University of Crete Faculty of Medicine Department of Forensic Science

# Doctoral Dissertation IDENTIFICATION OF SEX BASED ON DIGITAL RADIOGRAPHS OF THE SKELETON

Ο ΠΡΟΣΔΙΟΡΙΣΜΟΣ ΤΟΥ ΦΥΛΟΥ ΤΟΥ ΑΤΟΜΟΥ ΜΕ ΒΑΣΗ ΤΗΝ ΑΚΤΙΝΟΛΟΓΙΚΗ ΕΞΕΤΑΣΗ ΤΟΥ ΣΚΕΛΕΤΟΥ

# ELENA F. KRANIOTI



**HERAKLION, JULY 2009** 

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A DISSERTATION SUBMITTED TO THE MEDICAL SCHOOL AND THE COMMITTEE ON GRADUATE STUDIES OF THE UNIVERSITY OF CRETE IN FULFILLMENT OF THE REQUIREMENTS FOR THE DEGREE OF DOCTOR OF PHILOSOPHY

> ELENA F. KRANIOTI HERAKLION, JULY 2009

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# Summary in Greek

# Ο προσδιορισμός του φύλου του ατόμου με βάση την ακτινολογική εξέταση του σκελετού.

# Πεوίληψη

Η ταυτοποίηση ενός πτώματος αγνώστων στοιχείων αποτελεί πρωταρχικό στόχο κατά την Ιατροδικαστική διερεύνηση ενός θανάτου. Κάτι τέτοιο είναι σχετικά εύκολο σε περιπτώσεις που ο θάνατος έχει επέλθει κάποιες ώρες έως και λίγες μέρες πριν από την ιατροδικαστική έρευνα και διασώζονται ακόμη με ευκρίνεια τα χαρακτηριστικά του προσώπου ή ακόμη και τα αποτυπώματα ή κάποιοι ιστοί από τους οποίους μπορεί να γίνει γενετική ταυτοποίηση του αγνώστου πτώματος. Μετά την αποσύνθεση όμως, πολλά από τα αρχικά χαρακτηριστικά δεν είναι διαθέσιμα για αναγνώριση και ταυτοποίηση. Παρ 'ότι η ταυτοποίηση είναι σχετικά ευκολότερη διαδικασία σε περιπτώσεις που ανευρίσκεται ολόκληρος ο σκελετός, γίνεται εξαιρετικά δύσκολη έως και αδύνατη όταν διασώζονται μόνο μερικά οστά, και μάλιστα θρυμματισμένα και κατεστραμμένα, ενώ τα τμήματα του σκελετού τα οποία είναι ζωτικής σημασίας για την αναγνώριση του πτώματος εκλείπουν. Καθίσταται λοιπόν αναγκαία για την Ιατροδικαστική Επιστήμη, η ανάπτυξη νέων μεθόδων ταυτοποίησης σκελετικών υπολειμμάτων.

Θα πρέπει να υπογραμμιστεί πως η σκελετική ταυτότητα του κάθε ατόμου υπακούει σε κάποια συγκεκριμένα χαρακτηριστικά τα οποία είναι αντιπροσωπευτικά για την πληθυσμιακή ομάδα στην οποία ανήκει και κατά συνέπεια για τη φυλετική του καταβολή. Οι διάφοροι πληθυσμοί διαφέρουν μεταξύ τους εξαιρετικά, ιδιαίτερα όταν προέρχονται από γεωγραφικές περιοχές οι οποίες είναι απομονωμένες μεταξύ τους για εκτεταμένο χρονικό διάστημα. Το γεγονός αυτό καθιστά τον καθένα από τους απομονωμένους πληθυσμούς μοναδικό και εντελώς διαφορετικό από τους υπόλοιπους. Για το λόγο αυτό γεννάται η ανάγκη ανεύρεσης διαφόρων ειδικών τεχνικών με βάση τις οποίες θα γίνει εφικτή η δημιουργία μιας βάσης δεδομένων και η σύγκρισή της με τις αντίστοιχες βάσεις δεδομένων που έχουν δημιουργηθεί για άλλες πληθυσμιακές ομάδες.

Η Ελλάδα ως σύνολο και η Νότια Ελλάδα πιο συγκεκριμένα φαίνεται να αποτελεί μια γεωγραφικά σχετικά απομονωμένη περιοχή. Οι περισσότερες μεταναστεύσεις πληθυσμών περιορίστηκαν στην ηπειρωτική Ελλάδα και δεν κατάφεραν να επεκταθούν στα ελληνικά νησιά. Έτσι κατά το μεγαλύτερο μέρος της ιστορίας οι άνθρωποι των περιοχών αυτών παρέμειναν διαφορετικοί, γεγονός που πρέπει να λαμβάνεται σοβαρά υπόψη κατά την ιατροδικαστική διερεύνηση, πριν αποφανθεί κανείς σχετικά με την ταυτοποίηση ενός ατόμου αγνώστων στοιχείων. Ο σκοπός αυτής της έρευνας είναι ο σχεδιασμός μια τεχνικής για τον προσδιορισμό

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του φύλου, ειδικά σχεδιασμένης για τον πληθυσμό της νότιας Ελλάδας, και πιο συγκεκριμένα για τον πληθυσμό της Κρήτης..

Επιπρόσθετα τόσο οι Έλληνες γενικά όσο και οι Κρητικοί πιο συγκεκριμένα αποτελούν πληθυσμούς οι οποίοι ουδέποτε μελετήθηκαν ανθρωπολογικά στο παρελθόν και των οποίων τα μορφολογικά χαρακτηριστικά θα επιχειρηθεί να μελετηθούν και να καταγραφούν. Στόχος της εν λόγω ερευνητικής εργασίας είναι η ανάπτυξη συγκεκριμένων ανθρωπομετρικών τεχνικών για τον προσδιορισμό των χαρακτηριστικών του φυλετικού διμορφισμού στον υπό εξέταση πληθυσμό. Επιπρόσθετα τα αποτελέσματα της μελέτης δύναται να εφαρμοστούν και σε άλλες πληθυσμιακές μελέτες στην Ελλάδα και σε ολόκληρη την περιοχή των Βαλκανίων αλλά και σε μεσογειακά φύλα, προσδίδοντας τα δεδομένα για τον προσδιορισμό του φύλου ατόμων αγνώστων στοιχείων σε Ιατροδικαστικές υποθέσεις.

## Υλικό και Μέθοδοί

Το υπό εξέταση δείγμα αποτελείται από σκελετούς από τη συλλογή των οστεοφυλακίων του κοιμητηρίου του Αγίου Κωνσταντίνου και των Πατελών, του Ηρακλείου Κρήτης. Ο πληθυσμός μελέτης περιλαμβάνει σκελετούς Κρητικών ή ατόμων που γεννήθηκαν και έζησαν στην Κρήτη πάνω από τρείς γενεές. Στο δείγμα συμπεριλαμβάνονται άτομα που έζησαν και απεβίωσαν στην Κρήτη μεταξύ τέλη 19<sup>οο</sup> και αρχές 20<sup>οο</sup> αιώνα. Άτομα με καταγωγή από άλλα μέρη της Ελλάδος, μετανάστες από την Μικρά Ασία και άτομα με εμφανή παθολογία αποκλείστηκαν από τη μελέτη. Η ηλικία και η αιτία θανάτου ανεβρέθη από τα πιστοποιητικά θανάτου του Ληξιαρχείου Ηρακλείου, για το μεγαλύτερο μέρος του δείγματος ενώ το φύλο ήταν εμφανές από τα ονόματα στο εξωτερικό των οστεοθηκών. Η μέση ηλικία για τους άνδρες είναι 68.57+/-13.52 (N=61) και για τις γυναίκες 72.98+/-16.9 (N=58) έτη.

## Οστεομετοική μέθοδος

Κρανίο: Συνολικά 178 καλά διατηρημένα κρανία χρησιμοποιήθηκαν για τη μελέτη. Δεκαέξι μετρήσεις πραγματοποιήθηκαν σε σπλαχνικό και προσωπικό κρανίο. Η κάτω γνάθος αποκλείστηκε λόγω υπερβολικής φατνιακής απορρόφησης συνέπεια της προχωρημένης ηλικίας μεγάλου ποσοστού του δείγματος.

<u>Μακρά οστά:</u> Συνολικά 172 σκελετοί χρησιμοποιήθηκαν για τη μελέτη των μακρών οστών. Δώδεκα κλασσικές μετρήσεις ελήφθησαν από τα οστά του άνω άκρου (βραχιόνιο, ακτίνα, ωλένη) και δεκαοκτώ από τα οστά του κάτω άκρου (μηριαίο, κνήμη, περόνη).

Όλες οι μετρήσεις ελήφθησαν σύμφωνα με τη μέθοδο του Martin (Martin and Saller, 1959).

# Ακτινομετοική μέθοδος

Συνολικά 106 σκελετοί χρησιμοποιήθηκαν για την ανάπτυξη της ακτινομετρικής μεθόδου. Τέσσερα οστά (βραχιόνιο, ακτίνα, μηριαίο και κνήμη) ακτινογραφήθηκαν σε συγκεκριμένη ανατομική θέση.

Το ακτινολογικό μηχάνημα που χρησιμοποιήθηκε είναι ένα ψηφιακό μηχάνημα τύπου TCA 4R PLUS το οποίο αποθηκεύει τις λήψεις και μπορεί κανείς να τις μεταφέρει στον ηλεκτρονικό υπολογιστή που είναι προσαρτημένος στο μηχάνημα και να τις επεξεργαστεί ή να κάνει οποιαδήποτε μέτρηση. Αποτελεί μέρος του βασικού εξοπλισμού του εργαστηρίου Ιατροδικαστικών Επιστημών για την ακτινολογική εξέταση των ιατροδικαστικών περιστατικών, είναι εύχρηστο και δεν απαιτεί ειδική εκπαίδευση για το χειρισμό του η εξειδικευμένη γνώση και εμπειρία Ακτινολόγου, δεν απαιτεί τη χρήση αναλώσιμων πχ ακτινογραφικά φίλμς κτλ., όπως τα συμβατικά ακτινολογικά μηχανήματα, γεγονός που το καθιστά εξαιρετικά πρακτικό και

Δύο ακτινογραφίες ελήφθησαν από κάθε οστό για κάθε επίφυση ξεχωριστά. Ένας συγκεκριμένος αριθμός σημείων επιλέχθηκαν σε κάθε ακτινογραφία και όλες οι αποστάσεις μεταξύ όλων των σημείων υπολογίστηκαν. Οι αποστάσεις αυτές αποτέλεσαν τις μεταβλητές για την πραγματοποίηση της εν λόγω μελέτης. Χρησιμοποιήθηκε η ανάλυση μεταβλητότητας (ANOVA) για την επιλογή των μεταβλητών που διαφέρουν σημαντικά (p<0.05) μεταξύ των δυο φύλων. Η στατιστική μέθοδος που εφαρμόστηκε είναι η μέθοδος διακρίνουσας ανάλυσης (Discriminant function analysis). Επιπρόσθετα χρησιμοποιήθηκε η μέθοδος της βηματικής διακρίνουσας ανάλυσης (stepwise discriminant function analysis) ώστε να προσδιορισθούν οι παράμετροι που είχαν σημαντική στατιστικά διαχωριστική ικανότητα. Η ανοχή και οι τιμές του F (κριτήριο συμμετοχής στη διακρίνουσα συνάρτηση F> 3.84 και κριτήριο απόρριψης F<2.71). Για την ελαχιστοποίηση του σφάλματος με τη χρήση των διαφείνουσων συναρτήσεων, για κάθε συνάρτηση υπολογίστηκαν οι εκ των υστέρων πιθανότητες σωστής ταξινόμησης (posterior probabilities). Το σφάλμα μέτρησης μεταξύ του ίδιου και διαφορετικών παρατηρητών (inter- και intra-observer error) υπολογίστηκε με την δοκιμασία t (student 's t-test) για τη σύγκριση μέσων τιμών.

Η επιλογή των σημείων στις ακτινογραφίες και η μετρήσεις έγιναν με τη χρήση μιας σειράς λογισμικών (Tpsutil, Tpsdig2, Morpheus et al.). Η στατιστική επεξεργασία των δεδομένων έγινε με τη χρήση του στατιστικού προγράμματος SPSS 13 (Statistical Package for Social Sciences).

# Αποτελέσματα

# Οστεομετρία

Όλες οι μεταβλητές βρέθηκαν να διαφέρουν σημαντικά (p<0.05) μεταξύ των δυο φύλων. Τα μακρά οστά διαχώρισαν το φύλο με μεγαλύτερη επιτυχία από τις διαστάσεις του κρανίου. Ένας συνδυασμός 5 μεταβλητών του κρανίου διαχώρισε το φύλο με επιτυχία σε 88% του δείγματος. Ο καλύτερος συνδυασμός μεταβλητών για το βραχιόνιο οστό διαχώρισε το φύλο με επιτυχία σε 91% του δείγματος. Ο καλύτερος συνδυασμός όλων των μετρήσεων των οστών του άνω άκρου διαχώρισε το φύλο επιτυχώς σε 95% του δείγματος ενώ του κάτω άκρου σε 92% του δείγματος.

# Ακτινομετοία

Όλες οι μεταβλητές του βραχιόνου οστού, πλην μίας, βρέθηκαν να διαφέρουν σημαντικά (p<0.05) μεταξύ των δυο φύλων. Το ποσοστό σωστής ταξινόμησης ήταν παρόμοιο για την άνω και κάτω επίφυση του βραχιόνου. Οι μονές μεταβλητές ταξινόμησαν σωστά έως 86% του δείγματος ενώ διαφορετικοί συνδυασμοί μεταβλητών διαχώρισαν το φύλο με επιτυχία σε έως 89% του δείγματος.

Συνολικά 7/28 και 13/15 να διαφέφουν σημαντικά (p<0.05) μεταξύ των δυο φύλων για την άνω και κάτω επίφυση της ακτίνας αντίστοιχα. Η καλύτεφη μεταβλητή για την άνω επίφυση της ακτίνας ταξινόμησε σωστά 85% του δείγματος ενώ κανένας συνδυασμός μεταβλητών δεν κατάφεφε να υπεφβεί το 80% σωστής ταξινόμησης. Αντίθετα για την κάτω επίφυση ένας συνδυασμός 10 μεταβλητών ταξινόμησε σωστά 92% του δείγματος.

Η άνω και κάτω επίφυση του μηριαίου έδωσαν παρόμοια αποτελέσματα τόσο για τις μονές μεταβλητές όσο και για συνδυασμούς αυτών. Η καλύτερη μεταβλητή για την άνω επίφυση της μηριαίου ταξινόμησε σωστά 86% του δείγματος και για την κάτω επίφυση 85% του δείγματος. Οι συνδυασμοί των μεταβλητών και στις δυο περιπτώσεις διαχώρισαν το φύλο με επιτυχία σε 90% του δείγματος.

Η άνω επίφυση της κνήμης έδωσε καλύτερα αποτελέσματα σε σχέση με την κάτω επίφυση. Η καλύτερη μεταβλητή για την άνω επίφυση της κνήμης ταξινόμησε σωστά 88% του δείγματος ενώ κανένας συνδυασμός μεταβλητών δεν υπερέβη αυτό το ποσοστό.

# Συμπεράσματα

- Ο προσδιορισμός φύλου με τη χρήση γραμμικών μεταβλητών από ακτινογραφίες των μακρών οστών είναι δυνατή με σωστή ταξινόμηση έως και 95% του δείγματος.
- Η άνω και κάτω επίφυση έδωσαν παρόμοια αποτελέσματα σωστής ταξινόμησης για την ακτινομετρική μέθοδο σε όλες τις περιπτώσεις με εξαίρεση την ακτίνα.
- 3. Η ακτινομετQική μέθοδος μποQεί να χQησιμοποιηθεί ως εναλλακτική μέθοδος έναντι της οστεομετQικής σε πεQιπτώσεις ανεύQεσης σκελετικών υπολειμμάτων από μαζικές καταστQοφές ή διαμελισμένα θύματα ανθQωποκτονιών. Η πQαγματική υπεQoχή ή όχι της ακτινομετQικής μεθόδου έναντι της οστεομετQικής θα πQέπει να ελεγχθεί με μεταστατιστική ανάλυση.
- Η οστεομετρική μέθοδος είναι επιτυχής στον προσδιορισμό φύλου για τον Κρητικό πληθυσμό ο οποίος δεν είχε συμπεριληφθεί μέχρι τώρα στις γνωστές βάσεις δεδομένων.
- Ο σεξουαλικός διμορφισμός του Κρητικού πληθυσμού αντανακλάται περισσότερο στα μετρικά χαρακτηριστικά των μακρών οστών σε σύγκριση με το κρανίο.

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- Figure 8.2.1: a) Landmarks selected on the radiograph of the proximal humerus, b) Landmarks selected on the radiograph of the distal humerus.
- Figure 8.2.2: Plots of the first 2 principal components of PCA in proximal humerus: a) Shape-spaceb) Form-space and distal humerus c) Shape-space d) Form-space. Note that there is a clear separation of sexes in both proximal (b) and distal (d) end when form variables are used.
- Figure 8.2.3: Proximal Humerus: a)Deformation grid of the female configuration, b)Deformation grid of the male configuration c)Deformation grid adjusted to the mean female image e) Deformation grid adjusted to the mean overall image e) Deformation grid adjusted to the mean male image f) Mean female consensus g) Overall consensus h) Mean male consensus. All grids and mean images are exaggerated 5 times in order to visualize better the observed shape differences.
- **Figure 8.2.4:** Distal Humerus: a)Deformation grid of the female configuration, b)Deformation grid of the female configuration c)Deformation grid adjusted to the mean female image e) Deformation grid adjusted to the mean overall image e) Deformation grid adjusted to the mean male image f) Mean female consensus g) Overall consensus h) Mean male consensus. All grids and mean images are exaggerated 5 times in order to visualize better the observed shape differences.

Figure 8.3.1: ROC curves and cut-off values for the single variables of the humerus.

Figure 8.3.2: ROC curves and cut-off values for the single variables of radius and ulna.

# Part A

# **Chapter 1: Introduction**

When a human body is discovered, the primary goal in a forensic investigation is the identification of the deceased and the definition of the cause and the manner of death (Di Maio, 2001). The identification is quite an easy procedure in relatively recent deaths, where face and fingerprints are available. Quite often, though, individuals are found disfigured or in a highly decomposed state, without fingerprints. In mass disasters bones are usually commingled, charred and fragmented, thus identification is relayed in few components. The existing skeletal elements are partially exposed because of the remaining soft tissue; hence special techniques, like maceration, are needed in order to carry out the examination. Therefore, the necessity of developing new techniques for skeletal identification emerges.

The first and most vital biological characteristic under consideration is sex, since it reduces the number of possible matches in the population by fifty percent (İşcan and Loth, 1997; Loth and İşcan, 2000). The overall reliability depends on the method and on the specific population being examined. Of all demographic characteristics, sex differences have probably been studied the most. Almost every human bone has been analyzed in this regard (Stewart, 1979; Krogman and İşcan, 1986; İşcan, 2000). Scholars agree that sex diagnosis of adult skeletons can be performed easily and with high accuracy (Krogman and İşcan, 1986). Theoretically, sex assessment is easy to accomplish in puberty, when males and females diverge significantly so they can follow their distinct, genetically-determined forms and reproductive functions (Novotny et al., 1993). However, it becomes more complicated in adulthood when the sex discriminating traits become less marked.

The reliability of sex determination depends on the parts of the skeleton that are recovered as well as the conditions of preservation. Krogman and İşcan (1986) state that sex assessment in a collection of 750 skeletons was possible, with levels of reliability of 100% when the entire skeleton was present. However, in forensic investigations this is rarely the case, since the bones are usually recovered in a fragmentary state due to the effect of extreme environmental conditions and activities of carnivores and/or other scavengers. Normally in developed countries, as in the U.S. or Canada, forensic anthropologists are the experts who assess sex from recovered remains, using a variety of methods based on the skeletal characteristics specific to the regional population. The situation is different in Greece, where the discipline of forensic anthropology is in nascent state and there are only a few professionals, trained abroad, to be consulted when such cases emerge.

It has long been acknowledged that both cranial and postcranial characteristics of the skeleton are population specific and thus many studies have been carried out worldwide to develop population-specific methods. However, a lack of such investigations is noted in the Balkan area and in Greece in particular. Most pathologists use methods developed for other populations, hence the probability of a wrong estimate is higher. In addition, the recovered remains may be partially fleshed, charred or fragmented, thus the application of conventional techniques requires special techniques such as maceration to carry out the examination. In forensic cases, however, the remains are not always permitted to be macerated, or if so, the process can be time-consuming and slow down the investigation. An alternative way to study bones is the application of image processing techniques such as radiography and computer tomography.

The application of imaging methods allows the visualization of the bones independently of the state of the remains (semi-fleshed, mummified or charred), thus allowing immediate observation prior to autopsy. Moreover, radiographic equipment is routinely used in forensic departments and recently conventional radiographic machines have been replaced by digital ones, which have no additional cost for materials (ex. film). Digital radiographic equipment can produce and store the radiographs immediately, thus allowing a rapid evaluation of the skeleton in forensic cases. The hypothesis addressed here is the potential use of radiographs of the skeleton for identification of sex. Radiological identification was first introduced in 1926 by Culbert and Law and since then, it has been extensively used in diagnosing skeletal pathology and trauma as well as in positive identification (Krogman and Sassouni, 1957; Krogman and İşcan, 1986; Kahana and Hiss, 1997; Kahana et al., 1997; Kahana and Hiss, 1999). Nevertheless, its use in skeletal identification it has been, until recently, limited to classical radiographic methods (Riepert et al., 1996; Sağir, 2006; Petrovecki et al., 2007). Lately though, digital radiographs have been used in sex assessment of the femur with rather satisfying results (Harma and Karakas, 2007).

The recovery of fragmentary skeletal remains, in forensic investigations, requires easy and rapid techniques for biological profiling and reconstruction of scene history. The use of radiographs instead of the actual bones allows the identification of semi-decomposed bodies without the need for special preparation (ex. maceration), thus facilitating the whole forensic investigation. The current study aspires to accomplish a threefold purpose: to develop a sex determination technique using digital radiographs of long bones, to provide osteometric data on a contemporary population from Crete, Greece, and to introduce the discipline of forensic anthropology into modern multidisciplinary medico-legal investigation.

# Chapter 2: History of Forensic anthropology

# 2.1. Physical and forensic anthropology around the world

# 2.1.1. America

# 2.1.1.1 USA

The introduction of forensic anthropology in medico-legal practice took place, according to Stewart (1979), in 1878 when T. Dwight published a paper on the value of skeletal remains in forensic investigation. Yet there are reports of earlier cases, such as the identification of the military officer Warren Boston by the technician who repaired his denture (1776); and the expert witness testimony of two Harvard anatomists in the murder trial of Harvard chemistry professor John White Webster, who killed and dismembered George Packman (1850).

In the late nineteenth century anthropology gained some recognition marked by the foundation of American Anthropological Association (1888) and the American Journal of Physical Anthropology (1918). Many important scholars have contributed to the development of the field, such as Thomas Dwight, an anatomist with considerable research activities on age estimation of cranial sutures, sex determination from long bones and on skeletal variability (Stewart, 1979), and George Dorsey, the first person to earn a doctorate in anthropology from Harvard and one of the first expert witnesses in history (Stewart, 1978; Brickley and Ferllini, 2007). In the so-called Luetgert case, Dorsey opposed the evidence that was brought to court by two anatomists, providing the initial step towards the recognition of forensic anthropology in courtrooms (Brickley and Ferllini, 2007).

