics and heritability (Larsen 2003: 305; Pietrusewsky 2008:487; Pilloud and Hefner 2016:11), and that populations exchanging genetic information will become more phenotypically similar over time. Those populations that do not exchange genetic material will become dissimilar through genetic drift (Konigsberg 1988:471; Stojanowski 2003:216). Biodistance analysis thus can helpful to understand intra-cemetery structure based on

familial groups, post marital residence patterns, genetic drift, gene flow and bottlenecking in ancient populations (Stojanowski 2006:273; Konigsberg 1988:471; Konigsberg 1990:46). Dental and cranial metric and nonmetric traits have been linked to genetics, so analyses using these traits are performed in order to ascertain phenotypic, and therefore genotypic variation present in populations under study (Buikstra, J., S. Frankenberg and L. Konigsberg 1990: 1; Pietrusewsky, M. 2008:487; Pietrusewsky, M. 2014: 487; Pilloud and Hefner 2016:3). While genotypes play a large part in dental crown size and tooth morphology, environmental effects may alter tooth trait size and expression. Such environmental effects include nutritional deficiencies and disease (Bailit 1970:626; Stojanowski 2007:214). Other extrasomatic factors that can alter dentition include diet (grit in food), culture (tooth filing) and nutritional stress (hypoplasia). Polar teeth (M^1, P^1, C and I^1; M_1, P_1, C and I_2) are least susceptible to alterations from these environmental factors, with teeth in the second position in the dental arcade having more variation due to environmental factors and third molars having the most variation among other tooth classes (Scott 1997:176; Stojanowski 2003:223).

Thus, comparing the heritable trait of tooth size should identify any significant differences between Algonquian and Tuscarora populations to discern whether or not perceived linguistic and/or geographic differences served as barriers to gene flow in the North Carolina coastal plain. This will be complemented by analyzing differences in within-group variation to identify genetic drift due to reproductive isolation. This study assumes that the geographic distribution of the sites, those in the "inner coast" and those in the "outer coast", parallel the cultural influences identified by material culture and that the linguistic divisions observed in ethnohistoric data characterize the Late Woodland period in general. In addition, the geographically and culturally ambiguous site of Hollowell (31CO5), which lies near the border

identified by Colington versus Cashie artifact assemblages, and contains both Algonquian and Tuscarora material culture, will be assessed within the context of dental metric variation from both the outer and inner Coastal groups. This will determine whether or not Hollowell has more genetic similarity with either region, or with both regions.

Previous Biodistance Research in Eastern North Carolina

Levels of genetic relatedness between Algonquian and Tuscarora groups have been explored using cranial nonmetric traits and dental nonmetric traits. Kristina Killgrove (2002) utilized 25 nonmetric cranial traits within Late Woodland Tuscarora, Algonquian and Siouan cultural groups from 15 archaeological sites in order to explore between-group genetic differences. Killgrove found no discernable biological difference between the three cultural groups, suggesting that breeding between groups occurred. Killgrove's results were confirmed by Kakaliouras (2003), who used 37 dental nonmetric mandibular and maxillary traits of skeletal remains from 13 Late Woodland North Carolina archaeological sites also included in Killgrove's study.

Hypotheses

1.   The null hypothesis is that dental measurements will indicate similar levels of genetic variation exist between and within Algonquian and Tuscarora groups. If this pattern exists, then genetic material likely is being exchanged between both groups.

2.   For the alternative hypothesis, dental measurements will indicate greater genetic variability between Algonquian and Iroquoian groups than there is within each group. If this pattern exists, then there is little genetic admixture between Algonquian and Iroquoian groups on the North Carolina coast.

Summary

Utilizing biodistance analysis, this study compares Late Woodland Tuscarora and Algonquian populations in the North Carolina Coastal Plain in order to observe genetic similarity or differences between and within groups. Dental metrics are used here since phenotypes are inherited genetically from parents to offspring and can be passed on in the form of dental similarities. This study demonstrates that, despite linguistic differences between the Inner (Iroquoian) and Outer Coastal Plain (Algonquian), gene flow likely occurred. Also, this study adds to the collection of research (Harding and Sokal 1988; Hurles et al 1999; Kutanan 2014; Rosser et al 2000; Relethford 2012) showing that despite the linguistic differences between the two groups, their geographic proximity likely facilitated gene flow between the two groups. Building off of previous biodistance work in North Carolina, this study looks at the amount of variation within the Algonquian and Tuscarora populations.

Chapter 2 will cover methods used in this study to collect dental metric data, followed by statistical analyses used for the biodistance analysis. Chapter 3 presents results from the analysis. Chapter 4 addresses the implications of this study in regards to linguistic and geographic boundaries to gene flow. Finally, chapter 5 provides some suggestions for future research.

CHAPTER 2: METHODS

Human dental metric analysis uses measureable, continuous tooth dimensions to answer questions pertaining to population genetics, including migration and post-marital residence patterns (Pilloud and Hefner 2016:148). This section details the samples used for assessment of gene flow in the North Carolina coastal plain and the methods utilized for data collection.

Materials

This study focused on dental remains from Algonquian and Tuscarora populations curated at East Carolina University. Approval for this non-destructive study was received by the North Carolina Commission on Indian Affairs. Before data reduction for statistical analyses, dental remains (excluding loose, commingled teeth) included 170 observations from commingled adult individuals from the Outer Coastal Plain/Algonquian samples. These individuals were from Baum (31CK9), West Village #2 (31CK22), and Hatteras Village (31DR38). 53 observations from commingled adult individuals from the Inner Coastal Plain/Tuscarora samples were observed. These remains were from Jordan's Landing (31BR7), Sans Souci (31BR5) and Hollowell (31CO5) (Table 2). Hollowell, which displayed a mix of Algonquian and Tuscarora artifacts, was included in the Tuscarora population based on previous biodistance research (Killgrove, 2002). Loose, commingled teeth (N=97 for Algonquian, N=32 for Tuscarora) were included when available, with each loose tooth counted as a separate observation. Sex estimation was completed when possible based on cranial morphology as outlined in *Standards for Data Collection from Human Skeletal Remains* (Buikstra and Ubelaker 1994). The features observed included the nuchal crest, mastoid process, supraorbital margin, glabella and mental eminence, and were scored on a scale from one to five, with one being the most female expression of the

trait and five being the most male expressing. Individuals for whom sex estimation was not possible were identified as "indeterminate."

