Analysis of Sexual Dimorphism in Human Eye Orbits using Computed Tomography

by

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Abstract

A plethora of anthropological studies have been undertaken on the skull, including many analyses of sexual dimorphism. Sexual dimorphism reflected in the eye orbits has not always demonstrated consistent or reliable results. However, recent studies (Pretorius, Steyn, & Scholtz, 2006; Ji et al., 2010) suggest some positive results utilizing geometric morphometrics to predict sex. Utilizing 97 post-mortem CT (computed tomography) scans, established morphological and metric techniques for sex determination were assessed from 3D rendered models of the crania. In addition, landmark data were collected on the orbital margin to evaluate the accuracy of sex determination using geometric morphometric techniques. Traditional methods demonstrated poor levels of accuracy for prediction of sex, however, utilizing generalised procrustes analysis and discriminant function analysis on 3D landmark data resulted in 94.95% overall accuracy. Application of recent methodological advances, including geometric morphometrics, should continue to be developed as it increases the ability to assess sexual dimorphism which will allow for greater identification of unknown remains.

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Dedication

To Hailey Lidstone, my reason for doing anything and everything.

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Chapter 1: Introduction

The time frame of interest may differ between forensic anthropologists and bioarchaeologists but their basic questions remain the same. Both fields are interested in creating a biological profile for any human skeletal remains that are recovered. In doing so they must determine the sex of the individual. How this is achieved is based on the bones that are recovered and their state of preservation. Doing a complete analysis of the body is ideal but not always possible. When recovering skeletal forensic or archaeological remains, they can quite often be incomplete or even include fragmented bones. In those situations it is necessary to get the most accurate information from the bones that are available. From human skeletal remains the skull is easily identifiable and preserves fairly well. When analyzing the skull to determine sex, the primary basis for sexual dimorphism is in the robusticity and size difference between males and females.

This research will focus on the orbits of the skull to establish a better understanding of their role, if any, in sexual dimorphism. The benefit of using the orbits to assess sex is debated, but they continue to be a part of skeletal analyses (eg. Williams & Rogers, 2006; Pretorius, Steyn, & Scholtz, 2006). An in-depth examination of the orbits will be undertaken to determine if they are a useful component for the determination of sex from skeletal remains. Three different approaches to evaluating the orbits will be applied and evaluated against known sex to assess which methods have the most potential for discriminating sex in bioarchaeological and forensic situations. Three broad areas of analysis will be tested, using previously published methods: scoring of morphological traits; metrics and 3D landmark analysis.

Chapter 2 presents the role of forensic anthropologists and bioarchaeologists, and their interest in skeletal remains. A literary review of the approaches taken by researchers to assess sex from recovered bone is discussed, with special emphasis on the skull. The chapter highlights the relevant literature regarding a variety of methods for sex determination, as well as methodological advances through the use of CT data in anthropological studies. Chapter 3 presents details on the CT dataset used for the analysis, derived from post-mortem scans at the University of Copenhagen. Following this, each of the previously published methods of sex determination to be evaluated on the 3D rendered CT data are described. Morphological comparison is based on shape, position and margin sharpness, while measurements reflect width and height dimensions, and finally a morphometric approach evaluates the shape of the orbital margins (Buikstra & Ubelaker, 1994; Rogers, 2005; White & Folkens, 2005; Dayal, Spocter, & Bidmos, 2008) The potential level of error for each method applied will be considered.

Chapter 4 presents the overall results of the validation study for each method. Accuracy of predicted sex for each technique is compared to the known sex for the sample. Finally chapter 5 discusses the impact of the results for the field of physical anthropology. The benefits of the findings as well as concerns with research are discussed. The best approach for future research is recommended.

Chapter 2: Determination of Sex in Osteology

Introduction

When human skeletal remains are discovered, whether it be from a forensic or archaeological context, a variety of basic questions about these remains need to be addressed. The answers to these questions make up the biological profile and concern the age, sex, ancestry and stature of the individual. Within bioarchaeology the basis of the work on human skeletal remains is in the construction of a life history (in a sense a more detailed biological profile), the investigation of mortuary practices, paleopathology, paleodemography and a wider population based research focus. Age and sex structures of past populations are important, not only for a general understanding of the population itself through rates of mortality and fertility, but also for aspects of migration (Meindl & Russell, 1998). Forensic osteology, on the other hand, uses the analysis of remains and the creation of the biological profile to aid individual identification and help narrow the field of investigation for law enforcement. Methods utilized by forensic anthropologists are under close scrutiny to conform to the Daubert standards of expert evidence for admission in court (Christensen, 2004; Rogers & Allard, 2004). The Daubert standards came about after a US Supreme Court ruling in the *Daubert vs. Merrell-Dow Pharmaceuticals, Inc*. requiring evidence to display proven scientific methods (Daubert v. Merrell, 1993; Christensen, 2004; Rogers & Allard, 2004). The method must be scientifically tested with known error rates, and peer reviewed and accepted (Daubert v. Merrell, 1993; Christensen, 2004; Rogers & Allard, 2004). As a result, the biological profile and the accuracy of assessing osteological evidence is of particular importance for forensic anthropology and bioarchaeology.

Sex Determination

Biological Determinants

The assessment of sex of found human remains is a pivotal tool in the establishment of the biological profile. Sex differs from gender in that it is based on the physical expression of the genetic makeup of males and females. Gender, although sometimes reflective of the genetic differences, is a cultural construct and must be acknowledged as such (Mays and Cox, 2000). The determination of sex is primarily based on the concept of sexual dimorphism which is based on differing rates of growth for primary and secondary sexual characteristics during adolescence (Meindl & Russell, 1998). In general, females are expected to be approximately 8% smaller in size than males (Krogman, 1962; Krogman & Iscan, 1986; Byers, 2002) but the difference in dimensions can be as large as 20% (White & Folkens 2000, 2005). Because sexual characteristics do not manifest until puberty, most methods for sex determination are limited to application on adults following the completion of skeletal development (Keen, 1950; White & Folkens 2000, 2005; Byers, 2002; Oettlé, Pretorius, & Steyn, 2009).

Regardless of the specific methods used, determination of sex can be conducted on various aspects of the skeleton. Two areas of the skeleton that have been the focus of a great deal of research are the pelvic and cranial regions. Due to the pelvic region's direct role in the birth process, it is seen as the most reliable in determining sex (Meindl

& Russell, 1998; Burns, 1999; White & Folkens, 2000; Byers, 2002; Walker, 2005; Bruzek & Murail, 2006; Đurić, Rakočević, & Đonić, 2005; Bytheway & Ross, 2010). Females are seen to have smaller and less robust os coxae and sacrum, a wider greater sciatic notch, and larger subpubic angle (White & Folkens, 2000; Byers, 2002; Bytheway & Ross, 2010).

Sex determination of the skull is traditionally based around the size and degree of robusticity of cranial features (Meindl & Russell, 1998; White & Folkens, 2000). The degree of robusticity is also influenced by age, therefore as individuals age they develop increased robusticity of the cranial features (Meindl et al., 1985; Meindl & Russell, 1998; Buikstra & Ubelaker, 1994). In other words a more 'masculine' morphology is seen in older individuals. The degree of robusticity is seen to vary both within and between populations (Keen, 1950; Giles & Elliot, 1963; Meindl et al., 1985; Buikstra & Ubelaker, 1994; Meindl & Russell, 1998; Konigsberg & Hens, 1998; White & Folkens, 2000; Kemkes & Gobel, 2006; Ramsthaler et al., 2010). Although the concern for population influence is evident, some believe the significance is not great enough to restrict the applications of methods (Henke, 1977; Kimmerle, Ross, & Slice, 2008; Gonzalez, Bernal, & Perez, 2011). Although not a strict rule of thumb, many morphological studies utilize single populations (Maat, Mastwijk, & Van Der Velde, 1997; Graw, Czarnetzki, & Haffner, 1999; Walrath, Turner, & Bruzek, 2004; Rogers, 2005; Williams & Rogers, 2006) while some metric and morphometric studies combine multiple populations (Giles & Elliot, 1963; Johnson et al., 1989, 1990; Kimmerle, Ross, & Slice, 2008). Utilizing more than one method on the same sample could compensate for potential population influences (Ramsthaler, Kreutz, & Verhoff, 2007). There are many previous studies of sex determination conducted on the cranium (Giles & Elliot, 1963; Johnson et al., 1989, 1990; Maat, Mastwijk, & Van Der Velde, 1997; Steyn & İşcan, 1998; Graw, Czarnetzki, & Haffner, 1999; Walrath, Turner, & Bruzek, 2004; Rogers, 2005; Williams & Rogers, 2006; Pretorius, Steyn, & Scholtz, 2006; Ramsthaler, Kreutz, & Verhoff, 2007; Kimmerle, Ross, & Slice, 2008; Walker, 2008; Gonzalez, Bernal, & Perez, 2011). The focus for these studies, regardless of method used, has primarily been on combining multiple variables for sex determination.

To date, accuracy and precision of specific traits to discriminate sex has been the focus when developing and comparing methods (Rogers & Saunders, 1994; White & Folkens, 2000; Rogers, 2005; Williams & Rogers, 2006). The accuracy levels are indicative of how reliably the methods can predict the correct sex in a documented sample, and precision rates are the degree of consistency in obtaining those results (White & Folkens, 2000; Bruzek, 2002; Rogers, 2005; Williams & Rogers, 2006). When examining variables for sex determination it is critical that studies not only assess the accuracy levels, but also consider the level of intra and inter-observer error for such techniques. Some researchers have argued that the level of error, and in turn the accuracy, can be influenced by a predetermined bias, which in the case of sex determination methods tend towards males (Weiss, 1972; Meindl, et al., 1985; Rogers & Saunders, 1994; Konigsberg & Hens, 1998).

Accuracy for sex determination is 50% based on random distributions alone. As such, physical anthropologists typically look for methods that exceed 80% accuracy with high levels of precision and low levels of error. Methods utilizing the pelvic bones for sex determination have resulted in accuracy levels ranging from 89-98.5% (Rogers &

Saunders, 1994; Bruzek, 2002; Byers, 2002; Walker, 2005; Bytheway & Ross, 2010). The accuracy of sex determination using the cranium can be upwards of 80-90% for those with experience (White & Folkens, 2000). The continual development and improvement of methods, including technology, is necessary to stand up to professional, and legal, review and standards. This is important for forensic anthropologists who must conform to the Daubert standards of scientific evidence. The Daubert standards require the testimony to have been proven using scientific methods, methods that are reviewed and accepted by others in the profession, and display known rates of error (Daubert v. Merrell, 1993; Christensen, 2004; Rogers & Allard, 2004).

Preservation

The preservation of the skeletal remains can greatly affect the assessment of the biological profile. Preservation is affected by numerous taphonomic factors including the composition and acidity of the soil, the wetness or dryness of the environment and the affects of weathering. Preservation is also strongly correlated with the bone composition. As individuals age the density of their bones decrease, and in turn preservation of those remains decreases (Walker, Johnson, & Lambert, 1988; Walker, 1995; Stojanowski, Seidemann, & Doran, 2002). Despite the males' more robust nature, there has not been consistent evidence that preservation is influenced by sex differences (Walker, Johnson, & Lambert, 1988; Walker, 1995; Stojanowski, Seidemann, & Doran, 2002). Those differences observed are most notable when combining age and sex, finding older less dense females less preserved than males of similar age (Walker, 1995). As the degree of preservation is strongly affected by bone density, differential bone densities affect the completeness of the skeleton recovered. Elements with a greater proportion of cancellous bone will deteriorate faster, whereas dense cortical bone will preserve longer (Mays, 1991; Walker, Johnson, & Lambert, 1988; Walker, 1995; Stojanowski, Seidemann, & Doran, 2002). Overall, the analyses conducted are strongly influenced by those elements that preserve best and are available for study. It is because of this variability in density within the body that the skull is most often preserved compared to the thin cortical bone of the pelvis (Mays, 1991; Stojanowski, Seidemann, & Doran, 2002; Spennemann, 1992; Bytheway & Ross, 2010). Archaeological burials lack well preserved pelvic regions in some 80-90% of the time (Walker, 2005). Although the pelvic region is the most reliable in sex determination it is important to study other regions, like the cranium, due to low preservation of the pelvis (Weiss, 1972; Mays, 1991; Stojanowski, Seidemann, & Doran, 2002; Spennemann, 1992; Walker, 2005). Due to the potential influence of preservation on demographic reconstruction (and the assessment of the biological profile), research that focuses on those elements that are most likely to survive is an important and valid approach, making the refinement and investigation of new techniques using the skull an important endeavour.

Morphological Techniques

The skull is one of the most informative and widely studied elements within the human body as it can be used to assess sex, age at death, ancestry, and for individual reconstruction for the purposes of personal identification. As White and Folkens (2005:

75) state: “the skull is the most complex portion of the skeleton and is of major importance for human osteology. It is one of the keys to aging, sexing, and understanding the evolutionary history of hominids”. The bones of the skull, including the ear ossicles, account for 28 of the bones found in the human body (Steele & Bramblett, 1988; Burns, 1999; White & Folkens, 2005). The bones of the skull encase the brain and support many other organs including those responsible for chewing and for the senses (White & Folkens, 2005). Skull bones are easily identifiable on the whole, however fragmentary pieces are often difficult to distinguish from one another.

The growth of the cranium is primarily complete at the onset of puberty (Baughan & Demirjian, 1978). Most of the dimorphic traits seen in the skull are a result of the differing hormones, times and rates of puberty occurring in males and females (Mays & Cox, 2000). The result is a difference in size and robusticity for males, specifically around areas of muscle attachment. Traditionally physical anthropology was focused on classifying these observable anatomical features through descriptive methods. The classic osteological descriptions of the skull were made on the mandible, nasal aperature, orbits, zygomatic bones, supraorbital ridges, glabella, forehead shape, mastoid process, occipital region, and palate (Krogman, 1962; Krogman & İşcan, 1986). The zygomatic bones, supraorbital ridges/glabella, mastoid process, occipital region, and mental eminence of the mandible are all based on the robusticity of the observed feature, i.e. males are identified as being large or prominent (Krogman, 1962; Krogman & İşcan 1986; Mays & Cox, 2000). Although still influenced by increased bone growth, nasal aperature, orbits, and palate are described based on shape, i.e. elongated, squarer, broader (Krogman, 1962; Krogman & İşcan, 1986). While the males gain larger, more thicker skeletal features, females maintain a more gracile "infantile" form (Krogman, 1962; Mays & Cox, 2000). Classifying anatomical traits or features can be done in a dichotomous fashion, i.e. male-female, square-round, large-small, etc., or based on a scale system (as described in Buikstra and Ubelaker 1994). The scale system is mainly focused on those traits that differ in size or expression, such as mastoid process or nuchal crest. The trait is graded from least prominent (female) to clearly and largely expressed (male), a 3-point scale will have an indeterminate/ambiguous middle classification, or the more common 5-point scale allows for a 'questionable' female, ambiguous sex, and 'questionable' male classification (Buikstra & Ubelaker, 1994; Konigsberg & Hens, 1998; Đurić, Rakočević, & Đonić, 2005; Walker, 2008). Although the 5-point system could account for population diversity, not all traits can be defined by a grade, i.e. orbits - square/round, low/high. One way to determine where a skull fits morphologically into a population is through seriation, although this is not always feasible with large populations, multiple variables, or for isolated remains (i.e. forensic cases) (Konigsberg & Hens, 1998).

A review of the literature displays a range of studies looking at these traits, in varying combination, and is summarized in Table 1. Morphological methods have resulted in accuracies ranging from 70-96%, with the majority closer to ninety percent accuracy (Keen, 1950; Maat, Mastwijk, & Van Der Velde, 1997; Konigsberg & Hens, 1998; Graw, Czarnetzki, & Haffner, 1999; Đurić, Rakočević, & Đonić, 2005; Rogers, 2005; Williams & Rogers, 2006; Ramsthaler, Kreutz, & Verhoff, 2007; Walker, 2008; Ramsthaler, et al., 2010). The value at the low end of this range (70%) comes from Graw and colleagues' (1999) study on a single variable, the supraorbital margin. At the other end of the spectrum there is Ramsthaler and colleagues (2007, 2010), and Maat and colleagues (1997) with accuracy approaching 96%. Those studies try different combinations in order to obtain the greatest accuracy, and in the end the greatest accuracy is achieved by the full suite of traits. It should be noted that Maat and colleagues' (1997) study determined accuracy of the complete skull by comparing it to the pelvic analysis of sex, rather than known sex. Early on Stewart (1954) criticized this approach, maintaining that although the pelvic region has a high degree of sexual dimorphism there still remains a small margin for doubt as to the true sex.