Another important contribution to the field is attributed to Alex Hrdlicka, an anthropologist with various research activities who started one of the biggest osteological collections in the Smithsonian Institution. Hrdlicka was involved in many legal cases and established a continuous collaboration with the Federal Bureau of Investigations (FBI) during the 1930s-1940s (Brickley and Ferllini, 2007). Other important figures in the history of anthropology in the U.S. include Laurence Angel, T. Dale Stewart, Ellis R. Kerley, Wilton M. Krogman, etc., all with considerable achievements. Krogman's article (İşcan, 1988) entitled "Guide to Identification of Human Skeletal Remains" (1939) in the FBI Law Enforcement Bulletin served not only in numerous forensic cases but also in the identification process of World War II victims. It is essential to acknowledge the huge contributions of T. Wingate Todd and Robert J. Terry, the creators of the two most important osteological collections, housed in the Smithsonian Institution. The great majority of the early research on osteology was based on these collections (DiBennardo and Taylor, 1982; İşcan and Miller-Shaivitz,

1984; Berrizbeitia, 1989; Holman and Bennett, 1991) Even nowadays these remarkable osteological banks aid in the development of new methods by current forensic professionals (Ubelaker and Volk, 2002; Brown et al., 2007; Case and Ross, 2007a; Albanese et al., 2008).

World War II, the Korean War and the Vietnam War created the need for special teams to identify the victims, in which forensic anthropologists became an essential part. Furthermore, the collaboration of the FBI with scientists from the Smithsonian Institution like Krogman, Stewart and Angel, founded a new period in the development of forensic anthropology. Pretty soon the first books appeared (Krogman and İşcan, 1962; Bass, 1971); the first symposium took place, having only 4 speakers (1948); and the first seminars and graduate courses were established. Multivariate statistical analysis also emerged as a very important tool for anthropological research and practice.

Recent decades are marked by the participation of an increasing number of scientists in operations following mass disasters such as bomb casualties, fires, plane crashes and so on, as well as in the excavation of mass burial sites after wars. Even though education has started to spread in academic world, forensic anthropologists still have to enter the medico-legal world as physical anthropologists with an MSc or PhD in related subjects, and employment is available for only a limited number in military and government agencies.

The situation has begun to change lately with the incorporation of forensic anthropologists into medical examiners' offices. In big cities like New York or Vancouver, anthropologists are recruited for field and laboratory work. Although it is quite rare for crime scene investigation to be performed by anthropologists, they actively participate in the casework and they form their own reports concerning the findings of the examination in skeletal material cases. A very important thing to note is that there have been court cases in which the expert witness testimony of a forensic anthropologist was considered indispensable. In order to fulfil these requirements, anthropologists are submitted to continuous training and specialization in forensic anthropology, and an accreditation system has been developed in the United States in order to certify their capacity to express a professional opinion in legal cases.

The official incorporation of forensic anthropology in forensic medicine dates to 1972 with the foundation of the section of physical anthropology within the disciplines acknowledged by the American Academy of Forensic Sciences (Güleç and İşcan, 1994; Kennedy, 2000). In 1977 the American Board of Forensic Anthropology (ABFA) was formed with the goal to "encourage the study of, improve the practice of, establish and enhance standards for, and advance the science of forensic anthropology and to encourage and promote adherence to high standards of ethics, conduct, and professional practice in forensic anthropology". Certification is based upon the candidate's personal and professional record

of education and training, experience and achievement, as well as on results of formal examinations.

The ABFA requires a re-certification of all board-certified anthropologists in order to maintain their status. To achieve that, every diplomat has to fulfil several requirements, such as providing evidence of maintenance of the knowledge and the skills to practice forensic anthropology, keeping updated according to the current methods applied in forensic anthropology, and providing service to the community with respect to the ethical laws of the ABFA. Since its formation, the ABFA has improved its standards through the years. The annual update was created in 1984. The ethics policy was approved in 2001 and added to the re-certification process in 2003, along with an expanded section on continuing education. In 2002 ABFA applied for membership on the Forensic Sciences Accreditation Board and membership was granted in 2003. With this membership, the ABFA signals its ongoing commitment to the highest professional standards of practice and its intention to continue working to refine and improve the re-certification process.

# 2.1.1.2 Latin America

A considerable contribution to the development of forensic anthropology in Latin America is considered to be the foundation of the Equipo Argentino de Anthropologia Forense (EAAF) in 1984. EAAF is a non-governmental, non-profit, scientific organization that applies forensic anthropology and archaeology to the investigation of human rights violations in Argentina and worldwide. It was initially established to investigate the cases of at least 10,000 disappeared people in Argentina during the military government that ruled from 1976-1983. Currently EAAF has an international presence in South America, Europe, Africa, the Middle East and French Polynesia. Other forensic anthropology teams have been established in Chile (1989), Guatemala (Guatemala Forensic Anthropology Foundation, 1991), and Peru (Peruvian Forensic Anthropology Team 2001). Guatemala and Peru have endured violence, political oppression and human rights violations in recent decades, due to the extreme political instability and lawlessness that challenged the principles of democracy in much of Latin America between 1975 and 2000. The objectives of these organizations are focused on investigating and recording human rights violations, providing court evidence, assisting in identification of missing persons and contributing to the reconstruction of recent history. An integral part of the investigation consists of exhuming bones from mass graves or ossuaries, reconstructing their biological characteristics and attempting to match them with missing individuals.

From a forensic standpoint, the foundation of the Laboratory of Forensic Anthropology at the Morgue Judicial in Montevideo, Uruguay, and the assignment of forensic anthropologists as official consultant of coroners and legal authorities marks a new period of development for forensic anthropology (İscan and Olivera, 2000). During a seven-year time interval (1991-1997) a total of 189 cases, corresponding to 276 individuals, were brought to the Forensic Laboratory of Montevideo, from all judicial departments of Uruguay. 122 individuals were examined and of them 46 were positively identified. Facial reconstructions from the skull and electronic superimposition of a photograph have been employed frequently, consulting soft tissue thickness of other populations, due to lack of regional data. Similar techniques along with dental records were applied to various known cases such as of Pizarro (Maples et al., 1989), Mengele (Snow et al., 1984; Helmer, 1986), Sagredo (Solla and İşcan, 2001) and a mass suicide in Jonestown, Guyana (Thompson et al., 1987). Lately there has been some research activity in the field, concerning the population-specific standards that are required to assess biological characteristics in different populations of Latin America. Dental studies on eruption in children (Argentina Peru, Brazil and Cuba), dental wear (Paraguay) and dental size differences are reported (İşcan and Olivera, 2000). Sexual dimorphism was studied in the pelvis (Mexico), calcaneus and talus (Cuba), humerus, scapula and clavicle (Guatemala) (İşcan and Olivera, 2000; Frutos, 2005) and so on. Yet many aspects of the osteological characteristics of the majority of the South American countries have not been studied.

Despite the positive steps in the development of population specific standards for Latin America, no significant effort was made to formalize the education and training in forensic anthropology and the integration of specialists in the medico-legal system. Thus it is safe to conclude that more work should be done in terms of training and recognition of the need for forensic anthropologists in the organization of the medico-legal team, in most parts of South America.

#### 2.1.2 Australia

A short of the history of forensic anthropology in Australia is presented by Donlon (2008). According to the author Australia exhibits a short history in the field with slow development. Similarly to the United States, the discipline was initiated by anatomists, who were called by law enforcement agencies to assist in forensic cases around the 1950s. Some of the first anatomists involved in forensic work were Neil Mackintosh and Stan Larnach (University of Sydney), Les Ray (University of Melbourne), Fredrick Wood Jones and Andrew Arthur Abbie (University of Adelaide), David Allbrook and Len Freedman (University of Western Australia) and Walter Wood (University of Queensland) (Donlon, 2008). These anatomists began as physical anthropologists studying collections of Aboriginals and trained many students that are employed today as forensic anthropologists. The lack of non-

Aboriginal collections played an important role in the slow development of the field, since scientists were obliged to seek such collections in the U.S.

In many cases worldwide, anthropologists gained experience by identifying war victims. Australian legislation requires the identification and the reburial of their war dead from World War I and II to be made on site (Donlon, 2008). Since the Vietnam War, all Australian solders killed in battle have been repatriated. Australian anthropologists along with other professionals have formed part of the Australian Disaster Victim Identification team and they have been involved in the identification of victims of terrorist attacks, as in the Bali bombing (Lain et al., 2003).

The formation of the Australian Academy of Forensic Sciences (AAFS) in 1967, followed by the New Zealand Forensic Science Society (1971), attracted many different disciplines associated with forensics and offered the baseline for the development of forensic anthropology. The first journal appeared (Australian Forensic Science) and the first symposium with a session in forensic anthropology took place (Sydney, 1996). Another association related to anthropology is the Australia Society of Human Biology, formed in 1996, which has lately included forensic sessions in the conferences (Donlon, 2008).

Australia, along with many other countries, lacks the professional organization for forensic scientists and an official board for forensic anthropology found in the U.S. system (Briggs, 1998 in Donlon, 2008). Donlon (2008) reports that only one full-time forensic anthropologist is employed by the state. A few others are employed in forensic medicine departments or by the police, while a lot of work is still assigned to the departments of anatomy. A report of forensic cases in a period of 15 years (1992-2006) in the State of New South Wales shows an increasing number of forensic anthropologists in the past years with a peak in 2003 probably due to the Bali terrorist attack. Most of the time, they were requested by the coroner's office and sometimes directly by the police. Their work included excavation, recovery of the remains, biological profile, positive identification etc. It is worth noting that of the 153 cases of that report, only 5 times the forensic anthropologist was called in court.

Even though there seems to be a long way to go until Australian anthropologists can reach the American standards concerning the formation of an accreditation board and a professional association, the necessity of their involvement in forensic cases seems to be well acknowledged and the development of the field is increasing day by day.

# 2.1.3 Africa

Forensic anthropology in Africa became more relevant with the increase in the number of violent crimes and deaths, which resulted in a large amount of unidentified human bodies and skeletonized remains (Steyn et al., 1997; Franklin et al., 2007a; Franklin et al.,

2008a). The necessity of positive identification of these remains has led to the development of osteological standards for the identification of the African population. Dental records are not available, which makes any comparison of post-mortem records impossible. Illegal immigrants tend not to report missing people to the police, due to lack of proper identity documents, while even those who are African citizens do not declare missing persons easily (Steyn et al., 1997). More specifically for South Africa, these particular socioeconomic circumstances and the increase of violence created the necessity for the development of identification methods. Several scholars have contributed to the development of meristic and morphometric standards for identification of South Africans, such as Washburn (1949), Keen (1950), De Villiers (1968), Macho (1990b), Lundy and Feldesman (1987), Kieser et al. (1992), Loth & Henneberg (1996; 1998; 2001), Steyn and İşcan (1997; 1998; 1999), İşcan and Steyn (1999), Asala (2001; 2004), Oettlé & Steyn (2000), Patriquin et al. (2002; 2003b; 2005), Steyn and Smith (2007), Franklin and Cardini (2007), Franklin et al. (2007a; 2007b; 2008a; 2008b), Roelofse et al. (2008), Bidmos (2008), Bidmos and Dayal (2004), Barrier and L'Abbé (2008), Oettlé et al. (2005; 2009) and others. Although research activity is increasing, forensic anthropologists are few in number. More specifically there are only two professionals in Pretoria and one in Cape Town. In Pretoria a formal agreement with the authorities has been established and about 70 cases per year are brought to the forensic anthropology laboratory for examination. Although forensic anthropologists are not authorized to sign death certificates, they are regarded as expert witnesses and in some cases they are called for testimony in court (M. Steyn personal communication).

# 2.1.4 Asia

## 2.1.4.1 China

Forensic anthropology in Asia exhibits a different rhythm of development between the different countries. As has been the case elsewhere, the discipline has its roots in medicine, anatomy and physical anthropology. The rudiment of anthropology in China can be traced back to the classical work of traditional Chinese medicine "Huangdi Neijing" (Canon of Emperor) published in the date of the Warring state, 2500 years ago. This book recorded the physical characteristics of the people in different areas and the measurements of human skeletons and internal organs. Forensic medicine also has a long history in China, with the first manuscript appearing in 1246; however modern forensic pathology practice delayed until 1930 (Zhang and Pounder, 1998). Physical anthropology, on the other hand, was widely accepted as a discipline only after 1949, despite the early work of many foreign (F. Blumenback, A. Hrdlicka) and native (W. Dingliang, L. Hi) scientists, and exhibited rapid development only after the beginning of 1970. Many research papers have appeared since then with a focus on extensive morphological analysis of sex and age differences, which has contributed significantly to the development of forensic anthropology (Spencer, 1997). İşcan and Ding (1995) report one of the first manuscripts in forensic anthropology by C. Li in 1993, while some papers on Chinese populations (Wu et al., 1980) were published in Acta Anatomica Sinica (founded in 1953). Although the development of the field lags behind the developed Western countries, research activities have increased and forensic anthropology has become a subject of teaching in many Chinese universities. In the current medico-legal system, eight medical colleges and the Department of Forensic Sciences of the Ministry of Justice in Shanghai are active in China. The latter publishes the Chinese Journal of Forensic Medicine which includes a good number of articles in forensic anthropology in Chinese (Zhao et al., 2005; Zhang et al., 2006; Shi et al., 2008; Wang et al., 2008), while some scholars have made international contributions, among others in cranial-image superimposition (Chai et al., 1989; Lan, 1995), sex (İşcan and Ding, 1995) and age (Xu et al., 1991; Li and Ji, 1995; Xiping et al., 2008) assessment from teeth and different bones. The economic development in China has had a positive effect on the field, which is expected to develop further.

# 2.1.4.2 Turkey

In Turkey, distinguished pioneers in physical anthropology, such as Sevket A. Kansu and Muzaffer S. Senyürek, focused on the skeletal biology of the historic and prehistoric inhabitants of Anatolia (Güleç and Işcan 1994). The development of forensic medicine created the need for anthropological contribution to casework especially in establishing biological profiles and positive identifications. Although the necessity for forensic anthropology professionals is acknowledged, at the early steps of applying anthropological methods in medico-legal cases, Turkish professionals have adopted techniques developed in Europe and America (Güleç and İşcan, 1994). The foundation of the Adli Tip Dergisi (Turkish Journal of Forensic Sciences) in 1985 has brought together many forensic professionals and increased the interaction between traditional anthropologists and osteologists and forensic pathologists. Around the same time (1988) forensic anthropology was officially introduced to the Department of Forensic Medicine of the Institute of Legal Medicine and Forensic Sciences of Istanbul University with the incorporation of forensic osteology courses to the existing master's and PhD programs (Güleç and İşcan, 1994). In the following years the interest in the field increased significantly, with a large number of scientific contributions to international journals (Yavuz et al., 1998; Günay and Altinkök, 2000; Ozaslan et al., 2003; Ozden et al., 2005; Pelin et al., 2005; Uysal et al., 2005; Celbis and Agritmis, 2006; Ozer et al., 2006; Sağir, 2006; Büken et al., 2007; Harma and Karakas, 2007; Akansel et al., 2008; Hatipoglu et al., 2008). Today, research programs are under way in the Institute of Forensic Medicine in Istanbul and the Department of Physical Anthropology in Ankara to include the

collection of data on modern Turks (Güleç and İşcan, 1994). A number of research projects dealing with for example the development of age and sex determination standards for the Turkish population and other aspects of forensic anthropology are in progress (M.Y. İşcan personal communication). In addition, this field has attracted several graduate students (from medicine and archaeology) who are seeking a career in forensic anthropology. However, accreditation remains an issue since the potential forensic anthropologists rely on individual training or the limited workshops organized in Turkey without any official professional association, as seen in the majority of the non-U.S. countries.

# 2.1.4.3 India

In India, until the late 1960s and early 1970s, physical anthropology was concerned with anthropometric, dematoglyphic and serological studies. By the late 1980s and early 1990s, however, it became less laboratory-oriented, due to the lack of laboratory equipment to follow the western standards. Anthropologist focused on demographic studies and adopted several morphological and metric techniques from the West. The lack of museum collections for study and research has significantly affected the rate of development. During the past decade physical anthropologists have become unofficially involved in forensic cases. In India, as elsewhere, the examination of skeletal remains or individual bones was routinely undertaken by forensic pathologists of the corresponding forensic medicine department or the forensic science laboratory (Purkait, 2006). However, the increasing population and the multiracial profile of the country led the forensic departments to frequently ask assistance in cases of skeletonized remains from the anthropologists who were obviously more experienced in dealing with bones. The training opportunities have increased with the introduction of forensic anthropology as an independent course at the postgraduate level in five out of thirty anthropology departments in India, with Delhi University as the first in 1984 (Purkait, 2006). Forensic anthropology as a part of forensic science is also taught as a graduate course in five other universities. In parallel, Indian anthropologists have exhibited increasing research activity at the international level, publishing a series of papers in forensic anthropology (Purkait, 1996; Selvaraj et al., 1998; Jayaprakash et al., 2001; Purkait, 2001; Purkait, 2003; Purkait and Chandra, 2004; Purkait, 2005; Sahni et al., 2005; Nagesh et al., 2006; Agnihotri et al., 2007; Krishan and Sharma, 2007; Krishan, 2008; Moudgil et al., 2008; Sen and Ghosh, 2008; Zeybek et al., 2008). Despite the recent advances, though, the professional status of forensic anthropologists in India still remains without proper recognition.

# 2.1.5 Europe

In Europe the development of forensic anthropology varies significantly among the different countries (Brickley and Ferllini, 2007). Some countries present early research activity

in the field, as with France, Germany and Spain, while others are facing primitive situations concerning the study of human remains. In all cases, though, individuals that deal with skeletal remains derive from many different disciplines like archaeology, forensic medicine and biology and in no case do they have adequate training equivalent to the anthropologists in the U.S. (Brickley and Ferllini, 2007). In some countries such as the U.K., Denmark and Portugal, most forensic anthropologists are in academic institutions and their training experience and occupation vary (Brickley and Ferllini, 2007). Sometimes anthropologists are employed in forensic institutes as in the Netherlands, or in government organizations as in Spain and Hungary (Brickley and Ferllini, 2007; Prieto, 2009). Naturally without common training, no accreditation system exists between the countries of Europe.

# 2.1.5.1 Germany

In Germany, forensic anthropology has developed in different aspects. Since the question of whether recovered skeletal remains belong to humans often emerged, early investigations were concentrated on this task. Quantification of Haversian canals in bones' cross-sections was applied for that purpose (Rämsch and Zerndt, 1963; Schiwy-Bochat, 1991). Sex determination was based on morphological features of the skull (Leopold, 1978), or parts of it such as mastoid and temporal bone (Graw 1997 in Swiny-Bochat et al., 2004), while metric studies included mandible and mastoid. Image processing techniques were developed for the quantification of shape in different bony structures as orbit contour, mastoid and supraorbital margin (Schiwy-Bochat et al., 2004). Age determination initially was based on the expansion of marrow cavities and the quantification of osteons mainly in long bones, while currently the aspartic acid racemization technique seems to be the method of choice (Schiwy-Bochat et al., 2004). Considerable work has also been done in positive identification with the use of superimposition, computer and magnetic resonance tomography as well as trace elements analysis (Schiwy-Bochat et al., 2004). Finally, the development of age estimation methods on living individuals (Schmeling et al., 2000; 2003) is considered to be one of the most significant contributions of the German school of forensic anthropology.

# 2.1.5.2 France

The French school of anthropology rose with the foundation of the *Société de Anthropologie* de Paris in 1959 by Paul Broca. It is worth noting that the early identification method of Bertillon based on anthropometric measurements as a factor of individualization was only replaced by fingerprint identification in 1920 (Kennedy, 2000). Anthropologists were concentrated on research activities without any collaboration with pathologists until the mid-1980s (Baccino, 2009). An overview of the contributions of French scientists (anthropologists and medical doctors) to various fields of forensic anthropology is presented by İşcan and

Quatrehomme (1999). These include contributions to general anthropology (Olivier, 1959; Olivier et al., 1978), fetal and adult aging (Balthazard 1921, Derobert 1974 in İşcan and Quatrehomme, 1999), as well as histological and radiological methods (Barres and Durigon 1989, Rollet in Derobert 1974 in İşcan and Quatrehomme, 1999), functional stress, handedness and osteopathological markers, and trauma (İşcan and Quatrehomme, 1999). The most important recent accomplishment of the French school of anthropology is considered to be the two-criteria Lamendin technique on age estimation (Lamendin, 1978), which was introduced in 1978 and improved later in 1992 by Lamendin and coworkers (1992).

A very important step in the development of forensic anthropology in France was the foundation of the Brest Bone Collection (BBC), which started in Brest (Brittany, France) and later continued in Montpellier (France). It was initially made from hospital and forensic autopsy cases of known age, sex, ancestry, and stature, but it was restricted to forensic cases after 1994 when "bioethics laws" were issued in France, making collection of body parts from cadavers impossible. The BBC has now more than 400 individuals with pubic symphyses, fourth ribs, medial clavicles, iliac crests and teeth (Baccino, 2009).

About 50 forensic scientists, mostly pathologists, are dealing with the routine anthropological cases in France today (Baccino, 2009). Nevertheless, the training of the potential professionals is limited to international workshops, since there is no French university to offer specific graduate or post-graduate courses in forensic anthropology (Baccino, 2008). Unfortunately, there is still no official national recognition of forensic anthropology as an academic specialty in France since, as İşcan and Quatrehomme (1999) pointed out and Baccino (2009) reinforced, forensic anthropology still is considered within the duties of the forensic pathologist.

## 2.1.5.3 United Kingdom

Medicolegal investigations were performed atypically in the UK long before the development of distinct disciplines, which emerged in the 16<sup>th</sup> century and have become clearly defined in the past 25 years (Black, 2003). The increasing interest in forensic specialties has been attributed to both "the court's insistence on greater precision" and to the sudden "symptom of popularity" deriving from the media. Among the appealing forensic subspecialties, forensic anthropology holds one of the first places.

In the United Kingdom, forensic anthropology is practiced by a large variety of professionals. Some have a background in archaeology, or so-called osteoarchaeology, others are anatomists or forensic pathologists. The training system is complicated since archaeological universities may sometimes include anthropology as sub-discipline, or not, while in medical universities some departments of anatomy, as the one in Dundee, include a forensic anthropology formation. Several universities also have the option of postgraduate courses or master's degrees on the subject, attracting such a high number of participants that provoque saturation and accumulation of students in waiting lists. Research, on the other hand, is quite developed in many aspects, as for instance, in positive identification (Thompson, 2004; Berry et al., 2008; Meadows and Black, 2008; Wilkinson, 2008). Nevertheless, the use of forensic anthropologists in real cases is considered limited in UK. This is a general remark, since every area has its own autonomous practice. According to Black (2000, in Brickley and Ferllini, 2007), 98% of the British police that answered a questionnaire stated that they had never used a forensic anthropologist. In the same paper it is underlined that forensic cases concerning skeletal remains are undertaken mostly by pathologists with little or no experience in the field, as in many other European countries. In the past decade, the UK has developed an accreditation system which allows forensic anthropologists to undergo an evaluation process and register in one of the four forensic anthropology sub-specialties (general forensic anthropology, osteology, modelling and computed-based facial anthropology). More details on the accreditation system can be found in Black (2003). It is worth noting, though, that currently there are less than 10 board-certified forensic anthropologists in the UK (Black, 2008).

However, the job of forensic anthropologists has expanded in the last 10-15 years as they are more frequently asked to assist the international community in the investigation of war crimes, abuses of human rights and humanitarian repatriation. The mass graves of Rwanda, Yugoslavia and Iraq required their assistance as much as the disasters of the World Trade Centre, the Asian tsunami or the London bombings (Black, 2009).

# 2.1.5.4 Hungary

Hungary has a long history of physical anthropology which dates back to the 1960s with the work of many important researchers such as Nemeskeri, Harsaniy, Fasekas and Kosa and so on (Susa, 2007). The establishment of a democratic government in 1989 has led to an increase in research activity due to the necessity of investigating political victims. For that purpose many exhumations have taken place since 1989 and anthropologists were involved in the investigation of individuals that died between 1945 and 1962. It is important to note that political and war crimes in Hungary do not lapse and thus, upon positive identification of an individual, the family may claim compensation from the state. Seventy-one individuals who had died during political struggles were positively identified with the contribution of forensic archaeologists and anthropologists (Susa, 2007).