Table 2: Minimum number of individuals from all sites before data reduction

Site	Linguistic Affiliation	Total MNI	Dental Observations
Baum (31CK9)	Algonquian	205	61
West Village #2, #4, #5 (31CK22)	Algonquian	135	30
Hatteras (31DR38)	Algonquian	110	79
Jordan's Landing (31BR7)	Iroquoian	43	26
Sans Souci (31BR5)	Iroquoian	33	8
Hollowell (31CO5)	Iroquoian/ Algonquian	90	19

Total MNI data from Hutchinson (2002)

Methods

Dental crown measurements of buccolingual and mesiodistal breadths and crown height on well-preserved teeth from the six sites were taken using Mitutoyo CD-6" sliding digital calipers. Collected odontometric data (housed in the Joyner Library online data repository) for each individual along with sex estimations were inputted into an Excel spreadsheet using a Mitituyo input tool. The mesiodistal (MD) measurements were taken at the widest point of the crown, parallel to the occlusal surface following Moorrees and Reed (1954). The buccolingual (BL)

measurements were taken perpendicular to the mesiodistal plane at the widest point (Buiskstra and Ubelaker 1994). In addition, dental wear was recorded for each tooth following Smith's (1984) and Scott's (1979) methods outlined in Buikstra and Ubelaker (1994). Examples of dental wear stages are depicted in Figures 4 through 9 below.
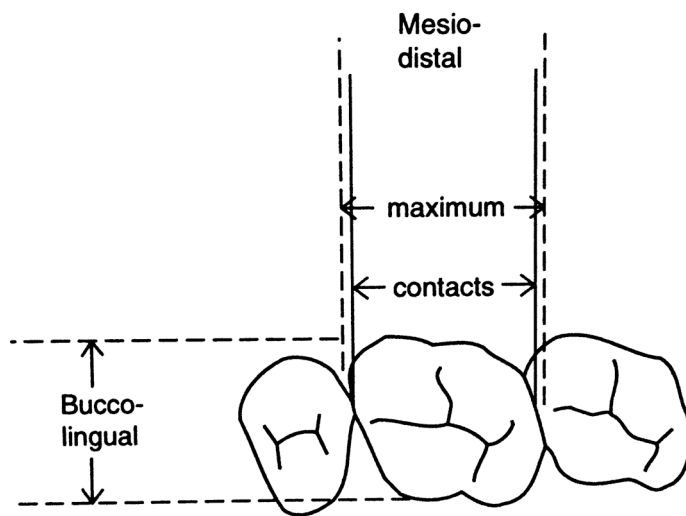
Figure 4: Mesiodistal and buccolingual dental measurements (from
        Buikstra and Ubelaker 1994)

The mesiodistal measurements of teeth with extensive dental wear (an incisor or canine wear score of 5 and above, or a premolar or molar wear score of 6 and above) were not included in the data analysis because of their potentially reduced tooth dimensions (Hillson 1996:70; Pilloud and Hefner 2016:142).
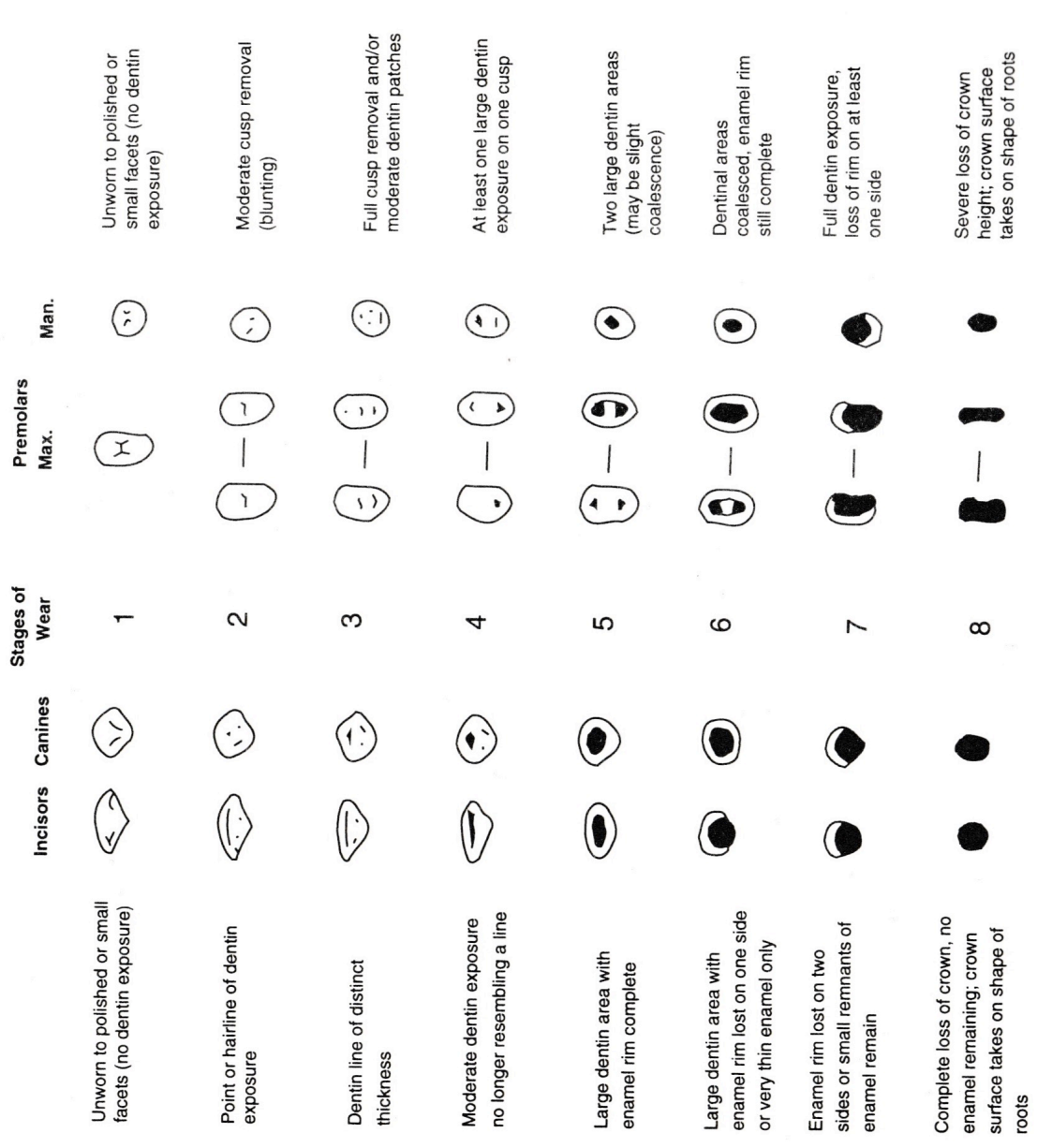
	Incisors	Canines	Stages of Wear	Premolars Max.	Man.	
Unworn to polished or small facets (no dentin exposure)			1			Unworn to polished or small facets (no dentin exposure)
Point or hairline of dentin exposure			2			Moderate cusp removal (blunting)
Dentin line of distinct thickness			3			Full cusp removal and/or moderate dentin patches
Moderate dentin exposure no longer resembling a line			4			At least one large dentin exposure on one cusp
Large dentin area with enamel rim complete			5			Two large dentin areas (may be slight coalescence)
Large dentin area with enamel rim lost on one side or very thin enamel only			6			Dentinal areas coalesced, enamel rim still complete
Enamel rim lost on two sides or small remnants of enamel remain			7			Full dentin exposure, loss of rim on at least one side
Complete loss of crown, no enamel remaining; crown surface takes on shape of roots			8			Severe loss of crown height; crown surface takes on shape of roots