Table 1: Cranial Morphological Sex Determination References

Reference	Sample	Feature/Trait	Accuracy	Error
Keen 1950	100 Cape Coloured	supraorbital ridges, occipital crest and nuchal lines, ridge at upper rim of auditory meatus	85%	NA
Maat et al. 1997	202 Netherlands	glabella, superciliary arch, frontal and parietal tubera, frontal inclination, mastoid process, nuchal plane, external occipital protuberance, temporal zygomatic bone, supramastoid crest, shape and sharpness of the rim of the orbit, robustness of the mandible, shape of the mentum, prominence and shape of the angle, inferior margin	96.2%	NA
Konigsberg & Hens 1998	138 Averbuch site	supercilliary arch form, mastoid form, superior orbtial margin form, nuchal cresting, chin form	83%	NA
Graw et al. 1999	108 German	Supraorbital margin	70%	3.4% (inter-observer)
Đurić et al. 2005	180 Balkan	Size of mastoid, size of occipital protuberance, nuchal cresting, sharpness of supraorbital margin, supercilliary arch form, prominence of the supramastoid ridge, robustness of the mandible, size of mental eminence, size of frontal tuber	70.56%	k = 0.9035 (inter-observer)
Rogers 2005	46 European	occipital condyle size (14%), tooth size (10.3%), size and architecture (38%), size of mastoid (44.7%), size of supraorbital ridge(60.9%), parietal eminences (28.9%), nuchal crest (53.3%), chin form (56.3%), mandibular symphysis & ramus size (51.1%), palate size and shape (36.6%), malar size and rugosity (68.4%), frontal eminences (31.9%), forehead shape (44.5%), zygomatic shape (70.3%), nasal size (52.8%), nasal aperature (76.6%), orbit shape and position (43.6%)	89.1%	11% (intra-observer)
Williams & Rogers 2006	50 Euro-American	size and architecture (88.0%), frontal eminences (64.0%), size of supraorbital ridge (86.0%), orbit margins (76.0%), nasal apperature (84.0%), size of nasals (68.0%), zygomatic extension (82.0%), size of mastoid (92.0%), occipital markings (58.0%), size and shape of palate (74.0%), mandible-symphysis height (58.0%), mandible-gonial angle (80.0%), mandible-gonial eversion (58.0%), chin form (72.0%)	92%	7.4% (intra-observer)
Ramsthaler, Kreutz & Verhoff 2007	98 Middle European	skull size and shape, supraorbital ridge, upper orbital rim, chin shape, tubera frontalia, mastoid process, occipital muscle ridges, zygomatic arch extension, mandible ramus	94%	k = 0.79 (inter-observer)
Walker 2008	460 Mixed ancestry	nuchal crest (71.4%), mastoid process (78.6%), glabella/supraorbital area (82.6%), supraorbital margin (68.8%), mental eminence (76.6%)	88%	96% (inter); 99.5% (intra)
Ramsthaler et al. 2010	50 central European	Glabella (81%), supraorbital ridge (85%), shape of orbits (56%), upper orbital edge (69%), decline/slope of frontal bone (67%), chin shape (61%), frontal eminences (65%), mastoid process (69%), supramastoid ridge (54%), external occipital protuberance (60%), occipital muscle ridges (66%), cheekbone shape (23%), cheekbone arch (42%), mentum (55%), condylar process (37%), gonial angle (49%), mandible lower side (42%)	96%	k = 0.83 (inter-observer)

Morphological analysis can be fairly subjective as it relies on a researcher's ability to visualize the differences between the bones. The application of morphological techniques can become more difficult if the variation is so slight as to be almost non-existent visually. Although the accuracy for morphological methods is comparable, if not slightly greater than metric techniques, their level of error is quite large with as high as 10.2-20% (Meindl, et al., 1985; Rogers, 2005; Williams & Rogers, 2006). This greater error is a product of the definitions and subjectivity of the traits in question, which causes difficulty in the assessment and subsequent comparisons (Walrath, Turner, & Bruzek, 2004; Rogers, 2005). A stronger and more reliable approach is displayed with combining the analysis of multiple traits rather than looking at individual traits (Rogers, 2005; Williams & Rogers, 2006). It is not always a guarantee as to which features will be available for analysis, so it is important that individual traits have high levels of accuracy on their own. Rogers (2005) had some extremely low accuracies, well below the 50% threshold from chance alone, including tooth size (10.3%), occipital condyle (14%), and parietal eminences (28.9%). Although not quite so low as Rogers, Ramsthaler and colleagues (2010) had some low individual values, including cheekbone shape (23%), condylar process (37%), and lower side of mandible (42%). Having high individual accuracy is especially important when analyzing incomplete skeletal remains where the full suite of traits may not be available.

Metric Techniques

In contrast to the more subjective morphological traits, metric data have also been examined for discrimination of sex. Measurements can be explicitly defined, with less interpretation and therefore produce less 'uncertain' individuals (Giles & Elliot, 1963; Weiss, 1972; Konigsberg & Hens, 1998; Kemkes & Gobel, 2006). Due to the reliance on standard landmarks, there are specific instructions and definitions in metric analysis which contrast the subjective nature of morphological analysis. Stewart argued that taking measurements instead of using morphological techniques would be a "waste of time and effort to measure such specimens simply to verify what the eye so quickly discerned" (1954: 389). The field of anthropology has developed quite a lot since Stewart's statement but the underlying objection to a metric approach still resonates. Obtaining information on individuals and/or populations should never be considered a "waste of time and effort" in research. A seasoned anthropologist with many years experience and vast knowledge of common variation may prefer to rely on their 'first impressions' when evaluating skeletal material. However, having methods that can identify differences that the eye cannot establish cannot be ignored. Having such methods also benefits those not as experienced, who may have more difficulty discerning those slight differences. In defense of a metric approach Giles (1966:86) states "the purpose of multivariate statistical analysis of both race and sex differences is scarcely to put the trained physical anthropologist out to pasture, but rather to put in his hands, and in the hands of others who must make such judgments, a tool that will elevate the level of objectivity at which he can operate". Although long before the development of the

Daubert standards for scientific evidence, this statement reflects the basis of what the standards are trying to instil. This requires the anthropologist to be accountable for how and what they do. Criticisms for metrics focus on potential errors and the lack of shape details provided in the numerical proportions (Stewart, 1954). Metric analysis has been restricted by the use of standard points and more importantly by many of the equations used in analysis being limited to those populations on which they were developed (Giles & Elliot, 1963; e.g. Ramsthaler, Kreutz, & Verhoff, 2007). However, metric methods have the benefit of being readily available for statistical analysis. This statistical backing supports a more 'scientific' appearance which is beneficial for forensic anthropologist when testifying in a courtroom (Reichs, 1986; Dayal, Spocter, & Bidmos, 2008). The standards set out by Daubert require the scientist to demonstrate the scientific validity through proven reliability and minimal error in the applied method (Christensen, 2004; Rogers & Allard, 2004).

Table 2: Cranial Metric Sex Determination References

Reference	Sample	Measurement	Accuracy
Keen 1950	100 Cape Coloured	**maximum length**, length of base, length of foramen magnum, maximum breadth, median sagittal arc, basion-bregma height, horizontal circumference, cranial capacity (cranial index, height-length index, base-maximum length, base-median sagittal arc index), total face height, **maximum bizygomatic diameter** (total face index), weight of mandible, angle of mandible, weight of cranium, nasion to bregma, bregma to lambda (parietal length), **depth of infratemporal fossa**, **length of mastoid process**, porion to superior temporal line, porion to vertex (index of temporal muscle extent), profile angle at nasion	85% (incl. bold)
Giles & Elliot 1963	408 Terry and Todd	glabello-occipital length, maximum width, basion-bregma height, basion-nasion, maximum diameter bizygomatic, basion-prosthion, prosthion-nasion height, palate -external breadth, mastoid length	82-89%
Kajanoja 1966	232 Finnish	maximum width, maximum bizygomatic diameter, glabello-occipital length, basion-bregma height, basion-prosthion, basion-nasion, prosthion-nasion height, nasal breadth	79%
Birkby 1966	104 American Indian	glabello-occipital length, bizygomatic diameter, basion-prosthion length, basion-nasion length, prosthion-nasion height	86%
Henke 1977	Westerhus	df 31: maximum skull length, basion-bregma height, bizygomatic breadth	88%
Snow et al. 1979	52 Forensic	cranial length, basion-nasion length, bizygomatic breadth, basion-prosthion length, prosthion-nasion length	88.5%
Johnson et al. 1989/1990	139 Modern mix	Caucasoids: bizygomatic breadth, maximum length glabella-opisthocranion, nasal breadth, subnasal height, palatal length, angle opisthion-basion-nasion; Mongoloids: angle opisthion-basion-nasion, maximum length glabella-opisthocranion, foraminal length, foraminal breadth, subnasal height, occipital chord	87% - 97%
Cunha & van Vark 1991	570 Coimbra	74 variables: 61 measurements and 13 angles	80.10%
Inoue et al. 1992	121 Japanese	39 craniometric points from radial lines crossing with lateral contour line	86%
Hsiao et al. 1996	100 Taiwanese	angular: GMSN, GMFH, GMBaN, GSgM, IOpSN, IOpFH, IOpBaN, OIOp; linear: SgGM, GSgN, FSHt, FSWd, IOpO, MaSN, MaFH, MaHt, MaWd; proportional: GPI	100%
Steyn & İşcan 1998	91 white South African	Cranial: maximum length, maximum frontal breadth, minimum frontal breadth, bizygomatic breadth, nasal height and breadth, basion-nasion length, basion-bregma height, basion-prosthion length, nasion-prosthion length, mastoid height, and biasterionic breadth; Mandibular: bicondylar breadth, bigonial breadth, minimum ramus breadth, gonion-gnathion length, and total mandibular length	80-86%
Konigsberg & Hens 1998	80 Averbuch site	maximum cranial length, maximum cranial breadth, frontal breadth, bizygomatic breadth, basion-bregma height, basion-nasion length	77.50%
Graw et al. 2005	410 Forensic	angle of meatus acusticus internus	66%
Franklin et al. 2005a	332 South African	cranial length, cranial breadth, basi-bregmatic height, upper facial height, bizygomatic breadth, alveolar-basion length, maxillo-alveolar breadth, mastoid length	77-80%
Kemkes & Gobel 2006	97 German; 100 Portuguese	porion, mastoidale, asterion (landmarks)	65%
Ramsthaler, Kreutz & Verhoff 2007	98 Middle European	maximum length, max cranial breadth, bizygomatic breadth, basion-bregma height, basion-nasion length, basion-prosthion length, biauricular breadth, nasal height, nasal breadth, frontal chord, parietal chord, occipital chord, nasion angle, basion angle, bregma angle	86% Fordisc
Dayal et al 2008	120 black South African	maximum cranial length, basi-bregmatic height, min frontal breadth, bizygomatic breadth, basion-nasion length, basion-nasospinale length, basion-prosthion length, nasal height, nasal breadth, orbital breadth, orbital height, palate breadth, palate length, upper facial height, total facial height, bicondylar breadth, bigonial breadth, ramus height, ramus breadth, total madibular length, gonion-gnathion length	80-85%
Kranioti et al. 2008	178 Cretan	cranial length, basion-nasion length, max vault breadth, max frontal breadth, min frontal breadth, bizygomatic breadth, foramen magnum length, foramen magnum breadth, basion-bregma height, basion-prosthion length, nasion-prosthion height, mastoid height, biorbital breadth, interorbital breadth, nose breadth, nose height	87.1-88.2%
Gapert et al. 2009	146 St. Bride's	max length of condyle, max width of condyle, max bicondylar breadth, min distance between condyles, max interior distance between condyles, external hypoglossal canal distance	53.4%-76.7%
Robinson & Bidmos 2009	230 South African of European descent	max length, max frontal breadth, bizygomatic breadth, nasal height, nasal breadth, basion-nasion length, basion-bregma height, bicondylar breadth, bigonial breadth, min ramus breadth, gonion-gnathion length, total mandibular length	72-95.5%

Like morphological descriptions of the skull, metric approaches endeavour to quantify the differences in size and shape based on robusticity. There are three ways to evaluate metric data; through raw measures, ratios or indices, and via discriminant function analysis (Reichs, 1986). Regardless of the method of metric comparison, the features being measured are similar to the morphological features analyzed. Buikstra and Ubelaker (1994) list 34 potential cranial and mandibular measurements, although few utilize the full suite of measurements in sex determination. Common measurements in cranial sex determination include maximum breadth, maximum length, skull height, facial breadth, full facial height, and upper facial height (Krogman, 1962; Krogman & İşcan, 1986). A review of previous studies utilizing metric data can be seen in Table 2, including univariate (e.g. Graw, Wahl, & Ahlbrecht, 2005) and multivariate approaches of up to 27 variables (e.g. Keen, 1950). Keen's (1950) early study based their analysis on the raw measurements, including indices and angles. From the original 27 variables, a recommendation of 4 measurements (maximum length, maximum bizygomatic diameter, depth of infratemporal fossa, and length of mastoid process) is made for future studies. Classification is based on the mean and standard deviation determined for each sex; i.e. for maximum length females are considered to be < 179mm and males are > 184mm, and any measurements between 179 – 184mm are considered indeterminate (Keen, 1950). The vast majority of the studies seen in Table 2 utilize discriminant function analysis to classify sex. Pioneers for discriminant function in anthropology were Giles and Elliot's (1963) and their study of cranial measurements. Their findings utilized 9 cranial measurements to create 21 discriminant functions, each containing a different combination of measurements (Giles & Elliot, 1963). Each function is essentially an equation where measurements can be plugged in, multiplied by the appropriate coefficient and added together to create a single value. This value is then compared to the sectioning point and standard deviation for that discriminant function, i.e. d.f. #1 has a sectioning point of 2676.39, with males greater than that and females less than (Giles & Elliot, 1963). Some have utilized these discriminant functions for their own research (Kajanoja, 1966; Birkby, 1966; Snow et al., 1979), while many others have used their own variables to create separate discriminant functions (Henke, 1977; Inoue, et al., 1992; Hsiao, Chang, & Liu,1996; Johnson, et al., 1989, 1990; Cunha & van Vark, 1991; Steyn & İşcan, 1998; Franklin, Freedman, & Milne, 2005a; Gapert, Black, & Last, 2009; Dayal, Spocter, & Bidmos, 2008; Kranioti, İşcan, & Michalodimitrakis, 2008). Metric methods have generated accuracy results of 53-100%, with greater focus around the mid-eighties (see Table 2). The lowest accuracy (53%) is seen in Gapert and colleagues (2009) analysis of measurements focused on the condyles, which they deemed to display low sexual dimorphism. The highest accuracy (100%) was based on discriminant function analysis of 18 variables (Hsiao, Chang, & Liu, 1996). The authors acknowledge the optimistic results are likely caused by the same dataset defining the discriminant function as evaluating it, a fact also acknowledged by Konigsberg and Hens (1998). This bias in the discriminant function can be diminished through cross-validation (Hsiao, Chang, & Liu, 1996). Any research involving measurements should also have an understanding of the measurement error present in the study, and is accomplished by performing multiple measurements per specimen or minimally for core group of specimens and calculating a standard deviation for those measurements (Hammer & Harper, 2006). The majority of the studies in Table 2 have not highlighted a measurement of error for the applied technique, and one study that did, displayed a high correlation between observers via a kappa statistic of 0.93 (Ramsthaler, Kreutz & Verhoff, 2007). Not having this data available hinders the ability to use such methods in legal matters, as they will not conform to the Daubert standards.