Nowadays forensic anthropologists in Hungary usually have a background in archaeology or medicine or they obtain a MSc in biological sciences after a three-year training (Susa, 2007). As an exception to the general rule, anthropologists in Hungary are often asked by the police to contribute to forensic cases concerning identification of skeletal remains,
excavation of mass graves and even identification of the living. In this last case they deal mostly with age determination of individuals involved in pornographic videos or photos, or of refugees that illegally enter the country.

# 2.1.5.5 Italy

In Italy, according to a report from the Institute of Legal Medicine in Milan alone, which performs over 1,000 autopsies every year, there are on average 30–40 cases per year which require application of anthropological or odontological techniques, either for defining individualization factors for positive identification, or for biological reconstruction (e.g. aging, sexing, facial approximation, etc.). The need for estimation of post-mortem interval and ancestry of recovered skeletal remains has also emerged (Cattaneo and Baccino, 2002). However the training system is quite poor since only the University of Milan offers postgraduate and master's courses in forensic anthropology, while some universities are limited to a few workshops (Cuhna and Cattaneo, 2006). Although research activity has increased over the years with several papers on sex and stature estimation (1993a; Introna et al., 1993b; Di Vella et al., 1994; Introna et al., 1997; Campobasso et al., 1998; Introna et al., 2005; 2006; Gualdi-Russo, 2007; Benazzi et al., 2008), still the implication of forensic anthropologists in medico-legal routine is limited.

### 2.1.5.6 Spain

Forensic anthropology in Spain formally appeared in 1865 with the foundation of the Spanish Anthropological Society and the Anthropological Museum in Madrid (1875) (Prieto, 2009). Soon after, the first scientific journals, the first reference collection of crania and the early research projects appeared (Prieto, 2009). In late 19th century, anthropology was associated with three different fields of forensic medicine; "Analyzing the relationship between human physical features and criminal conduct, identification of the living and identification of skeletal remains" (Prieto, 2008). Soon after the Spanish juridical system appointed by law forensic pathologists as official advisors on medical and biological issues in court, the necessity of having anthropological expertise emerged, thus anthropological issues were included in the forensic pathologist evaluation exam (Vibert 1916, in Prieto 2008). The Spanish Civil War negatively contributed to the development of anthropology since it resulted in the destruction of many institutions including the National Institute of Physical-Natural Science, where the Anthropological Museum was incorporated (Otero Carvajal 2001, in Prieto 2008). The modern period of forensic anthropology in Spain is marked by the foundation of the Laboratory of Forensic Anthropology and Paleopathology in the Madrid Legal Medicine School (Reverte 1991, in Prieto 2008). The first book in Spanish entitled Forensic Anthropology introduced the current techniques used in the United States to the

forensic professionals, and anthropological skills were required from all forensic pathologists in order to practice.

Lately some laboratories were incorporated in routine forensic cases in order to assist in cases of unidentified skeletonized bodies. There are nine such laboratories in Spain now with about 200 cases in total per year. Forensic pathologists continue to be the ones handling skeletonized cases but anthropological reports are supplementary to these forensic cases. Furthermore, the teaching of anthropology in Spain now forms part of the forensic medicine training and is offered also in universities as undergraduate and postgraduate courses, master's degrees and PhDs.

Nevertheless, research remains limited by the lack of reference collections and most of the ongoing activity takes places in institutions affiliated with forensic medicine departments. Unfortunately state grants on biomedical research do not include forensic medicine or forensic anthropology, which is one more limiting factor to the development of the field.

# 2.1.5.7 Portugal

The history of anthropology in Portugal starts back in 1885 with foundation of the disciplines anthropology, paleoanthropology and pre-history at the Coimbra Institute of Anthropology (Cuhna and Pinheiro, 2007). The main activities of the Institute of Anthropology (1903-1927) concerned anthropometric measurements and other methods for identification of living individuals. After that anthropological cases were taken by the recently founded (1920) Forensic Medicine Institutions (Coimbra, Lisbon, Oporto) (Cuhna and Pinheiro, 2007).

Even though forensic anthropology in a preliminary form appeared early in Portugal, the establishment of the discipline in its current form did not come until 1990 with the foundation of the National Institution of Legal Medicine from the fusion of three autonomous forensic institutes (Cuhna and Pinheiro, 2007). NILM is an autonomous administrative entity in the city of Coimbra that answers directly to the Ministry of Justice and is composed of three delegations (Cuhna and Pinheiro, 2007). Despite the fact that no department of anthropology officially exists in NILM, all cases concerning decomposed and skeletonized bodies are undertaken by anthropologists that collaborate with the Forensic Pathology Department of one of the three delegations (Cuhna and Pinheiro, 2007). Anthropologists are also called at times by regional forensic units (medico-legal offices) in cases that require their assistance. It has been estimated that in Portugal (12 million inhabitants) with a total mortality rate of 105,000 individuals per year, the forensic anthropology cases for 2004 were 30. The cases of anthropological interest include recovery of animal or human bones as, for instance, modern cemetery disturbances, archaeological remains, missing elderly people, homicides and age estimation of living adolescents (Cuhna and Pinheiro, 2007). In these cases by law only a forensic pathologist can sign a death certificate; nevertheless the anthropologist assisting in the case usually prepares an anthropological report and if asked also attends court for testimony (E. Cuhna personal communication).

In the past years there seems to have been a quick evolution in the field with increasing research activity and establishment of postgraduate courses in universities and the involvement of forensic anthropologists in crime scene investigation. Nonetheless, in very few cases a forensic anthropologist is called to a crime scene, especially in rural areas of Portugal where forensic work is handled by graduates of medicine without any special training (J. Pinheiro personal communication).

#### 2.1.5.8 Balkan Peninsula

The Balkan area has constantly suffered political instability, wars and catastrophes that have negatively affected the living conditions of the people and the development of the region for centuries. The recent wars in Croatia-Serbia, Bosnia-Herzegovina and Kosovo are examples of the instability of the region, which resulted in numerous war victims including citizens and soldiers of the neighboring countries. Executions, mass burials of civilians along with military personnel and a large number of missing persons are common characteristics in all war conflicts of the Balkan region (Šlaus et al., 2007). Obviously the need for identification of the victims and repatriation emerged. In some countries like Croatia special teams were formed (commission of imprisoned and missing individuals) to proceed with the identification of the skeletal remains exhumed from the mass burials (Šlaus et al., 2007).

Obviously the demanding need for identification brought many professionals to the field to contribute their skills. This resulted in the formation of multidisciplinary teams from many parts of the world cooperating with the local expertise in the identification of the remains. Because of the lack of ante-mortem an essential part of the identification process required anthropological techniques. The recovered skeletal remains of the Balkan area have given a great amount of information on the skeletal characteristics of the local populations, which were not represented so far in the existing databases. Research focused on craniofacial characteristics of different ethnic groups (Ross, 2004), postcranial elements for sex (Šlaus et al., 2003; Šlaus and Tomičić, 2005) and stature estimation (Ross and Konigsberg, 2002; Petrovecki et al., 2007; Jantz et al., 2008b). Forensic anthropology methods were applied successfully in a large number of cases (Brkić et al., 2004; Šlaus et al., 2007). The rest of the Balkan countries seem to lack osteometric data on the local populations, at the level of published works in international journals. Some research has been presented on various

occasions to the meetings of the Balkan Academy of Forensic Sciences, which indicates an initial step towards the development of the discipline of forensic anthropology.

# 2.1.5.9 FASE

As a result of the obvious need for forensic anthropology experts in Europe, an effort to promote and deal with this need was made with the foundation of the Forensic Anthropology Society in Europe (FASE). FASE was formed in September 2003 in Milan as an official subdivision of the International Academy of Legal Medicine. Its aim consists of bringing together anthropologists, forensic pathologists, odontologists, geneticists and other experts in the fields of forensic medicine and forensic science in the scientific and academic promotion and development of the discipline of forensic anthropology across Europe. Its main objectives as stated on many occasions are " to encourage the study of, to promote the practice of, to establish and enhance the standards for Forensic Anthropology and to promote training and create a board of trained professionals." Within a period of the three years since its foundation FASE counts almost 50 members in the whole of Europe with rising number of activities such as workshops, intensive courses and conferences in order to attract more scientists and to educate and train a new generation of forensic anthropologists. Literature and research is growing significantly but still there are countries in Europe in which forensic anthropology remains an almost infantile discipline.

# 2.2 Physical and forensic anthropology in Greece

# 2.2.1 From ancient history to the Middle Ages

The history of anthropology in Greek culture is described in detail by Agelarakis (1995) in his article entitled as *An Anthology of Greeks involved in the field of Physical Anthropology*. He placed the foundation of anthropology unofficially in the 6<sup>th</sup> century B.C. when the disciplines of physiology, anatomy, medicine and biology started to form. Hippocrates, the founder of medicine, appears to have been the first to study the structure of the skeleton and described the treatment of fractures, trauma and surgical operations, setting the foundation of osteology. Aristoteles, on the other hand, was the creator of comparative anatomy and scientific zoology, setting the basis of posterior Linnean taxonomy (Agelarakis, 1995).

During the Hellenistic period (300BC-100AD) the foundation of the Alexandria library, which collected a huge amount of scientific manuscripts, facilitated the development of physiology and anatomy, which advanced considerably with the practice of dissection and embalming of bodies in ancient Egypt.

The Greco-Roman époque is marked by an important contribution to scientific legacy: that of Gaius Plinius Segundus in forming the Naturalis Historia, an encyclopedia of the time with tremendous amount of data on anatomy, zoology, medicine and anthropology.

Another medical encyclopedia by Cornelius Celsus kept a record of the Alexandria surgery techniques.

The Middle Ages were marked by the absence of scientific action until the beginning of the Byzantine Empire. During that time the scientific field was reinvigorated by daring Hellenized scholars. However, during the ruling of Justinianos, the University of Athens was closed as an attempt to forbid the activities of the pagan philosophers and to enhance Christianity.

# 2.2.2 From the 19<sup>th</sup> century until the present

Clon Stephanos, a medical doctor, was the one that reopened the field of physical anthropology with the foundation of the Anthropological Museum at Athens University, in 1886. He excavated prehistoric cemeteries in Syros and Naxos and a part of the recovered skeletal remains were transferred to the Anthropological Museum. Nevertheless, his work was mostly of archaeological interest. His best achievement was the creation of the department of physical anthropology at the University of Athens in 1913. The chair of the department passed then to professor I. Koumaris. He was trained in Berlin by Fischer and in an effort to reinforce the discipline he founded the Greek Anthropological Association. Despite his active presence for 49 years he concentrated on fieldwork, failing to establish a training system in anthropology.

Another important scientist who contributed in the field was Aris Poulianos, who was trained in both the U.S. and USSR and returned to Greece to establish the first department of anthropology and biology, at the University of Patras (1965). Two years later he was arrested and put in jail by the military junta government and the department of anthropology was shut down forever. After his release he founded the Greek Anthropologikh Etairia in 1970 and he devoted his time to research and publishing activities. He carried out the most extensive study of modern Greeks, taking more than seventy anthropometric measurements from a large sample of Greeks from different parts of the country. He concluded that both Greeks and their neighboring populations are basically a mixture of Aegean and Epirotic (Dinarics) tribes deriving from the ancient inhabitants of their lands (Poulianos, 1960: The origin of the Greeks).

Several studies on Greek populations were conducted by the American anthropologist J. Laurence Angel (1946; 1966; 1971). In his studies he often dealt with the problem of bad preservation of the skeletal material and the fact that male individuals are outnumbered compared to females in almost all studied populations (Angel 1943, 1971). An exception is the Lerna population and the Athenian Agora, where the numbers are comparable for both sexes, probably due to the better preservation of the skeletal material (Halstead 1977 in Liston

1993). Some studies dealt with paleopathology of different populations of late Bronze and Iron Age (Angel 1944, 1971, 1973, 1975, Angel and Burns 1973; McGeorge 1980 in Liston 1993). Dentition is one of the best-preserved parts of the skeleton in archaeological samples, so many of the studies focused on ancient populations' dental health (Angel, 1944). According to this there is a decline of dental health in late Helladic period III, compared to I and II, with the predominance of dental caries and absorbed alveoli in the population. Slightly later in the Submycenaean Late Bronze Age, the Cephallenia population (N=44) demonstrates a good dental status. Skeletal trauma was interestingly reported in 32.2% of this population, contrary to others where it was almost absent (Angel, 1943).

More recent publications were based on limited skeletal material of archaeological interest, making population studies impossible (Musgrave, 1984; Prage et al., 1984; Sakellarakis and Sapouna-Sakellaraki, 1981; Xirotiris and Langennscheidt, 1981 in Liston 1993).

Halstead (1977, in Liston 1993) studied pathology and life span in Bronze Age Minoans and populations from the mainland. Interestingly, Minoans demonstrate a significantly higher life span for both sexes during Middle Helladic period compared to mainlanders who present high mortality during childbearing in women and more frequently traumatic lesions in men. These differences were attributed to the different socioeconomic conditions of the different populations (Halstead 1977, in (Liston, 1993). A change occurred during Late Bronze age with a reduction of the traumatic evidence for male mainlanders while there is an increase of childbearing mortality for Minoan women especially during the Late Minoan III.

McGeorge (1988) studied a Cretan population (N=76) dated to the Late Minoan Bronze Age from Armenoi. Her work focused on pathology and dietary stress markers. She described a great frequency of enamel hypoplasia and Harris lines on tibias, suggesting seasonal nutritional deficiencies as a possible explanation. He also observed signs of anemia expressed as cranial porosity.

Manolis (1991) studied the craniofacial characteristics of different southern populations of the Bronze Age including Minoans, populations from Messinia, Attica and Argolida, and gave a morphological type of the craniofacial skeleton of each population. He concluded that there was less contact with significantly different populations. The study also presents the mean age on the different populations of Middle and Late Helladic, but interestingly the results do not agree with previous studies. More specifically, no differences are noted in the life span between Minoans and mainlanders during the Middle Helladic Period, while the situation improved during the Late Helladic period for both males and females. This is contrary to Halstead's observations (Halstead 1977 in Liston 1993). These differences, though, could be a result of the limited number of individuals in Manolis's study.

Tsivilakos and co-workers (2002) investigated the incidence of periodontitis in a Mycenaean population of the Late Bronze Age unearthed at the cemetery of Aghia Triada, West Peloponnese, Greece. In a total of 172 individuals the mean age is estimated to be about 38 years. A notable percentage of the individuals (24%) lost three or more teeth during their lifetime and a total 53% of the population had antemortem tooth loss. It was concluded that ancient jaws present a high proportion of antemortem tooth loss, attrition and deep caries, whereas the frequency of periodontitis does not seem to differ from that of other prehistoric samples.

It is worth noting that very little information is available concerning children in the literature mainly because of bad preservation and burial habits. According to Desborough (1964, in Liston 1993) children on the mainland were buried in simple pit graves or slabcovered cists while Garland states that from the Geometric period to Roman times pots or coffins were used for that purpose (Garland 1985, in Liston 1993). An exception is the study of a population found in Franchthi Cave in the Argolid peninsula, which contained 38 subadult individuals. The population is dated from the Upper Paleolithic to the Final Neolithic and also included 22 adults with mean age 32.3 years. The examination of the skeletons revealed pathological conditions such as the high incidence of cribra orbitalia (45%, 10/22) and porotic hyperostosis (20%, 14/71), trauma (15%, 9/60), industrial wear of the front teeth (12% 16/131), antemortem tooth loss and osteoarthritis, and relatively low incidence of dental caries (2.4% 11/458), LEH (6.8% 31/458) and infection. A paleodietary reconstruction has been performed by means of isotope analysis of human bone collagen and carbonate apatite on eighteen individuals. The results point to terrestrial, predominantly C3 diet focused primarily on plant resources. Both analyses suggest that the site was occupied by agriculturalists with a land-based subsistence (Papathanasiou, 2005).

Nikolaos Xirotiris, more recently, surveyed Greek skeletal material and a number of genetic and anthropometrical studies on modern Greeks (Schneider et al., 1975; Xirotiris et al., 1979). Several population studies in the Balkan area dealt with genetic material, mtDNA and Y chromosomes in order to study the similarity between neighboring populations (Huckenbeck et al., 2001; Scheil et al., 2001). He and his colleagues concluded, as did many others, that there has been racial continuity in Greece from prehistory, through classical and medieval, to modern times (Huckenbeck et al., 2001; Scheil et al., 2001; Scheil et al., 2001; Scheil et al., 2001; Scheil et al., 2001; Scheil et al., 2001; Scheil et al., 2001; Scheil et al., 2001; Scheil et al., 2001). His dissertation dealt with the population of Pomakoi in Northern Greece (Ξυροτήρης, 1971).

A few roentgenometric studies on cephalo-dentofacial morphology of contemporary populations have also been recorded (Argyropoulos and Sassouni, 1989; Argyropoulos et al.,

1989). Argyropoulos and associates (1989), in a cephalometric study by means of radiography, concluded that craniofacial characteristics in Greeks have remained unaltered for the past 4000 years.

Panagiaris and associates (1994) have studied the cephalometric characteristics of three groups of Sarakatsanoi, a Hellenic population which was considered nomadic until the 1970-80s. The groups were geographically isolated in Epirus, Central Macedonia and Peloponnesus. Nine cephalometric variables were measured and subjected to statistical analysis. 55% of the variables indicate a statistically significant difference but only the Peloponnesian population resulted to be well differentiated, while the other two groups were less distinct. More specifically the Peloponnesian population was found quite distinct in minimum frontal breadth, auricular height and morphological facial height.

Loukopoulou and Pentzos-Daponte (1995) conducted a craniofacial morphological study in children between 6 and 12 years old from Thessaloniki (northern Greece). The sample consisted of 689 males and 664 females from primary public school and is regarded as representative of the social status of the city. The authors used principal component analysis to manage the data and results indicate a quite homogenous population with small shape changes mainly attributed to vertical and ear variables. In a later work using the same data set, the authors studied the craniofacial sex dimorphism (Loukopoulou and Pentzos-Daponte, 1999). Results indicated that sexual dimorphism is expressed more in head variables than in facial characteristics. Furthermore before puberty is reached, it seems that sexual dimorphism is attributed to size differences rather than shape, in the given population.

As seen so far, the vast majority of the anthropological studies in Greece during the 20<sup>th</sup> century were based on archaeological material. Modern skeletal remains were not investigated most likely because of religious and local superstition. The Greek Church does not allow human remains to be removed or studied. Cemeteries are now all being "rented" for a couple of years. Bones are exhumed and later destroyed and put in a mass grave without any individual identity (Eliopoulos et al., 2007). However, a positive step towards the utilization of this remarkable osteological bank was the formation of the Athens collection (see below) completed in 2003 (Eliopoulos et al., 2007). Around the same time authorization was given to the Department of Forensic Sciences, University of Crete in order to analyze a certain number of skeletons from two cemeteries in Heraklion, Crete (see below).

# 2.2.3 The foundation of the first forensic anthropology lab in Greece

The initiative point for the development of Forensic anthropology in Greece in a practical sense was set with the foundation of the first forensic anthropology lab, in the Department of Forensic Medicine and Toxicology of the University of Athens. The foundation of the laboratory dates recently in 1999 under the leadership of K. Moraitis. The

laboratory deals with mass disasters, forensic and archaeological cases which involve skeletal remains, possesses the necessary equipment for maceration and examination of skeletal material (ex stereomicroscope) and undertakes part of the training of graduate and postgraduate students as well Forensic pathology residents (K. Moraitis personal communication). Aditionally contributes to the forensic community by a series of reseach achievements presented in international conferences and journals (Moraitis et al., 2006; Mitsea et al., 2009; 2009).

### 2.2.4 The Athens collection

The description of the Athens reference collection was made in detail by Eliopoulos et al. (2007). According to the authors the Athens collection was built in two phases. The first part of the collection, known as the "Wiener Lab Collection" (Pike, 1997), was built at the Wiener Laboratory of the American School of Classical Studies at Athens between the years 1996 and 1997 by A. Lagia. This collection consists of 72 documented skeletons that were acquired from cemeteries in the Athens area. In 1998, the collection was donated to the Department of Animal and Human Physiology, at the University of Athens (Roberts et al., 2005). An additional 153 skeletons were prepared by C. Eliopoulos, between the years 2001 and 2003, bringing the total number of specimens to 225. Of those, 114 (114 males and 100 females) have a complete record of age, sex, occupation, cause of death and place of birth. Mean age is 54.7 years for males and 55.5 years for females, while subadults are few in number. The vast majority of the samples derive from individuals that died between 1960 and 1996 and represent lower to middle socioeconomic classes. The origin of the individuals covers almost all the regions of the country.

# 2.2.5 The Cretan collection

The Cretan collection comprises of skeletons selected from among the available material of both Cemeteries of St. Konstantinos and Pateles, Heraclion, Crete. The collection consists mostly of Cretans or individuals that lived in Crete for more than three generations. More specifically it includes individuals who were born in Crete between 1867 and 1956, and died between 1968 and 1998. A number of people who may have migrated from Turkey (18), islands (1) and mainland Greece (3) are also included in the collection. Sex was available for all individuals while age, occupation, marital status and cause of death were accessed for most of the skeletons from the death certificates in the City Hall's archives.

This collection was initially created in order to be used in a study of long bones (current dissertation), therefore skeletons were selected with the criterion of the integrity of at least one set (left or right) of long bones. Most of the skeletons are quite well preserved, but, in a number of cases, pathology and trauma are present. A significant number of edentulous individuals and others with excessive alveolar resorption is observed. Cause of death is recorded in 77 cases, where a death certificate was available. A limited number of cases were submitted to autopsy for accidental deaths or homicides, as it was observed from the state of the skeleton but autopsy reports were not always available.

The Cretan collection has been been primarily created for the current dissertation, however it has been made available to several scholars (M.Y. İşcan, M. Steyn) and students (B. Mergen) for research purposes.

# 2.2.6 Recent anthropological research

The creation of two large osteological collections the last decade in Greece has resulted in increasing research activity with special emphasis in forensic identification. The most important contributions are listed below. In a preliminary study by Fox and associates (2003) a sex determination technique based on the morphology of the pituitary fossa was attempted. The sample consisted of 23 males and 9 females (with craniotomies) from the Wiener Laboratory Modern Human Skeletal Collection of the University of Athens in Athens, Greece. The authors suggested that females tend to have a more pronounced tuberculum sella (Turkish saddle) compared to males (English saddle). A blind test of the technique resulted in correct sexing as accurate as 88%.

Papaloucas and colleagues (2008a) looked at sexual dimorphism on 200 pelvises and femora from the Athens collection. Four dimensions were measured and two indices were calculated. Single dimensions performed well giving up to 95% classification accuracy. This method succeeded in identifying 99% of the original sample using only the ratio of the distance from the pubic tubercle to the anterior acetabular rim over the acetabular diameter.

Steyn and İşcan (2008) conducted an osteometric study on the sexual dimorphism of the pelvis. The material used derived from the Cretan collection and consisted of 199 dry skeletons with complete pelvises. The authors took 17 classical measurements and managed to correctly classify 95% of the individuals.

Papaloucas and colleagues (2008b) studied the asymmetry in length of right and left humerii in the Athens collection and concluded that there is a statistically significant difference (p < 0.001) between the mean length of right and left humerus in Greeks with higher predominance in males.

İşcan and colleagues (İşcan et al., 2009) studied dental health and odontometric characteristics of a population, who lived during the 15<sup>th</sup> -13<sup>th</sup> centuries B.C. (Mycenaean era) at the site of Apatheia, Galatas, in the northeast Peloponnese. The Galatas population is represented in a fragmented condition with considerable postmortem tooth loss. Examination is based on 245 teeth, on which many dental health problems, such as dental caries and abscesses, are noticed. It was observed that their nutrition was slightly based on grainy food. There is evidence of occasional use of their teeth in occupations like fishing and

sailing. Hypoplasia is notable on most of the incisors, as a result of nutritional stress and fever. Compared to modern Greeks, teeth dimensions in the Galatas population are smaller.