Figure 5: Incisor, canine and premolar wear scoring stages (Buikstra and Ubelaker 1994)

Score	Description
0	No information available (tooth not occluding, unerupted, antemortem or postmortem loss, etc.)
1	Wear facets invisible or very small
2	Wear facets large, but large cusps still present and surface features (crenulations, noncarious pits) very evident. It is possible to have pinprick size dentine exposures or dots which should be ignored. This is a quadrant with much enamel.
3	Any cusp in the quadrant area is rounded rather than being clearly defined as in 2. The cusp is becoming obliterated but is not yet worn flat.
4	Quadrant area is worn flat (horizontal) but there is no dentine exposure other than a possible pinprick sized dot.
5	Quadrant is flat, with dentine exposure one-fourth of quadrant or less. (Be careful not to confuse noncarious pits with dentine exposure.)
6	Dentine exposure greater: more than one-fourth of quadrant area is involved, but there is still much enamel present. If the quadrant is visualized as having three sides (as in the diagram) the dentine patch is still surrounded on all three sides by a ring of enamel.
7	Enamel is found on only two sides of the quadrant.
8	Enamel on only one side (usually outer rim) but the enamel is thick to medium on this edge.
9	Enamel on only one side as in 8, but the enamel is very thin—just a strip. Part of the edge may be worn through at one or more places.
10	No enamel on any part of quadrant—dentine exposure complete. Wear is extended below the cervicoenamel junction into the root.
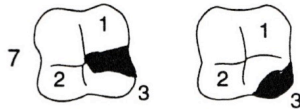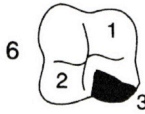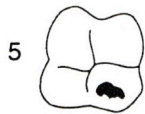
Figure 6: Molar wear scoring stages (from Buikstra and Ubelaker 1994)

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Figure 7: Canine dental wear stages 1 through 4 (from left to right). Canine 1: Accession #329/31DR38/ NAGPRA Box 5-in bag TEETH, Canine 2: Accession #225/31DR38/ Box 5-Vial 7, Canine 3: Accession #225/31CK9/Burial 7P, Canine 4: Accession 225/31CK9/ Burial 1Y.

Figure 8: Premolar dental wear stages 1 through 5 (from left to right). Premolar 1: Accession #329/31DR38/Box 5-in bag teeth (33)-burial 1, Premolar 2: Accession #225/31CK9/Burial 1E, Premolar 3: Accession #225/31CK9/Crania DDD, Premolar 4: Accession #329/31DR38/ Burial 1-Jaws and Teeth-B-AY, Premolar 5: Accession #329/31DR38/ Burial 1-Jaws and Teeth-

Figure 9: Molar dental wear stages 1 through 5 (from left to right). Molar 1: Accession #329/31DR38/ Box 5-in bag teeth (33)-Burial 1, Molar 2: Accession #225/31CK9/ Burial 1-Cranium X, Molar 3: Accession #225/31CK9/ Crania DDD, Molar 4: Accession #309/31CK22/ Bur 2-Crania C-15-Sect B-3, Molar 5: Accession #225/31CK9/ Burial 1Y.

P

Polar teeth (I^1, I_2, C', C, PM^1, PM_1, M^1, M_1) are less affected by environmental and epigenetic factors, and thus more strongly reflect narrow-sense heritability (Alvesalo and Tigerstedt 1974:316; Dahlberg 1963:173; Stojanowski, 2005:85). Therefore only these teeth were included in the analysis. In addition, strong correlation between left and right antimeres (Stojanowski et al. 2017:8) meant that for statistical analysis only measurements of the left side of the dental arcade were retained, and if missing, replaced by its antimere if available.

Finally, the commingled nature of the samples utilized here meant that, in most cases, mandibles and maxillae, and loose, commingled teeth, could not be individuated (that is, linked together as belonging to one person). As a result, an unlinked maxilla would contain missing values for the mandibular dentition, and an unlinked mandible would contain missing values for maxillary dentition, resulting in numerous missing values in the database. Stojanowski and colleagues (2017:9) recently confirmed that the maxillary dentition has higher narrow-sense heritability than the mandibular teeth. Thus, to reduce the amount of missing values in the dataset, which can impact multivariate data analysis in particular, only the maxillary dentition were included in the univariate and multivariate statistical analyses.

Controlling for Sexual Dimorphism

The dental measurements from each region (inner and outer coasts) were standardized in order to control for the effects of sexual dimorphism. JMP Pro 13 (SAS Institute, Cary, NC) was used to compute z-scores within male and female samples from each region. In order to standardize the measurements of teeth with an indeterminate sex estimation the mean and standard deviation of the combined female and male z-scores in each region were be used to standardize the indeterminate sex sample, with the assumption that the sex ratio of males and females in the indeterminate group is the same as the known males and females.

Univariate Analyses

Data were tested for normality using the Shapiro-Wilk W test and outlying values removed as necessary to create normal distributions. Univariate assessment of between-region variation utilized a student's t-test with a Bonferroni correction for multiple comparisons to compare the mean of each dental measurement between the Inner Coastal Plain and the Outer Coastal Plain using JMP Pro 13. This statistic would identify any general difference between the linguistic groups. Bonferroni correction reduces the chance of receiving a Type I error, or false positive, which increases with each successive t-test on a particular sample (Kao and Green 2008). . Bonferroni Correction resets the p-value so the threshold for a statistically significant result occurs less frequently (Kao and Green 2008:4). Levene's test and Bartlett's test were then used to compare variability in the form of unequal variances. Levene's test is a one-way analysis of variance F-test, which was used to evaluate the variance homogeneity of the samples (Lim and Loh 1995). Bartlett's test is another test for homogeneity of variances of the samples. Bartlett's test is more sensitive if the data departs from normality.

Multivariate Analyses

Despite attempts to limit missing data by focusing only on maxillary dentition and replacing missing values from the left side of the arcade with those on the right as described above, many observations still contained missing data. First, any variable missing greater than 50% of its observations were removed. Then, any observation containing data on only one measurement also was removed. Missing values in the remaining data were imputed in JMP Pro 13 utilizing the program's multivariate normal imputation (MVNI) program with shrinkage, which relies on least squares prediction of missing values based on present values in the dataset.