Morphometric Techniques

Buikstra and Ubelaker (1994) recommend that sex determination be based on both morphological analysis and metric calculations, an approach that is also supported by Ramsthaler, Kreutz and Verhoff (2007), and originally done by Keen (1950). Gaining popularity is geometric morphometrics, which provides the reliability of metrics, with a more revealing morphology and less restriction on the location of analysis than strict metrics. Taxonomy, microevolution, ontogeny, intraspecific variation (e.g. polymorphism, sexual dimorphism), and asymmetry are some classic applications of morphometrics (Hammer & Harper, 2006). Intraspecific variation (i.e. variation within a single species) is directly applicable to studies by bioarchaeologists and forensic anthropologists on human remains. The variation displayed in the skeleton is routed in the genetic makeup of the individual. As a whole, human remains can display multiple variations (polymorphism) reflective of the ancestral lineage from which they come and a dichotomous variation (dimorphism) representative of the male/female genetic differences visible in the skeleton (Jurmain et al., 2000; Byers, 2002). Regardless of the number of expressions possible for each skeletal feature, the basis for the skeletal differences is in the form and shape. Morphometrics "refers to the measurement of the shape and size of organisms or their parts, and the analysis of such measurements" (Hammer & Harper, 2006: 78). Measurement of objects can be categorized into four basic groups: univariate, multivariate, outlines, and landmarks. Morphometrics that evaluate distances, distance ratios, and/or angles through multivariate statistics but lack geometric information relating to biological structures are known as 'traditional' or 'multivariate morphometrics' (Slice, 2005). The term "geometric morphometrics" is in reference to the analysis of the position of landmarks or outlines on an object. That is, it is the analysis of shape while still retaining all of the geometric information contained within the data (Slice, 2005). An important aspect of this type of analysis is how easily the results are visualized and interpreted, unlike more "traditional" morphometric methods (Hammer & Harper, 2006). A classic variable utilized in morphometrics is distance. Distance does not rely on orientation or position, however maintaining the endpoints relative locations connected to shape becomes difficult (Slice, 2005). A way to overcome this difficulty is to define the points of interest based on Cartesian coordinates. When these coordinates are associated with an anatomically defined location they are referred to as 'landmarks' (Slice, 2005). Applications utilizing landmarks benefit the investigator by being able to reference specific points on a biological form versus traditional morphometric methods (Lele & Richtsmeier, 1991; Richtsmeier, Cheverud, & Lele, 1992; Lele, 1993; Hammer & Harper, 2006). Landmark data have three classifications; anatomical landmarks (homologous between specimens), mathematical landmarks (based on geometric property), and pseudo-landmarks (constructed) (Hammer & Harper, 2006). Landmark based approaches have been questioned on their stated use of biological homologous points. Homologous points are expected to "be consistently and reliably located with a measurable degree of accuracy on all forms considered" (Lele & Richtsmeier, 1991: 415). However, biological homology generally applies to whole structures and not single geometric points utilized in 'landmark' methods (MacLeod, 1999).

While landmark methods are beneficial, the primary goal of research utilizing these methods is to be able to relate the calculated difference to an observable location, be it for growth, ontogeny or phylogeny (Corner & Richtsmeier, 1991; Richtsmeier, et al., 1993; Cole & Richtsmeier, 1998; Lague & Jungers, 1999; Richtsmeier, DeLeon, & Lele, 2002). The methods that incorporate landmarks are often divided into two categories; coordinate-based and coordinate-free (Lele, 1991; Richtsmeier, Cheverud, & Lele, 1992; Richtsmeier, et al., 1993). When applying a coordinate system to an anatomical specimen and its points, there are no real set of axes that have biological significance, and therefore an arbitrary axis is often defined in these circumstances (Richtsmeier, Cheverud, & Lele, 1992; Richtsmeier et al., 1993; Slice, 2005). In order to avoid this arbitrary registration coordinate-free methods compare the landmarks of a form to each other. The form of an object is constant and will not change despite translation, rotation and reflection (Lele, 1991; Lele & Richtsmeier, 1991; Richtsmeier, Cheverud, & Lele, 1992; Lele & Richtsmeier, 1995). The analysis of shape is also not influenced by orientation or location, but differs from form because size is also removed (Lele, 1991; Lele & Richtsmeier, 1991; Richtsmeier, Cheverud, & Lele, 1992; Slice, 2005). In addition to landmarks, outlines of the object can be generated and compared. These methods may be considered pseudo-landmark methods as coordinate points are digitized along the curve of interest (MacLeod, 1999). Criticisms of earlier outline methods claim a lack of homologous points for comparison. Recent outline methods attempt to correct for this, as is seen with Elliptical Fourier analysis and Eigenshape analysis, using an initial homologous starting point for all objects (e.g. Chen, Lestrel, & McColl, 2000; MacLeod, 1999).

As the field of anthropology progresses, so must the number of methods and the technology available to apply those methods. With advancing technology, measurements are made more accurate and easier, expanding on the traditional tools of rulers, calipers and goniometers (Hammer, 2002; Bytheway & Ross, 2010). In contrast to classic metrics, digital measurements allow more variables to be included and are able to capture more information including variability (Rohlf, 1990; Kimmerle, Ross, & Slice, 2008; Bytheway & Ross, 2010). Geometric morphometrics does not take anything away from the traditional data but rather adds a level of increased sensitivity to the data that allows subtle differences in shape to be identified (Franklin, et al., 2007; Bernal, 2007). This increased sensitivity is more appropriate for modern populations that appear to display lower levels of variation (Cunha & van Vark, 1991; Kemkes & Gobel, 2006; Bernal, 2007). This advancement has generated many recent studies in anthropology utilizing geometric morphometric techniques (Pretorius, Steyn, & Scholtz, 2006; Kimmerle, Ross, & Slice, 2008; Oettlé, Pretorius, & Steyn, 2009; Bytheway & Ross, 2010; Gonzalez, Bernal, & Perez, 2011). The two most common morphometric methods associated with landmark data are Procrustes and Euclidean Distance Matrix Analysis (EDMA). The older and more widely used Procrustes method is a coordinate-based method which adjusts for size and orientation through superimposition (Lele, 1991; Richtsmeier, Cheverud, & Lele, 1992; Richtsmeier, et al., 1993; Lele & Richtsmeier, 1995;

Richtsmeier, DeLeon, & Lele, 2002; Slice, 2005). EDMA, in comparison, avoids registration methods and does not rely on a location within a coordinate system (i.e. is coordinate-free). It is interested in form (size and shape) rather than merely the shape of the object (Lele, 1991, 1993; Richtsmeier, Cheverud, & Lele, 1992; Richtsmeier, et al., 1993; Lele & Richtsmeier, 1995; Richtsmeier, DeLeon, & Lele, 2002; Slice, 2005).

Accuracies of sex determination from studies utilizing geometric morphometrics approaches have ranged from 68-89% for cranial methods, seen in Table 3, and 90-99% from postcranial methods (Pretorius, Steyn, & Scholtz, 2006; Kimmerle, Ross, & Slice, 2008; Oettlé, Pretorius, & Steyn, 2009; Bytheway & Ross, 2010; Gonzalez, Bernal, & Perez, 2011). Similar to what was observed in the morphological and metric studies, the lowest geometric morphometric accuracy (68%) was observed for a single trait, the ramus flexure (Pretorius, Steyn, & Scholtz, 2006). The highest level of accuracy was observed by Kimmerle and colleagues' (2008) in their study of 16 craniofacial landmarks, which produced 89% accuracy. Three of the studies involving geometric morphometrics collected their data from photographs of the bone that were then digitized (Pretorius, Steyn, & Scholtz, 2006; Oettlé, Pretorius, & Steyn, 2009; Gonzalez, Bernal, & Perez, 2011). Kimmerle and colleagues (2008) used a digitizer to locate and record the landmarks. Information provided by photographs is limited by the choice of location and angle of the photo and future research will be restricted to that information. Both of these approaches restrict the use of further analysis in the future. If the correct photograph or landmarks were not obtained at the time of data collection, future research will need to obtain the original sample in order to proceed. This in essence defeats the purpose of maintaining a digital archive. In order to analyze the data once the landmarks were collected, most studies relied on a Generalized Procrustes Analysis, followed by a Principle Components Analysis and/or Discriminant function analysis (Kimmerle, Ross, & Slice, 2008; Oettlé, Pretorius, & Steyn, 2009; Gonzalez, Bernal, & Perez, 2011).

Table 3: Cranial Geometric Morphometric Sex Determination References

Reference	Sample	Feature/Landmark	Accuracy
Pretorius et al. 2006	191 black South African	Greater Sciatic Notch, Mandibular Ramus Flexure, Orbit Shape	87.1-93.1%(GSN), 67.8-69.9%(RF), **73.3-80.0%(O)**
Kimmerle et al. 2008	118 W.M. Bass	Alare, Basion, Bregma, Frontomalare anterior, Frontomalare temporale, Lambda, Maximum malar projection, Nasion, Opisthocranion, Opisthion, Subsubspinale, Frontotemporale	86.65-89.66%
Oettlé et al. 2009	74 black South African	Mandibular gonial eversion	71-74%
Gonzalez et al. 2011	125 Coimbra	Frontex, Nasion, Frontomalare anterior, Frontomalare temporale, Infraorbitale, Zygomaxillare anterior, most superior point on suture between zygomatic process and temporal process, most inferior point on suture between zygomatic process and temporal process, Auriculare, lateral aspect of inferior border of zygomatic process, anterior point on root of mastoid process, posterior point on root of mastoid process, 25 semilandmarks	77.86% discrimin; 72.15% k-means

The focus of this research was inspired from Pretorius and colleagues' (2006) study utilizing geometric morphometrics. Pretorius and colleagues' (2006) research examined the shape of the greater sciatic notch, mandibular ramus flexure, and shape of the orbits with geometric morphometrics. Their primary focus was on the usability of geometric morphometrics in assessing sexual dimorphic characteristics. Their methods involved photographing the traits/features in a standardized position and viewing the information as an electronic image in jpg format. Landmarks were then assigned to their images as the points of comparison. Analysis of these landmarks involved relative warps, thin-plate splines, and canonical variates analysis (CVA). The greater sciatic notch was expected to display sexual dimorphism and performed as expected, providing the greatest separation between the sexes. The orbits were not expected to fare well in this analysis and ended up with surprising results. Although it did not perform the best it did perform better than ramus flexure, specifically in association with CVA (80.0% females and 73.3% males correctly placed with the orbits, versus 67.8% females and 69.9% males placed correctly with the ramus flexure). The researchers recommended further analysis in this area in order to determine if the "orbit shape is more sexually dimorphic than previously expected, or that biological reality is not reflected by this technique" (Pretorius, Steyn, & Scholtz, 2006: 64). They encouraged using as many techniques as possible in order to explore the results and determine which area is best suited to demonstrate differences. Studies on sex determination using geometric morphometrics have been conducted within the last decade, indicating that this method is a fairly new endeavour in anthropology and requires further investigation in order to obtain its true potential with different skeletal elements. One way to help establish the contribution of geometric morphometrics in anthropology is to apply the technique alongside more traditional methods of sex determination. Refining the applicable methods in order to be as precise and accurate as possible is of great importance.

CT Data in Osteology

Traditional osteological analyses have been conducted and developed directly on bones. Human skeletal remains are recovered, either from an archaeological site or more recent forensic crime scene, and analysis is promptly performed on those remains. Direct assessment of the bones is not always the most appropriate or practical. Modern digital technology helps to overcome potential obstacles when dealing with mummies or sensitive material. Having a digital archive of the data also allows for multiple methods to be applied at present or in future research. Additionally, the ability to do research globally becomes possible without the need for travel.

Medical advances have provided cross-sectional slices of scanned individuals in the form of computed tomography (CT). Increasingly, anthropological studies have made use of data from computed tomography (CT) scans (Park, et al., 2006; Ramsthaler, et al., 2010). When CT data are provided in the primary 2D slice format these are similar to plain x-rays that are commonly used in physical anthropology (Robinson, et al., 2008). However, in x-rays, radiographed structures are overlapped and are not clearly visible (Park, et al., 2006). Algorithms, based on soft tissue and bone, are used to reconstruct the scanned CT data of the subject (Horton, et al., 2002). Complicated graphics and a variety of mathematics can be applied quite easily to such data (Hildebolt & Vannier, 1988; Corner, Lele, & Richtsmeier, 1992; Park, et al., 2006). Skeletal anatomy and imaging of such "features" as tumours, trauma, and infection is greatly improved with the use of 3D CT imaging (Horton, et al., 2002). Inner structures and particular features or organs can be highlighted, separated and rotated (Hildebolt & Vannier, 1988; Park, et al., 2006). The negative drawbacks of such a tool would be cost and radiation (Hammer, 2002; Park, et al., 2006). Radiation is less of a concern for anthropological research as we are not utilizing living subjects. As technology becomes more widespread, the cost for using such technology will continue to decline.

The use of CT scans benefits researchers in many ways. CT scans become a valuable tool with the non-destructive ability to display internal structures of bone with no harm to the actual bone (White & Folkens, 2000; Ramsthaler, et al., 2010). The benefits to the bones themselves are not only for preservation, but also aid researchers in analyzing remains with decomposing tissues where health and safety issues are a concern (Robinson, et al., 2008). Traditional anthropological examination of bone is conducted once the flesh has been removed. The implementation of CT allows for measurements to be acquired quicker as the need to remove the flesh first is no longer necessary (Robinson, et al., 2008; Ramsthaler, et al., 2010). Not only is the analysis available to fleshed or contaminated remains, obtaining CT images brings the analysis to the international level, for peer review or large international collections. Also, as new bone collections are not as frequent, creating digital archives becomes a greater need (Ramsthaler, et al., 2010).

Despite some of the positive benefits for using CT scans there are some concerns in research when obtaining landmarks and measurements from the data. Richtsmeier and colleagues (1995) identify three types of error; the quality of the scanner/instrument producing the image, the observer recognizing the landmarks, the use of software correctly to locate and record landmarks. During reconstructions density thresholds are applied to the image in order to display the area of interest, and it is the choice of threshold that should also be considered a potential source of error when obtaining data from CT images (Williams & Richtsmeier, 2003). As coordinates are rounded to nearest pixel, it is important to set the equipment to the maximum allowed resolution (Hammer, 2002). The location of the landmark is restricted by the scanned points available in the dataset, and will be approximated to those available (Hammer & Harper, 2006). In a study where some error was observed it must be noted that those landmarks utilized were identified using the 2D sliced images (Richtsmeier, et al., 1995). There is no more information added with a 3D reconstruction, however the authors make note that it looks similar to the object itself which may make the identification easier and allow for a greater number of landmarks to be identified (Richtsmeier, et al., 1995; Williams & Richtsmeier, 2003). This will be especially beneficial for newer researchers who are not familiar with the technology but are knowledgeable of the bones themselves. The use of both the axial slices and 3D reconstructions increased the accuracy of obtaining landmarks as it decreased any difference found between direct vs. digital collection of data (Williams & Richtsmeier, 2003). Three dimensional data and CT scanned data are seen to provide appropriate information and data for morphological comparisons and traditional measurement analysis (Franklin, Freedman, & Milne, 2005b; Ramsthaler, et al., 2010). Another aspect of 3D reconstructions that affects error is the ability to consistently orient the reconstruction in order to identify and locate the landmarks (Corner, Lele, & Richtsmeier, 1992). Although physically handling the bone is not possible, traditional Frankfort orientation is still relied on to maintain consistency. Hildebolt and Vannier (1988) found in their study that, when compared with landmark measurements using calipers, 3D measurements of those landmarks were not significantly different from the traditional ones, and were performed quickly and without potential anatomical obstacles. Also, Robinson and colleagues' study (2008) did not determine any significance in the difference between measurements based on the technique applied. Both CT and direct physical measurements had a range of accuracy from 67-73%, where both methods had a part in inter-observer variability when identifying the landmarks (Robinson, et al., 2008). This indicates that measurements made using the CT scans are comparable to those made using dry bone. Studies have found that the level of error in the location and utilization of landmarks to be within an acceptable range and of benefit in research (Corner, Lele, & Richtsmeier, 1992; Valeri, et al., 1998; Williams & Richtsmeier, 2003). An unacceptable level of error when calculating the distance between landmarks is that greater than 5% (Corner, Lele, & Richtsmeier, 1992). It is necessary for landmarks to be identified and located with ease in order to be useful in studies, while still maintaining high accuracy and precision (Richtsmeier, et al., 1995; Valeri, et al., 1998). Developing standards for CT measurements should be done separately from those of traditional landmark measurements and therefore would require their own validation (Robinson, et al., 2008). Overall, studies using CT data are valid, as they are found to be precise and accurate, however, it is recommended that they be used independently and not in conjunction with direct bone measurements or other various sources (Lele & Richtsmeier, 1991; Richtsmeier, et al., 1995).