Recently the Institute for Aegean Prehistory (INSTAP) has funded a research project which will deal with the craniofacial morphology and characteristics of an ancient Greek population, excavated in Ilida (North-West Peloponese). The study will be conducted in collaboration with the Department of Forensic Sciences, University of Crete (E.F. Kranioti, M. Michalodimitrakis), the Max Planck Institution for Evolutionary Anthropology (K. Harvati) and the Z' Ephoreia of Prehistoric and Classical Antiquities of Olympia (X. Arapogianni, G. Rambach). The purpose of this work is to document the biological characteristics of the craniofacial complex of a population dated in Protohelladic I period (3600-2800 BC), a unique osteological material for that time in the Peloponnese region and to assess this morphological pattern in the context of global geographic human variation, as well as in the context of local morphological change through time among populations inhabiting the broader region of Greece, including classical and recent period groups. The processes that influence the expression of the particular morphological pattern (population history, selection pressures to climate and dietary requirements, health and pathology) will be evaluated.

While research is making small steps of progress, the training system is absent. No training opportunities exist for postgraduate students in the form of a master's degree or PhD. Most of the physical anthropologists are biologists trained outside the country, who deal mostly with archaeological material. The medico-legal system does not involve anthropologists in investigation since osteological examination is considered within the duties of the forensic pathologist. Yet the forensic pathology training program does not include any tasks of forensic anthropology, thus pathologists are evaluating skeletal remains mostly using handbooks and standards produced from different populations. No protocol of examination exists; hence, it is in the jurisdiction of each pathologist to decide how to manage the investigation. An exception to this general rull is the foundation of the first forensic anthropology lab in Athens (1999) with only one however forensic anthropologist for the entire Greece. The system needs to establish major changes in order to bring anthropologists abroad. However, to create a new discipline, a potent training system is demanded. The desperate need for research material and expertise led to the design of this study, which aspires to serve as a catalyst for the advancement of the field in Greece.

# Chapter 3: Previous research on sex identification

# 3.1 Morphological Methods

# 3.1.1 Skull

The skull is the single most studied bone in physical anthropology and much of our knowledge of human evolution is based on cranial remains (Krogman and İşcan, 1986). It has been widely used for age and sex determination with the evaluation of morphological features. The reliability of cranial traits for sex and age estimation is discussed widely by many authors (Krogman, 1955; Perizonius, 1984; Krogman and İşcan, 1986; Novotny et al., 1993; Walrath et al., 2004; Rogers, 2005; Williams and Rogers, 2006)

In 1955, Krogman (1955) introduced 13 traits capable of distinguishing male from female skulls and in 1986 concluded that 92% accuracy could be achieved (Krogman and İşcan, 1986) Some scientists tested a number of traits suggested by Krogman (Williams and Rogers, 2006), while others used isolated parts of the human skull such as the glabella region, the mandible and the mastoid process with diverse results (Krogman and İşcan, 1986). In general, the features of the facial cranium performed better than those of the calvarium, a fact that was verified in more recent studies (Williams and Rogers, 2006).

In a recent study of a mass murder grave in Serbia (Durić et al., 2005), cranial traits were used for sex estimation. The sample, consisting of individuals of Albanian descent killed in the recent Kosovo war, was sexed with an accuracy rate that did not exceed 71.0%.

Some authors emphasized the significance of estimating inter- and intra-observer error in scoring non-metrical traits for sex estimation, suggesting that experience and methodological standardisation are of great importance (Gualdi-Russo et al., 1999; Walrath et al., 2004). Williams and Rogers (2006) suggested six highly specific traits for sex estimation, highlighting the value of zygomatic extension and nasal aperture, traits not recognized to any great extent so far in the literature.

# 3.1.2 Pelvis

The pelvis has long been recognized as the most sexually dimorphic bone due to reproductive roles (Novotny et al., 1993). Thus it is considered the best skeletal element for the assessment of sex from skeletal remains. Several techniques for the visual evaluation of traits of the hip bone have been reported (Phenice, 1969; Houghton, 1974; Kelley, 1978; İşcan and Derrick, 1984; MacLaughlin and Bruce, 1986; Suri and Tandon, 1987; Sutherland and Suchey, 1991; Fernandez Camacho et al., 1993; Rogers and Saunders, 1994; Luo, 1995; Bruzek, 2002; Patriquin et al., 2003a; Ginesse A. Listi, 2006)

Phenice (1969)used three traits on the pubis. He suggested that the presence of the ventral arc in the surface of the pubis, the sub-pubic concavity and the medial aspect of the ischio-pubic ramus are female characteristics. He achieved an accuracy of 95% using this method of morphological observation. Sutherland and Suchey tested two of the three variables suggested by Phenice in a larger forensic sample (N=1984). They found 94% accuracy for the ventral arc but only 70% for the ischiopubic ramus. Uberlaker and Volk (2002) have also tested the method in a sample of 198 individuals from the Terry collection. Sex was correctly assigned in 88.4% of the cases, with better accuracy in females. Consideration of additional traits raised classification accuracy to 96.5%, with higher classification in males (Uberlaker and Volk, 2002).

The method of Ferembach et al. (1980) suggests sexing the entire pelvis through an evaluation of eleven traits. İşcan and Derrick (1984) worked on the posterior pelvis, achieving correct group assignment in 90% of the cases. Bruzek and Ferembach (1992) found 93% correct sex assignment, using a set of eight variables of the hip bone. Bruzek (2002) studied individual traits on the pelvis and provided 60–80%, correct classification, but when multiple traits were combined, accuracy increased to 95%. Rogers and Saunders (1994) tested accuracy and reliability in a set of 17 individual traits on a small sample of 49 pairs of innominates, achieving up to 83% accuracy by combining the scoring of three traits.

Patriquin and associates (2003) studied morphological features on 400 pelvises from South Africans Blacks and Whites. Overall pubic bone shape was the easiest to assess and the most reliable morphological indicator of sex. Pubic bone shape and sub-pubic concavity form were found to be the most reliable traits in Whites, with 88% correct group assessment.

#### 3.1.3. Long bones

Sexual dimorphism of the humerus has been studied so far in terms of size. One must consider, however, that sexual dimorphism is also expressed in shape and in that concept there is a lack of evidence on this topic. An exception is a shape analysis of the humerus in a Portuguese sample using transformed indices deriving from osteometric data (Carretero et al., 1995). The authors conclude that excluding size (which explains 80% of the observed variability), in the given population men tend to have shorter humeri with voluminous epiphyses, while women have longer shafts with smaller epiphyses.

Rogers (1999) developed a method of sex identification based on four visual traits of the distal humerus (thochlear constriction, trochlear symmetry, olecranon fossa shape and depth and angle of the medial epicondyle). He reported high classification in a sample of Whites from the Toronto Grant Collection. A test of the method on three other samples (Bass and New Mexico collection in America and St Bridge collection in UK) was successful but no test was attempted on African-Americans (Ceri et al., 2005; Klepinger, 2006). It has been suggested that the olecranon perforation of the distal humerus appears significantly more often in females than in males, which was attributed to differences in humerus robustness (Benfer and McKern in Klepinger, 2006). Nevertheless, this characteristic is not a very reliable sex indicator, according to other scholars (Klepinger, 2006).

Godycki (1957), in an early morphological study on sexual dimorphism of long bones, cited Martin, who had suggested that there is a division in the surface of the sigmoid notch in 66.2%, a partial division in 10.3%, and no division in 25.5% of individuals. He suggested that a divided notch is a male characteristic and a non-divided one a female characteristic, with 95% correct classification for males and 85% for females. Nonetheless, his results were not verified by other authors (Neto, 1959)

### 3.2 Anthropometric methods

### 3.2.1 Skull

Metric studies of the skull include the measurement of a large number of dimensions on the face and the calvarium, such as maximum cranial length, maximum cranial breadth, cranial height, facial breadth and height, bigonial breadth, etc. In the majority of the studies, accuracy didn't exceed 88% (Krogman and İşcan, 1986).

One of the earliest craniometric studies is the one of Giles and Elliot (1963) on 408 American Black and White crania of known sex. They found several dimensions such as cranial height, maximum bizygomatic diameter and mastoid length to differ significantly between the sexes. Classification accuracy yielded up to 85.5%.

Song and collaborators (1992) measured 41 dimensions in a sample of 80 Chinese skulls. A stepwise procedure selected 14 measurements which resulted in 100% correct group assessment. Hanihara (1959) worked on Japanese skulls and found 89.7% accuracy in diagnosing the sex correctly. In a more recent study, İşcan and Ding (1995) found accuracies of 84.1% (entire skull) and 83.7% (cranium only) in Japanese skulls.

Steyn and İşcan (1998) took 13 standard cranial measurements on South Africans. Correct group membership reached 98% in some cases. In another study by the same authors, correct sexing in White South Africans was achieved with an accuracy rate of 85.7% for crania and 80.2% for bizygomatic breadth alone. Franklin and co-workers (2007a) studied sexual dimorphism in the crania of indigenous, Bantu-speaking, South Africans. They used a number of linear measurements derived from three-dimensional data, comparable to those in use in classical osteometry. Facial width was found to be the strongest discriminating variable, followed by cranial length and basion-bregma height. Slaus and colleagues (2004) analyzed metric characteristics of medieval crania in 215 individuals from 39 European and 5 Iranian sites. Both principal component and discriminant function analysis were used. Results demonstrate significant differences between Central and South-East European medieval populations. The authors also focused on the development of effective discriminant functions for distinguishing early medieval Croats from individuals belonging to the Bijelo Brdo culture. The accuracies achieved by these functions reached up to 98%.

Deshmukh and Devershi (2006) studied sexual dimorphism on 74 adult crania of Indian origin. Correct classification using univariate analysis did not exceed 33%, with maximum cranial circumference found to be most reliable variable. Multivariate analysis performed considerably better (88%).

The petrous bone is one of the most often recovered parts of the fragmented skull because of its density, and therefore it has been tested previously for sex assessment (Kalmey, 1996; Graw et al., 1999; Wahl and Graw, 2001; Graw et al., 2003; Norén et al., 2005; Lynnerup et al., 2006b; Akansel et al., 2008). Some of the methods used, however, included complicated and difficult procedures, thus restricting their applicability. Graw and associates (Graw et al., 2003) measured the lateral angle of the petrous bone in the left and right internal acoustic canals of 205 individuals from an archaeological site. They concluded that the lateral angle is smaller in males as compared to females, while the opposite was observed for the medial angle. Although differences were found to be significant between the sexes, classification accuracy was poor. Lynnerup and co-workers (2006b) have tested the validity of the petrous bone for sexual identification using one single dimension: the diameter of the internal acoustic canal. The sample originated from South Germany and consisted of 173 adult individuals. Although an easy and simple measurement was employed, classification accuracy was very low (70%).

A comparative study of the ethnic groups' databases with the established norms of the North America Whites (NAW) was made in order to facilitate surgical restoration of the craniofacial morphology (Farkas et al., 2005). The study group consisted of 1470 healthy subjects from Europe, the Middle-East, Asia and Africa. A total of 14 measurements were taken. In the regions with single measurements, identical values to Whites in forehead height, mouth width, and ear height were found in 99.7% in both sexes, while in those with multiple measurements, vertical measurements revealed a higher frequency of identical values than horizontal ones. In the Middle Eastern groups, nose width was identical to those of NAW but the height was significantly greater.

Although most researchers focused on the morphological features of the pelvis, metric characteristics have also been studied in regard to sexual dimorphism. Early research suggested that acetabular dimensions were of significance in gender identification (Schulter-Ellis et al., 1983; 1985; Krogman and İşcan, 1986).

DiBennando and Taylor (1983) studied a sample of 260 innominates and femora from the Terry collection. Out of 32 measurements, 15 were selected by a stepwise procedure and three functions were produced. Results indicated a high discriminatory value for sex and race, reaching 95%. Principal component analysis permitted the separation of size and shape elements in the sample.

Nine dimensions were measured in a sample of African Black and Whites (Patriquin et al., 2005). Of all measurements, ischial length (in Whites) and acetabular diameter (in Blacks) were the most dimorphic. Classification accuracy reached 86%. Thirteen dimensions were assessed from the same population in order to explore racial affinities (Patriquin et al., 2002). It is noteworthy that two of the four dimensions chosen as best discriminators of race (pubic length, greater sciatic notch posterior width, iliac breadth/total height, acetabulum diameter) were also found to be good gender markers.

Rissech and Malgosa (2005) studied a sample of 327 individuals (51 juvenile and 176 adult) from the London, Coimbra, Lisbon and Barcelona collections. Six measurements from the ilium were taken in order to estimate sex and age in relation to ilium growth rate. The female Iberian series and the males of the Barcelona collection have higher mean values for ilium length than the Britannic series, thus an independent analysis of the ilium length was conducted for these groups. Four dimensions were used to discriminate sex. Sexual dimorphism in this bone was based on the different growth rate between the males and females. The earliest age where sex differences can be detected is about 12 years in acetabular diameter and 15 or later in iliac length and width. A considerable contribution to this paper also rests on the observation that age estimation can be accomplished by absolute measurements of the ilium.

### 3.2.3 Long bones

Apart from the skull and pelvis, many scientists have studied postcranial skeletons as well, and in some cases achieved sex prediction even in non-complete skeletal remains. Recent metrical and morphological studies on various elements, some of which are on populations with no previous data, have included long bones such as the humerus (Singh and Singh, 1972; Dittrick and Suchey, 1986; Carretero et al., 1995; İşcan et al., 1998; Rogers, 1999; Steyn and İşcan, 1999; Albanese et al., 2005; Frutos, 2005), radius (Berrizbeitia, 1989; Celbis and Agritmis, 2006; Barrier and L'Abbé, 2008), ulna (Steel, 1972; Singh et al., 1974; Introna et al., 1993; Purkait, 2001; Grant and Jantz, 2003; Matzon et al., 2006; Barrier and L'Abbé, 2008; Cowal and Pastor, 2008), femur (İşcan and Ding, 1995; King et al., 1998; Seidemann et al., 1998; Mall et al., 2000; Asala, 2001; Asala et al., 2004; Albanese et al., 2008), tibia (Hanihara, 1958; İşcan and Miller-Shaivitz, 1986; Holland, 1991; Kieser et al., 1992; Işcan et al., 1994; Steyn and İşcan, 1997; González-Reimers et al., 2000; Sakaue, 2004; Šlaus and Tomičić, 2005) and fibula (Sacragi et al., 1993). Some of the most important studies are summarized here.

### 3.2.3.1 Humerus

Given that osteometric methods for sex identification are population-specific, many researchers from around the world have conducted studies on the humerus in order to establish group specific standards of assessment for many different populations.

In Asia, Singh and Singh (1972) studied 290 humeri of an Indian population and suggested that maximum length is a good indicator of sex. İşcan and co-workers (1998) studied 82 Chinese, 79 Japanese and 104 Thai humeri using canonical discriminant function statistics on six standard dimensions. From the variables selected by a stepwise procedure, only epicondylar breadth and vertical head diameter were shared by all groups. The best single variable was vertical head diameter for the Chinese (80.5%) and epicondylar breadth for the Japanese (89.9%) and Thais (93.3%). Another study was carried out on a Japanese sample using in total 9 dimensions, of which the width of the distal articular surface (95%) and epicondylar breadth (94%) were the most effective dimensions. Classification results yielded up to 97% (Sakaue 2003).

Steyn and İşcan (Steyn and İşcan, 1999) worked with a South African sample from the Raymond Dart and Pretoria collection. A stepwise procedure selected epicondylar breadth and vertical head diameter for Whites (accuracy 89.1% for males and 95.8% for females) and vertical head diameter and maximum length for Blacks (accuracy 95.1% for males and 91.1% for females). The best single variable for Blacks was head diameter and for Whites epicondylar breadth.

In North America, Holmann and Bennett (1991) analyzed a random sample of 302 individuals from the Terry collection. Five measurements of the long bones of the upper limb were chosen. Interestingly, humeral length was found to be of high discriminatory value in Whites in combination with ulnar semistyloid breadth, giving 84.6% correct classification for males and 92.3% for females.

In South America, Frutos (2005) studied a Guatemalan sample of 118 humeri deriving from victims of the internal conflict underway in that country. Head diameter, minimum midshaft diameter and epicondylar breadth entered the analysis using the stepwise procedure (98.5% classification accuracy) while head diameter was found to be the best discriminatory variable. In this study, the sample was classified using standards of Chinese, Japanese, Thai, German and Spanish populations with diverse results.

In Europe, osteometric studies of the humerus were conducted in German and Portuguese populations (Carretero et al., 1995; Mall et al., 2001). Mall and co-workers studied a German sample of 143 individuals, and sex was correctly classified in 93% of the cases when the stepwise procedure was used. Interestingly, maximum length enters the equation along with head diameter and epicondylar breadth. Head diameter results in the best group membership, followed by epicondylar breadth. Standards for the Portuguese population (154 individuals) are accomplished using 12 variables. A stepwise procedure selects transversal diameter of the head and epicondylar breadth with 93.6% correct membership for males and 94.7% for females.

Some studies have a more archaeological context. In 1986, Dittrick and Suchey (1986) studied prehistoric remains in California using 9 humeral dimensions. The best discriminatory variables were, in order of reference, transversal diameter of the head (89.5%), followed by vertical head diameter (89%) and epicondylar breadth (85.2%). Furthermore, Albanese and co-workers (2005) developed standards for the humerus from an archaeological sample of Beneville (Canada) with satisfactory results.

### 3.2.3.2 Ulna

Although not as popular as the humerus, the ulna as well has been the subject of several osteometric studies. Steel (1972), for instance, measured three dimensions (length, coronoid height, distal width) in a small set of complete ulnae (17 males and 24 females). However, his results were probably biased due to the small sample size.

A later study by Singh and co-workers (1974) selected three ulnar measurements (length, midshaft circumference and distal breadth) for their study. Interestingly, almost 100% correct classification was achieved in the original sample, while the method was proven successful for 99.75% of the Indian population. A more recent study in a contemporary Indian sample from Madhya Pradesh, India, included three original measurements (olecranon-coronoid angle, length, and width of inferior medial trochlear notch) from the proximal epiphysis (Purkait 2001). Direct analysis revealed the olecranon-coronoid angle as the best single parameter. Discriminant functions deriving from several combinations of the variables correctly classified 80-96% of the population.

In a study of the Terry collection by Holmann and Bennett (1982), five measurements of the long bones of the upper limb were chosen and among them was ulna length and semistyloid ulnar breadth (SBB). SBB was defined as the distance between the most medial point of the head and the most lateral point of the styloid process, in right angle to the long axis. The combination of length and SBB of the ulna results in higher accuracy for Black (84.6% in both sexes) as compared to White (84% for males and 72% for females) Americans. Grant and Jantz (2003) used three different measurements (notch length, olecranon width and coronoid height) in order to predict sex from the proximal ulna. The sample comprised 217 individuals of European–American and African–American descent from three different collections, and findings were analysed by means of discriminant statistics. The results produced an accuracy of 100% for Black males and White females. Four White males (94.4%) and one set of Black females (97.1%) were misclassified. This discrepancy was attributed to the width of the olecranon process, which was the most strongly weighted variable (Grant and Jantz, 2003).

In Europe, a set of twelve ulnar measurements were taken on 80 skeletons deriving from a Southern Italian population (Introna et al., 1993). Several combinations of the measured distances were used to produce discriminant functions. The highest percentage of correct sex classification (95%) was obtained by the association of the minimum circumference and the maximum length. Mall and co-workers (2001) applied three measurements (maximum length, maximum proximal breadth and maximum distal breadth) in a contemporary forensic sample from Germany consisting of 143 individuals. Discriminant function analysis using all dimensions resulted in 90.58% correct sex assignment. Maximum length was found to be the most effective single dimension (87.05%), while the other two did not exceed 80% when used singularly.

Some authors tested the efficacy of circumferential measurements on long bones in gender identification (Safont et al., 2000). In this study, four different populations were used to test standards deriving from a Roman period population of today's Tarragona (Spain). Among the selected measurements, minimum circumference of the ulna was included. Interestingly, classification results yielded at 91.1% when it was used as single dimension. In cross-validation using populations of different ancestry, correct group membership varied between 75.9 and 81.8%. Naturally, ancient populations were more successfully assigned as compared to contemporary groups.

In a study of a modern forensic sample (80 males, 47 females) from Turkey, the authors measured ulnar and radial length in order to access sex and stature (Celbis and Agritmis, 2007). Discriminant functions were produced using the combination of the two variables, as well as each of them separately. Classification accuracy for both original and cross-validated data rose to 95.7% when only ulnar length was used.

Matzon and associates (2006) studied the morphology of the proximal ulna in a sample of 35 individuals. A three-dimensional system was used in order to digitise nineteen anatomical landmarks. Many distances and angles were measured, and mean differences and standard deviations were presented. Among them four different coronoid height were included, using different definitions. Differences between the sexes were found to be significant between males and females in all cases.

Barrier and L Ábbe (2008) took seven measurements in a sample of 400 South African radii. Classification accuracy reached 89% when all measurements were applied, while minimum midshaft diameter was the best discriminatory variable (83%). Interestingly, the combination of minimum and maximum midshaft diameter gave an 86% correct group membership.

### 3.2.3.3 Radius

Berrizbeitia (1989) analysed a sample 1108 radii (left and right) corresponding to Black and White North Americans from the Terry Collection, in respect to the radial head. According to her results, an individual is assigned as female when the maximum head diameter is equal to or less than 21mm and as a male when maximum head diameter is greater than 24mm. A sample of 50 pairs of radii was used for cross validation, resulting in 92% correct group membership for the left radius, 94% for the right radius, and 96% accuracy when both radii were used jointly.

Among the five measurements taken from the forearm by Holman and Bennett (1991), maximum length and semistyloid breadth (SBB) are included. The SBB of the radius was measured from the most lateral point on the styloid process to the deepest point of the ulnar notch, at a right angle to the long axis of the bone. When only these 2 measurements were used, classification accuracy yielded 72% in males and 92% for females. Radius SBB and Ulna SBB (defined previously) resulted in 82% correct classification, while when radial length was added, correct sex assignment rose to 84%.

Mall and co-workers (2001), in a study of the upper extremity of a contemporary German population, included three radial dimensions (maximum length, maximum head diameter and distal width). The best discriminatory variable for the radius was found to be maximum length (89.1%), followed by maximum head diameter (88.6%), while distal width gave poorer results (78.3%) It is noteworthy that classification results deriving from the discriminant function analysis for the radius (94.9%) provided better results than both humerus and ulna.

In a study by Safont and collaborators (2000) using the circumferences of long bones, radial tuberosity circumference was found to be the second most effective single dimension, with a classification accuracy of 92.8%. In the stepwise functions, minimum circumference of the radius and circumference at the nutrient foramen of the tibia are the variables most frequently selected. This choice demonstrates the high discriminatory value of the radius and tibia in sex determination, for the sample listed below.

In a later study on a Turkish population, radius and ulna length were measured in order to produce standards for sex and stature (Celbis and Agritmis, 2006). The sample consisted of 80 males and 47 females. Radial length gave 90.6% correct group membership, while in combination with ulnar length classification accuracy reached 91.3%.

Sakaue (2004) studied the radius, among other long bones, aiming to assess sex in a contemporary Japanese sample (N=64). Length, sagittal and transverse head diameter, distal maximum breadth, sagittal diameter of ulnar notch and midshaft area were measured for this purpose. Sagittal head diameter and distal breadth were found to be the more effective single dimensions with accuracy reaching the level of 92%. A stepwise procedure results in 95% correct group assignment using a combination of these variables.

Barrier and L Ábbe (2008) took 9 measurements in a sample of 400 South African radii. Classification accuracy reached 88% when all measurements were applied, while minimum midshaft diameter was the best discriminatory variable (86%). Cut-off value was set to 11 mm.