Exploring potential sources of variation in the multivariate dataset utilized stepwise linear discriminant function, canonical discriminant analysis (CDA), principal components analysis (PCA), Van Valen's test and Quadratic Discriminant Function Analysis in JMP Pro 13. These analyses can identify any variables or groups of variables that may be driving variation within the dataset in order to better interpret multivariate statistical results. Stepwise discriminant function analysis isolates variables or a set of variables that best discriminate the groups being analyzed. This can be used as a data reduction technique to remove non-significant variables from future analyses. CDA can identify a variable or set of variables that maximize between-group difference and within-group homogeneity (Zhao and Maclean 2000:842). PCA isolates correlated groups of variables and calculates how much they contribute to the overall variation in the sample (Kucharczyk et al. 2017:111). In addition, PCA controls for correlation between the variables that may skew multivariate results by combining groups of correlated variables into principal components (PCs), which then can be statistically analyzed if inter-variable correlation is an issue.

Multivariate testing for unequal variances between Inner Coastal and Outer Coast was conducted with MANOVA and Van Valen's test. MANOVA tests for mean vector differences between two or more groups with the use of multiple dependent variables, and can detect patterns between these dependent variables.

Van Valen's test is a multivariate version of Levene's test that detects small amounts of variation in two or more samples when the means are unknown and may be unequal (Manly 2006:1). The data first are standardized around the sample mean using the following formula (calculated using Excel):

$$d_{ij} = \sqrt{\left\{ \sum_{k=1}^{p} \left(x_{ijk} - \bar{x}_{jk} \right)^2 \right\}}$$

where *d* is the distance, *i* is the *i*th individual, *j* is the sample, x^{ijk} is the is the value of the variable, and x^{jk} is the is the mean of the same variable (Manly and Alberto 2016:43). The equation extracts the distance of the individual from the center of the sample, and the sample means of the values (*d* value) can be compared using Wilcoxon Rank Sums t-test in JMP Pro 13 to test for unequal variances.

Next, log covariance matrix determinants were compared between the Inner Coastal Plain and the Outer Coastal Plain (Stojanowski 2003:224). This was achieved through Quadratic Discriminant Function (QDF) analysis with shrinkage to take into consideration the differing sample sizes. QDF is a modification of Linear Discriminant Analysis (LDA), which assumes variables are normally distributed. It assumes, unlike LDA, that each class has a unique covariance matrix and it does not assume the covariance matrix is equal within groups. QDR develops discriminant functions that will discriminate between the categories of dependent (categorical) variables using linear combinations of independent (continuous) variables. This will allow examination of possible differences that may exist between populations (Lani 2017).

The calculation of log covariance matrix determinants for each group (inner and outer coast) was then bootstrapped 1000 times, and the ratio of the actual log covariance matrix determinants were compared to the resampled determinants produced through bootstrapping. This analysis ascertains if statistically significant results obtained in analysis were a result of actual variation between and within the two linguistic populations or due to a Type 1 or Type 2 error.

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Prediction of Hollowell classification in either the inner coast or outer coast was conducted using ANOVA, a Student's t-test and K-nearest neighbor (KNN). KNN was run to predict membership of Hollowell observations to either the inner coastal groups or outer coastal groups. To achieve classification of Hollowell observations to either group, KNN assigns unclassified observations to a particular group, based on similarity of measurements to other, classified observations.

CHAPTER 3: RESULTS

Multiple univariate and multivariate statistical tests were used to test the two hypotheses (Chapter 1) related to this study. Univariate analysis of inner and outer coastal between-group variability of each measurement utilized a simple t-test, and differences in within-group variability relied on Levene's and Bartlett's test. Multivariate analysis of between-group variability focused on only three measurements that effectively distinguished between the inner and outer coastal samples, first premolar buccolingual measurement (PM1BL), first molar buccolingual measurement (M1BL), and first molar mesiodistal measurement (M1MD) and a MANOVA test was used to evaluate between-group differences. Finally, multivariate analysis of heterogeneity in within-group variances was conducted using Van Valen's test in addition to comparing the ratio of log covariance determinants in each group versus a bootstrapped sample of ratios. Finally, Hollowell was compared to the inner and outer coastal groups to identify the more closely-related sample.

Table 3: Number of observations, mean and standard deviation for dental measurements of the Inner and Outer Coast groups

Measurement	Group	Number	Mean	SD
I1BL	Inner	0	0	0
	Outer	21	6.937	0.620
I1MD	Inner	0	0	0
	Outer	9	8.200	0.535
CBL	Inner	9	8.033	0.535
	Outer	44	8.044	0.542
CMD	Inner	0	0	0
	Outer	13	8.215	0.483
PM1BL	Inner	12	9.052	0.790
	Outer	41	9.537	0.556
PM1MD	Inner	4	6.823	0.180
	Outer	23	7.505	0.800
M1BL	Inner	12	11.773	0.662
	Outer	57	11.560	0.560
M1MD	Inner	5	11.390	0.802
	Outer	27	11.029	0.967

Univariate statistical analyses were utilized on data collected for this biodistance analysis study between Algonquian and Tuscarora populations. A total of 17 loose teeth and 84 maxillae containing dentition were included in the univariate analyses, with five loose teeth and 15 maxillae from the Inner Coast sample and 12 loose teeth and 69 maxillae from the Outer Coast sample. The Bonferroni-corrected alpha for multiple comparisons was set at a significance level of 0.0063. The results of univariate comparisons using a t-test found that no measurement showed a significant difference at the p<0.0063 level between the two linguistic groups (t=1.9188, p=0.0376).

Testing for differences in with-group variability by tooth measurement discovered that the Outer Coast group had more variability in the canine buccolingual measurement (CBL), while the inner coastal sites had more variability in the first premolar mesiodistal measurement (PM1MD). Levene's and Bartlett's tests found similar results, not surprising since the normal distribution of the samples would not generate false results from the Bartlett's Test, which is sensitive to departures from normality (Table 4, Figure 10).