Chapter 3: Materials and Methods

The bones that articulate to create the eye sockets, known as the orbits, consist of 7 skull bones. The seven bones that create each orbit are the frontal, maxilla, ethmoid, lacrimal, zygomatic, palatine and the sphenoid (Burns, 1999; White & Folkens, 2005). The basic assumptions of size and robusticity in sex determination of the overall skull can be applied specifically to the orbits. Males are larger, more robust, and more specifically seen to have squarer orbits, with females wider and flatter (Meindl & Russell, 1998; White & Folkens, 2000; Hennessy, Kinsella, & Waddington, 2002). Generally, morphological, metric and the current morphometric methods rely on the presence of the complete or essentially complete cranium. The orbits are important because as part of the skull they tend to preserve well and can be included in the biological profile. Studies on the orbits are also useful because the unique shape allows researchers to implement all three methodological approaches onto one skeletal feature. The orbits have documented morphological aspects, standard biological landmarks located on the orbit margin, and the shape of the orbit margin permits itself to geometric morphometric analysis.

The main scope of this study touches on previous work performed by Pretorius, Steyn and Scholtz (2006). The purpose of their research was to determine if geometric morphometrics provided a better assessment of common morphological characteristics for sexual dimorphism. Although the orbits were not their primary focus, their research did demonstrate some potential, including the potential for using the shape of the orbits as sexually dimorphic characteristics. The present research will focus on this characteristic, and will additionally include metric and morphological analysis of the orbits. In contrast to the findings by Pretorius and colleagues (2006), Williams and Rogers (2006) and Rogers (2005) did not find the shape of the orbits to be of much significance in displaying sexual dimorphism. Evaluating the sharpness of the orbit margin has not demonstrated consistent results, accuracies have ranged from 28-76% (Đurić, Rakočević, & Đonić, 2005; Williams & Rogers, 2006; Walker, 2008). It must be noted that a difference in methodology is present in these studies, morphometric analysis (Pretorius, Steyn, & Scholtz, 2006) versus gross anatomical visualization (Rogers, 2005; Đurić, Rakočević, & Đonić, 2005; Williams & Rogers, 2006), and it could be this difference that is the cause in the varying results. Williams and Rogers (2006) did demonstrate a difference in correlation with sexual dimorphism when analyzing the orbits through general shape and location versus the appearance of the orbit margin. The orbit shape and location provided a higher rate of error (12%) compared to the margin of the orbit (10%), and was therefore not included in continued analysis of Williams and Rogers (2006) study. The high rate of error demonstrates difficulty in analyzing the visual shape based on the current morphological descriptions. This may be an area where the detailed and statistical comparison of geometric morphometrics will increase the identification of such differences, and decrease the level of error. Although not directed at sex determination specifically, Ji and colleagues' (2010) study on 3D reconstructions of the orbits did find males and females display noticeable differences. Research displaying a dimorphic difference in sex only enforces the present author's belief that an appropriate method of analysis for the orbits is possible in anthropological research. A large component of this research is to determine if geometric morphometric data are capable of displaying, if any, sexual dimorphism. This research will help explore the value of the orbits, as well as the use of geometric morphometrics, for determination of sex on a documented, modern post-mortem sample of CT scan data. This information is of particular importance in creating a biological profile of individuals, an application important for archaeological and forensic investigators.

Materials

The data used for this study was taken from a randomly selected sample of 104 adult post-mortem, computed tomography (CT) scans collected by the Department of Forensic Medicine, University of Copenhagen, Denmark. These data contain adult individuals of documented sex and age. This collection will provide a test of the range of variability between individuals within a single population when analyzing the orbits of the skull. From the original 104 scans, only 101 individuals were able to be included in the following analysis; 2 individuals could not be used for any analysis due to frontal damage, and 2 case files were duplicates of one individual, so case file #52 was also removed from comparison. Of the 101 individuals utilized in this study not all were able to have each method applied; 2 individuals could not be used in metric analysis due to incomplete or abnormal scanned images, 1 individual could not be used in metric or morphometric analysis due to zygomatic damage, and one additional individual could not be used for morphometric analysis due to an incomplete scan. This leaves a total of 97 individuals available for the full suite of analysis and comparison. The data were collected using a Siemens/Sensation 4 scanner, with a 512 x 512 matrix, 120 KV, and 169.5 mAs. The thickness of the slices ranged from 0.5mm to 3mm. The average age of the sample is 51, ranging from 19 to 88 years old. With approximately equal distribution of males and females, the average age for each sex is 46.6 and 54.6, respectively. Case information for each individual, including slice thickness, age, and sex can be found in the Appendix.

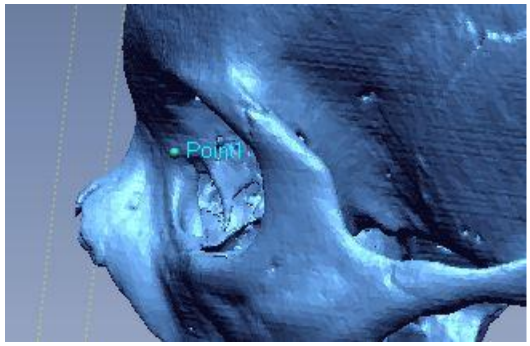
Methods

CT data of the cranium have been tested and utilized to validate the representation of true form, and the measurements that can be obtained from the object (Hildebolt & Vannier, 1988; Richtsmeier, et al., 1995; Williams & Richtsmeier, 2003; Park, et al., 2006; Ji, et al., 2010). As Pretorius and colleagues (2006), and Williams and Rogers (2006) demonstrate, there is some debate as to the value of the orbits themselves, and for this reason cannot be ruled out of analysis. As such, this research will explore the accuracy and reliability of sex determination from geometric morphometrics of the orbits using 3D rendered models from computed tomography (CT) scan data. The reliability, as indicated by the level of error, ensures that the methods can be reproduced with great precision multiple times and/or from multiple investigators. In addition to the geometric morphometrics, this research will examine standard metric and morphological comparisons of the orbits. This will allow for comparison of current published methods on a single individual and potentially provide either a greater or more selective range of methods for future investigators. Applying metric and morphological methods together on a single dataset allows for proper comparison of each method as well as the usability of the image format in analysis. Being able to compare the methods conducted on a single sample will highlight the more valuable and informative approach, and will hopefully demonstrate whether the eye orbits do in fact display sexually dimorphic characteristics. Overall, the primary objective of this research is to determine if geometric morphometrics can be used on human eye orbits to assist in identifying sexual dimorphism. This will be achieved by attempting to identify male and female patterns of orbital shape using a series of landmarks along the margins of the orbits. As demonstrated in previous studies (Corner, Lele, & Richtsmeier, 1992; Rogers & Saunders, 1994; Rogers, 2005; Williams & Rogers, 2006), this research will rely on 80% as the appropriate level of accuracy, and will accept a level of error ≤ 10% for classification and ≤ 5% for landmark placement, in order to display validity. The proposed application has implications for legal matters in forensic cases, and cultural matters in archaeological finds.

Data Acquisition

The CT data were rendered in 3D models using MIMICS imaging software. All data collection necessary for each method was subsequently carried out using INUS Rapidform XOR3 64 reverse engineering software. The first objective within this study was to isolate the eye orbits on the CT data from the rest of the skull. The 3D data were oriented onto the Frankfort horizontal plane from the left side and then rotated on the horizontal plane until the orbits were centered on the screen. The model was then zoomed in so as to minimize any influence from other skull features while still retaining necessary features of the orbits needed for analysis. In order to maintain consistent placement of the Dacryon landmark, these were assigned on the skull while in the lateral view, prior to rotation and zooming, see Figure 1.

Figure 1: Placement of Dacryon



Once in position the morphological characteristics were assessed and recorded for each individual. Morphological characteristics of the orbits previously used in sex determination include orbit shape and position, and the orbit margin (Table 4). The orbit margin was evaluated via a cross-section. This was obtained with a vertical line section originating from the Orbitale. In order to maintain consistency, and not have to re-align the images more than once, this step was conducted after all data were collected for the final two techniques.

In order for the remaining methods to be performed, a total of sixteen landmarks were indentified and located on the 3D data using the left orbits. Four of the landmarks are standard biological landmarks often utilized and the remaining twelve landmarks are pseudo-landmarks. Figure 2 displays the biological landmarks identified for both left and right orbits, while Figure 3 displays the order of the morphometric landmarks located on the left orbit. The pseudo-landmarks were obtained by applying a spline (a line of digitized points fitted along the curve of interest) to the margin between each biological landmark, and limiting the number of points to three (a total of 5 including a biological landmark at each end). A list of the biological and morphometric landmarks and their definitions can be seen in Table 5 and Table 6, respectively. Initial measurements were obtained from the standard landmarks. These standard orbit measurements (Buikstra & Ubelaker, 1994), seen in Table 7 and visualized in Figure 4, were inputted into Fordisc 2.0 (Ousley & Jantz, 1996). Fordisc generates a classification and probability of sex for each individual based on the orbit measurements.

The final analysis of sex determination on the orbits will include a comparison of data of all sixteen landmarks along the left margin. These data were analyzed using the morphometric statistical software program PAST (PAleontological STatistics) (Hammer, et al., 2001). A Procrustes analysis was conducted on both the two-dimensional and three-dimensional data to transform it under a common coordinate. These transformed data were analyzed using discriminant function analysis with a leave-one-out approach in PAST as well. This analysis provides a classification for each individual based on the discriminant function determined for the full sample while leaving out the one individual to be classified.

The total analysis of the CT data will compare the morphological assessment with the results generated by the measurements inputted into Fordisc 2.0 and PAST. These findings will be compared to the known data to determine the accuracy of each method. Accuracy greater than or equal to 80% will display the orbits as a useful indicator of sex.

Error Testing

The data collected were repeated for 20 random individuals two weeks after the original collection was completed. This will determine an intra-observer error for the methods applied. Intra-observer error less than or equal to 10% will be considered an acceptable level of precision for the methodology. Landmark placement of less than or equal to 5% mean difference will be considered accurate. The data will also be compared using PASW Statistics 17.0 software to determine the significance of the various traits and measurements. The morphological features were compared using Spearman's Correlation Coefficient, while the measurements for both the standard metric analysis and those between the observed landmarks were compared using a standard t-test.

Table 4: Morphological Orbit Criteria from Sex Determination

Morphology	Male	Female	References
Orbit Shape	square, low	round, high	Burns 1999, Rogers 2005, Williams and Rogers 2006, White and Folkens 2005
Margin Sharpness	Rounded, thick	Sharp, thin	Bass 1995, Burns 1999, Rogers 2005, Williams and Rogers 2006, White and Folkens 2005, Buikstra and Ubelaker 1994, Walker 2008, Byers 2002, Graw *et al.* 1999

Figure 2: Biological Landmarks

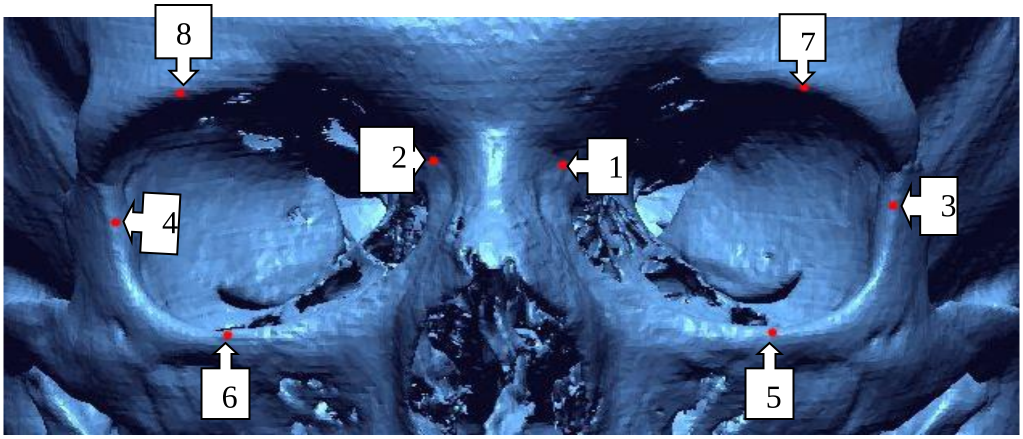


Table 5: Biological Landmark Definitions

Landmark	Definition
Point 1 & 2 = Dacryon*	paired point at intersection of the anterior lacrimal crest and frontomaxillary suture
Point 3 & 4 = Ectoconchion*	paired point at the outer edge of eye orbits
Point 5 & 6 = Orbitale*	paired point at the lowest part of orbital margin
Point 7 & 8 = Superior Orbital Border*	perpendicular to the natural horizontal axis of the orbit

*(taken from Burns 1999,pg43)

Figure 3: Morphometric Landmarks for the left orbit

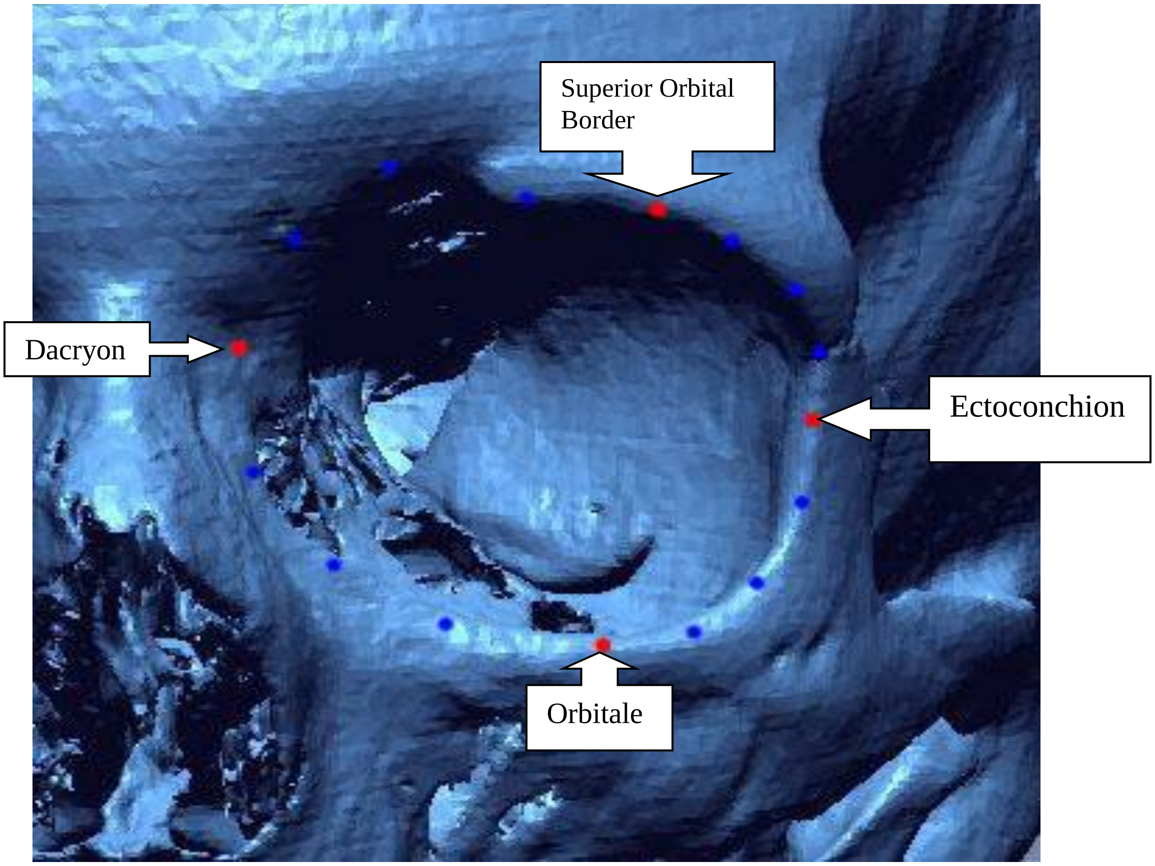


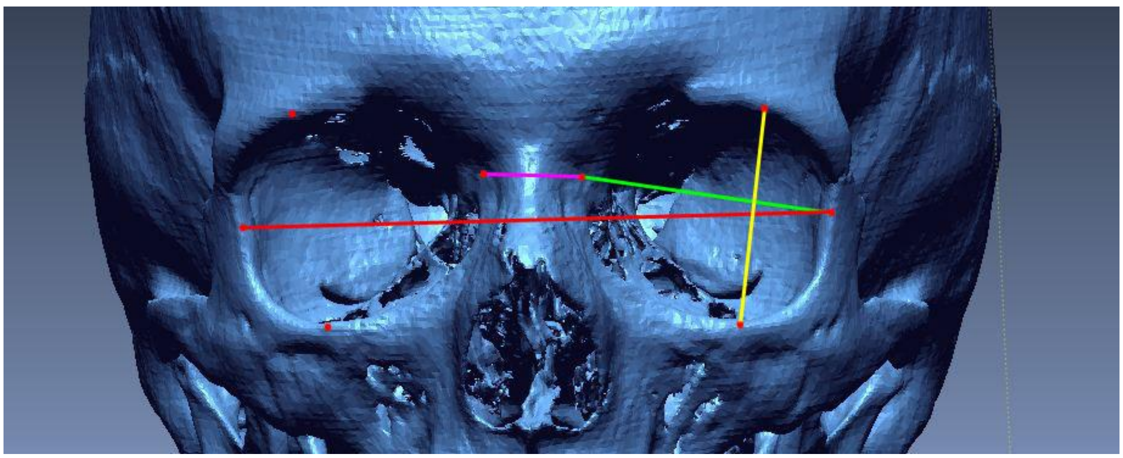
Table 6: Morphometric Landmarks

Landmark	Definition
Point 1	Dacryon
Point 2-4	along orbital margin between dacryon and superior orbital border point
Point 5	Superior Orbital Border
Point 6-8	along margin between superior orbital border and ectoconchion
Point 9	Ectoconchion
Point 10-12	along margin between ectoconchion and orbitale
Point 13	Orbitale
Point 14-16	along margin between orbitale and dacryon