# 3.2.3.4 Femur

The femur is practically the longest and heaviest bone in the human skeleton, surrounded by the largest limb muscle mass. Because of its strength and density, it is frequently recovered in forensic and archaeological contexts (White 1991). Therefore, many studies have been conducted over the years using the femur in archaeological and modern skeletal material (DiBennardo and Taylor, 1982; Taylor and Dibennardo, 1982; Wu, 1989; İşcan and Ding, 1995; Steyn and İşcan, 1997; King et al., 1998; Seidemann et al., 1998; Mall et al., 2000; Safont et al., 2000; Alunni-Perret et al., 2003; Purkait, 2003; Šlaus et al., 2003; Asala et al., 2004; Purkait and Chandra, 2004; Purkait, 2005).

Steyn and İşcan (1997) took six femoral measurements in a cadaver sample of 106 African Whites (Dart collection). A stepwise procedure selected 3 of them (head diameter, transverse and distal breadth) with an accuracy of 88.6%. Distal breadth was found to be the single most discriminatory variable (90.5%), while the combination of femoral and tibial measurements gave 91.4% correct group assessment.

Mall and co-workers (2000) studied the femur in a contemporary German population. Out of six measurements, transverse head diameter classified the specimens most accurately (89.6%). With all measurements subjected to a stepwise procedure, 91.7% of cases were classified correctly by midshaft diameter and head circumference.

Since the integrity of the femoral bone in forensic cases can not be assured, different fragmentary models can be assumed. In that aspect, some authors tested the validity of single femoral variables in sex determination (Seidemann et al. 1998, Safont et al. 2000, Alunni-

Perret et al. 2007) while others created single diaphysis patterns assuming that only one of the distal ends was preserved (Asala 2001, Asala et al. 2004; Purkait 2003, Purkait and Chandra 2004, Purkait 2005).

Seidemann and co-workers (1998) indicated that supero-inferior femoral neck diameter is a reliable sex predictor for American skeletal samples, with percentages rising from 87% (for African-Americans) to 92% (for Caucasians). Safont and partners, on the other hand, achieved poor results testing the efficacy of femoral midshaft (83%) and subtrochanteric (88%) circumference. Most recently, biepicondylar breadth was studied in a contemporary French population, yielding higher classification accuracy (94.4%) than head diameter (Alunni-Perret et al. 2007).

Purkait (2003) studied four dimensions of the femoral case in an Indian population, indicating maximum vertical and horizontal diameter as the best single discriminators and attaining an accuracy of 92.1% each. In a later study (Purkait 2005), the same author introduced an imaginary triangle resulting in 86.4% accuracy combining all three dimensions. Brown and co-workers (2007) tested the previous method in 200 samples from the Terry collection. The sample was further partitioned between African Americans and European Americans. The measurement from the point projecting most medially on the greater trochanter and the highest point on the lesser trochanter, was determined to be valuable in estimating sex using the proximal end of the femur, particularly in combination with the maximum vertical diameter of the head. Using the discriminant function of the combination of these two variables, the accuracy was 90%.

# 3.2.3.5 Tibia

Hanihara (1958; 1981) developed a sex estimation method using length, anteroposterior diameter at midshaft, least circumference and width of proximal end that correctly classified a series of Japanese skeletons with 96% accuracy. Likewise, Singh et al. (1975) employed length, circumference, and both proximal and distal tibia widths in their work, although with only 62-66% accuracy.

İşcan and Miller-Shaivitz (1984a) developed a new technique using the tibia shaft, particularly shaft circumference, to assess sex. Using 159 adult tibiae from the Terry Collection at the Smithsonian Institution, the authors recorded three measurements at the level of the nutrient foramen antero-posterior diameter, transverse diameter, and circumference, as well as maximum length. Discriminant functions derived from these measurements correctly sexed their original sample with up to 78.5% (Whites) and 83.8% (Blacks) accuracy (İşcan and Miller-Shaivitz, 1984a).

Tibial condyles from 100 individuals in the Hamann-Todd collection were studied by Holland (1991). Measurements included biarticular breadth, medial condyle articular width and length, lateral condyle articular width and length. Biarticular breadth proved the most diagnostic dimension with 95% accuracy. Sex was predicted with 86-95% accuracy using regression equations. Using the standards deriving from the original sample, a test sample of 20 individuals also from the Hamann-Todd Collection was classified with 85-100% accuracy. A second sample of 20 prehistoric individual bones was classified with 85-100% agreement. The author remarked that knowledge of race does not significantly enhance the predictive power of the equations, and no clear racial bias with regard to the equations' accuracy is notable.

Kieser and co-workers (1992) investigated sexual allocation of the proximal tibia. Data for the present investigation were derived from the Raymond Dart Collection and consisted of 100 tibiae of Caucasoids (50 males, 50 females) and 102 tibiae of South African Blacks (50 males, 52 females). Five measurements were taken on the proximal end of each tibia as defined by Holland (1991). Biarticular breadth was found to be the most useful, with a percentage correct classification of 94% in Caucasoid males and 92% in males while females presented slightly lower group assessment. High levels of correct classification (84.62 - 92%) were matched by high levels of reallocation.

Bruzek (1995) studied a series of 95 adult tibiae from the collection of the Coimbra Anthropological Museum in Portugal. Eight measurements were selected according to previous studies (İşcan and Miller-Shavitz, 1986; Kieser et al., 1992). When each variable was used singularly, classification varied between 68% and 86%, with the anteroposterior diameter of the lateral articular surface found to be the most effective single dimension. A stepwise procedure selected six variables, reaching accuracy of 88%.

Steyn and İşcan (1997) took seven tibial measurements in a cadaver sample of 106 African Whites of the same collection. A stepwise procedure selected five of them with an accuracy of 90.6%. Distal breadth was found to be the single most discriminatory variable (88.7%), followed by proximal breadth (86.8%).

France (1998) looked at 135 tibiae in Blacks and Whites from the Tennessee data bank. According to her, proximal epiphysis was found more dimorphic with accuracies that reached 95%.

Gonzalez-Reimers and associates (1999) studied 59 complete skeletons housed in the Museo Canario of the city of Las Palmas, belonging to pre-Hispanic individuals from diverse archaeological sites on the island Gran Canaria. The study included seven measurements, of which the first parameter which entered the discriminant function was lateral diameter, and the second one, minimum shaft perimeter. Classification ranged from 94.9% to 98.3% when different fragmentary patterns were assumed.

Sakaue (2004) studied the tibia in a recent Japanese sample, taking 11 dimensions. He found proximal epiphyseal breadth to be the single most effective dimension (94%), followed by transverse diameter of the lateral articular surface (92%) and mid-shaft area (solid cross-sectional area at the middle point of the length) (91%). A stepwise procedure resulted in 97% correct classification.

Slaus and Tomičić (2005) studied 7th century tibial remains collected from several medieval cemeteries in Croatia and the eastern Adriatic coast. Their study was composed of 96 males and 84 females. Following the technique carried out by İşcan and Miller-Shaivitz (1984a), the authors recorded the tibial length and five epiphysis dimensions in order to determine sex from the complete as well as fragmentary bones. They found that sex determination was possible with an accuracy of 93% when all six dimensions were used, and 85.6 to 81.7% for single dimensions in a presumably fragmented condition. The best discriminatory variable was found to be maximum epiphyseal breadth.

#### 3.2.3.6 Fibula

The fibula is one of the least studied of the long bones because of the fact that is rarely recovered intact from crime and archaeological scenes. Nevertheless, some studies dealt with that bone as well in terms of sex identification. Sacragi et al. (1994) studied 106 Japanese fibulas and found 90.6% of accuracy using 5 measurements of the distal end.

### 3.2.4 Other bones

Sex estimation techniques were also developed for several other bones like the scapula (Di Vella et al., 1994; Frutos, 2002; Ozer et al., 2006), clavicle (McCormick et al., 1991; Rogers et al., 2000; Frutos, 2002), hand (Scheuer and Elkington, 1993; Lazenby, 1994; Falsetti, 1995; Smith, 1996; Wilbur, 1998; Stojanowski, 1999; Zanella and Brown, 2003; Barrio et al., 2006; Case and Ross, 2007b) and foot bones (Steele, 1976; Riepert et al., 1996; Introna et al., 1997; Robling and Ubelaker, 1997; Smith, 1997; Bidmos and Asala, 2004; Bidmos and Dayal, 2004), patella (Introna et al., 1998; Bidmos et al., 2005; Dayal and Bidmos, 2005; Kemkes-Grottenthaler, 2005; Gualdi-Russo, 2007; Mahfouz et al., 2007) and ribs (İşcan, 1985; Wiredu et al., 1999). Some of the more important studies are summarized here.

In a contemporary Italian sample, seven scapular parameters were taken and the combination of three (max. distance acromion-coracoid, maximum length of coracoid and length of glenoid cavity) gave 95% correct sexing (Di Vella et al., 1994). Similarly, a study on Anatolian medieval population gave the same classification results with the best

discriminatory dimension being the maximum scapular height (Ozer et al., 2006). Scapula dimensions were also employed in the estimation of stature using linear regressions, with encouraging results (Campobasso et al., 1998).

The patella was quite neglected in sex identification, despite the fact that it articulates with the highly dimorphic femur and tibia (İşcan, 2005). Recently though, it has gained more attention. Introna and co-workers (1998) mention three studies with volumetric and classical metric methods, giving encouraging results. Similar results were achieved in a later study by applying this methodology on a sample from the early medieval period in Germany (Kemkes-Grottenthaler, 2005). O'Connor (1996) demonstrated a statistically significant dimorphism in patellae measurements collected from the Terry Collection and radiographs of college students. His method resulted in 82.5% classification accuracy in females and 78.6% in males. Tatarek and Lease (1996) reanalyzed O'Connor's work and reported an accuracy of 67–80% using a discriminant function from the patella measurements. Another paper by Bidmos and associates (2005) dealt with the problem using six measurements. An overall accuracy of 83% using a linear discriminant analysis was reported. In a sample of South African Blacks, classification results reached 85%, which agrees with the previous studies (Dayal and Bitmos, 2005). There is also a report of a study on a Guatemalan population (Frutos, 2002).

Ribs were first studied for age determination purposes a couple of decades ago, when the rib phase method for age estimation was introduced (İşcan et al., 1984a, b; İşcan, 1985; İşcan and Loth, 1986). Yet ribs were found useful for sex and race assessment as well (İşcan et al., 1987). Several authors discussed the value of ribs as a gender marker and their impact in different populations (İşcan, 1985; Cöloğlu et al., 1998). In this regard, Cöloğlu and associates (1998) attempted to produce a discriminant function using two variables taken from the costochondral end of the rib (maximum superior-inferior height and maximum anteriorposterior breadth) on a modern Turkish sample. An accuracy of 86-90% was achieved when both dimensions were used, yet maximum superior-inferior height was found to be more effective when used singularly.

In a study carried out with a cadaveric sample of British origin, six measurements from five metacarpals and the first proximal phalanx were taken and tested on 20 specimens (Scheuer and Elkington 1993). Results provided an accuracy rate ranging from 74% to 94%, with the first metacarpus demonstrating the highest discriminatory value. In a study testing their formula for the 2<sup>nd</sup> metacarpal, bilateral asymmetry along with the secular trend of declining bone robustness were observed (Lazenby 1994). Falsetti also used dimensions defined in an earlier paper (Scheuer and Elkington 1993) plus anterioposterior and mediolateral midshaft breadths (Falsetti 1995). In the latter study, the Terry collection was used and tested for differences between Americans of European and African descent. A comparison of the  $1^{st}$  digit with the  $3^{rd}$  exhibited different dimorphism between the sexes. Accurate classification ranged from 77% for the  $2^{nd}$  digit and 80% for the  $4^{th}$  to 85% for the  $5^{th}$ .

Not all methods provided high accuracies for sex determination. For instance, in a study by Smith (1996), accuracy ranged from 67% to 82%. Sex differences were also studied in Native Americans using foot and hand bones (Wilbur, 1998). Accuracy was low for many of the metacarpal bones, especially the 3<sup>rd</sup> one (72%). Yet it was higher (87%) for the same bone in Morton (a Mississippian group of central Illinois) and Arikara (a protohistoric group in 18<sup>th</sup> century South Dakota) samples.

Other researchers also took part in the sexual analysis of the hand (Stojanowski et al., 1999), developing 35 functions with the aim of determining the sex of individuals with pathological conditions and preservation problems. Sex accuracy ranged from 75-95% with metacarpal IV providing the highest degree of sexing. Barrio and co-workers (Barrio et al 2006) also investigated metacarpal bones in a contemporary Spanish population and obtained up to 91% classification accuracy, with the highest rate being for the left metacarpus II.

A validation study of some previously mentioned methods (Scheuer and Elkington 1993; Falsetti 1995; Stojanowski 1999) used a small sample (N=23) of recent White American skeletons (Burrows et al 2003). The discouraging results verify the existence of population specific differences in osteometric values.

Questioning the effectiveness of robustness dimensions in identification of sex due to activity-induced changes, a study based on metacarpal lengths was carried out (Case and Ross 2007), obtaining an accuracy of 80%. Interestingly, phalanges were found to be better in discriminating sex than metacarpals.

Tarsal bones have even been proven to be sexually dimorphic, with improved accuracy when combining multiple measurements rather than singular. The calcaneus is a compact bone that is able to withstand high tensile forces. Some of its parameters have been used for sex determination in several populations (Murphy, 2002a; Introna et al., 1997; Bitmos and Asala, 2003, 2004; Riepert et al., 2004; Gualdi-Russo, 2007). The importance of the talus as a gender indicator has also been discussed by some authors (Murphy, 2002b; Gualdi-Russo, 2007).

Murphy (2002a; 2002b) used discriminant function analysis for sex determination of the calcaneus and talus in a prehistoric New Zealand Polynesian population. Five measurements of each bone were taken and the accuracy of sex determination for the discriminant functions derived ranged from 88.4% to 93.5% for the calcaneus and from 85.1% to 93.3%, for the talus. Reduction in error over random assignment by sex did not exceed 87% in both cases.

Measurements of the calcaneus have been shown to be sexually dimorphic in both South African Blacks and Whites (Bitmos and Asala, 2003; 2004). One hundred and sixteen (116) Black and 113 White calcanei were selected from the Dart osteological collection by a simple random sampling technique, and nine measurements were taken. All measured parameters showed significant sexual differences. Individual variables ranged from 64–79% in Blacks and 73-86% for Whites. A stepwise procedure produced better results for both African Blacks (86%) and Whites (91%) (Bitmos and Asala, 2003; 2004). The talus was studied as well in the same population. A total of nine dimensions were measured in a sample of 60 South African Whites and 120 South African Blacks (Bitmos and Dayal, 2003, 2004). In both samples, dimensions were found dimorphic and classification accuracy reached 88% and 89% respectively.

The calcaneus was also studied in North and South Italians (Introna et al., 1997; Gualdi-Russo, 2007). Eight measurements, taken on the right calcaneus of a contemporary Southern Italian skeletal population (N=80), were used to determine sex by multivariate discriminant analysis. Correct sex determination reached 85%. A later study on a modern North Italian sample (N=118) from the Frasseto Collection dealt with nine dimensions of the talus and nine dimensions of the calcaneus (Gualdi-Russo, 2007). The accuracies of sex determination based on the talar measurements were higher. A test of the method showed that is not applicable to South Italian populations.

### 3.3 Virtual Anthropology and Geometrics-Morphometrics

Geometric morphometrics is a relatively novel field of multivariate statistical biometric analysis, which allows the quantification of the shape and size component of morphological variation. These are techniques with great statistical power, offering various choices for visualisation of the results. Virtual Anthropology allows the study of specimens in 3 dimensions through the use of medical imaging techniques, such as CT scans. This approach is ideal for application to the study of fragile and precious archaeological skeletal material, and enables the study of internal features without damaging the specimens in any way. Because of these properties, virtual anthropology methods have become the standard in the reconstruction and study of highly valuable and fragile fossil skeletal material, and, increasingly, also recent skeletal remains. Both 3-D coordinate data, such as those used in geometric morphometric analyses, and standard linear measurements, such as those used in more traditional craniometric analyses, can be obtained non-invasively from CT scan data, as well as additional useful measurements such as volumes and surfaces. Therefore many studies have applied this new methodology in physical and forensic anthropology (Lague and Jungers,

1999; Rosas and Bastir, 2002; Pretorius et al., 2006; 2007a; Franklin et al., 2007b; 2008a; Kimmerle et al., 2008b; Wilson et al., 2008; Franklin et al., 2009).

Lague and Jungers (1999) dealt with the shape of hominoid distal humerus using geometric morphometrics. Although not the principal goal of this study, sexual dimorphism was mentioned in the results. It was found that the sexes of the American Whites and African-Americans showed a mixed pattern of affinities, with the males of each group closer in shape to the females of the other group. Yet these results were not proven feasible in establishing shape criteria for assessment of sex.

Rosas and Bastir (2002) investigated allometry and sexual dimorphism through 2D geometric morphometrics in a modern Portuguese population. Twenty-nine threedimensional (3D) craniofacial and mandibular landmark coordinates were recorded from a sample of 52 adult females and 52 adult males of known age and sex. The landmarks were digitized using a MicroScribe 3DX digitizer and InScribe software for personal computers and then they were transformed into a 2D data set. Once size is eliminated, a series of morphological features are directly related to sex. These features are present both in the skull (subnasal prognathism, nasoglabellar profile, projection of the mastoids, orientation of the occipital clivus, and the differential relationship of the relative proportions of the occipital squama and nuchal area of the occipital bone) and mandible (curvature of the anterior symphysis, development of the pre-angular notch, and flexion of the ramus). The authors stated that no difference in the influence of size on shape between the sexes could be identified.

In a more recent study, Pretorius et al. (2006) test the efficacy of geometric morphometrics in anthropological studies using a sample of African Blacks from the Pretorian collection. Digital photograph of the orbits, the mandibular ramus and the ischiatic notch were obtained. Ten landmarks were assigned to quantify the shape of the orbits and eleven were selected on the mandibular ramus and five on the ischiatic notch. The authors reported preliminary findings that the shapes of the eye orbits are more sexually dimorphic than the commonly used mandibular ramus. As expected, ischiatic notch shape was found to be an effective gender marker.

Kimmerle and associates (2008b) digitalised 3D coordinates of 16 standard craniofacial landmarks from a sample of 112 American Blacks and Whites in order to investigate the implication of size and sex to craniofacial shape in different populations. Standard geometric morphometric techniques were applied and discriminant analysis using the principal components of shape and form was performed. Correct group membership was found to be 77.9% for Blacks and 76.7% for Whites, when using only the shape variables. Yet accuracy increased to 89.7% and 86.7% respectively when CS was included in the

discriminant function. Consequently, size seriously affects shape differences in these populations.

Braga and Trail (2007) studied computed tomography scans of 127 children (54 boys, 73 girls) of mixed origin living in the area of Toulouse (France), ranging in age from a few days to 18 years. Geometric morphometric methods were used to calculate age from centroid size of the face and basicranium, derived from the three-dimensional coordinates of eight anatomical landmarks. A conventional least square linear model was used for this purpose. Results indicated that centroid size of the facial cranium can be used successfully as an age marker even with increasing age.

Franklin and collaborators (2007b) digitised 38 landmarks on subadult mandibles using a Microscribe, attempting to assess sex. Results indicate that population differences are more pronounced than sexual dimorphism in the subadult mandible. However, when interlandmark distances were generated and regressed with age, highly accurate standards were obtained employing ramus height (Franklin and Cardini, 2007). In a similar study on South African adult mandibles, the authors demonstrated that the mandible is highly dimorphic in shape and size, especially in the condyle and ramus in several different populations (Franklin et al., 2008b). As a continuation of the previous study, the authors generated linear distances from the 3-D coordinates of the landmarks and classification accuracy reached 84% (Franklin et al., 2008a).

Wilson and co-workers (2008) employed morphometric methods to assess sex from juvenile ilia (N=25). Six metric criteria were tested and 96% accuracy was accomplished using the shape of ischiatic notch as a discriminating variable for sex. The method showed significant improvement with increasing age for several criteria. A more recent study (Gonzalez et al., 2009) performed a 2D morphometric analysis on the ischiopubic region aiming to develop an accurate sex estimation method. The authors employed discriminant function analysis and k-mean clustering for both shape and form variables concluding that shape variables give better classification results especially in the case of ischiatic notch.

# 3.4 Histological Methods

Quantitative bone histology was introduced by Balthazard and Lebrum (1911 in Robling and Stout, 2000) as a new method to estimate age from bone cross-sections. Since then, histomorphometric methods have been used for the prediction of age in forensic archaeological and paleontological specimens. Different methodologies, reference samples and bone elements were used in this regard with encouraging results.

Several authors investigated the association between age and the prevalence of primary osteons (Kerley, 1965; Ericksen, 1991), while others developed methods to quantify

secondary osteons (Type I) in a specific area or per unit as an age predicting variable (Robling and Stout, 2000). Later studies included the quantification of secondary osteon fragments in their age determination methods, since the fragmentation is positively related to increased age (Thomson, 1979; Stout and Paine, 1992). Some authors (Ericksen, 1991; Yoshino et al., 1994) suggested that osteons type II increase in number with age, while others reported no correlation (Richman et al., 1979). Double zonal osteons have also been considered for age determination but several studies produced contradictory results (Robling and Stout, 2000). Another methodological approach employed the quantification of all secondary osteon types as an age predicting variable (Ahlqvist and Damsten, 1969; Ericksen, 1991; Lynnerup et al., 1998).

Most histomorphometric studies dealt with the femur (Kerley, 1965; Singh and Gunberg, 1970; Kerley and Ubelaker, 1978; Thomson, 1979; Ericksen, 1991; Lynnerup et al., 2006a; Chan et al., 2007). Kerley (1965) and Kerley and Uberlaker (1978) studied cross sections of the midshaft in American Black and White femora. They considered four variables (osteons, osteon fragments and non-Haversian canals, and estimating the percentage of lamellar bone in four selected 100 power fields in the outer third of the cortex) and obtained satisfactory results. Nevertheless, some authors expressed difficulties in microstructure identification (Bouvier and Ubelaker, 1977; Stout and Gehlert, 1980; Walker et al., 1994).

Significant work has been done on other long bones as well. Some of the abovementioned authors repeated their methods on the tibia (Kerley, 1965; Singh and Gunberg, 1970; Kerley and Ubelaker, 1978), ulna (Thomson, 1979), fibula (Kerley, 1965, Kerley and Uberlaker, 1978) and humerus (Thomson, 1979). Others used the clavicle (Stout and Paine, 1992; Stout et al., 1996), ribs (Stout and Paine, 1992; Stout et al., 1994; Crowder and Rosella, 2007), metacarpals (Kimura, 1992), and mandible (Singh and Gunberg, 1970).

Some authors tested the methods for sex differences (Kerley, 1965; Stout and Paine, 1992; Stout et al., 1994; 1996) with negative results. Thomson (1979) seems to be the first author to present sex-specific formulae. Ericksen (1991) found that sex-specific equations perform better as compared to the ones deriving from the pooled sample. She emphasized that females accumulate intact osteons up to the sixth decade of life, while males up to the tenth. Additionally, sex differences in fragmentary osteons are noted. Some studies (Burr et al., 1990) suggest that in females osteons appear to increase in size with advancing age, while others support the exactly opposite (Broulik et al., 1982). This could be attributed to population differences. Other recent studies suggest no sex differences in osteon size (Pfeiffer, 1998). The sex-related variation on bone remodeling seems to be a factor that must be taken into account when creating age predicting formulae using histomorphometric characteristics, but histology has not yet proven feasible for separating the sexes.