Table 4: Levene's and Bartlett's results for differences in sample
variation within the Inner Coast versus the Outer Coast samples

Measurement	Test	F Value	DF	P Value
CBL	Levene's Test	8.6644	1	**0.0049**
	Bartlett's Test	5.4823	1	**0.0192**
PM1BL	Levene's Test	0.6404	1	0.4273
	Bartlett's Test	0.7332	1	0.3918
PM1MD	Levene's Test	65.5607	1	**<0.0001**
	Bartlett's Test	23.0167	1	**<0.0001**
M1BL	Levene's Test	0.9135	1	0.3426
	Bartlett's Test	0.7949	1	0.3726
M1MD	Levene's Test	2.7615	1	0.1070
	Bartlett's Test	2.1543	1	0.1422
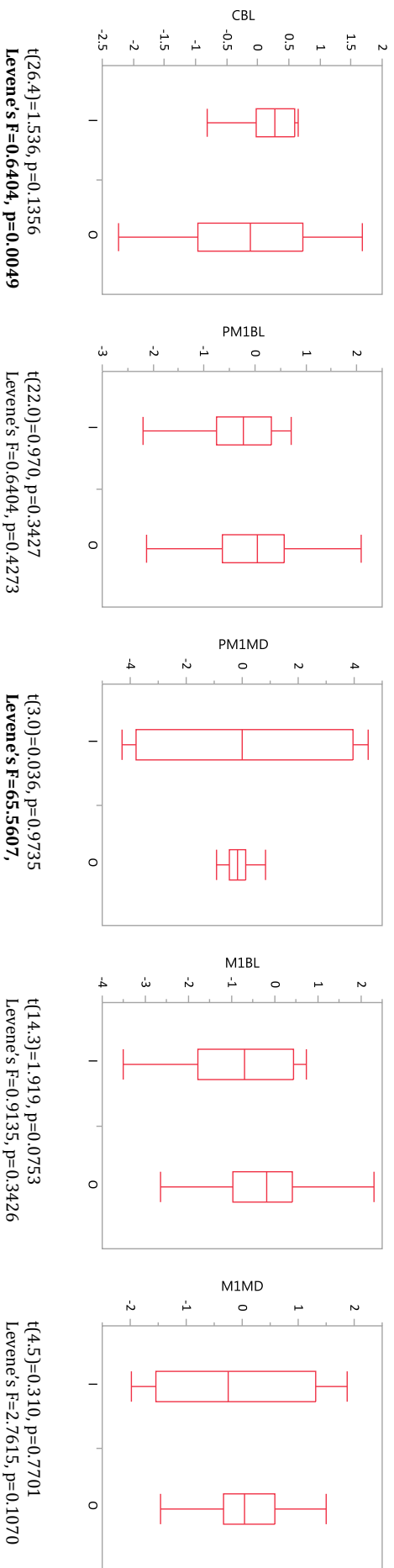
Figure 10: Boxplot of each measurement with t-test and Levene's results
(I=inner coast, O=outer coast; Bonferroni-corrected p-value = 0.0063)

CBL

t(26.4)=1.536, p=0.1356
Levene's F=0.6404, p=0.0049

PM1BL

t(22.0)=0.970, p=0.3427
Levene's F=0.6404, p=0.4273

PM1MD

t(3.0)=0.036, p=0.9735
Levene's F=65.5607,

M1BL

t(14.3)=1.919, p=0.0753
Levene's F=0.9135, p=0.3426

M1MD

t(4.5)=0.310, p=0.7701
Levene's F=2.7615, p=0.1070

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Multivariate Analyses

Various multivariate statistical analyses were also utilized to test between-population and within-population variability. As described in the methods, observations with only one measurement and variables missing more than 50% of observations were deleted, resulting in a significantly trimmed sample of seven loose teeth and 69 maxillae for multivariate tests, with three loose teeth and 13 maxillae in the Inner Coast sample and five loose teeth and 56 maxillae from the Outer Coastal Plain. In addition, three variables, first incisor buccolingual measurement (I1BL), first incisor mesiodistal measurement (I1MD), and canine mesiodistal measurement (CMD) had over 50% missing values, and were not included in the multivariate testing. In addition, correlations were found between PM1BL and first incisor buccolingual measurement (I1BL) (r 0.5420, p<0.0001), CBL and I1BL (r 0.4720, p<0.0001), PM1BL and CBL (r 0.6467, p<0.0001), and M1MD and PM1MD (r 0.4084, p=0.0003). However, each set of correlated variables had one variable removed in the analysis, as described below, minimizing the effects of correlated variables. In order to identify the variables responsible for variation between the groups, the eight dental measurements underwent stepwise selection for linear discriminant function analysis. A combination of two variables, PM1BL M1BL best discriminated the Inner Coast and the Outer Coast groups. While PM1BL did have significant correlation with I1BL and CBL, these measurements did not vary notably between groups and therefore were not included in analyses of between-population differences. Similar results were obtained from canonical discriminant analysis, with M1BL having the most discriminatory power, followed by PM1BL based on the canonical scoring coefficients (Table 5, Figure 11). In addition, principal components analysis (PCA) was run on covariance matrices of the inner and outer coastal groups in order to determine which variable or combination of variables were responsible for creating

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variance in the sample. Significant principal components were identified as having eigenvalues above or near 1.0 (Table 6). Here the results confirmed the variables identified above. PC1 only included M1BL, which explained 35.96% of the variation, followed by PC2 (PM1MD) for 26.74% of the variation (Table 6, Table 7).

Table 5: Canonical coefficients identifying the sources of variation between the inner and outer coastal groups

	PM1BL	M1BL
Canon 1	0.3481215	0.9080195
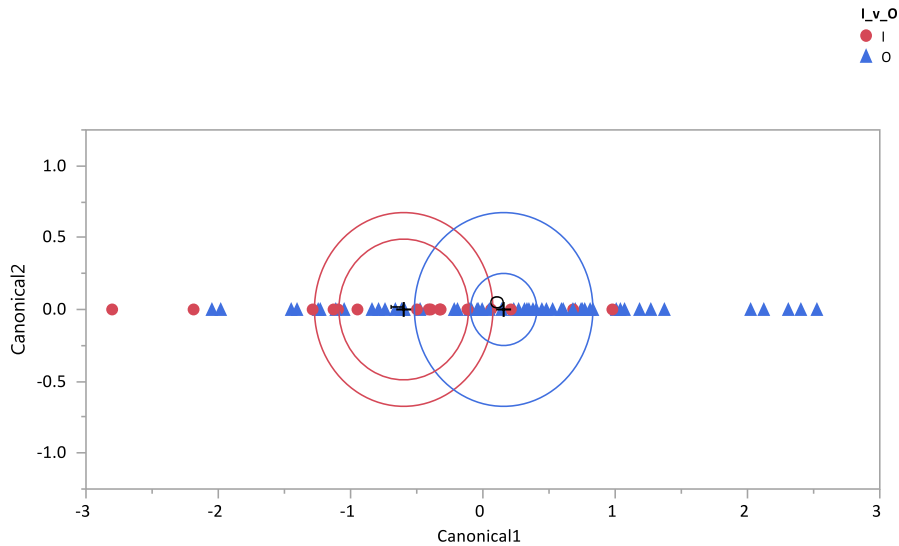
Figure 11: Discriminant canonical plot including PM1BL and M1BL showing discrimination between Inner Coast and Outer Coast

Table 6: Principal component analysis depicting significant eigenvalues and percentage of variation in the sample attributed to each variable

Number	Eigenvalue	Percent		Cum Percent
1	1.2478	35.961		35.961
2	0.9278	26.739		62.700
3	0.7227	20.829		83.529
4	0.3046	8.779		92.307
5	0.2669	7.693		100.00

Table 7: Eigenvectors of the principle components

Measurement	Prin1	Prin2	Prin3	Prin4	Prin5
CBL	0.27145	0.00534	0.50501	-0.32873	0.75046
PM1BL	0.28245	-0.10334	0.76754	0.23400	-0.51543
PM1MD	0.23834	0.88377	-0.10878	-0.33294	-0.22570
M1BL	0.87518	-0.27337	-0.39244	-0.03296	-0.06496
M1MD	0.15422	0.36539	-0.03849	0.85161	0.34056

The results of a MANOVA test comparing between-group difference utilizing only

PM1BL and M1BL found a significant difference between inner and outer coastal samples

(Table 8).