Table 7: Orbit Measurements

Measurements	Cranial Locations
Orbital Width (ORBR)	Dacryon (d) to Ectoconchion (ec)
Orbital Height (OBH)	Superior Margin to Inferior margin/Orbitale
Biorbital Width (BIOB)	Ectoconchion (ec) to Ectocochion (ec)
Interorbital Width (INTB)	Dacryon (d) to Dacryon (d)

Figure 4: Orbit Measurements



Chapter 4: Results

A total of 101 individuals, 50 males and 51 females, were analyzed based on morphological expression. The gross morphological assessment consisted of three variables; orbit shape, orbit position and margin sharpness. All individuals in the sample were assessed for sex based on each of these traits. The results, presented in Table 8, Table 9 and Table 10 show the overall distribution of accurate prediction of sex for each technique. Orbit shape demonstrated an accuracy of 58.4%, with 56% correctly classified as male and 60.8% as female. While orbit position displayed an accuracy of 69.3%, with 64% correctly classified as male and 74.5% as female. The final morphological feature, margin sharpness, had an overall accuracy of 65.3%, with accuracies of 62% for males and 68.6% for females. In each of the three variables, the females were correctly classified more often than males. Overall, all three methods were poor predictors of sex in this sample and do not surpass the 80% requirement.

Table 8: Known Sex vs. Orbit Shape

Known Sex * Orbit Shape			Orbit Shape		
			male	female	Total
Known Sex	male	Count	28	22	50
	female	Count	20	31	51
	Total	Count	48	53	101

Table 9: Known Sex vs. Orbit Position

Known Sex * Orbit Position			Orbit Position		
			male	female	Total
Known Sex	male	Count	32	18	50
	female	Count	13	38	51
	Total	Count	45	56	101

Table 10: Known Sex vs. Margin Sharpness

Known Sex * Margin Sharpness			Margin Sharpness		
			male	female	Total
Known Sex	male	Count	31	19	50
	female	Count	16	35	51
	Total	Count	47	54	101

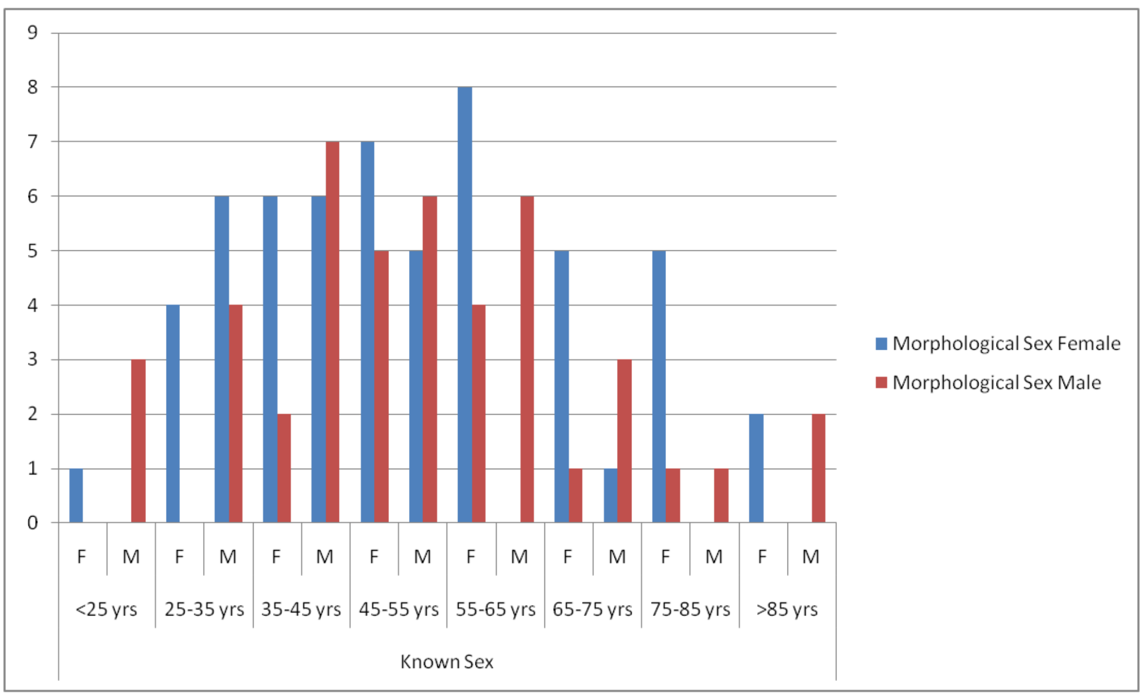
The majority of morphological studies combine multiple variables when obtaining their final classification of sex. Combining the three variables for a multivariate gross morphological approach, resulted in 38 (74.5%) individuals being correctly identified as female and 32 (64%) correctly identified as male. Determining a final classification of sex using all three variables, more females were correctly identified than males. All three morphological traits agreed with each other on the classification of sex for 54 individuals (24 males and 30 females). Of those, 39 (18 males and 21 females) were correct classifications. When comparing the multivariate classification with the known sexes, 70 were correctly identified for an accuracy of 69.3%. The breakdown of the male and female distribution can be seen in Table 11. In order to look at how potential age-related changes may impact the morphological analysis, a chart (Figure 5) displaying the distribution of sex classification based on ten year age groups was created from the data.

The known sex of individuals in each age group was compared to the multivariate classification of those individuals. A chart displaying the classification of sex obtained from each method for each individual in the sample can be found in the Appendix.

Table 11: Known Sex vs. Multivariate Morphology

Known Sex * Multivariate Morphology			Multivariate Morphology		
			male	female	Total
Known Sex	male	Count	32	18	50
	female	Count	13	38	51
	Total	Count	45	56	101

Figure 5: Age distribution of morphological sex classification



The traditional measurements were evaluated on 98 individuals from the sample (48 males and 50 females), using the Fordisc database to compare and predict sex. The morphological multivariate assessment did not agree with the Fordisc evaluation in 43 individuals, nearly half of the sample. In contrast to the morphology, the metric evaluation resulted in 17 (34%) females and 43 (89.6%) males correctly identified. More males were correctly identified utilizing the metric assessment than females. This is in direct contrast to the morphological approach, which classified females correctly more often than males. Bar charts representing the morphological multivariate assessment and the Fordisc evaluation provide a visual representation of the differences (Figure 6 & Figure 7). The probabilities associated with the Fordisc evaluations ranged from 50.1-91.3% for females (average 69.1%) and 51.1-99.9% for males (average 84.5%). When compared to the known sex data an overall accuracy of 61.2% was obtained, and can be seen summarized in Table 12.

Figure 6: Distribution of Males and Females classified by a combined multivariate morphology

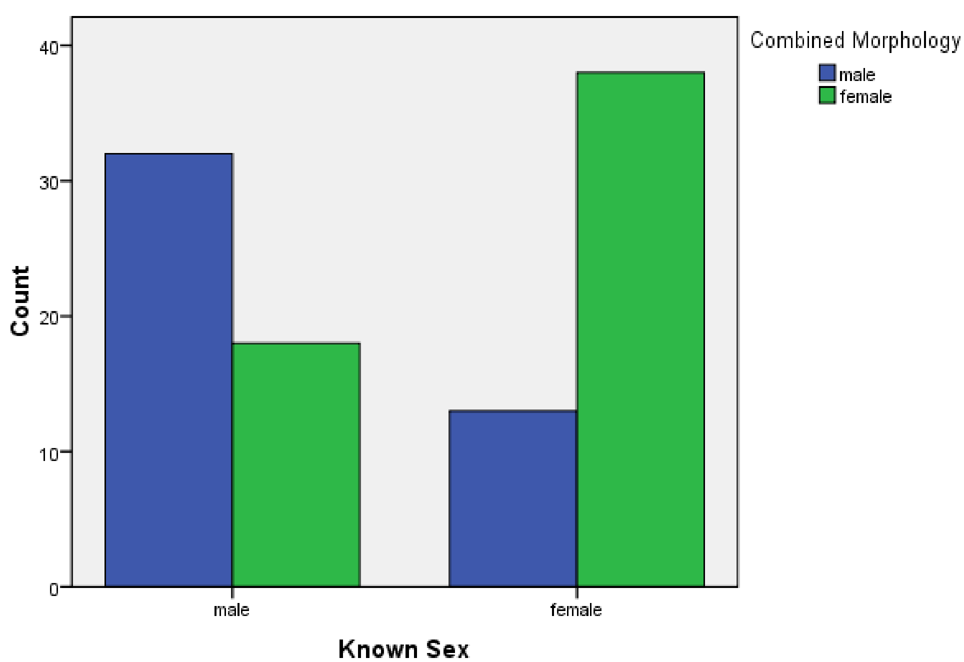


Figure 7: Distribution of Males and Females classified by Fordisc

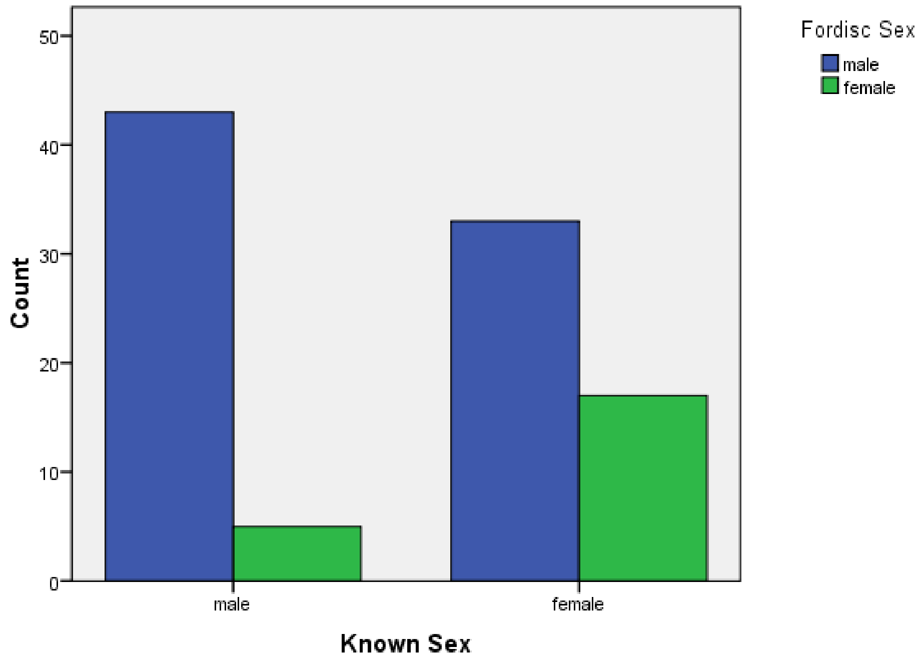


Table 12: Known Sex vs. Fordisc Sex

Known Sex * Fordisc Sex			Fordisc Sex		
			male	female	Total
Known Sex	male	Count	43	5	48
	female	Count	33	17	50
	Total	Count	76	22	98

The final method of sex determination was applied to 99 individuals (48 males and 51 females). The geometric morphometric analysis was run with both two dimensional and three dimensional landmark data. After a Procrustes 2D transformation, discriminant function analysis with a leave-one-out approach was conducted in PAST. Of the 99 individuals included in the 2D morphometric analysis, 41 (80.4%) were correctly identified as female and 41 (85.4%) were correctly identified as male.

Following the 3D Procrustes transformation the discriminant analysis resulted in 46 (90.2%) females and 48 (100%) males being correctly identified. The 2D analysis resulted in an overall accuracy of 82.8%, while the 3D data provided an accuracy of 94.95%, summarized in Table 13 and Table 14, respectively.

Table 13: Known Sex vs. 2D Morphometric Sex

Known Sex * 2D Morphometric Sex			2D Morphometric Sex		
			male	female	Total
Known Sex	male	Count	41	7	48
	female	Count	10	41	51
	Total	Count	51	48	99

Table 14: Known Sex vs. 3D Morphometric Sex

Known Sex * 3D Morphometric Sex			3D Morphometric Sex		
			male	female	Total
Known Sex	male	Count	48	0	48
	female	Count	5	46	51
	Total	Count	53	46	99

Comparison of the relationship between each methodology and the known sex was done using Spearman's correlation coefficient. The results of this, along with the statistical significance, can be found in Table 15. All correlations between methodology and known sex were statistically significant except for orbit shape. Orbit position, margin sharpness, multivariate morphology, and metric sex displayed statistically significant low correlations. Only the morphometric (both 2D and 3D) comparisons show statistically significant high correlations to known sex. The geometric morphometric analysis being conducted in 2D and 3D gave a unique opportunity to compare and observe if combining the results from each method would provide a multi-method combined sex classification and provide greater aid in identification. As the individual morphological traits did not yield greater accuracies than the multivariate approach, the multivariate conclusion was utilized when combining the results from the additional methods. The multi-method approach was based on various combinations of the multivariate classification, traditional metric classification, 2D morphometric classification, and 3D morphometric classification. A full comparison of the predicted sex from each method, and combination thereof, for each individual can be seen in Appendix with the full data worksheet, as well as the cross-tabulation tables of accuracy for each multi-method combination. Combining the results from all four analyses to produce a single sex determination lead to inconclusive results (i.e. two approaches indicate female and the other two indicate male), and produced an accuracy (77.3%) lower than selecting either the 2D or 3D classification for the multi-method comparison (79.4% and 85.6%, respectively). Combining the 2D and 3D classifications with either the morphological or metric classification resulted in an accuracy of 91.8% and 89.7%, respectively.