# Chapter 4: Applications of radiology in forensic medicine

Forensic radiology is a sub-specialization of forensic medicine, defined as the discipline that "utilizes the interpretation of medical radiological examinations to answer legal questions" (Walsh et al., 2004). The importance of radiographic methods has been long acknowledged in medico-legal practice (Knight, 1984; Evans and Knight, 1986; Krogman and İşcan, 1986; Kahana and Hiss, 1997; Kahana et al., 1997; Brogdon, 1998; Kahana and Hiss, 1999; Brogdon, 2006). Nowadays it includes both clinical and post-mortem radiology. Despite the fact that the most frequent application surveys positive identification, it has been widely used in biological profiling of the deceased, determination of cause and manner of death, medical negligence, non-accidental trauma and smuggling (Kahana and Hiss, 1997, 1999; Brogdon, 2006).

The benefits of a non-invasive technique are obvious in many cases in which postmortem examination is imperative. This chapter focuses on the various applications of forensic radiology in everyday medico-legal practices, with implications for clinical forensic medicine.

### 4.1. Historical cases

Walsh and associates (2004), in an extended review article, date the first use of forensic radiology to the attempt murder case of Elizabeth Ann Hattley, in 1897, in England. Even though Mrs. Hattley took 4 bullets in the head by her husband, she did not die from the attack, hence the local general practitioner ordered an X-ray to see if the bullets could be located (Brogdon and Lichtenstein, 1998; Walsh et al., 2004). However, Brogdon and Lichtenstein (1998) report an earlier application in Montreal (1895). It concerned the shooting in the leg of T. Cunning by G. Holder. Any effort to locate the bullet by probing proved unsuccessful, the wound healed but it remained symptomatic. A radiograph was requested by Cunning's surgeon and the flattened bullet was located between the tibia and the fibula. The radiograph was brought to court as evidence for attempted murder and Holder was convicted to 14 years in the penitentiary. The first case of malpractice in which radiographs were accepted in court as evidence was the case of Smith against Grant, a well-known surgeon of the time, who was accused of misdiagnosing a fracture on the femoral head of the patient, resulting in limb shortening and disability. Judge Lefevre, after a long consideration of the nature of the evidence decided to accept radiographs in court (Brogdon and Lichtenstein, 1998). The first criminal case in the U.S. involving X-rays was the murder trial of Haymen (1897) in Watertown, N.Y. (Brogdon and Lichtenstein, 1998). The victim was shot in the jaw,

but the examination discovered a second foreign object in the back of the head. The question that emerged was whether this was a second bullet or a fragment of the first. Dr Cannon was called into court as an "expert witness" to testify on the radiograph and he excluded the second bullet theory. His testimony was accepted by Judge Wright. These first cases opened the field of forensic radiology, and the acceptance of radiographic evidence and expert witness testimony slowly began.

Other potential applications of X-rays have emerged at an experimental level since the very beginning. Some examples are discussed by Brogdon and Lichtenstein (1998) in a detailed review of the history of the field: In 1897, T. Bordas published an article in the "Annales d'Hygines Publiques et de Medecine Legale", suggesting the potential use of X-rays in the identification of explosive devices in suspicious packages; the Bureau de Douanes (1897) used a fluoroscope to examine luggage at the Pavillon de Rohan and the Gare de Nord; Bertillon's anthrometric method for positive identification was supplemented by frontal and lateral radiographs; Levinsohn suggested that direct measurements of the skeleton through radiographs and Beclere added nail configuration. The examination of mummies by means of X-rays was introduced in 1897 as an adequate non-invasive technique to assess age, sex, pathology and trauma.

### 4.2. Scope of forensic radiology

As Brogdon and Lichtenstein (1998) wisely concluded: "Professor Röntgen furnished the tool. His contemporaries showed us how to use it. Realization of the full scope of forensic radiology was to depend on the imagination and the industry of new scientists, and the indulgence or approval of the courts."

### 4.2.1 Detection of foreign objects

A post-mortem radiological examination allows the detection of metallic foreign bodies like bullets or bullet fragments in the body (Brogdon, 1998; Kahana and Hiss, 1999; Brogdon, 2006; Stein and Grünberg, 2009). This is of particular value in cases of highly decomposed bodies, where the necropsy gives limited information on the circumstances of death. Such evidence is necessary for the reconstruction of the incident as well as for a court testimony. Thus it is used as a routine examination after necropsy in order to record the location of the bullet or the bullet fragments, the bullet track, the type of bullet (high or low velocity) and its position relative to the possible entrance wound, since not rarely it can migrate and be recovered further from its original apposition due to movement of the body after the incident or during its transportation to the forensic lab.(Kahana and Hiss, 1999; Brogdon, 2006) Bullet type especially is easily recognizable by the radiographic pattern; for example, high velocity hunting rifles create a characteristic image of "snowstorm" after striking a bone (Brogdon, 2006). There are reports of recovering old bullets from previous shooting. In that case it is not feasible to distinguish the old bullet from the new one. However a CT may allow the observation of a dense layer of surrounding scar tissue (Brogdon, 2006).

Radiography can be extremely helpful in the identification and retrieval of bomb fragments when there are individuals in the vicinity of the explosion and the fragments are often found embedded in their tissue (Walsh et al., 2004). Pieces of glass or plastic deriving from traffic accidents or mass disasters cannot be easily detected with X-rays. Other foreign bodies, such as opaque poisons, can be seen in the stomach. Drugs in plastic bags carried by smugglers are detectable in radiographs.

#### 4.2.2. Mass disasters

Radiography has been proven very useful in mass disasters like airplane crashes or bomb explosions where there is no information of whether there are individuals involved (Brogdon, 1998; Walsh et al., 2004). Radiographic means allow the identification of human remains, mainly bones, which can give indication of the number of victims and additionally separate human from animal bones, in cases where visual examination is not possible. In some cases of mass fatalities such as explosions, the recovered remains are completely dismembered and it is very difficult to gather and match the different parts. Kahana and Hiss (1999) report the creation of an identification team specialized in the recovery of human body parts in suicide bombings in Israel. The identification process is mainly based on radiographic methods.

Murphy and associates (Murphy et al., 1980) suggest that in some cases of mass fatalities, radiography can be three times more effective than DNA and five more effective than dental records. The superiority of radiography versus DNA in mass disasters has also been supported by others (Binda, 1999). Walsh and co-workers (2004) cited Nye for his work on the Oklahoma City bombing. According to the Oklahoma City protocol, radiography was set as the optimal method for the investigation because of the considerable quantities of lead found in the bodies (The Murrah Federal Building construction was based on a great amount of lead glass).

Radiology is used as well for survivors of a mass disaster, to identify the exact extent of their injuries (Lichtenstein, 1998; Walsh et al., 2004). The mechanism, the extent of the injury and its relation to the fatal environment are to be defined in order to accurately define the circumstances of the incident. There is a special need to identify those in control of the environment, as for instance pilots in plane crash or terrorists in a fatal attack. Patterns of injuries of the hands or the feet of an aircraft crew can give information on their status and control as well as their reactions and movements during the event (Lichtenstein, 1998).

### 4.2.3. Charred bodies

In many forensic cases, the recovered bodies are burned as a result of a fire (domestic or traffic accident) or an explosion resulting in fire (work accident or bombing). A radiographic examination of charred remains that cannot be identified otherwise may reveal the existence of humans or animals. Often immolation is incomplete and the remaining skeleton or even the bony fragments can determine if the victim was juvenile or adult. The presence of epiphyseal fusion in a complete bone will indicate maturity. Moreover, the identification of small bone fragments with fused ends points to a small adult animal rather than a human infant (İşcan and Loth, 1997).

In some cases the fire constitutes an attempt to destroy the evidence and the body of a murder victim. Bogdon (2006) describes the case of a woman burned beyond recognition in a domestic fire. The remains were radiographed for comparison with ante-mortem thoracic X-rays of the occupant. The post-mortem study led to a positive identification of the body. Additionally, it revealed coils of wire ligature around the neck, indicating a homicide covered by fire.

Other cases may involve victims of gunshots or blunt force trauma, set on fire to eliminate the evidence. Post-mortem radiography can identify the existence of a bullet or a fatal skull fracture, setting a murder investigation in motion.

## 4.2.4 Positive identification

### 4.2.4.1 Comparison with ante-mortem records

Cases of highly decomposed, mutilated, incinerated or skeletonized bodies are impossible to identify by conventional means such as facial features, fingerprints, birthmarks or scars (Brogdon, 2006). Positive identification can be accomplished with the comparison of ante-mortem and post-mortem radiographs. Dental records (Pretty and Sweet, 2001; Pretty, 2007), former fractures, surgical work (Hogge et al., 1995; Dean et al., 2005; Simpson et al., 2007; Šlaus et al., 2007), calcification of tissues or stones or bladder-stones are mentioned in the literature (Murphy et al., 1980; Kahana and Hiss, 2002; Brogdon, 2006). Although every single part of the body has been used for positive identification, scholars suggest that the most popular are the radiographs of the teeth, skull, chest and abdomen (Murphy et al., 1980). Several bones have been used for identification purposes including the skull (Campobasso et al., 2007), vertebrae (Mundorff et al., 2006; Valenzuela, 1997) and hand bones (Koot et al., 2005). Cranial radiographs can provide information on anatomical features, pathology, previous trauma and surgical operations (Kahana et al., 1997; Sudimack et al., 2002). Scholars report numerous cases of positive identification using dental records (Nicopoulou-Karayianni et al., 2007), and different cranial features such as frontal sinuses (Marlin et al., 1991; Quatrehomme et al., 1996; Kirk et al., 2002; Wood, 2006; Tang et al., 2008), anatomical structures (Messmer and Fierro, 1986; Jablonski and Shum, 1989; Rhine and Sperry, 1991) and trabecular architecture (Kahana and Hiss, 1994; Kahana et al., 1998).

The comparison of post-mortem and ante-mortem radiographs has proven to be the most adequate in cases of mass disasters where faces are disfigured and fingerprints are not available, while DNA examination is a more expensive and time-consuming method. However, the application of this method requires the existence of efficient ante-mortem documentation of the deceased, which is not always the case, especially in countries with low quality health care systems (Brogdon, 2006).

# 4.2.4.2 Biological profiling

# Age estimation

Classical radiographic techniques can be helpful in assessing biological features from different bones of the human skeleton. Age estimation using dental radiographs is reported extensively in the literature (Kvaal et al., 1995; Brogdon, 1998; Maber et al., 2006; Thevissen et al., 2009a; Thevissen et al., 2009b). Before obstetrical ultrasonography, radiology was the only method to establish fetal maturity based on the appearance of distal femoral and proximal tibia epiphyses (Brogdon, 1998). Furthermore, the closure of the epiphyses in radiographs appears up to six months before it can be observed in the dry bones (Paterson, 1929), which can be very helpful in cases of age estimation in juvenile individuals. The last epiphysis to close is the medial end of the clavicle during the third decade of life. Many scholars have employed the mineralization of costal cartilages on the "chest plate" for age estimation (McCormick and Stewart, 1988; Barrès, 1989). Degenerative changes of the skeleton point to elder individuals.

#### Stature estimation

Stature can be approximated by measuring the length of long bones, especially of the lower limbs. The same measurements can be taken in radiographs. Some recent studies on stature estimation relied on measurements taken on radiographs of upper limb bones (Zhou et al., 2007), tibia and fibula (Fan et al., 2008). Muñoz and co-workers (2001) found that the most valuable long bone for stature estimation in their radiographic study was the femur. Sağir (2006) developed a stature estimation method based on radiographs of metacarpals. Zhang and collaborators (2008) suggested a new stature estimation technique using measurements on the cervical vertebrae taken on X-ray films from CT scans.
# Sex estimation

Radiography can be quite successful in sex identification, apart from its acknowledged value on positive identification. Brogdon (1998) gives an example of what he calls "the absolute roentgenographic indicator of sex" in one of the victims of the Air India crash (Flight 182, July, 2000). Many of the recovered victims had viscera displaced into the thoracic cavity, resulting in the accidental discovery of an 18-22 week fetus in the chest radiograph of a young female.

Sex can be identified by the shape and the size of the pelvis, the cranial features and the size of the long bones (Brogdon, 1998; Bass, 2006 in Spitz). It has long been established that there is a distinct difference in sex patterns in costal cartilage calcification (McCormick and Stewart, 1988). Calcification of tracheobronchial cartilage occurs rarely but it exhibits a female predominance, while thyroid cartilage ossification occurs more often in males (Brogdon, 1998). For human remains, where some soft tissue is retained, a radiograph of the chest plate can provide a useful method of sexing (McCormick and Stewart, 1983; McCormick et al., 1985; Pao and Pai, 1988; Rejtarová et al., 2004). Other investigators have used chest plate radiographs from which they measured dimensions of the sternum and ribs (Torwald and Hoppa, 2005). Riepert and associates (1996) studied sexual dimorphism in radiographs of the calcaneus, achieving 80% correct group membership. Patil and Mody (Patil and Mody, 2005) accomplished sex identification from lateral cephalograms with an accuracy of 99%. Abdel Moneim and collaborators (2008) developed a sex estimation method based on patella and foot measurements on radiographs.

## 4.2.5. Physical abuse

#### 4.2.5.1 Child abuse

Another application of radiographic methods is the examination of children with the suspicion of child abuse, since fractures happen in more that 50% of the cases (Loder and Bookout, 1991). According to Brown (1995 in Kahana and Hiss, 1999) more than 80% of the child-abuse injuries identified in US are detected by means of medical imaging. Evidence from the literature notes the effectiveness of the method in investigations where assault injuries are detected in children under three years old that can not give information on the history of the incident (Kemp et al., 2006; Kemp et al., 2008). The repeatability of the abuse is reflected in multiple fractures of different age, while information on the distribution of the fractures is also significant for any expert testimony on potential abuse. Scholars mention skeletal surveys and bone scintigraphy as adequate methods for investigating occult fractures on children, while a recent review on the subject suggests that both methods have a tendency to miss occult fractures when used alone (Kemp et al., 2006). According to another review

(Kahana and Hiss, 1999), scintigraphy is considered highly sensitive in the detection of rib, spinal and diaphyseal fractures, while it exhibits low sensitivity on cranial injuries. For that type of injuries, forensic professionals suggest various types of image techniques including CT and MRI. Subdural injuries deriving from the violent shaking of the child (shaken baby syndrome or whiplash shaken syndrome) are better detected with MRI, subarachnoid haemorrhages with CT (Kahana and Hiss, 1999). An increasing number of pediatric hospitals are using computer radiography or direct digital radiography, but no study so far has evaluated their effectiveness in detecting occult fractures (Kemp et al., 2006). Nevertheless, Kleinman et al. (2002) found digital techniques to be comparable with the conventional ones, which were also suggested in earlier studies (Langen et al., 1993).

## 4.2.5.2 Partner abuse

Domestic violence mostly involves women in marriage or cohabitation to a greater extent than battered children. The face, the neck and the hands are considered common targets in battered women with high incidence of mandibular body-angle and ramus fractures. Sometimes ribs can be fractured laterally. In cases of prenatal child abuse, the breast and the abdomen are likely to be struck (Brogdon, 2006). Women often delay going to the hospital and they hardly ever report the incidents to the police. External injuries are healed and sometimes only radiographs are available when the medical doctor or the victim decides to call the police. Radiography provides evidence that can be taken to court in both fatal and non-fatal cases.

## 4.2.5.3 Abuse of the elderly

Similarly to the battered children and women, the elderly can suffer abuse by family members or the medical staff of nursing homes. The traumatic lesions expected in such cases are maxillofacial injuries, intracranial damage, defensive injuries such as fending fracture of the hands or forearms and trauma due to squeezing and physical restraint (Brogdon, 2006). As a general rule, the radiographic identification of battered elder individuals is difficult due to pathological conditions such as osteopenia or osteoporosis that make the bones extremely fragile and sensitive. Moreover, the elderly individuals hardly ever report the abuse, fearing a repeated and more hostile attack.

## 4.2.6. Age estimation of the living

Another aspect of forensic radiology is the estimation of the age of a living individual who claims to be younger (ex. individuals facing criminal, civil or asylum procedures) or older (ex. individuals claiming pensions) than his or her real age (Braga and Treil, 2007; Schmeling et al., 2007; 2008). Additionally to the physical examination and dental status performed in these cases, radiographic techniques are employed in order to correctly estimate age in young

adults claiming to be underaged. Brogdon (1998), reports as an example the murder case of J. Adamson by an African native. The perpetrator was released from the death penalty by hanging due to the court's decision, against the radiologist's testimony, that he was underaged when he committed the crime.

In Germany, X-ray examinations for age estimation are only provided for in criminal law and subject to the legal order of a magistrate under Section 81 of the Code of Criminal Procedure (StPO) (Schmeling et al., 2007). An X-ray of the left hand is recommended as well in the estimation of age in individuals under 18 (Schmidt et al., 2008). When the question of whether a person has reached the age of 21 arises and the hand bones are fully developed, the ossification of the medial clavicular epiphyses is considered (Schmeling et al., 2004; 2007; Schulz et al., 2008).

## 4.2.7. Securing evidence

The importance of radiology lies not only in the detection of foreign objects before autopsy or the recognition of human body parts in mass disasters, but it also constitutes a very important tool in securing evidence that can be used in the future whenever this is necessary. The recording of the exact position of a foreign object, a bone fracture or a traumatic lesion can be permanent evidence for court even years after the incident, when the body or the bones are no longer available. This potential usefulness of radiology is, as correctly stated by Stein and Grünberg (2009), in its infancy in many parts of the developed world.

## 4.2.8. Alternative modalities in forensic radiology

An increasing number of new imaging techniques are been introduced in the forensic sciences. Wheatley (2005) studied the proximal femur by means of dual energy X-ray absorb geometry in a sample of 31 individuals from Alabama. He measured the minimum femoral head diameter and diameter directly below the lesser trochanter and also the bone mineral density in the neck and lesser trochanter. This data was also quantified with Ward's triangle using X-ray absorb geometry. 94% correct group membership was obtained using all the bone mineral densities and femoral neck diameter for sex determination. Despite the encouraging results, the equipment is not commonly available in forensic anthropology laboratories, thus making the application of the method infeasible.

Computer tomography has also been used in a forensic context. Harma and Karakas (2007) predicted sex with 84.6% accuracy by using CT scans of femora deriving from hospital patients. Mahfouz and associates (2007) studied sexual dimorphism in the patella using high resolution CT. 228 patellas were CT-scanned and the data were segmented, a set of geometric

features was automatically extracted, normalized and ranked. A feature vector of dimension 45 for each subject was then constructed, in order to be used by a neural network to classify the sex. The authors also tested different classification methods and concluded in favor of linear discriminant classification (90.3%). When neural network was applied to the full 45 features, an overall accuracy of 93.5 was accomplished. Furthermore, multislice computed tomography (MLCT) has been reported in positive identification of charred bodies (Thali et al., 2002; Dedouit et al., 2007) and mass fatalities (Sidler et al., 2007).

Magnetic Resonance Imaging (MRI) is routinely used for the detection of nontraumatic cranial injuries like subdural hematomas, concussive and shear injuries (Kahana and Hiss, 1999). Lately it has been applied to survivors of attempted manual and ligature strangulation or forearm chokeholds (Yen et al., 2007). Victims were brought to forensic experts in order to evaluate to what extent incidents were life-threatening and to provide court evidence. First they were submitted to classical forensic examination and secondly to MRI. Findings of MRI included hemorrhaging in the subcutaneous fatty tissue of the neck (10 cases), hemorrhages of the neck and larynx muscles, the lymph nodes, the pharynx, and larynx soft tissues. Based on the classical forensic strangulation findings with MRI, eight of the cases were declared life-endangering incidents. It is noteworthy that in four of the cases signs of impaired brain function due to hypoxia were identified without any petechial hemorrhage.

# Chapter 5: Aim of the study

The introduction of sophisticated imaging tools in forensic investigation has been widely discussed in the previous chapter, with encouraging results mainly in the aspects of positive identification, recovery of evidence of the body and diagnosis of trauma. Yet the establishment of biological features is mainly based on classical osteometric methods, with some exceptions of conventional radiographic applications.

The employment of digital radiography as a routine examination in forensic cases provides many advantages compared to classical radiographic techniques and computed tomography. Radiographs can be easily taken after external inspection and stored in a computer for further examination. Moreover, digital X-ray machines are part of the standard equipment of the forensic laboratory, which makes their use for sex identification easy, rapid and non-costly.

The recovery of fragmentary skeletal remains in forensic investigations requires easy and rapid techniques for biological profiling and reconstruction of scene history. The use of radiographs instead of the actual bones allows the identification of semi-decomposed bodies without the need of special preparation (ex. maceration), thus facilitating the whole forensic investigation.

The current study aspires to accomplish a threefold purpose:

- to develop a sex determination technique using digital radiographs of long bones applicable in cases of commingled, charred and fragmented remains as in mass disasters or criminal cases,
- 2. to provide cranial and postcranial osteometric data on a contemporary population from Crete, Greece, that has not been represented so far in the existing databases, and
- to introduce the discipline of forensic anthropology as an integral part of modern multidisciplinary medico-legal investigation in Greece.

# Part B

# Chapter 6: Materials and methods

## 6.1 History of Crete

The purpose of the study is to develop a sex determination technique using osteometric data from remains exhumed from two contemporary Cretan cemeteries in Heraklion, Crete. The population of Crete is thought to have a complicated political history with many different civilizations ruling its ever-changing people. The island, surrounded by myths and legends, has an intriguing history in both ancient and modern times. After the collapse of the ancient Minoan civilization (1400 B.C.), its administration was taken over by different civilizations (Hood and Smyth, 1981; Evans, 1909). These include Myceneans coming from the mainland (1400-1100 B.C.) (D'a Desborough, 1964; Hallager, 1977), followed by Dorians, also from the mainland (1100-67 B.C.) (Willets, 1974). The Romans ruled from 67 B.C. until their decline at the end of the 4th century A.D. Crete was then incorporated into the Byzantine Empire and was ruled from Constantinople (today's Istanbul in Turkey) (Glykatzi – Ahrweiller, 1961).

The rule of the island was taken over by Arabs in 824 A.D., who built their capital city El Khandak (today's Heraklion) in order to prevent invasion by Byzantines. Byzantines retook the island in 961 A.D., creating a second Byzantine period. The capture of Constantinople by the Fourth Crusade resulted in the division of the empire. Crete was given to Boniface of Monferat, who later sold the island to Venice in 1204. Venetian rule lasted until 1669, when Turks replaced them and Crete became part of the Ottoman Empire (Miles, 1964; Thiriet, 1977; Murphey, 1993). In 1898, the island was taken away from the Turkish Empire to be ruled by an international administration. While the administration of the island changed, its population remained relatively intact until 1923, when the island was officially transferred to Greece and its Turkish people were relocated to Turkey under a general population exchange agreement (Buckley, 1977). The last stage of Cretan population history was the control of the island by Germans from 1941 until 1945. At the end of World War II the island was given to Greece.

Based on this brief history, it seems implausible to state that the native Cretan islanders remained relatively uninfluenced by the populations of forces that ruled and administered the island. However according to Tomadakis (in Detorakis, 1990) there was no significant ethnic alteration of the Cretans despite the many populations that ruled the island. The Greek language was never extinct and the cultural heritance remains alive up today. Limited alterations are recorder in urban areas while the rural population has remained homogenous. Therefore the Cretan population is considered homogenous in the current study.

## 6.2 Burial habits in Greece

The availibility of skeletal material representing modern Eurepeans to carry out population based studies is very limited. The situation is different in Greece, where the remains are also buried and then removed after a few years (3 years or so). In some of the cases the exhumation process is delayed because of factors such as embalming, or enviromental conditions that delay soft tissue decomposition. The exhumed bones are gathered, cleaned (in the vast majority) and placed in wooden or metal boxes which are stored in a special room (ossuary) all together or in family tombs if they exist. According to an old religious tradition, when the bones are exhumed, close relatives of the deceased clean the bones carefully with wine and wrap them with a white sheet as a last act to honor their beloved one. Unless living members of a deceased person can afford to keep them in the tomb with a "rental" fee, to be destroyed (Eliopoulos et al., 2006). Bones are gathered and emptied in a large underground pit, usually located in the back of the cemetery, where they are cremated.