Table 8: MANOVA results on variation between inner and outer coastal groups using
PM1BL and M1BL only

Test	Value	Exact F	NumDF	DenDF	Prob>F
Wilks' Lambda	0.9119977	3.5703	3	74	0.0331
Pillai's Trace	0.0880023	3.5703	3	74	0.0331
Hotelling-Lawsley	0.096494	3.5703	3	74	0.0331
Roy's Max Root	0.096494	3.5703	3	74	0.0331

Van Valen's test then served to provide multivariate assessment of differences in with-

region variance using the entire sample and just the measurements selected by PCA as being the

most variable (PM1MD and M1BL). The means of the log-transformed d-values for each region were tested using Wilcoxon Rank Sums t-test, which found no differences (Z=1.45012, p=0.1470) in Inner Coast versus Outer Coast within-group variation using two most variable measurements (PM1MD and M1BL) (Figure 12). In addition, no significant difference in within-region variance was found using all of the variables (Z=0.86003, p=0.3898).
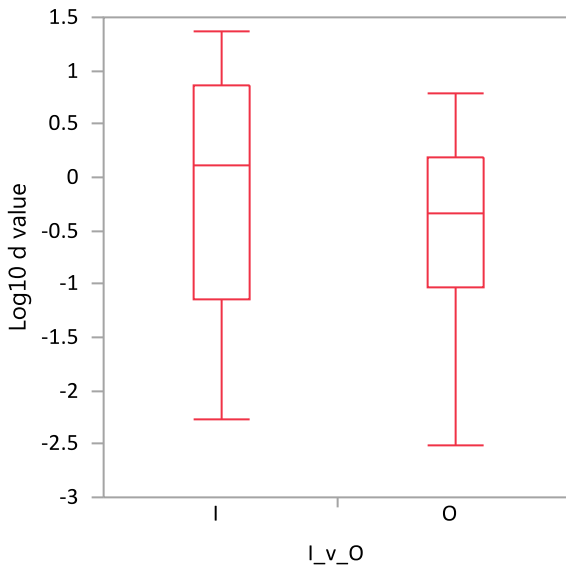
Figure 12: Boxplot of Inner versus Outer Van Valen d scores using only
PM1MD and M1BL (I=inner coast, O=outer coast)

However, the results of the log covariance determinants using all dental measurements found a significant difference in within-region variation. The ratio of the Outer Coast determinant (-5.354222) and the Inner Coast determinant (-7.593504) is 0.705106, which fell outside the 95% confidence interval of the bootstrapped sample of log covariance matrices (CI 95% = 0.3814861, 0.402003) (Figure 13). This suggests that the Inner Coast group has significantly more variation than the Outer Coast.
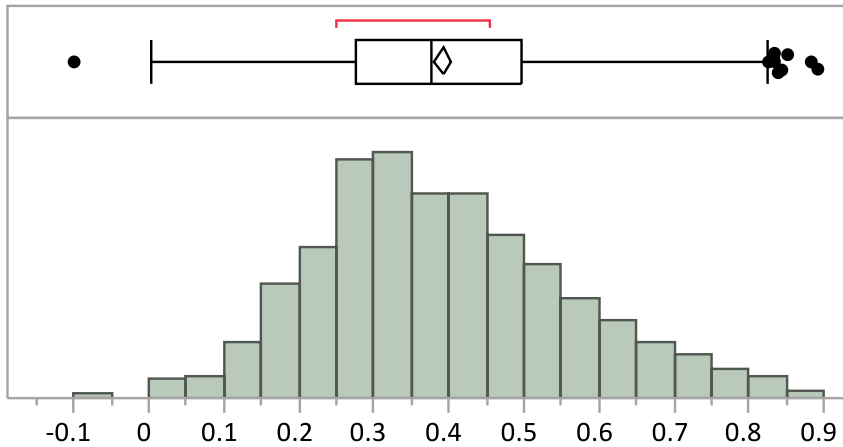
Figure 13: Bootstrapped distribution of the log covariance matrix determinants between inner and outer coastal groups

Hollowell. For the above analyses, dentition from the Hollowell site was placed into the Inner Coast sample based on its geographic location. In order to see whether Hollowell has dental metrics similar to Inner or Outer Coastal groups, the three groups were compared via ANOVA and the means compared for each pair using a Student's t-test. Hollowell showed significant differences from the Inner Coast group in the PM1MD (t=-1.99254, p=0.0085), M1BL (t=1.99254, p=0.0124) and M1MD (t=1.99354, p=0.0120), suggesting that those buried at this site most genetically resemble the Outer Coast population. Multivariate predictive modeling also was run using K-nearest neighbor (KNN), which included only non-Hollowell observations to train and validate the model to predict membership in the Inner or Outer Coast groups based on PM1MD, M1BL, and M1MD. Running the model to predict membership of the Hollowell observations in inner coastal vs. outer coastal samples classified all ten observations as Outer Coast members. These results confirm that of the univariate analyses that they more resemble the Outer versus the Inner Coast groups.

Summary

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Statistical analyses for this study on collected odontometric data have not indicated significant differences between the Algonquian and Tuscarora populations. Previous biodistance research on North Carolina Coastal Plain Late Woodland populations utilizing non-metric dental and cranial traits performed by Kakaliouras (2003) and Killgrove (2002), have indicated no genetic differentiation between Algonquian and Tuscarora human remains, indicating homogeneity between populations as well.

In this study, data infer greater genetic variability within the Inner Coastal groups than within the Outer Coastal population. Hollowell, a site of ambiguous cultural affiliation, is shown to have a closer affinity to the Outer Coast groups than to the Inner Coast, which is supported by Kakaliouras' non-metric dental data, but not Killgrove's study of craniometrics differences.