Table 15: Comparison of known sex to individual methods predicted sex using Spearman's rho correlation

Known Sex compared with	Correlation Coefficient	Sig. (2-tailed)	N
Orbit Shape	0.168	0.093	101
Orbit Position	0.387*	<0.001	101
Margin Sharpness	0.307*	0.002	101
Multivariate Morphology	0.387*	<0.001	101
Metric Sex	0.283*	0.005	98
2D Morphometric	0.658*	<0.001	99
3D Morphometric	0.904*	<0.001	99

*. Correlation is significant at the 0.05 level

Intra-observer error was calculated for each methodology utilized. The intra-observer error tests revealed varying results. The morphological intra-observer error was looked at for each individual variable, as well as the resulting multivariate classification. Orbit shape had the greatest amount of change between observations, 7 of the 20 (35%) individuals evaluated had a change of classification of sex from Trial 1 to Trial 2. Of these, 5 changed observation to the correct classification of sex. Orbit position and margin sharpness both saw a change in 3 different individuals (15%). Of these changes by orbit position, 2 were to the correct classification. All three of the margin sharpness changes were from an originally "correctly" classified female to an incorrect male. We can now look at how those individual variable changes affected the outcome in the multivariate approach. The second observation of morphological variations saw a change in identification for four of the 20 individuals, representing an error rate of 20%. All four individuals went from being incorrectly classified during the first set of observations to being correctly classified. Based on the sample used in the intra-observer error test, the change in classification resulted in an increase in accuracy of 20% for the multivariate morphological evaluation. The comparison of results between the first and second series of metric analysis displays one individual that changed sex identification based on Fordisc assessment. The sex and probability for this identification changed from female with 61.3% to male with 58.0%. These results for the metric comparison indicate a 5% error rate when measurements and Fordisc are conducted again. The intra-observer component of the morphometric analysis involved performing a Procrustes transformation and discriminant function analysis on the full sample with the second observations. The two dimensional comparisons of the second observations resulted in 5 of the 20 differing from the original classification, indicating a 25% error rate in the analysis, with only one of these individuals changing to a correct classification. The three dimensional comparison of the second trial resulted in 5 of the 20 individuals differing from the original classification, indicating a 25% error rate in the analysis. As with the 2D, only one individual changed to a correct classification. For all methods the only one to remain under the 10% error constraint after the intra-observer test of predicted sex, was the metric measurement evaluation provided by Fordisc.

In addition to re-testing the outcome from each individual method for the small sub-sample, statistical significance was observed in the data collected for each method. The morphological analysis involved evaluating and ranking the individual traits utilized in the method. The Spearman's correlation coefficient between the trials indicates that orbit position is most correlated (0.739), followed by margin sharpness (0.707), and the lowest correlation between trials was for orbit shape (0.373). When comparing the correlation between each trait and the actual known sex for each trial and incorporating the above ranks for each trait, the highest rank would be for orbit position, followed by margin sharpness, and lastly orbit shape. The detailed comparison of trial 1 and trial 2 for each morphological trait and known sex can be seen in Table 16.

Table 16: Morphological Intra-observer comparison using Spearman's rho correlation

			Trial 2				
			Orbit Shape	Orbit Position	Margin Sharpness	Multivariate Morphology	Known Sex
Trial 1	Orbit Shape	Correlation Coefficient	0.373	0.373	0.126	0.373	-0.312
		Sig. (2-tailed)	0.105	0.105	0.597	0.105	0.181
		N	20	20	20	20	20
	Orbit Position	Correlation Coefficient	0.739*	0.739*	<0.001	0.739*	0.375
		Sig. (2-tailed)	<0.001	<0.001	1.000	<0.001	.103
		N	20	20	20	20	20
	Margin Sharpness	Correlation Coefficient	0.082	0.492*	0.707*	0.492*	0.250
		Sig. (2-tailed)	0.731	0.027	<0.001	0.027	0.288
		N	20	20	20	20	20
	Multivariate Morphology	Correlation Coefficient	0.616*	0.616*	0.290	0.616*	0.082
		Sig. (2-tailed)	0.004	0.004	0.215	0.004	0.731
		N	20	20	20	20	20
	Known Sex	Correlation Coefficient	0.123	0.533*	<0.001	0.533*	1.000
		Sig. (2-tailed)	0.605	0.015	1.000	0.015	.
		N	20	20	20	20	20

*. Correlation is significant at the 0.05 level (2-tailed).

The metric and morphometric methods rely on measurements and therefore differ from morphological analysis when comparing repeated trials. For the traditional metrics, each measurement (ORBR, OBH, INTB, and BIOB) was conducted twice in order to assess intra-observer error between the measurements. The measurement from Trial 1 was then compared to the measurement from Trial 2 using a Paired samples t-test to determine if the mean difference between the two measurements was significantly different than zero (i.e. there is no intra-observer error for the measurement). The values for the metric comparison can be found in Table 17. The mean difference for the set of measurements ranged from -0.586220 to 1.2198650. The measurement showing a statistically significant difference between Trial 1 and Trail 2 is orbital height (OBH), with a mean difference of 1.2198650. Based on the mean measurements for OBH, provided in Appendix, a 5% difference would be 1.6mm, for which 1.22 is well below. There was no statistical significance in the mean difference for the remaining measurements observed in Table 17.

Table 17: Metric Intra-observer Paired T-test

Measurement difference between Trial 1 and Trial 2	Paired Differences					t	df	Sig. (2-tailed)
	Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
				Lower	Upper			
ORBR1 - ORBR2	0.242250	1118712	0.250152	-0.281323	0.765823	0.968	19	0.345
OBH1 - OBH2	1219865	0.844541	0.188845	0.824607	1615123	6.460	19	<0.001
INTB1 - INTB2	-0.586220	1481871	0.331357	-1279757	0.107317	-1769	19	0.093
BIOB1 - BIOB2	0.431760	2.835848	0.634115	-0.895458	1758978	0.681	19	0.504

The 16 morphometric landmarks were collected twice in order to assess intra-observer error. Measurements were taken from Landmark 1 to each individual landmark for a total of 15 measurements per individual. These 15 measurements from Trial 1 (A) were compared to the measurements obtained from the landmarks in Trial 2 (B) using a Paired t-test. The values for the 2D and 3D morphometric intra-observer comparison can be seen in Table 18 and Table 19, respectively. In the 2D comparison, landmarks 3, 4, 5, 8, 12, 13, 14, 15, and 16 were all found to have statistically significant mean differences between Trial 1 and Trial 2. The largest mean and standard deviation is seen in landmark 13, with values of -0.751000 and 1.10938, respectively. In the 3D comparison, landmarks 4, 5, 12, 13, 14, 15, and 16 were found to have statistically significant mean differences. Again the largest mean and standard deviation is seen in landmark 13, with values of -0.70210 and 1.05409, respectively.

Table 18: 2D Morphometric Intra-observer Paired T-test

Landmark Measurements between Trial 1 (A) and Trial 2 (B)	Paired Differences					t	df	Sig. (2-tailed)
	Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
				Lower	Upper			
A1-2 - B1-2	-0.089640	0.362335	0.081021	-0.259218	0.079938	-1.106	19	0.282
A1-3 - B1-3	-0.250950	0.508530	0.113711	-0.488949	-0.012951	-2.207	19	0.040
A1-4 - B1-4	-0.473850	0.616636	0.137884	-0.762444	-0.185256	-3.437	19	0.003
A1-5 - B1-5	-0.516400	1.025251	0.229253	-0.996232	-0.036568	-2.253	19	0.036
A1-6 - B1-6	-0.099150	0.921441	0.206040	-0.530398	0.332098	-0.481	19	0.636
A1-7 - B1-7	0.229400	0.666101	0.148945	-0.082345	0.541145	1.540	19	0.140
A1-8 - B1-8	0.318550	0.532012	0.118961	0.069561	0.567539	2.678	19	0.015
A1-9 - B1-9	0.178050	0.636360	0.142295	-0.119776	0.475876	1.251	19	0.226
A1-10 - B1-10	-0.023650	0.512298	0.114553	-0.263413	0.216113	-0.206	19	0.839
A1-11 - B1-11	-0.211700	0.605182	0.135323	-0.494934	0.071534	-1.564	19	0.134
A1-12 - B1-12	-0.477650	0.834727	0.186651	-0.868314	-0.086986	-2.559	19	0.019
A1-13 - B1-13	-0.751000	1.110938	0.248413	-1.270935	-0.231065	-3.023	19	0.007
A1-14 - B1-14	-0.656450	0.911469	0.203811	-1.083031	-0.229869	-3.221	19	0.004
A1-15 - B1-15	-0.558450	0.720870	0.161191	-0.895828	-0.221072	-3.465	19	0.003
A1-16 - B1-16	-0.317005	0.482387	0.107865	-0.542769	-0.091241	-2.939	19	0.008

Table 19: 3D Morphometric Intra-observer Paired T-test

Landmark Measurements between Trial 1 (A) and Trial 2 (B)	Paired Differences					t	df	Sig. (2-tailed)
	Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
				Lower	Upper			
A1-2 - B1-2	0.117125	0.437361	0.097797	-0.087566	0.321816	1.198	19	0.246
A1-3 - B1-3	-0.048450	0.576029	0.128804	-0.318040	0.221140	-0.376	19	0.711
A1-4 - B1-4	-0.410950	0.547558	0.122438	-0.667215	-0.154685	-3.356	19	0.003
A1-5 - B1-5	-0.515500	1.008519	0.225512	-0.987502	-0.043498	-2.286	19	0.034
A1-6 - B1-6	-0.129350	0.994560	0.222390	-0.594819	0.336119	-0.582	19	0.568
A1-7 - B1-7	0.178650	0.851905	0.190492	-0.220054	0.577354	0.938	19	0.360
A1-8 - B1-8	0.318750	0.720648	0.161142	-0.018524	0.656024	1.978	19	0.063
A1-9 - B1-9	0.238850	0.770353	0.172256	-0.121686	0.599386	1.387	19	0.182
A1-10 - B1-10	0.030300	0.578003	0.129245	-0.240214	0.300814	0.234	19	0.817
A1-11 - B1-11	-0.170650	0.616380	0.137827	-0.459125	0.117825	-1.238	19	0.231
A1-12 - B1-12	-0.431400	0.788266	0.176262	-0.800320	-0.062480	-2.447	19	0.024
A1-13 - B1-13	-0.702100	1.054093	0.235702	-1.195431	-0.208769	-2.979	19	0.008
A1-14 - B1-14	-0.618550	0.877430	0.196199	-1.029200	-0.207900	-3.153	19	0.005
A1-15 - B1-15	-0.571100	0.698087	0.156097	-0.897815	-0.244385	-3.659	19	0.002
A1-16 - B1-16	-0.316925	0.429455	0.096029	-0.517916	-0.115934	-3.300	19	0.004

Chapter 5: Discussion

Although there has been debate over the value of the orbits of the skull in determining sex from human skeletal remains, advancing technology and methods indicate there may be more information available than originally thought. The present research attempted to focus on this particular feature and apply multiple methods, both new and traditional. Maintaining a focus of only the orbit ensures that, if possible, the best method for displaying sexually dimorphic characteristics of the orbit is ascertained.

The traditional gross morphological analysis displays results that suggest the methods poorly predict sex. Based on these findings alone the author would recommend not using the morphological assessments of the orbits for determination of sex in producing a biological profile. The accuracy for the multivariate approach (69.3%) did not surpass the 80% minimum accuracy for significant methods, neither did any of the individual variable accuracies. The low values obtained here are comparable to those observed in Table 1 specifically regarding accuracies of the orbit (Rogers, 2005; Williams & Rogers, 2006; Walker, 2008; Ramsthaler et al. 2010). This low accuracy would indicate that the traits observed do not reflect any dimorphism present in the orbits. The intra-observer comparison of the morphological traits does not support further use of the method either. The intra-observer error of 20% greatly exceeds the acceptable level of 10%. This also was observed in previous studies that incorporated the orbits in the evaluation of sex (Rogers, 2005; Williams & Rogers, 2006). When taking a closer look at the individual traits utilized in the methodology there is a difference in their individual strengths. The trait with the worst results is the orbit shape, where males are said to be square and females round. Between trials, orbit shape had the worst initial accuracy (40%) but had the greatest increase for trial 2, displaying 55% accuracy, an improvement of 15%. The amount of difference caused by this change results in an error of 35% (7/20), highest of all three traits. The greatest correlation between the trials is seen in the Orbit position, with one of the lowest error rates (15%) and highest accuracies (70% trial 1 and 80% trail 2). The increase in accuracy between both trials (for both traits) displays an increase in understanding and a learning curve involved in identifying appropriate differences. The high error rates indicate that the apparent differences are not as clearly defined and identifiable as one would hope. Of all the traits examined, margin sharpness is often the one relied on and maintained in complete skeletal analysis, even by those who do not support orbit shape and position as valid sex difference identifiers (Maat, Mastwijk, & Van Der Velde, 1997; Konigsberg & Hens, 1998; Graw, Czarnetzki, & Haffner, 1999; Đurić, Rakočević, & Đonić, 2005; Williams & Rogers, 2006; Walker, 2008). The present research finds that margin sharpness falls below orbit position and above orbit shape. An error rate of 15% caused a decrease in accuracy between trial 1 and 2. It must be noted that traditional analysis on dry bone requires the physical examination of the margin and its sharpness, which is not possible on the present CT scans where instead evaluation of sharpness is based on a cross-section of the scanned data. More defined and detailed descriptions of the margin cross-section have the potential to increase the value of this trait but not likely to valid levels. Examination of the combined morphological classifications of sex separated by age displays somewhat surprising results. The expectation of older females being classified as males due to increased robusticity, and conversely younger males being misclassified as females, does not seem to be of great significance with this sample. There does appear to be almost equal misclassification of individuals in the middle (45-55) age group. This is surprising as one would expect this age group to be most representative of the potential sex differences. Although there may be a change in robusticity as individuals age these morphological traits do not appear to significantly reflect this. Moreover, this distribution according to age supports the observation that the morphological traits do not accurately reflect sexual dimorphism. The present researcher is by no means an expert in skeletal analysis and has limited experience with such examinations. This lack of experience is likely a contributing factor in the low morphological results displayed here, however, they do not differ greatly from more experienced researchers' observations, as seen in Table 1. The demonstrated learning curve indicates that with practice higher accuracies and likely lower error rates would be accomplished. However, the author is not convinced that the increase would be great enough to reach the 80% accuracy and 10% error level of acceptance. Based on the above findings one would not recommend pursuing this method in archaeological or forensic contexts. This is in support of previous researchers that dismiss the evaluation of the orbits.

The lowest accuracy for all methods was seen in the metric evaluation (61.2%). In contrast, utilizing Fordisc provided the lowest error rate (5%), the only one within the acceptable range. This demonstrates that although the method through Fordisc is consistent in providing the same results, those results that are produced are not accurate. There is a definite male bias in the results likely influenced by the comparative sample of the software, which has almost double the amount of males to females (170 vs. 99, respectively). This would indicate that although the methodology is strong it does not represent appropriate sex differences in the sample. Although the comparison was only made with "white" individuals in the Fordisc database, they may not represent the true form of the Denmark individuals found in this sample. This is in agreement with Ramsthaler and colleagues (2007) finding that Fordisc does not represent the European population accurately. Looking at the measurement differences between the trials, a statistically significant difference is seen in orbital height. This measurement is weighted very low in Fordisc (1.4%) and should have no major influence in inaccuracies despite this measurements larger range of difference. Overall, this method can be applied more consistently then the morphological evaluation, even by an inexperienced assessor. However, the population is not well represented in the comparative sample to produce accurate results.

The highest accuracies, and the only ones able to pass the appropriate level of acceptance, where those of the geometric morphometric analysis. Both 2D and 3D accuracies (82.8% and 94.95%, respectively) were well above the 80% limit. These are quite desired levels and contrast greatly with the traditional methods. These values are even higher than those obtained by Pretorius and colleagues (2006). These levels would indicate that sexual dimorphism within the orbits is measurable and quantifiable. Although the traditional methods do not display the dimorphism to such a level, and actually oppose the use of the orbits for such information, the morphometric analysis tells a different story. These results bring the orbits back into a tangible position within the field of anthropology. They cannot and should not be dismissed for lack of sexual dimorphism when evaluating human remains. The use of landmarks provides information on areas of the orbit that neither morphological evaluation can recognize nor metric analysis can capture. The fact that the 3D data performed higher than the 2D data, again supports the idea that more information is most beneficial. As sexual dimorphism of the skull is based on size and robusticity, the protrusion of bone all along the orbit including the brow, zygomatic and nasal regions will influence the form of the orbit in all dimensions. The 'z' data (in the x,y,z coordinates) provides the third dimension running in the sagittal plane, and landmarks reflective of this dimension will differ from those only found on a single coronal plane of the skull. This can be visually seen as the "flatness" of female orbits. The difference can be depicted from a lateral view of orbits, see Figure 8 and Figure 9.