## 6.3 Permissions and limitations

For any osteological study of skeletons kept in cemeteries in Greece, a standard procedure is required. According to Greek law, a permit can be given only by a district attorney. For the skeletons that are stored in the ossuary for a fee, permission of the family members is also required. For those that are to be destroyed, the D.A.'s permit is sufficient to carry out the study. To move the skeletons from the cemeteries to the osteological lab, an additional permit from the health service is necessary. The health department requires a full "decontamination" of the skeletons before they are moved from the cemeteries. Osteometric studies also require maceration, because some soft tissue may still be present in a number of individuals. This procedure takes place in the facilities where the collection is stored.

Usually the skeletal material to be destroyed consists of individuals that lived in the previous century and demographic information is not always available. Sometimes, several individuals are comingled in the same box and this material cannot be used for any study. The sex of the individuals can be inferred from the names written on the boxes that contained the remains. Age and cause of death are not available in the cemetery archives, but they can be obtained from the City Hall census archives. The link between the cemetery archives and the City Hall census archives is a reference number written on the boxes, from which one can find the date of exhumation. Then one must calculate about 3-5 years before that date to find the date of burial in which the birth date is mentioned, and after that the demographic information is obtained from the census office of the city where the individual was buried or

the city where he or she was born. Inconsistencies in the archives sometimes create problems in achieving the correct demographic information.

Such a procedure was followed in the past for the creation of the Heraklion osteological collection (Cretan collection). Skeletal material of 200 individuals was obtained after permission from the D.A was given to the Department of Forensic Sciences, University of Crete, in order to be used in anthropological studies.



Fig 6.1 Sex and age distribution (by age groups)

# 6.4 Study population

The skeletal material was selected among the bones (N=200) of both the Cemetery of St Konstantinos and the Cemetery of Pateles, Heraclion, Crete. The study population consists mostly of Cretans or individuals that lived in Crete for more than three generations, who lived and died between the end of the 19th century and the beginning of the 20th. A number of people who may have migrated from Turkey, other islands and mainland Greece are excluded from the study. All individuals with obvious bone pathology are also removed from the sample. Age and cause of death are obtained from the Heraklion City Hall census archives for only part of the skeletal material while sex is obvious from the names written on the boxes that contained the remains. Mean age for males is 68, 57 +/- 13.52 (N=61) and for females 72, 98 +/-16, 90 (N=58). Sex and age distribution (by age groups) is illustrated in Fig 6.1. The skeletons were chosen according to the following criteria:

- Good preservation of the majority of the skeletal elements with emphasis on the skull and long bones.
- > Representation of all age groups for both sexes (to the extent possible).
- Minimum existence of trauma or visible pathological alterations.
- Minimum existence of soft tissue.
- Confirmed Cretan origin.

# 6.5 Methodology

# 6.5.1.Osteometry

# 6.5.1.1 Osteometric equipment

- 1. Sliding caliper
- 2. Spreading caliper
- 3. Anthropometric inelastic measuring tape.
- 4. Osteometric board.

# 6.5.1.2 Measurements

# Cranial skeleton

Biometric definition of cranial landmarks and measurements

There are several landmarks from which measurements can be taken. Biometric landmarks include (De Villiers 1968, Knussman, 1988, Moore-Jansen et al. 1994, Buikstra and Ubelaker, 1994):

- 1. *Alare (al)*: Instrumentally determined as the most lateral points on the nasal aperture in a transverse plane.
- Basion (b): The midpoint of the anterior margin of the foramen magnum, most distant from the bregma.
- 3. Bregma (br): The intersection of the coronal and sagittal sutures, in the midline.
- 4. **Dacryon** (*da*): The point on the medial wall of the orbit, at the junction of the lacrimomaxillary suture and the frontal bone.
- 5. Euryon *(eu)*: The two points on the opposite sides of the skull that form the termini of the lines of greatest breadth, i.e., the most widely separated points on the two sides of the skull.
- 6. Frontotemporale temporale (fmt): The most laterally positioned point on the fronto-malar (fronto-zygomatic) suture.
- Frontotemporale (*ft*): The most medial point on the curve of the temporal ridge. These points lie on the frontal bones just above the zygomaticofrontal suture.

- 8. **Glabella** (g): The most forward projecting point in the midline of the forehead at the level of the supra-orbital ridges and above the nasofrontal suture.
- 9. Gnathion (gn): The lowest median point on the lower border of the mandible.
- 10. Gonion (go): The most lateral point on the posterior inferior angle of the mandible.
- 11. Lambda (1): The intersection of the sagittal and lambdoid sutures, in the midline.
- 12. Nariale (na): The lowest point on the inferior edge of the nasal opening on either side of the nasal spine.
- 13. Nasion (n): Intersection of the nasofrontal suture with the midsagittal plane.
- 14. Opisthion (ops): The midpoint of the posterior margin of the foramen magnum.
- 15. **Opisthocranion** *(op)*: The most posterior point on the skull (not the same as the external occipital protuberance). It is the point furthest from the glabella.
- 16. Orbitale (or): The lowest point on the lower margin of the orbit.
- 17. Porion (po): The most superior point on the margin of the external auditory meatus.
- 18. Prosthion (pr): The most anterior point in the midline on the upper alveolar processes.
- 19. Zygion (zy): The most lateral point on the zygomatic arch.

Cranial measurements (see <u>Table 6.1</u> for abbreviations and instruments used):

- 1. Maximum cranial length (CL): Greatest length in median sagittal plane from glabella to opisthocranion (g-op). *Instrument*: spreading caliper.
- 2. Basion-Bregma Height (B-Br): Distance from basion to bregma.
- 3. Maximum Vault Breadth or Crania Breadth (CB): Greatest biparietal breadth taken at right angles to the mid-sagittal plan. Distance from euryon to euryon (eu-eu). *Instrument*: spreading caliper.
- 4. Maximum Frontal Breadth (MaxFrB): Distance between the two external points on the frontomalar suture (fmt). *Instrument*: spreading caliper.
- 5. **Minimum Frontal Breadth (MinFrB):** From frontotemporale to frontotemporale (ft-ft). *Instrument*: sliding caliper.
- 6. Foramen Magnum Length: From opisthion to basion. Instrument: sliding caliper.
- 7. Foramen Magnum Breadth: Distance perpendicular to length of foramen magnum. *Instrument*: sliding caliper.
- 8. Bizygomatic Breadth: From zygion to zygion (zy-zy). Instrument: sliding caliper.
- Mastoid Height: The height of the mastoid process from its tip to the Frankfort plane. The measurement is perpendicular to the Frankfort plane. *Instrument*: sliding caliper.

- 10. Basion-Nasion Length (Ba-N): Distance from basion to nasion. *Instrument*: spreading caliper.
- 11. **Basion-Prosthion Length (Ba-Pr):** Distance from basion to prosthion. *Instrument:* spreading caliper.

| Measurement             | Abbreviation | Instrument        |
|-------------------------|--------------|-------------------|
| Cranial length          | CL           | Spreading caliper |
| Basion-bregma           | Ba-br        | Sliding caliper   |
| Nasion-bregma           | Na-br        | Sliding caliper   |
| Bregma-lambda           | Br-la        | Sliding caliper   |
| Lambda-opisthion        | La-op        | Sliding caliper   |
| Cranial breadth         | СВ           | Sliding caliper   |
| Max frontal breadth     | MaxFrB       | Spreading caliper |
| Min frontal breadth     | MinFrB       | Spreading caliper |
| Bizygomatic breadth     | BizyB        | Sliding caliper   |
| Foramen magnum length   | FML          | Sliding caliper   |
| Foramen magnum breadth  | FMB          | Sliding caliper   |
| Mastoid height          | МаН          | Sliding caliper   |
| Basion-nasion length    | Ba-na        | Sliding caliper   |
| Basion-prosthion length | Ba-pr        | Sliding caliper   |
| Nasion-prosthion height | Na-pr        | Sliding caliper   |
| External palatal length | ExtPL        | Sliding caliper   |
| Exernal palatal breadth | ExtPB        | Sliding caliper   |
| Biorbital breadth       | BiB          | Sliding caliper   |
| Interorbital breadth    | IntB         | Sliding caliper   |
| Nose breadth            | NB           | Sliding caliper   |
| Nose height             | NH           | Sliding caliper   |

Table 6.1: Measurements of the cranium, abbreviations and instruments.

- 12. Nasion-prosthion Length (N-Pr): Distance from nasion to prosthion. *Instrument*: sliding caliper.
- 13. Interorbital Breadth (IntB): From dacryon to dacryon. (da-da). Instrument: sliding caliper.
- Biorbital Breadth (BB): (ec-ec): Direct distance between right and left ectoconchion (ec). *Instrument*: sliding caliper.
- 15. Nose Height (NH): From nasion to nariale (n-na). Instrument: sliding caliper.
- 16. Nose Breadth (NB): The maximum breadth of the nasal aperture (al-al). *Instrument*: sliding caliper.

Postcranial measurements (see Table 6.2 for abbreviations and instruments used)

## Humerus

The following measurements that are easy to assess in skeletonized bodies were taken:

- 1. Maximum Length (HL): Direct distance from the most superior point on the head of the humerus to the most inferior point on the trochlea. Humerus shaft should be positioned parallel to the long axis of the osteometric board. *Instrument*: osteometric board.
- 2. Vertical Head Diameter (HVD): Direct distance between the most superior and inferior points on the border of the articular surface. *Instrument*: sliding caliper.
- 3. Maximum Midshaft Diameter (HMaxMid): Maximum diameter at midshaft. Instrument: sliding caliper.
- 4. Minimum Midshaft Diameter (HMinMid): Minimum diameter of midshaft. Instrument: sliding caliper. Instrument: tape.
- 5. Midshaft Circumference (HmidCirc): Circumference measured at the level of the midshaft.
- 6. Epicondylar width (HEW): Distance of the most laterally protruding point on the lateral epicondyle from the corresponding projection of the medial epicondyle. *Instrument*: osteometric board.

## Ulna

- 1. Maximum Length (UL): Distance from the most superior point on the olecranon to the most inferior point on the styloid process. *Instrument*: osteometric board.
- 2. Coronoid Height (UCH): Seen in profile the upper part of the posterior border of the ulna is a straight line. The perpendicular distance of the tip of the coronoid process is measured. *Instrument*: sliding caliper.
- 3. Distal epiphyseal breadth (HDB): The distance between the radial articular surface and the medial surface slightly proximal to the styloid process. *Instrument*: sliding caliper.

## Radius

- 1. Maximum Length (RL): The distance from the most proximally positioned point on the head of radius to the tip of the styloid process without regard for the long axis of the bone. *Instrument*: osteometric board.
- 2. Maximum Head Diameter (HVD): The maximum diameter of the femur head, wherever it occurs. *Instrument*: sliding caliper.
- 3. Distal Breadth (RDB): The limbs of the small sliding caliper are held parallel to the long axis of the of the shaft so one is trangential to the lower end of the lateral

boarder and the other passes through the middle of the distal articular facet for the ulna or the medial surface of the bone immediately above it. *Instrument*: sliding caliper.

## Femur

- 1. Maximum length (FL): Maximum distance from the medial condyle to the most prominent proximal part of the head. On the osteometric board, the posterior side of the bone must face downwards, with the condyle at the solid vertical board. *Instrument*: osteometric board.
- 2. Maximum Head Diameter (FMaxHD): The maximum diameter of the femur head. *Instrument*: sliding caliper.
- **3.** Anterior-Posterior (Sagittal) Sub-trochanteric Diameter (FsubTap): The anterior-posterior diameter under the lower trochanter. *Instrument*: sliding caliper.
- 4. Medial-Lateral (Transverse) Sub-trochanteric Diameter: The medial-lateral diameter under the lower trochanter. *Instrument*: sliding caliper.
- 5. Anterior-Posterior (Sagittal) Midshaft Diameter: Distance between anterior and posterior surfaces measured approximately at the midpoint of the diaphysis, at the highest elevation of linea aspera. *Instrument*: sliding caliper. *Comment*: The sagittal diameter should be measured perpendicular to the anterior bone surface.
- 6. Medial-Lateral (Transverse) Midshaft Diameter: Distance between the medial and lateral surfaces at midshaft, measured perpendicular to the anterior-posterior diameter. *Instrument*: sliding caliper.
- 7. Midshaft Circumference: Circumference measured at the level of the midshaft. *Instrument*: measuring tape.
- 8. Biepicondylar width: Distance between the two most laterally projecting points on the epicondyles. *Instrument*: osteometric board.

## Tibia

- 1. Maximum Length: Distance from the superior articular surface of the lateral condyle to the tip of the medial malleolus. *Instrument*: osteometric board. *Comment*: Place the tibia on the board, resting on its posterior surface with the longitudinal axis parallel to the instrument. Place the lip of the medial malleolus on the vertical endboard and press the movable upright against the proximal articular surface of the lateral condyle.
- 2. Proximal Epiphyseal Breadth: Maximum distance between the two most laterally projecting points on the medial and lateral condyles of the proximal articular region (epiphysis). Tibia diaphysis should parallel the upright of the osteometric board. *Instrument:* osteometric board.

- **3. Maximum Diameter at the Nutrient Foramen**: Maximum distance between the anterior crest and the posterior surface at the level of the nutrient foramen. *Instrument*: sliding caliper.
- 4. Minimum Diameter at the Nutrient Foramen: Maximum distance between the anterior crest and the posterior surface at the level of the nutrient foramen. *Instrument*: sliding caliper.

| Measurement                                      | Abbreviation | Instrument        |
|--|--------------|-------------------|
| Humerus maximum length                           | HML          | Osteometric board |
| Humerus head vertical diameter                   | HVD          | Sliding caliper   |
| Humerus maximum midshaft                         | HMaxMid      | Sliding caliper   |
| Humerus minimum midshaft                         | HMinMid      | Sliding caliper   |
| Humerus midshaft circumference                   | HMidCirc     | Таре              |
| Humerus biepicondylar breadth                    | HBB          | Sliding caliper   |
| Ulna maximum length                              | UL           | Osteometric board |
| Ulna notch height                                | UNH          | Sliding caliper   |
| Ulna distal breadth                              | UDB          | Sliding caliper   |
| Radius maximum length                            | RL           | Osteometric board |
| Radius head diameter                             | RHD          | Sliding caliper   |
| Radius distal epiphysis breadth                  | RDB          | Sliding caliper   |
| Femur bicondylar lenght                          | FBL          | Osteometric board |
| Femur max length                                 | FMaxL        | Osteometric board |
| Femur head maximum diameter                      | FHMaxD       | Sliding caliper   |
| Femur subtrochanteric anterior-posterior breadth | FSubTap      | Sliding caliper   |
| Femur subtrochanteric transverse breadth         | FSubTtr      | Sliding caliper   |
| Femur midshaft anterior-posterior breadth        | FMidap       | Sliding caliper   |
| Femur midshaft transverse breadth                | FMidtr       | Sliding caliper   |
| Femur midshaft circumference                     | FMidCirc     | Таре              |
| Femur distal epiphyseal breadth                  | FDB          | Sliding caliper   |
| Tibia length                                     | TL           | Osteometric board |
| Nutrient foramen maximum diameter                | NFMax        | Sliding caliper   |
| Nutrient foramen minimum diameter                | NFMin        | Sliding caliper   |
| Nutrient foramen circumference                   | NFCirc       | Таре              |
| Minimum circumference                            | TMinCirc     | Таре              |
| Upper epiphysis breadth                          | TUB          | Sliding caliper   |
| Lower epiphysis breadth                          | TLB          | Sliding caliper   |
| Fibula length                                    | FibL         | Osteometric board |

Table 6.2: Measurements of the long bones, abbreviations and instruments.

5. Circumference at the Nutrient Foramen: Circumference measured at the level of the nutrient foramen. *Instrument:* measuring tape.

6. Distal Epiphyseal Breadth: Maximum distance between the two most laterally projecting points on the medial malleolus and the lateral surface of the distal epiphysis. *Instrument*: osteometric board.

## Fibula

1. Maximum length: The maximum distance between the most superior point on the fibula head and the most inferior point on the lateral malleolus. *Instrument*: osteometric board.

## 6.5.2 Radiometry

## 6.5.2.1 Radiographic equipment

A digital x-ray machine (Technic TCA 4R PLUS) was used for taking the radiographs of the long bones. The digital acquisition system DIP2000 (Digital Image Processor) of the TCA 4R PLUS is an advanced and flexible device for acquisition, processing and image treatment. The system adjusts settings automatically according to the density of the projected object. The possibility to interface the system with video printers, VCRs and the device DICOM allows the acquired images to be sent for easy reference and quick storage. Thus data are quickly accessed from the digital X-ray machine and stored as bitmap images that are easy to manipulate.

## 6.5.2.2 Definition of landmarks

Landmarks were selected with the objective of being readily distinguishing from a non-professional observer and to form variables that are of known significance for sex variation.

## Humerus

Standard orientation of the bones has been achieved by letting the humerus balance on the horizontal plane, with the anterior surface facing the X-ray camera.

**Proximal Humerus:** Five landmarks (A-E) are selected on the radiograph of the proximal humerus and 10 generated distances (PH1-PH10), representing all possible combinations of these marks, are calculated (Table 6.4).

**Distal Humerus:** Seven landmarks (A-G) are selected on the radiograph of the distal epiphysis and 21 generated distances (DH1-DH21), representing all possible combinations of these marks, are calculated. The selected landmarks for both proximal and distal humerus are defined in <u>Table 6.3</u> and illustrated in <u>Figure 6.2</u>.

## Radius

Standard orientation of the bones has been achieved by letting the radius balance on the horizontal plane, with the anterior surface facing the X-ray camera.

**Proximal Radius:** Eight landmarks (A-G) are selected on the radiograph of the proximal radius and 28 generated distances (PR1-PR28), representing all possible combinations of these points, are calculated (Table 6.6). All landmarks are illustrated in Figure 6.3.

**Distal Radius:** Six landmarks (A-G) are selected on the radiograph of the distal epiphysis (<u>Table 6.5</u>) and 15 generated distances (DR1-DR15), representing all possible combinations of these landmarks, are calculated (<u>Table 6.6</u>).



Fig 6.2 Selection of landmarks. a) Proximal b) Distal humerus

Table 6.3 Definition of landmarks for both proximal and distal humerus.

#### **Proximal humerus**

- A The projection of the medial and inferior part of the head.
- B The projection of the superior part of the anatomical neck
- C The sectioning point on the humeral head outline, of the orthogonal projection of the middle point between landmarks A and B.
- D The maximum curvature point of the greater tubercle
- E The most lateral point that defines the maximum distance from landmark A.

#### **Distal humerus**

- A The incision point between the medial epicondilus and medial part of the trochlea.
- B The maximum curvature point projected in the distal surface of the medial trochlea.
- C The incision point in the distal surface of the troclear groove.
- D The maximum curvature point in the distal surface between the capitulum and the trochlea.
- E The incision point of the capitulum and medial epicondylus.
- F The most lateral point of the projection of the lateral epicondilus
- G The most medial point of the projection of medial epicondilus.

#### Table 6.4 Definition of variables for the proximal and distal humerus

| Proximal  | humerus  | Distal humerus |          |           |          |  |  |  |
|-----------|----------|----------------|----------|-----------|----------|--|--|--|
| Variables | Distance | Variables      | Distance | Variables | Distance |  |  |  |
| PH1       | AB       | DH1            | AB       | DH11      | BG       |  |  |  |
| PH2       | AC       | DH2            | AC DH12  |           | CD       |  |  |  |
| PH3       | AD       | DH3            | AD       | DH13      | CE       |  |  |  |
| PH4       | AE       | DH4            | AE       | DH14      | CF       |  |  |  |
| PH5       | BC       | DH5            | AF       | DH15      | CG       |  |  |  |
| PH6       | BD       | DH6            | AG       | DH16      | DE       |  |  |  |
| PH7       | BE       | DH7            | BC       | DH17      | DF       |  |  |  |
| PH8       | CD       | DH8            | BD       | DH18      | DG       |  |  |  |
| PH9       | CE       | DH9            | BE       | DH19      | EF       |  |  |  |
| PH10      | DE       | DH10           | BF       | DH20      | EG       |  |  |  |
|           |          |                |          | DH21      | FG       |  |  |  |



Figure 6.3 Selection of landmarks. a) Proximal b) Distal radius

Table 6.5 Definition of landmarks for both proximal and distal radius

#### **Proximal radius**

Point under the lateral projection of radial tuberosity A

- В Point so that the distance AB is vertical to the axis of the radial shaft.
- C and D Points on the radial neck so that the distance CD represents the minimum radial diameter on the radiograph
- E and F Points on the radial head so that the distance EF represents the maximum radial diameter on the radiograph.
  - Point on the most lateral projection of the radial tuberosity G
  - Н Point on the radial shaft so that the distance GH is vertical to the radial shaft.
    - **Distal radius**
  - Point on the most medial projection of the distal radial epiphysis A В
    - Point on the most distal projection of the styloid process
  - С Point on the most lateral projection of the styloid process
  - Point on the most inferior and medial border of the articular facet and the medial border of the styloid D process.
  - Point of intersection between the posterior border of the articular facet and the medial border of the styloid Е process.
  - F Point of insertion of brachioradialis

#### Table 6.6 Definition of variables for the proximal and distal radius.

|           | Proxima   | Distal    | Distal radius |           |           |
|-----------|-----------|-----------|---------------|-----------|-----------|
| Variables | Distances | Variables | Distance      | Variables | Distances |
| PR1       | AB        | PR16      | CF            | DR1       | AB        |
| PR2       | AC        | PR17      | CG            | DR2       | AC        |
| PR3       | AD        | PR18      | СН            | DR3       | AD        |
| PR4       | AE        | PR19      | DE            | DR4       | AE        |
| PR5       | AF        | PR20      | DF            | DR5       | AF        |
| PR6       | AG        | G PR21 D  |               | DR6       | BC        |
| PR7       | AH        | PR22      | DH            | DR7       | BD        |
| PR8       | BC        | PR23      | EF            | DR8       | BE        |
| PR9       | BD        | PR24      | EG            | DR9       | BF        |
| PR10      | BE        | PR25      | EH            | DR10      | CD        |
| PR11      | BF        | PR26      | FG            | DR11      | CE        |
| PR12      | BG        | PR27      | FH            | DR12      | CF        |
| PR13      | BH        | PR28      | GH            | DR13      | DE        |
| PR14      | CD        |           |               | DR14      | DF        |
| PR15      | CE        |           |               | DR15      | EF        |



Figure 6.4 Selection of landmarks on a) Proximal b) Distal femur

Table 6.7 Definition of landmarks for both proximal and distal femur.

#### **Proximal femur**

- A Point under the lower end of lesser trochanter in continuance with the vertical axis of the shaft. Points selected on the femoral neck at the points where the curvature changes forming the head so that B and E the distance from B to E is the minimum neck diameter.
- Points on the femoral head, so that the distance C-D is the maximum femoral diameter parallel to the B-C and D E.
  - F Point on the most superior projection of the greater trochanter
  - G Point on the most lateral projection of the proximal epiphysis of the greater trochanter.Landmark in the longitudinal axis of the shaft with the distance A-H (representing the sub-trochanteric
  - H diameter in the radiograph), vertical to the axis of the shaft.