CHAPTER 4: DISCUSSION

Two linguistically-different groups, the Algonquian and Tuscarora populations, lived on the North Carolina Coastal Plain during the Late Woodland period (AD 800-1650). The two groups likely made contact during this 850-year span, and two other studies (Kakaliouras 2003; Killgrove 2002) using different genetically-linked data found that gene flow occurred between them. This study utilizes dental metric data from Algonquian and Tuscarora skeletal samples in order to confirm whether or not these genetically-linked traits differed significantly between the two groups, and whether or not one group displayed more within-group genetic variability than the other. Univariate analysis comparing each dental measurement found little difference between the linguistic groups, except the canine buccolingual (CBL) measurement containing more variation in the Outer Coastal sample and the first premolar mesiodistal (PM1MD) measurement containing more variation within the Inner Coastal sample. Multivariate analysis found that the first molar buccolingual measurement (M1BL) in addition to first premolar buccolingual (PM1BL) measurement showed more variation in the inner coastal/Tuscarora group. Van Valen's multivariate analysis using the two highly-discriminating measurements discovered through principal component analysis, the M1BL and PM1MD, found homogeneous variance between the two samples. A further test of the log covariance matrix determinants for each population found that, when compared to the result produced by a bootstrapped sample, the inner-group contained more variation than the outer-group.

Very small differences in dental metric variation were seen between the groups. The Algonquians arrived in the North Carolina coastal plan supposedly in AD 600 (Loftfield 1990:121), migrating from the region of New York State, and thus likely originally was a genetically distinct population from the Tuscarora (Langdon 1995:357). The greater genetic

variation in the Tuscarora sample identified by the log covariance matrix determinants ratio could derive from a number of factors. First, the inner coast Tuscarora could have been more regularly interbreeding with other groups, such as groups further west in the Piedmont region, while the outer coast Algonquians only chose to interbreed with the Tuscarora, or had less mate choice due to geographic constraints.

Next, the differences in variation could result from postmarital residence practices. The Tuscarora and Algonquian, along with the majority of groups in the southeastern U.S., were matrilineal (Heath and Swindell 2011:9; Perdue 2003:21; Pritzker 1998), and thus more Algonquian males moved into Tuscarora communities and had offspring than Tuscarora males moving to Algonquian areas. Finally, the reduced genetic variation in the Algonquian groups could result from founder effect within the Algonquian groups due to a relatively small group migrating to the Outer Coast, effectively lowering dental variation. Since the differences in variation are slight, the reduced variation in the Algonquian populations due to genetic drift over time had been ameliorated by gene flow occurring between the two groups since their arrival into the region.

Genetic homogeneity between the two regions suggests that perceived linguistic barriers in the coastal plain did not prevent gene flow. The idea that languages of prehistoric North Americans hindered gene flow, and thus skeletons can be grouped into linguistic typologies based on skeletal characteristics clearly misrepresents not only the level of genetic homogeneity within these populations, but also the issues involved with producing typologies based on biological variables. For instance, the use of cranial size for establishing ethnic or linguistic typologies or identities remained the focus in many parts of the world well into the 20th century (Cook 2006), even though it became clear from the work of Franz Boas (1912) that cranial

shapes are subject to both the environment, particularly diet and stress during growth, and heredity. In addition, specific ethnicities or linguistic groups can actually have significant within-population genetic variation that is greater than any genetic variation between these groups, one population may not represent succinctly a geographic region or specific culture as a whole (Templeton 2014:270). For example, a greater capacity to move between two linguistically different populations as opposed to other groups speaking the same language will result in greater gene flow between the better-connected yet linguistically diverse populations (Relethford 2012:239-241). As a result, introduced genetic variants into one subset of the linguistic group created greater genetic variability within the entire group as a whole than expected based on linguistic boundaries.

Numerous geographically and temporally diverse studies on modern populations have found that gene flow is more influenced by geography than language differences. For instance, analysis of Y chromosome variation in Neolithic Europe and Northern Africa found that the Mediterranean Sea along with other geomorphological features within Europe served as a greater barrier to gene flow than perceived linguistic differences (Rosser et al 2000). This has been explored more specifically in the Y chromosomes of Catalan and Basque populations, who diverged linguistically farther back in time than they diverged genetically, and thus continued to have genetic exchange even though the populations eventually spoke different languages (Hurles et al 1999). A broader study of modern European linguistic and genetic diversity also found that 20.3% of genetic variation correlated with geographic distance, while language only was linked to 1.0% of genetic variation (Harding and Sokal 1988). Similar results have emerged in studies of mtDNA. In Northeastern Thailand, mtDNA studies found that haplotypes could be grouped

based on geographic location, but had no correlation with linguistic classifications (Kutanan 2014).

Ethnohistoric accounts indeed have shown Algonquian and Tuscarora populations in North Carolina spoke a variety of languages (Lawson 1903[1714]:134). The multilingualism of the region and purported dominance of the Tuscarora and their language in terms of trade in the 18th century emerges in John Lawson's *History of North Carolina*:

> [T]he Difference of Languages, that is found amongst these Heathens, seems altogether strange. For it often appears, that every dozen Miles, you meet with an Indian Town, that is quite different from the others you last parted withal; and what a little supplies this Defect is, that the most powerful Nation of these Savages scorns to treat or Trade with any others (of fewer numbers and less power) in any other Tongue but their own, which serves for the Lingua of the Country, with which we travel and deal; as for Example, we see that the Tuskeruro's are most numerous in North Carolina, therefore their Tongue is understood by some in every Town of all the Indians near us (1903[1714]:134).

In addition, Mnemonic devices, such as smoke signals, beads, knots, and pictures (Swanton 1924:446), could have superseded spoken language to conveying meaning during inter-linguistic interactions.

Bilingual individuals in each village also may have made it possible for marriage partners to be obtained, thereby increasing gene flow (Jobling, Hurles and Tyler-Smith 2004:377). In addition, ethnohistorical documents of the Carolinas also include accounts of slaves being acquired through war (Fischer 2002:79; Hrdlička 1916:113; Swanton 1924:705). Adult slaves were adopted into the village as part of the group (Fischer 2002:79; Hrdlička 1916:113; Swanton 1924:705; Wallace 1972:46). Historical accounts indicate that slaves, once accepted as a

tribesperson, could attain a high distinction (Swanton 1924:705). In addition, the Tuscarora population, captives could marry within the group, thereby increasing genetic variation (Wallace 2012, 169).

Therefore, the most basic explanation for the observed pattern in between- and within-population variation seen is that the Algonquian group arrived in the outer coastal region with less genetic diversity than expected due to genetic drift. Over time, interbreeding with the Tuscarora (and closely related groups) led to gradual genetic synchrony with regional populations, but with retention of less genetic diversity. In addition, the genetic homogeneity between the groups implies that both groups interbred and perhaps practiced exogamous marriage. Ethnohistorical sources in fact provide evidence for this practice during the colonial period, when marriage between female Native Americans and European males occurred in order to establish terms of trade between the two groups (Fischer 2002:72; Swanton 1924:705). The matrilineal and matrilocal nature of the Native American groups meant if a colonial man married a southeastern Native American woman, he would be accepted into her clan and considered an acceptable trade partner with that clan's contacts. Colonial men who did not marry Native American women, on the other hand, had difficulty conducting trade (Fischer 2002:72). The importance of exogamous marriage to open trade relationships between groups could have existed during the Late Woodland period, suggesting intermarriage (and presumably resulting gene flow), would have been common. This correlates with the interpretations by Kakaliouras (2003) and Killgrove (2002) on differences in non-metric cranial and dental trait frequencies they used to compare inner and outer coastal populations.