Figure 8: Male lateral view displaying unevenness of the orbital margin along the "z" dimension

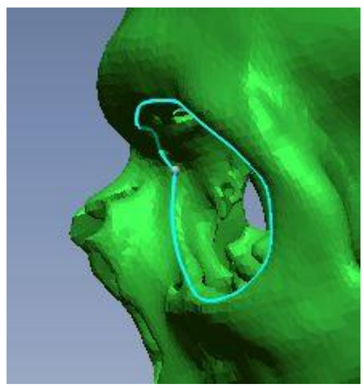
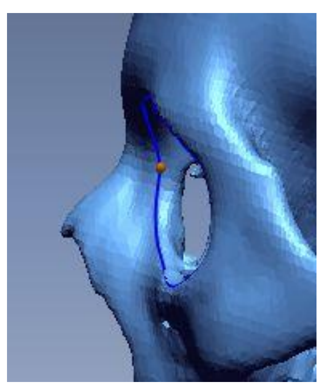


Figure 9: Female lateral view displaying relative flatness of the orbital margin along the "z" dimension



The t-test performed on the measurements from landmark 1 to all others for the data in the morphometric intra-observer trials indicates that the landmarks obtained display slight inconsistencies. As previously noted, measurement differences between landmarks greater than 5% are deemed to be unacceptable (Corner, Lele, & Richtsmeier, 1992). For the present research this would result in a mean difference of between 0.45mm and 2mm, depending on the specific landmark measurement evaluated. The largest difference for both 2D and 3D data was seen in Landmark 13, and for this measurement a difference of 1.70mm would be at the 5% level. Both the mean difference and standard deviation is well below this value, -0.751000 and 1.10938, respectively for 2D data and -0.70210 and 1.05409, respectively for 3D data. This demonstrates and supports the current use of CT data for the analysis of skeletal remains. Landmark 13 is the standard biological landmark Orbitale, defined as the lowest point on the inferior border of the margin. The curvature is gradual and almost horizontal along the inferior border, so there is some degree of error of subjectivity in the investigator that would be associated with the determining the exact 'bottom' point. As this is a standard biological landmark, the location was also utilized in the standard measurements for the orbital height measurement (OBH). This measurement was one that demonstrated statistically significant differences during the intra-observer comparison for the standard metrics. The placement of landmark 13 not only affects the OBH measurement but also impacts the pseudo-landmarks that surround it. The placement of landmark 13 determines the placement of the spline which applies the pseudo-landmarks to the margin, as seen in the statistical significance of landmarks 12-16, for both 2D and 3D. Landmark 5 also displayed statistical significance in the morphometric intra-observer comparison; this is the biological landmark for the superior border of the margin. The placement of landmark 5 is also influenced by landmark 13 as this is also used to create the OBH measurement. Experience is likely to contribute to the placement of landmarks as well, although it doesn't appear to impact the outcome to the same degree as is seen in the morphological analysis. Although the landmarks obtained are acceptable, the results for the intra-observer comparison did change for 5 individuals (25% error) with the 2D data, and the 3D data, when the second trial was run through PAST. This high level of error does not appear to be a question in obtaining consistent data, but rather is an issue with producing a discriminant function utilizing the same sample. The present morphometric analysis is not based on comparing an unknown to a known database (as is seen with Fordisc), however it uses the information from the present data to create a standard and then apply each individual to that standard. In the case of the intra-observer data, when the second trial was added it changed ever so slightly the sample population from which the information was being gathered. This is the greatest drawback of fully incorporating morphometrics into the present field of anthropology. With geometric morphometrics still in the early stages of implementation into physical anthropology, the data from different collections has been limited. Collecting more data from as many populations and samples as possible to create a morphometric database, similar to the Fordisc database, is a key step to being able to implement this method in mainstream anthropology.

Applying multiple methods to a sample, especially with the ease of collection on 3D data, is a recommended approach to skeletal analysis (Buikstra & Ubelaker, 1994; Franklin, Freedman, & Milne, 2005b; Ramsthaler, Kreutz, & Verhoff, 2007). In an effort to see if combining the traditional methods with geometric morphometrics would produce a collective determination of sex higher than any individual method, many combinations were examined. A combined result for all four approaches (morphological, metric, 2D and 3D geometric morphometric) did not yield results (78.4%) sufficient to be considered acceptable (≥ 80%), and was also the lowest accuracy of all the combinations. The full combination also resulted in inconclusive results (18.6%) which are not ideal in a forensic or bioarchaeology analysis. The highest combination (93.8%) was achieved by combining the morphological assessment with the 2D and 3D geometric morphometric classification. This result is no better than the classification obtained by using the individual 3D morphometric analysis. The author feels that combining the 2D and 3D analysis is rather redundant of an approach. The best approach is to rely solely on the 3D geometric morphometric classification. Being able to achieve such a high degree of separation between the sexes from the 3D data using geometric morphometrics supports the use of 3D rendered CT scans as an appropriate medium for analysis. The use of CT images also allows future research to use the same individuals, even if the orbits are not the area of interest. This would not be possible with previous geometric morphometric studies that relied on digitizing photographs or specific landmarks for the data (Pretorius, Steyn, & Scholtz, 2006; Kimmerle, Ross, & Slice, 2008; Oettle, Pretorius, & Steyn, 2009; Gonzalez, Bernal, & Perez, 2011).

This research focused on the orbits and their display of sexual dimorphism. Traditional morphological examination of the orbit shape did not show significant results indicating an accurate portrayal of the sexual dimorphism. However, utilizing the geometric morphometrics, at 94.95% clearly display that the sexual dimorphism is significant. This would imply that the shape difference may be a little more complicated than just 'square' or 'round'. Applying future geometric morphometric techniques, like EDMA, which can isolate the landmarks and locate where the differences are occurring, would help identify how accurate the traditional morphological assessment is at depicting sexual dimorphism. Although the multi-method approach cannot be recommended in this circumstance, it would be beneficial for future research to examine combining a multivariate geometric morphometric approach. This would require obtaining data for two or more features at a time and running morphometric analysis on all of the data and producing a single classification, similar to the multivariate morphological approach. Based on this, further research is needed to determine the best possible features that display significant sexual dimorphism when utilizing a geomorphometric approach.

Conclusion

The present research clearly displayed sexual dimorphism in the orbits of the skull, by utilizing geometric morphometrics on the orbit outline. Traditional methods were not able to identify this dimorphism to a significant degree, actually making it appear that the orbits lack much dimorphism. However, the increased power and sensitivity of geometric morphometrics was necessary to depict this dimorphism with great significance. Before geometric morphometrics can be utilized by fields such as forensics and bioarchaeology, a known database (similar to Fordisc®) must be created.

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Appendix

Denmark Collection

Case Number	File Name	Pixel Size (mm)	Field of View (cm)	# of Slices	Slice Thickness(mm)	Sex	Age
1	08151058	0.434	22.20	104	2	f	49
2	08281235	0.441	22.60	110	2	f	29
3	08281236	0.465	23.80	119	2	m	45
4	08291401	0.391	20.00	121	2	f	41
5	08291405	0.477	24.40	120	2	m	60
6	08291406	0.512	26.20	121	2	m	36
7	08291410	0.469	24.00	117	2	m	52
8	08291430	0.508	26.00	117	2	m	68
9	08291433	0.465	23.80	117	2	m	43
10	08281228	0.484	24.80	112	2	m	57
11	08281230	0.422	21.60	103	2	f	58
12	08281218	0.512	26.20	121	2	m	41
13	08281222	0.480	24.60	118	2 (abnrml zyg)	m	51
14	08291439	0.473	24.20	124	2	m	50
15	08291440	0.434	22.20	108	2	f	53
16	08291441	0.469	24.00	120	2	m	55
17	08291443	0.391	20.00	117	2	f	48
18	08291450	0.434	22.20	114	2	m	42
19	08291452	0.441	22.60	240	1	f	58
20	08291457	0.492	25.20	105	2	m	41
21	09031128	0.391	20.00	109	2 (R-zyg -INC*)	f	56
22	09051201	0.434	22.20	121	2	f	74
23	09051203	0.430	22.00	121	2	m	29
24	09090926	0.504	25.80	107	2	m	53
25	09090927	0.484	24.80	99	2	m	21
26	09090928	0.473	24.20	121	2	f	33

Case Number	File Name	Pixel Size (mm)	Field of View (cm)	# of Slices	Slice Thickness(mm)	Sex	Age
27	09090930	0.504	25.80	105	2	m	39
28	08291502	0.426	21.80	100	2	f	72
29	09011339	0.477	24.40	100	2	m	73
30	09011340	0.504	25.80	111	2	m	87
31	09021211	0.453	23.20	247	1	m	29
32	09021234	0.449	23.00	108	2	m	42
33	09021236	0.516	26.40	120	2	m	27
34	09021245	0.492	25.20	105	2	m	41
35	10221306	0.406	20.80	129	2	m	46
36	10221307	0.414	21.20	134	2	m	51
37	10221308	0.375	19.20	111	2	f	38
38	10221310	0.379	19.40	128	2	m	33
39	10221311	0.395	20.20	128	2	f	58
40	10221312	0.465	23.80	102	2 (no mrphmt-INC)	m	48
41	10221313	0.477	24.40	102	2	f	57
42	10231228	0.418	21.40	130	2	m	26
43	10231229	0.469	24.00	125	2	m	57
44	10231230	0.434	22.20	112	2	m	40
45	10231251	0.406	20.80	115	2	m	51
46	9101255	0.453	23.20	112	3	m	60
47	10221223	0.395	20.20	128	3	f	55
48	10221224	0.391	20.00	104	3	f	55
49	10221225	0.430	22.00	112	3	f	86
50	10221240	0.391	20.00	104	3	m	40
51	10221242	0.441	22.60	126	2	m	52
52	10221248	0.371	19.00	113	2	f	39
53	10221249	0.371	19.00	113	2 (duplicate)	f	39
54	10221252a	0.406	20.80	115	2	f	52
55	10221252b	0.426	21.80	132	2	f	38

Case Number	File Name	Pixel Size (mm)	Field of View (cm)	# of Slices	Slice Thickness(mm)	Sex	Age
56	10221246	0.418	21.40	126	2	m	42
57	10221221	0.438	22.40	96	2	f	60
58	11181218	0.457	23.40	115	2	f	54
59	11181225	0.484	24.80	111	2	m	70
60	11181201	0.418	21.40	110	2	f	85
61	11181202	0.422	21.60	110	2	m	88
62	11181203	0.477	24.40	239	1 (trauma)	m	73
63	11181204	0.402	20.60	107	2	f	58
64	11181214	0.465	23.80	108	2	f	81
65	11181226	0.457	23.40	103	2 (INC- holes)	m	24
66	11181140	0.391	20.00	127	2	f	59
67	11181142	0.418	21.40	129	2	m	38
68	11181143	0.445	22.80	116	2	f	75
69	11181145	0.449	23.00	507	0.5	m	31
70	11181146	0.461	23.60	125	2	m	47
71	11031255	0.441	22.60	118	2	m	60
72	11031257	0.418	21.40	479	0.5 (trauma)	m	55
73	11031258	0.430	22.00	123	2	m	84
74	11181129	0.477	24.40	121	2	m	41
75	11181133	0.379	19.40	112	2	f	34
76	11031225	0.426	21.80	108	2	f	52
77	11031227	0.492	25.20	125	2	m	24
78	11031228	0.449	23.00	103	2	f	44
79	11031230	0.441	22.60	227	1	f	51
80	11031231	0.547	28.00	230	1	m	24
81	11031232	0.453	23.20	127	2	m	28
82	11031243	0.438	22.40	112	2	m	31
83	11181227	0.441	22.60	107	2	m	32
84	11251046	0.449	23.00	210	1	f	83

Case Number	File Name	Pixel Size (mm)	Field of View (cm)	# of Slices	Slice Thickness(mm)	Sex	Age
85	11251047	0.477	24.40	128	2	m	33
86	12789100	0.432	22.10	412	1	f	67
87	12789102	0.391	20.00	467	1	f	78
88	12789120	0.543	27.80	497	1	f	53
89	12798106	0.387	19.80	503	1	f	43
90	12798107	0.367	18.80	505	1	f	37
91	12798112	0.594	30.40	487	1	f	47
92	12798118	0.547	28.00	473	1	f	69
93	12879103	0.406	20.80	547	1	f	78
94	12879113	0.508	26.00	471	1	f	78
95	12879119	0.508	26.00	483	1	f	60
96	12897101	0.426	21.80	417	1	f	72
97	12897105	0.375	19.20	531	1	f	52
98	12897109	0.414	21.20	443	1	f	28
99	12897111	0.523	28.80	499	1	f	19
100	12897114	0.684	35.00	457	1	f	46
101	12978116	0.559	28.60	501	1	f	65
102	12987104	0.363	18.60	479	1	f	36
103	12987108	0.379	19.40	473	1	f	60
104	12987117	0.559	28.60	475	1	f	47

*INC = incomplete

Data Worksheet for all Methods

Case Number:	Morphology				Metric		Morphometric		**Final**					**Actual Sex**	
	Shape	Position	Margin	Total	Fordisc	Prob.	2D	3D	**no mrph**	**no met**	**w/2D**	**w/3D**	**All 4**	**Sex**	Age
1	m	f	f	f	m	0.944	m	m	**m**	**m**	**m**	**m**	**m**	**f**	49
2	m	f	f	f	m	0.698	f	f	**f**	**f**	**f**	**f**	**f**	**f**	29
3	f	f	m	f	m	0.975	m	m	**m**	**m**	**m**	**m**	**m**	**m**	45
4	f	f	f	f	f	0.913	f	f	**f**	**f**	**f**	**f**	**f**	**f**	41
5	m	m	m	m	m	0.990	m	m	**m**	**m**	**m**	**m**	**m**	**m**	60
6	f	m	f	f	m	0.859	m	m	**m**	**m**	**m**	**m**	**m**	**m**	36
7	f	f	f	f	m	0.985	m	m	**m**	**m**	**m**	**m**	**m**	**m**	52
8	m	m	m	m	m	0.996	m	m	**m**	**m**	**m**	**m**	**m**	**m**	68
9	m	m	f	m	m	0.952	m	m	**m**	**m**	**m**	**m**	**m**	**m**	43
10	m	m	m	m	m	0.946	m	m	**m**	**m**	**m**	**m**	**m**	**m**	57
11	f	f	f	f	m	0.938	f	f	**f**	**f**	**f**	**f**	**f**	**f**	58
12	f	m	m	m	m	0.734	m	m	**m**	**m**	**m**	**m**	**m**	**m**	41
13	m	m	m	m			f	m						**m**	51
14	m	m	m	m	m	0.872	m	m	**m**	**m**	**m**	**m**	**m**	**m**	50
15	f	f	f	f	f	0.872	m	f	**f**	**f**	**f**	**f**	**f**	**f**	53
16	m	m	m	m	f	0.559	m	m	**m**	**m**	**m**	**m**	**m**	**m**	55
17	f	f	f	f	f	0.525	f	m	**f**	**f**	**f**	**f**	**f**	**f**	48
18	f	f	f	f	m	0.747	m	m	**m**	**m**	**m**	**m**	**m**	**m**	42
19	m	m	m	m	m	0.709	f	f	**f**	**f**	**m**	**m**	**inc**	**f**	58
20	f	m	m	m	m	0.967	m	m	**m**	**m**	**m**	**m**	**m**	**m**	41
21	m	f	f	f			f	f						**f**	56
22	f	f	m	f	m	0.864	f	f	**f**	**f**	**f**	**f**	**f**	**f**	74
23	m	m	f	m	m	0.626	m	m	**m**	**m**	**m**	**m**	**m**	**m**	29
24	m	m	m	m	m	0.865	m	m	**m**	**m**	**m**	**m**	**m**	**m**	53
25	m	m	m	m	m	0.999	m	m	**m**	**m**	**m**	**m**	**m**	**m**	21
26	f	f	f	f	m	0.770	f	f	**f**	**f**	**f**	**f**	**f**	**f**	33
27	m	m	m	m	m	0.992	m	m	**m**	**m**	**m**	**m**	**m**	**m**	39