One site included within the Outer Coast group for statistical analysis is the Hollowell (31CO5) site. This village has a mix of Algonquian and Tuscarora artifacts, particularly

ceramics, and is considered to be a border site between Algonquian and Tuscarora populations and perhaps a locus of inter-regional trade. Genetically, Hollowell is most likely from the Outer Coastal Plain due to data analysis from this study. While these results confirm those from Kakaliouras (2003:140) based on dental non-metric traits, it contrasts with the results from Killgrove (2002:120) based on cranial non-metric traits. Killgrove found the Hollowell site burial population was more closely genetically related to the Tuscarora.

While both metric and non-metric dental data agree on genetic relatedness of the Hollowell burial population, cranial non-metric data do not agree. One possible reason for the disagreement could be small the sample sizes used for Killgrove's analysis. For example, in regards to the West #2 site, she mentions only 4 crania out of 32 could be reliably scored for the infraorbital suture. Due to already small sample sizes, trait reduction was not performed. Therefore, Killgrove explained that cautious interpretation of the conclusions is in order due to small sample sizes. While sample sizes were small enough to require trait reduction in this study (e.g. I1BL, I1MD and CMD) for statistical analysis, enough data were still available to reach the same conclusion across multiple analyses. Regardless of population affiliation, Hollowell demonstrates that trade did occur in prehistory between the two linguistic groups, so it can be assumed communication took place.

Summary

Thus, the combined evidence of gene flow and slight differences in genetic variability suggests that interbreeding, and likely intermarriage, did occur between the Algonquian and Tuscarora linguistic groups. This gene flow not only led to similar dental metric data, but also likely increased the variability in the Algonquian migrants originally having low genetic variation due to genetic drift. Ethnohistoric data suggests that intermarriage was an important

link to establish trade between different communities. In addition, these early historic data also mention the exchange of young individuals between them in order to learn the unfamiliar language and serve as a translator (Fausz 1987:42). Hollowell, with its ambiguous population affiliation, proximity to both linguistic groups, and evidence of trade may have been a center for trade between the Algonquian and Tuscarora populations.

Studies have determined that other factors (e.g. geography) have impeded gene flow more than language. Anthropologists in the past have assumed a correlation between linguistics and cultural identity (Coe et al 1982; Hrdlička 1916; Loftfield 1990). Due to research in the North Carolina Coastal Plains, it has been shown that supposed linguistic and cultural boundaries between these groups did not create enough physical variation within groups to be seen with biodistance analysis. One cannot determine cultural and linguistic affinity by examining human remains in ossuaries in the North Carolina Coastal Plain. Despite linguistic differences in North Carolina, contact, trade and mate exchange did occur, resulting in homogeneity between and amongst Algonquians and Tuscarora.

CHAPTER 5: CONCLUSION

The Late Woodland period in the North Carolina Coastal Plain saw the emergence of semi-sedentary settlements and the use of agriculture. Hereditary chiefdoms were common. Two linguistic groups—the Algonquian and Tuscarora—populated the northern Coastal Plain. Both cultures used ossuary burial, though with a few differences. For Algonquians, numbers of individuals interred in an ossuary varied from 20-60 individuals with very little to no grave good inclusion. In contrast, Tuscarora ossuaries contained two to five individuals with grave good inclusion.

Currently, geography, subsistence and material culture, particularly ceramics, are being used to assign cultural affiliation of archaeological remains on the North Carolina Coastal Plain. Previous research, however, has failed to distinguish between Algonquian and Tuscarora burial practices based on skeletal morphology alone (Kakaliouras 2003; Killgrove 2002).

Recent research in biodistance analysis using non-metric cranial traits (Killgrove 2002), non-metric dental traits (Kakaliouras 2003) has failed to distinguish between Algonquian and Tuscarora burial practices based on skeletal morphology These previous studies, as well as this study, are in agreement that no correlation has yet been found between linguistic dissimilarity and skeletal differences.

In this study, the question of genetic similarity within and between the Algonquian and Tuscarora populations in the North Carolina Coastal Plains during the Late Woodland Period was examined. In order to determine biological convergence or divergence between the two linguistic populations, univariate and multivariate statistical analyses were conducted. No discernable skeletal or dental biological difference between the two linguistic groups was determined. Without significant biological differences between Algonquian and Tuscarora

populations, it can be inferred that language differences were not a hindrance to gene flow. Furthermore, one inference from this study is that exogamous marriage took place due to high genetic similarities between the two groups. If endogamous marriage were practiced, there would be genetic divergence between the two populations and lower genetic variability within (Kumar et al 2006; Oota et al 2001).

Future directions

Archaeologists will continue to assign cultural affiliation to archaeological remains based upon site location and artifact assemblages. Advances in aDNA have great potential to address questions of cultural contact and population migrations from the lower Great Lakes. Promising techniques for determining genetic convergence within and between populations are mtDNA and Y chromosome DNA. mtDNA and Y chromosome DNA are passed though the female and male lines respectively. Due to characteristics of mtDNA and Y chromosome genetic material, population history can be seen (Kemp and Schurr 2010). Differences between haplogroups, defined as a phylogenetic population with mutations in the coding region, can be determined, even in recently diverged groups (Kemp and Schurr 2010). aDNA may answer the question of how much contact the Algonquian and Tuscarora populations had prehistorically, and how long ago this contact began.

Another avenue of study would be biological distance analyses of prehistoric Algonquian and Iroquoian populations from the lower Great Lakes region. Very little research has been produced in this area on the topic (Langdon 1995). While it appears there is convergence of biological distance on the North Carolina Coast Plain during the Late Woodland, not much can be said for the Great Lakes region. While it is assumed gene flow occurred between lower Great Lakes Algonquian and Iroquoian populations in prehistory, a biological distance analysis would

help piece together population history of these two linguistic groups. Eventually, a skeletal morphology analysis between the Algonquian and/or Tuscarora populations from the lower Great Lakes and North Carolina Coastal Plain could be conducted to observe the amount of skeletal change due to environment and diet that took place. How much did dietary changes and migration from a colder, drier climate to one more humid and warm have on the skeletal morphology of these populations, if any?

There is still much to be learned in North Carolina and in North America about prehistoric Algonquian and Iroquoian population interactions, and how much those interactions altered population genetics between and among the groups. By conducting research into migration, diet and environmental changes, and how much these changes altered skeletal morphology, a clearer picture of prehistoric life-ways can be seen.

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