Case Number:	Morphology				Metric		Morphometric		**Final**					**Actual Sex**	
	Shape	Position	Margin	Total	Fordisc	Prob.	2D	3D	**no mrph**	**no met**	**w/2D**	**w/3D**	**All 4**	**Sex**	Age
28	f	f	f	f	f	0.841	f	f	f	f	f	f	f	f	72
29	f	m	f	f	m	0.812	m	m	m	m	m	m	m	m	73
30	m	f	m	m	m	0.921	m	m	m	m	m	m	m	m	87
31	f	f	m	f	f	0.703	f	m	f	f	f	f	f	m	29
32	f	f	f	f	m	0.581	m	m	m	m	m	m	m	m	42
33	m	m	f	m	m	0.919	f	m	m	m	m	m	m	m	27
34	m	m	m	m	m	0.963	m	m	m	m	m	m	m	m	41
35	f	f	m	f	f	0.713	m	m	m	m	f	f	inc	m	46
36	f	f	f	f	m	0.983	m	m	m	m	m	m	m	m	51
37	f	m	f	f	m	0.525	m	f	m	f	m	f	inc	f	38
38	f	f	f	f	m	0.799	f	m	m	f	f	m	inc	m	33
39	f	f	m	f	f	0.501	m	m	m	m	f	f	inc	f	58
40	f	f	f	f	m	0.822								m	48
41	f	f	m	f	m	0.988	f	f	f	f	f	f	f	f	57
42	f	f	m	f	m	0.943	m	m	m	m	m	m	m	m	26
43	m	m	m	m	m	0.995	m	m	m	m	m	m	m	m	57
44	f	f	m	f	m	0.552	f	m	m	f	f	m	inc	m	40
45	m	m	f	m	m	0.939	m	m	m	m	m	m	m	m	51
46	m	m	f	m	m	0.876	m	m	m	m	m	m	m	m	60
47	m	m	f	m	m	0.655	f	f	f	f	m	m	inc	f	55
48	f	m	f	f	m	0.950	f	f	f	f	f	f	f	f	55
49	f	f	f	f	f	0.874	f	f	f	f	f	f	f	f	86
50	f	f	m	f	m	0.887	m	m	m	m	m	m	m	m	40
51	f	m	m	m	m	0.991	m	m	m	m	m	m	m	m	52
53	f	f	f	f	f	0.850	f	f	f	f	f	f	f	f	39
54	m	f	m	m	m	0.701	m	m	m	m	m	m	m	f	52
55	m	m	m	m	m	0.967	f	f	f	f	m	m	inc	f	38

Case Number:	Morphology				Metric		Morphometric		**Final**					**Actual Sex**	
	Shape	Position	Margin	Total	Fordisc	Prob.	2D	3D	**no mrph**	**no met**	**w/2D**	**w/3D**	**All 4**	**Sex**	Age
56	m	m	m	m	m	0.993	f	m	**m**	**m**	**m**	**m**	**m**	**m**	42
57	m	m	m	m	f	0.705	f	f	**f**	**f**	**f**	**f**	**f**	**f**	60
58	m	f	f	f	m	0.734	m	f	**m**	**f**	**m**	**f**	**inc**	**f**	54
59	m	m	m	m	m	0.981	m	m	**m**	**m**	**m**	**m**	**m**	**m**	70
60	f	f	f	f	m	0.651	m	f	**m**	**f**	**m**	**f**	**inc**	**f**	85
61	m	m	f	m	m	0.843	m	m	**m**	**m**	**m**	**m**	**m**	**m**	88
62	m	m	m	m										**m**	73
63	m	f	f	f	m	0.675	f	f	**f**	**f**	**f**	**f**	**f**	**f**	58
64	f	f	f	f	f	0.577	m	m	**m**	**m**	**f**	**f**	**inc**	**f**	81
66	f	m	m	m	f	0.534	f	f	**f**	**f**	**f**	**f**	**f**	**f**	59
67	m	m	m	m	m	0.880	m	m	**m**	**m**	**m**	**m**	**m**	**m**	38
68	f	f	m	f	f	0.882	f	f	**f**	**f**	**f**	**f**	**f**	**f**	75
69	m	m	m	m	m	0.633	m	m	**m**	**m**	**m**	**m**	**m**	**m**	31
70	f	m	m	m	m	0.996	m	m	**m**	**m**	**m**	**m**	**m**	**m**	47
71	m	f	m	m	m	0.969	m	m	**m**	**m**	**m**	**m**	**m**	**m**	60
73	m	m	f	m	m	0.784	m	m	**m**	**m**	**m**	**m**	**m**	**m**	84
74	f	f	f	f	m	0.876	m	m	**m**	**m**	**m**	**m**	**m**	**m**	41
75	f	f	f	f	f	0.690	f	f	**f**	**f**	**f**	**f**	**f**	**f**	34
76	m	f	f	f	m	0.511	m	f	**m**	**f**	**m**	**f**	**inc**	**f**	52
77	m	m	f	m	m	0.982	m	m	**m**	**m**	**m**	**m**	**m**	**m**	24
78	m	m	m	m	m	0.819	f	f	**f**	**f**	**m**	**m**	**inc**	**f**	44
79	f	f	f	f	f	0.691	f	f	**f**	**f**	**f**	**f**	**f**	**f**	51
80	m	m	m	m	m	0.935	m	m	**m**	**m**	**m**	**m**	**m**	**m**	24
81	f	f	m	f	f	0.680	f	m	**f**	**f**	**f**	**f**	**f**	**m**	28
82	f	f	f	f	m	0.905	m	m	**m**	**m**	**m**	**m**	**m**	**m**	31
83	m	m	m	m	m	0.803	m	m	**m**	**m**	**m**	**m**	**m**	**m**	32
84	m	f	m	m	m	0.970	f	f	**f**	**f**	**m**	**m**	**inc**	**f**	83

Case Number:	Morphology				Metric		Morphometric		**Final**					**Actual Sex**	
	Shape	Position	Margin	Total	Fordisc	Prob.	2D	3D	**no mrph**	**no met**	**w/2D**	**w/3D**	**All 4**	**Sex**	Age
85	f	f	f	f	f	0.585	m	m	**m**	**m**	**f**	**f**	**inc**	**m**	33
86	f	f	f	f	m	0.856	f	f	**f**	**f**	**f**	**f**	**f**	**f**	67
87	m	f	f	f	m	0.716	f	f	**f**	**f**	**f**	**f**	**f**	**f**	78
88	f	m	m	m	m	0.851	f	f	**f**	**f**	**m**	**m**	**inc**	**f**	53
89	f	f	f	f	m	0.662	f	f	**f**	**f**	**f**	**f**	**f**	**f**	43
90	f	m	f	f	m	0.672	f	f	**f**	**f**	**f**	**f**	**f**	**f**	37
91	m	m	m	m	m	0.990	f	f	**f**	**f**	**m**	**m**	**inc**	**f**	47
92	f	f	f	f	m	0.866	f	f	**f**	**f**	**f**	**f**	**f**	**f**	69
93	f	f	f	f	m	0.646	f	f	**f**	**f**	**f**	**f**	**f**	**f**	78
94	f	f	f	f	m	0.986	f	f	**f**	**f**	**f**	**f**	**f**	**f**	78
95	f	f	f	f	m	0.995	f	f	**f**	**f**	**f**	**f**	**f**	**f**	60
96	f	f	m	f	f	0.613	f	f	**f**	**f**	**f**	**f**	**f**	**f**	72
97	m	f	m	m	m	0.902	f	f	**f**	**f**	**m**	**m**	**inc**	**f**	52
98	f	f	f	f	m	0.827	f	f	**f**	**f**	**f**	**f**	**f**	**f**	28
99	f	f	f	f	m	0.720	f	f	**f**	**f**	**f**	**f**	**f**	**f**	19
100	m	m	f	m	f	0.554	f	f	**f**	**f**	**f**	**f**	**f**	**f**	46
101	m	m	m	m	f	0.626	f	f	**f**	**f**	**f**	**f**	**f**	**f**	65
102	m	f	f	f	f	0.604	m	f	**f**	**f**	**f**	**f**	**f**	**f**	36
103	m	f	f	f	m	0.814	f	f	**f**	**f**	**f**	**f**	**f**	**f**	60
104	f	f	f	f	m	0.606	f	f	**f**	**f**	**f**	**f**	**f**	**f**	47

Legend

= Female Classification

= Method Not Applied

inc = Inconclusive Classification

Intra-Observer Worksheet

Case Number	Trial 1							Trial 2							Actual
	Morphology				Metric	Morphometric		Morphology				Metric	Morphometric		
	Shape	Position	Margin	Combined		2D	3D	Shape	Position	Margin	Combined		2D	3D	
3	f	f	m	f	m	m	m	f	m	m	m	m	m	f	m
6	f	m	f	f	m	m	m	m	m	f	m	m	m	m	m
11	f	f	f	f	m	f	f	m	f	f	f	m	f	f	f
12	f	m	m	m	m	m	m	m	m	m	m	m	f	m	m
19	m	m	m	m	m	f	m	m	m	m	m	m	f	m	f
20	f	m	m	m	m	m	m	m	m	m	m	m	m	f	m
32	f	f	f	f	m	m	m	f	f	f	f	m	m	m	m
41	f	f	m	f	m	f	f	f	f	m	f	m	f	f	f
47	m	m	f	m	m	f	f	m	m	f	m	m	f	f	f
49	f	f	f	f	f	f	f	f	f	m	f	f	f	f	f
50	f	f	m	f	m	m	m	f	m	m	m	m	f	m	m
53	f	f	f	f	f	f	f	f	f	m	f	f	f	f	f
54	m	f	m	m	m	m	m	m	m	m	m	m	f	f	f
60	f	f	f	f	m	m	f	f	f	m	f	m	m	f	f
70	f	m	m	m	m	m	m	m	m	m	m	m	m	m	m
75	f	f	f	f	f	f	f	m	f	f	f	f	f	f	f
83	m	m	m	m	m	m	m	m	m	m	m	m	f	f	m
91	m	m	m	m	m	f	f	m	m	m	m	m	m	m	f
96	f	f	m	f	f	f	f	f	f	m	f	m	f	f	f
97	m	f	m	m	m	f	f	f	f	m	f	m	f	f	f

Legend

= Change between trials

Crosstabulation Charts for Combined Methodology

Known Sex * Fordisc, 2D, and 3D combined			No Morphology		Total
			male	female	
Known Sex	male	Count	45	2	47
		% of Total	46.4%	2.1%	48.5%
	female	Count	8	42	50
		% of Total	8.2%	43.3%	51.5%
	Total	Count	53	44	97
		% of Total	54.6%	45.4%	100.0%

Known Sex * Morphology, 2D, and 3D combined			No Fordisc		Total
			male	female	
Known Sex	male	Count	43	4	47
		% of Total	44.3%	4.1%	48.5%
	female	Count	4	46	50
		% of Total	4.1%	47.4%	51.5%
	Total	Count	47	50	97
		% of Total	48.5%	51.5%	100.0%

Known Sex * Morphology, Fordisc, and 2D combined			With 2D		Total
			male	female	
Known Sex	male	Count	41	6	47
		% of Total	42.3%	6.2%	48.5%
	female	Count	14	36	50
		% of Total	14.4%	37.1%	51.5%
	Total	Count	55	42	97
		% of Total	56.7%	43.3%	100.0%

Known Sex * Morphology, Fordisc, and 3D combined			With 3D		Total
			male	female	
Known Sex	male	Count	43	4	47
		% of Total	44.3%	4.1%	48.5%
	female	Count	10	40	50
		% of Total	10.3%	41.2%	51.5%
	Total	Count	53	44	97
		% of Total	54.6%	45.4%	100.0%

Known Sex * All 4 Methods			All 4 Methods			Total
			male	female	Inconclusive	
Known Sex	male	Count	41	2	4	47
		% of Total	42.3%	2.1%	4.1%	48.5%
	female	Count	2	34	14	50
		% of Total	2.1%	35.1%	14.4%	51.5%
	Total	Count	43	36	18	97
		% of Total	44.3%	37.1%	18.6%	100.0%

Metric Measurement Paired T-test Values

Trial 1 & 2	Mean	N	Std. Deviation	Std. Error Mean
ORBR1	41.803120	20	2.4634685	0.5508483
ORBR2	41.560870	20	2.4619889	0.5505174
OBH1	33.498655	20	2.3706670	0.5300973
OBH2	32.278790	20	2.2683346	0.5072150
INTB1	19.865895	20	2.1438063	0.4793697
INTB2	20.452115	20	2.3371549	0.5226037
BIOB1	98.223495	20	4.3561312	0.9740606
BIOB2	97.791735	20	4.4124941	0.9866637

2D Landmark Measurement Paired T-test Values

Trial 1 (A) & Trial 2 (B)	Mean	N	Std. Deviation	Std. Error Mean
A 1-2	9.37470	20	1.085589	0.242745
B 1-2	9.46434	20	1.086130	0.242866
A 1-3	17.99390	20	1.517276	0.339273
B 1-3	18.24485	20	1.514608	0.338677
A 1-4	24.62620	20	1.941914	0.434225
B 1-4	25.10005	20	2.203947	0.492818
A 1-5	32.00905	20	2.702849	0.604375
B 1-5	32.52545	20	3.071860	0.686889
A 1-6	35.89205	20	2.650081	0.592576
B 1-6	35.99120	20	2.990589	0.668716
A 1-7	38.72040	20	2.713898	0.606846
B 1-7	38.49100	20	2.950689	0.659794
A 1-8	39.60315	20	2.872598	0.642333
B 1-8	39.28460	20	3.016395	0.674487
A 1-9	39.82460	20	3.090047	0.690955
B 1-9	39.64655	20	3.023801	0.676142
A 1-10	40.66860	20	3.128764	0.699613
B 1-10	40.69225	20	3.050771	0.682173
A 1-11	40.4174	20	3.049960	0.681992
B 1-11	40.62910	20	3.142898	0.702773
A 1-12	37.87775	20	2.942286	0.657915
B 1-12	38.35540	20	3.166170	0.707977
A 1-13	34.08040	20	2.865709	0.640792
B 1-13	34.83140	20	3.101144	0.693437
A 1-14	25.94060	20	2.318541	0.518442
B 1-14	26.59705	20	2.447560	0.547291
A 1-15	17.87375	20	1.637029	0.366051
B 1-15	18.43220	20	1.738091	0.388649
A 1-16	9.195115	20	0.900093	0.201267
B 1-16	9.51212	20	0.870710	0.194697

3D Landmark Measurement Paired T-test Values

Trial 1 (A) & Trial 2 (B)	Mean	N	Std. Deviation	Std. Error Mean
A 1-2	10.65489	20	0.927301	0.207351
B 1-2	10.53776	20	0.832283	0.186104
A 1-3	18.92995	20	1.428430	0.319407
B 1-3	18.97840	20	1.332592	0.297977
A 1-4	25.08725	20	1.799152	0.402303
B 1-4	25.49820	20	2.026435	0.453125
A 1-5	32.50065	20	2.410452	0.538993
B 1-5	33.01615	20	2.852926	0.637934
A 1-6	36.56115	20	2.365778	0.529004
B 1-6	36.69050	20	2.721148	0.608467
A 1-7	39.79405	20	2.363570	0.528510
B 1-7	39.61540	20	2.566550	0.573898
A 1-8	41.36410	20	2.342733	0.523851
B 1-8	41.04535	20	2.436776	0.544880
A 1-9	41.80305	20	2.463582	0.550874
B 1-9	41.56420	20	2.465302	0.551258
A 1-10	42.43220	20	2.566429	0.573871
B 1-10	42.40190	20	2.582776	0.577526
A 1-11	41.64170	20	2.700131	0.603768
B 1-11	41.81235	20	2.826236	0.631966
A 1-12	38.69610	20	2.812893	0.628982
B 1-12	39.12750	20	3.012199	0.673548
A 1-13	34.7196	20	2.811531	0.628678
B 1-13	35.4217	20	3.043435	0.680533
A 1-14	26.4828	20	2.211017	0.494398
B 1-14	27.10135	20	2.395660	0.535686
A 1-15	18.43700	20	1.449227	0.324057
B 1-15	19.00810	20	1.676410	0.374857
A 1-16	9.654855	20	0.705871	0.157838
B 1-16	9.97178	20	0.845431	0.189044