#### **Distal femur**

- A Point on the most lateral projection of the lateral epicondyle
- B Point on the most medial projection of the medial epicondyle
- C Point on the groove between the projection of lateral condyle and epicondyle
- D Point on the groove between the projection of medial condyle and epicondyle
- E Point on the maximum curvature between the inferior projections of the condyles

|           | Proxima   | Distal femur |          |           |           |  |
|-----------|-----------|--------------|----------|-----------|-----------|--|
| Variables | Distances | Variables    | Distance | Variables | Distances |  |
| PF1       | AB        | PF15         | CE       | DF1       | AB        |  |
| PF2       | AC        | PF16         | CF       | DF2       | AC        |  |
| PF3       | AD        | PF17         | CG       | DF3       | AD        |  |
| PF4       | AE        | PF18         | CH       | DF4       | AE        |  |
| PF5       | AF        | PF19         | DE       | DF5       | BC        |  |
| PF6       | AG        | PF20         | DF       | DF6       | BD        |  |
| PF7       | AH        | PF21         | DG       | DF7       | BE        |  |
| PF8       | BC        | PF22         | DH       | DF8       | CD        |  |
| PF9       | BD        | PF23         | EF       | DF9       | CE        |  |
| PF10      | BE        | PF24         | EG       | DF10      | DE        |  |
| PF11      | BF        | PF25         | EH       |           |           |  |
| PF12      | BG        | PF26         | FG       |           |           |  |
| PF13      | BH        | PF27         | FH       |           |           |  |
| PF14      | CD        | PF28         | GH       |           |           |  |



Figure 6.5 Selection of landmarks on a) Proximal b) Distal tibia

Table 6.9 Definition of landmarks for both proximal and distal Tibia

#### Distal tibia

- A Point on the most medial projection of the articular surface of the medial condyle
- B Point on the tip of the lateral intercondylar tubercle
- C Point on the tip of the medial intercondylar tubercle
- D Point on the most superior lateral projection of the lateral condyle
- E Point on the most lateral projection of the lateral condyle
- F Point on the most medial projection of of the articular surface of the the medial condyle

#### Distal tibia

- A Point on the most medial projection of the medial alveolus
- B Point on the most inferior projection of the medial alveolus
- C Point on the maximum curvature on the posterior border of the articular surface for the talus
- D Point on the most lateral projection of the distal epiphysis of the tibia
- E Sectioning point between the projection of the posterior and anterior border of the articular surface for the talus

|           | Proxin   | Distal tibia |          |           |          |
|-----------|----------|--------------|----------|-----------|----------|
| Variables | Distance | Variables    | Distance | Variables | Distance |
| PT1       | AB       | PT11         | CE       | DT1       | AB       |
| PT2       | AC       | PT12         | CF       | DT2       | AC       |
| PT3       | AD       | PT13         | DE       | DT3       | AD       |
| PT4       | AE       | PT14         | DF       | DT4       | AE       |
| PT5       | AF       | PT15         | EF       | DT5       | BC       |
| PT6       | BC       |              |          | DT6       | BD       |
| PT7       | BD       |              |          | DT7       | BE       |
| PT8       | BE       |              |          | DT8       | CD       |
| PT9       | BF       |              |          | DT9       | CE       |
| PT10      | CD       |              |          | DT10      | DE       |

Table 6.10 Definition of variables for the proximal and distal tibia

## Femur

The bone is orientated with the anterior surface facing the X-ray table and the epicondyles resting on the horizontal plane.

**Proximal femur:** Eight landmarks (A-H) are selected in the radiograph of the proximal femur and 28 distances (PF1-PF28), representing all possible combinations of these marks, are generated. The selected landmarks are shown in <u>Figure 6.4</u> and described in <u>Table 6.7</u>.

**Distal femur:** Five landmarks (A-E) are selected in the radiograph of the proximal femur and 10 distances (DF1-DF10), representing all possible combinations of these marks, are generated. The selected landmarks are shown in <u>Figure 6.4</u> and described in <u>Table 6.8</u>.

## Tibia:

To take the image, the bone is orientated with the anterior surface facing the X-ray table and the distal epiphysis perpendicular to the axis of the camera.

**Proximal Tibia:** Six landmarks (A-F) are selected in the radiograph of the proximal tibia and 21 distances (PT1-PT21), representing all possible combinations of these marks, are generated. The selected landmarks are shown in <u>Figure 6.5</u> and described in <u>Table 6.9</u>.

**Distal tibia:** Five landmarks (A-E) are selected in the radiograph of the proximal femur and 10 distances (DT1-DT10), representing all possible combinations of these marks, are generated. The selected landmarks are shown in Figure 6.5 and described in <u>Table 6.9</u>. All variables for both epiphyses are presented in <u>Table 6.10</u>.

## 6.5.2.3 Cases excluded from the study

A limited number of specimens did not allow the observation of the selected landmarks and therefore there were excluded from the study. Such cases are presented in Figure 6.6. On the left side it is illustrated the proximal epiphysis of a femur on which the lower trochanter is not projected on the radiograph which means that landmarks A and H (Fig 6.4) can not be located. On the right side it can be observed the proximal epiphysis of a radius on which the radial tuberosity is not visible on the radiograph. In that case the landmarks A, B, G and H could not be placed and therefore the specimen was not used in the study.

#### 6.5.2.4 Inter-landmark distances

For this analysis several software have been employed such as tps series and Morpheus et al. Tps util was used to create the databases from the radiographs. TpsDig2 was used to digitize the selected landmarks and to incorporate the scaling factor. Morpheus et al. was used to generate the distances from the selected landmarks.



Figure 6.6 Left: Proximal femur without the lesser trochanter, Right: Proximal radius without the radial tuberosity. Both cass are excluded from the study.

## **6.5.3 Statistics**

## 6.5.3.1 Estimation of error

Sixty specimens (30 males and 30 females) were randomly selected and measured by the same observer over a period of 1 month to estimate the intra-observer error. The same specimens were measured by a second observer (intra-observer error) and the means were compared with the first measurements of the first observer (inter-observer error) using a student's T-test.

The paired t test provides an hypothesis test of the difference between population means for a pair of random samples whose differences are approximately normally distributed. So, if *d* represents the difference between observations, the hypotheses are:

 $H_0: d = 0$  (the difference between the two observations is 0)

 $H_a: d 0$  (the difference is not 0)

The test statistic is *t*:

$$t = \frac{\overline{d}}{\sqrt{s^2 / n}}$$

where d is the mean difference between the paired observations,  $s^2$  is the sample variance, **n** is the sample size and **t** is a Student t quantile with n-1 degrees of freedom. If the p-value associated with **t** is low (< 0.05), there is evidence to reject the null hypothesis. The standard

error over which this mean is tested is the the standard error of the mean difference (Sokal and Rohlf, 1998; Wheatley, 2005).

## 6.5.3.2 Discriminant Function Analysis (DFA)

Discriminant function analysis is a powerful descriptive classification method developed by Fisher (Fisher, 1936). It is used to select the optimal combination of variables and to calculate specific formulae in order to classify cases in preexisting groups according to the similarities between each case and the other cases belonging to the same group (Brown and Wicker, 2000). With this method it can be determined which variables are more useful for separating one group from another and if different sets of variables perform equally well. Comparisons of percentages of diagnostic accuracy indicate which variables or combination of variables produce a greater separation of groups and, in this particular case, the characteristics of sexual dimorphism. Discriminant function statistics can also pinpoint extreme cases within the groups that differ from the others (outlines). A one-way ANOVA is used in order to calculate the means and the standard deviations for each measurement.

The discriminant function is constructed by assigning a discriminant score to each case. Depending on the variable and combination of variables for a function, the score changes from case to case. A sectioning point (SP) is created by using the mean male and female discriminant scores, which are also known as the group centroids. Therefore, each function has a different sectioning point, which is based on the variables entered in the function. Unstandardized discriminant coefficients are used for building the formula. The standardized (Fisher's) coefficients are used to compare the relative importance of the independent variables (Gapert et al., 2009). A discriminant function is built as follows:

# $\mathbf{P} = \mathbf{a}\mathbf{1} \times \mathbf{x}\mathbf{1} + \mathbf{a}\mathbf{2} \times \mathbf{x}\mathbf{2} + \dots + \mathbf{a}\mathbf{n} \times \mathbf{x}\mathbf{n} + \mathbf{b}$

where **a1** through **an** are the discriminant coefficients, **x1** through **xn** are the discriminating variables and **b** is the constant. To assign the case to either male or female sex, the product P is compared to the sectioning point derived by the discriminant function. A value higher than the sectioning point was deemed to be male and a value below it deemed to be female.

Stepwise discriminant function analysis is used to select the combination of variables that best discriminate sexes. In stepwise discriminant function analysis, a model of discrimination is built step-by-step. Specifically, at each step all variables are reviewed and evaluated to determine which one will contribute most to the discrimination between groups. That variable will then be included in the model, and the process starts again. The stepwise procedure is "guided" by the respective F to enter and F to remove values. The *F* value for a

variable indicates its statistical significance in the discrimination between groups, that is, it is a measure of the extent to which a variable makes a unique contribution to the prediction of group membership. In this analysis, F to enter is set to 3.84 and F to remove to 2.71.

## 6.5.3.3 Cross-validation

A jack-knife or leave-one-out classification procedure is applied in order to demonstrate the accuracy rate of the original sample and the one created by cross-validation. In this procedure, one specimen is systematically held out and DFA is performed to the remaining sample. Then the excluded case is classified in one of the groups according to the discriminant function extrapolated by the analysis. The procedure is repeated until each case in the sample has been held out and classified (Brown and Wicker, 2000). Than the classification accuracy is computed and compared to the classification accuracy for the original sample. The closer the cross-validated to the original accuracy, the higher the reliability of the discriminant function.

## 6.5.3.4 Posterior probability (PP)

The normal curve models of the predictor variables for each group can be used to provide probability estimates of a particular score given membership in a particular group. They are calculated from the Mahalanobis distance, i.e. the distance between a specimen and the centroid of the distribution of all specimens in a multi-dimensional space made up of the variables taken into account in the DFA (Mardia et al., 2000; Murail et al., 2005). In DFA, the area in the tails under a normal curve model (Fig. 6.7) for a given group between points equally distant from  $\mu$  (mean) is the probability of either point given that group.



Figure 6.7 Normal curve Gray areas represent the 95% confidence intervals.

The computation of posterior probabilities is made with an equal prior probability for males and females. Data analysis was carried out using discriminant function subroutines of SPSS 13.0.

# **Chapter 7: Results**

#### 7.1. Osteometry

## 7.1.1. Cranial Skeleton

A total of 178 well preserved, adult skulls (90 males and 88 females) of Cretan origin are measured. Sixteen dimensions are taken from the neural and facial portion of the skull. These dimensions are maximum cranial length, basion-nasion length, maximum vault breadth, maximum frontal breadth, minimum frontal breadth, bizygomatic breadth, foramen magnum length, foramen magnum breadth, basion-bregma height, basion-prosthion length, nasionprosthion height, mastoid height, biorbital breadth, interorbital breadth, nose breadth and nose height.

A comparison is made with several populations geographically and time wise distant from Cretans. The data are from the early 20<sup>th</sup> century White Americans (Terry collection) and South Africans Whites (Dart and Pretoria collections) [21, 22] all gathered by the author İşcan. Archaeological data are obtained from a published work [18] and derive from the remains of Middle (1900 B.C.-1600 B.C.) and Late Helladic (1600 B.C.-1100 B.C.) periods in Crete.

Descriptive statistics of 16 skull measurements and associated univariate F-ratio to measure the differences between the sexes are shown in Table 7.1.1.1. All but interorbital breadth, are found significantly different between the sexes. Mean age difference is not significant (mean age for men= 68.94 +/- 13.41, N=66; for women= 73.21 +/-16.77, N=66). Table 7.1.1.2 provides various discriminant functions statistics where the sex of an unknown skull can be determined. These functions are constructed so that different preservation conditions can be considered to make identification. Function 1 (CF1) is designed to analyze a complete skull which is commonly seen in a protected area, not so seriously damaged, thus many dimensions can be measured. The table shows the result of a stepwise discriminant function analysis using 15 dimensions. Function 2 (CF2) assumes that face is not fully available for measurement. Eight dimensions (maximum cranial length, maximum vault breadth, maximum frontal breadth, minimum frontal breadth, bizygomatic breadth, foramen magnum length, foramen magnum breadth, basion-bregma height, mastoid height) are entered into another stepwise analysis and five of them are selected (Table 7.1.1.2). Forming CF3-CF8, cranial length, basion-bregma height, basion-nasion length, bizygomatic breadth, biorbital breadth and nose height are used with direct discriminant function procedure (Table <u>7.1.1.2</u>).

|                         | Mal    | es   | Fema   |      |         |
|-------------------------|--------|------|--------|------|---------|
| Dimensions              | Mean   | SD   | Mean   | SD   | F-ratio |
| Max cranial length      | 181.07 | 6.63 | 172.89 | 6.48 | 64.92   |
| Basion-bregma height    | 139.70 | 4.87 | 132.47 | 6.83 | 62.14   |
| Max vault breadth       | 137.64 | 6.63 | 133.92 | 5.85 | 14.84   |
| Max frontal breadth     | 122.46 | 5.79 | 118.99 | 5.42 | 16.03   |
| Min frontal breadth     | 96.33  | 4.52 | 93.23  | 4.50 | 19.63   |
| Bizygomatic breadth     | 130.54 | 5.13 | 122.07 | 4.57 | 126.57  |
| Foramen magnum length   | 36.19  | 2.80 | 34.49  | 2.31 | 18.38   |
| Foramen magnum breadth  | 31.37  | 2.80 | 28.85  | 2.51 | 37.60   |
| Mastoid height          | 31.69  | 3.71 | 28.56  | 3.50 | 31.50   |
| Basion-nasion length    | 102.01 | 3.85 | 96.25  | 6.54 | 48.36   |
| Basion-prosthion length | 93.11  | 5.05 | 88.76  | 5.70 | 27.33   |
| Nasion-prosthion height | 69.38  | 6.56 | 64.12  | 6.40 | 27.44   |
| Biorbital breadth       | 97.86  | 4.25 | 93.14  | 4.17 | 52.41   |
| Nose breadth            | 23.98  | 2.54 | 23.16  | 2.11 | 5.17*   |
| Nose height             | 51.60  | 3.04 | 48.20  | 2.98 | 53.03   |

 Table 7.1.1.1 Descriptive statistics of cranial dimensions and univariate F-ratio of the differences between the sexes.

<u>Table 7.1.1.3</u> summarizes the accuracy rate for both the original data and "leave one out classification" in all functions. This classification provides a test to determine the sex of an unknown individual. The highest accuracy rate is obtained using CF1 (88.2%) followed by CF2 (83%). Correct group membership reaches 82% when bizygomatic breadth (CF3) is used alone and 75% in the case of basion-bregma height (CF4) and biorbital breadth (CF5).

The sex can be calculated from these functions by multiplying the values of the cranial dimensions by the corresponding coefficients plus the constant. If the resulting discriminant function score is greater than zero it is classified as male. In the situation that only one dimension is used for the analysis the sex can be simple determined by evaluating the measurement of the unknown according to the demarking point which in the case of bizygomatic breadth is 126.19 (mean of both sexes). For example, a skull of an unknown person with a bizygomatic breadth 120 mm will be classified as female.

The "leave one out classification" statistic surveys to a comparison of accuracy rate between the original sample and the one created by cross validation. Figure 7.1.1

<sup>\*</sup> Significant at p<0.05. all others significant at p< .001, df=1.165

demonstrates the probability levels of correct sexing according to the discriminant scores of each individual.

| Step    | Variables entered       |         |       |                                      |
|---------|-------------------------|---------|-------|--------------------------------------|
|         | CF1: Total cranium      | Exact F | df    | Raw coefficient                      |
| 1       | Bizygomatic breadth     | 129.48  | 1.168 | 0.073045                             |
| 2       | Max cranial length      | 83.57   | 2.167 | 0.1495                               |
| 3       | Nasion-Prosthion height | 60.55   | 3.166 | 0.063251                             |
| 4       | Mastoid height          | 47.39   | 4.165 | 0.039003                             |
| 5       | Nose breadth            | 39.37   | 5.164 | -0.096953                            |
|         | Constant                |         |       | -34.024003                           |
|         |                         |         |       |                                      |
|         | CF2: Neurocranium       |         |       |                                      |
| 1       | Max cranial length      | 70.41   | 1.176 | 0.088869                             |
| 2       | Basion-bregma height    | 52.78   | 2.175 | 0.059045                             |
| 3       | Mastoid height          | 41.41   | 3.174 | 0.047681                             |
| 4       | Foramen magnum breadth  | 34.62   | 4.173 | 0.117936                             |
| 5       | Max vault breadth       | 30.08   | 5.172 | 0.081853                             |
|         | Constant                |         |       | -36.208088                           |
|         |                         |         |       | Demarking point                      |
| CF3: Bi | zygomatic breadth       | 132.17  | 1.175 | Female< 126.19 <male< td=""></male<> |
| CF4: Ba | asion-bregma height     | 52.64   | 1.176 | Female<135.81 <male< td=""></male<>  |
| CF5: Bi | orbital breadth         | 54.27   | 1.176 | Female< 95.42 <male< td=""></male<>  |
| CF6: No | ose height              | 55.92   | 1.169 | Female< 49.87 <male< td=""></male<>  |
| CF7: Ba | sion-nasion length      | 49.10   | 1.176 | Female< 99.1 <male< td=""></male<>   |
| CF8: M  | ax cranial length       | 70.41   | 1.176 | Female< 176.80 <male< td=""></male<> |

 Table 7.1.1.2 Discriminant function statistics, F-ratios and statistical significance in Cretans. Sectioning point was set to zero for both CF1 and CF2.

Initially the posterior probability values for each function are produced using a discriminant subprogram of SPSS, than misclassified cases are removed and probability of correct classification for both sexes is combined. Plotting the data with Excel program for Windows resulted in the diagram presented in Figure 7.1.1 For example if a discriminant score based on the neurocranial measurements (CF2) is -1.40337 (x coordinate) the posterior probability of that individual to be female is 93.03% (y coordinate).

A comparison of the modern Cretans is made with American and South Africans Whites (Caucasoids) of approximately the same period (<u>Table 7.1.1.4</u>). One would note that Cretans are closer in size to American Whites in most dimensions and furthest from African Whites. African Whites demonstrate a significantly larger cranial length (over 7 mm for males and over 6mm for females) while means for maximum frontal breadth are greater in Cretans for both sexes. Mean values for cranial length are greater in White (Terry) Americans as well but all other dimensions are very close to contemporary Cretans.

| Cranial dimensions and    |       |       |       |       |       |
|---------------------------|-------|-------|-------|-------|-------|
| functions                 | M     | lale  | Fen   | nale  | Total |
| F1: Total cranium         | N     | %     | N     | %     | %     |
| Original                  | 75/86 | 87.21 | 75/84 | 89.29 | 88.20 |
| Cross-validated           | 75/86 | 87.21 | 73/84 | 86.90 | 87.10 |
| F2: Neurocranium          |       |       |       |       |       |
| Original                  | 77/90 | 85.56 | 71/88 | 80.68 | 83.10 |
| Cross-validated           | 77/90 | 85.56 | 70/88 | 79.55 | 82.60 |
| F3: Bizygomatic breadth   |       |       |       |       |       |
| Original                  | 71/90 | 78.89 | 74/87 | 85.06 | 81.90 |
| Cross-validated           | 71/90 | 78.89 | 74/87 | 85.06 | 81.90 |
| F4: Basion-bregma height  |       |       |       |       |       |
| Original                  | 68/90 | 75.56 | 66/88 | 75.00 | 75.30 |
| Cross-validated           | 68/90 | 75.56 | 66/88 | 75.00 | 75.30 |
| F5: Biorbital breadth     |       |       |       |       |       |
| Original                  | 67/90 | 74.44 | 67/88 | 76.14 | 75.30 |
| Cross-validated           | 67/90 | 74.44 | 67/88 | 76.14 | 75.30 |
| F6: Nose height           |       |       |       |       |       |
| Original                  | 63/86 | 73.26 | 64/85 | 75.29 | 74.30 |
| Cross-validated           | 63/86 | 73.26 | 64/85 | 75.29 | 74.30 |
| F7: Basion-nasion breadth |       |       |       |       |       |
| Original                  | 68/90 | 75.56 | 60/88 | 68.18 | 71.90 |
| Cross-validated           | 68/90 | 75.56 | 60/88 | 68.18 | 71.90 |
| F8: Max cranial length    |       |       |       |       |       |
| Original                  | 62/90 | 68.89 | 63/88 | 71.59 | 70.20 |
| Cross-validated           | 62/90 | 68.89 | 63/88 | 71.59 | 70.20 |

# Table 7.1.1.3 Classification accuracy on cranial dimensions in Cretan population.

Middle to late Helladic population of Crete is also compared with the cemetery sample. Due to the lack of sufficient sample size, only 6 measurements (maximum cranial length, maximum vault breadth, basion-bregma height, maximum frontal breadth, minimum frontal breadth and bizygomatic breadth) are available for comparison and it is observed that the archaeological Cretans are relatively smaller than the recent descendents in all dimensions but cranial length. Mean values for cranial length are almost 5 mm greater in Helladic males and 7 mm in Helladic females compared to modern Cretans.

In order to test the efficacy of the equations deriving of modern Cretans it is attempted to classify the archaeological sample using the most effective single dimension; bizygomatic breadth. This measurement is available in 46 of the 126 Helladic crania and correct group membership is found 83.3% for females, 64.3% for males and 71.7% in total. Classification results yield about 10% less than in the original sample.



Figure 7.1.1 Probability levels of correct sexing according to the discriminant scores of each individual. Negative discriminant scores correspond to females and positive discriminant scores correspond to males.

|              |    | Cretans  |      |    | White Americ | ans   |    | White Africa | <b>1</b> 5 | H  | elladic popula | tion | ion t-Test differ  |                    | Cretans and            |
|--------------|----|----------|------|----|--------------|-------|----|--------------|------------|----|----------------|------|--------------------|--------------------|------------------------|
| Male         | N  | Mean     | SD   | N  | Mean         | SD    | N  | Mean         | SD         | N  | Mean           | SD   | White<br>Americans | White<br>Africans  | Helladic<br>population |
| CL           | 90 | 180,82   | 6,54 | 46 | 182,28       | 6,95  | 44 | 187,77       | 5,44       | 82 | 185,07         | 9,19 | -1,207             | °-6,09             | <sup>b</sup> -3,52     |
| Ba-br        | 90 | 139,19   | 5,89 | 43 | 133,42       | 9,28  | 44 | 136,84       | 4,06       | 36 | 127,81         | 6,73 | ° 4,35             | *2,38              | ° 9,41                 |
| CB           | 90 | 137,73   | 6,50 | 46 | 142,76       | 5,20  | 44 | 139,59       | 5,83       | 82 | 133,82         | 4,83 | °-4,55             | -1,61              | °4,45                  |
| MaxFrB       | 90 | 122,46   | 5,66 | 46 | 123,93       | 5,87  | 44 | 119,45       | 4,92       | 71 | 113,27         | 6,23 | -1,425             | <sup>b</sup> 3,01  | ° 9,79                 |
| MinFrB       | 90 | 96,24    | 4,44 | 46 | 96,13        | 5,49  | 44 | 97,89        | 3,84       | 66 | 94,83          | 4,35 | 0,129              | * -2,1             | *1,99                  |
| Bizy breadth | 90 | 130,30   | 5,07 | 46 | 130,80       | 4,69  | 44 | 128,93       | 4,37       | 28 | 126,32         | 6,72 | -0,563             | 1,53               | <sup>b</sup> 3,35      |
| FML          | 90 | 36,19    | 2,78 | 45 | 37,02        | 2,74  | 44 | 37,68        | 2,59       |    |                |      | -1,649             | <sup>b</sup> -2,99 |                        |
| FMB          | 90 | 31,30    | 2,74 | 45 | 31,49        | 2,98  | 44 | 31,57        | 1,81       |    |                |      | -0,371             | -0,59              |                        |
| MaH          | 90 | 31,68    | 3,62 | 46 | 30,59        | 3,07  | 44 | 33,95        | 3,35       |    |                |      | 1,745              | °-3,51             |                        |
| Ba-na        | 90 | 101,82   | 3,82 | 46 | 101,22       | 6,83  | 44 | 102,48       | 4,48       |    |                |      | 0,663              | -0,88              |                        |
| Ba-pr        | 89 | 92,99    | 4,96 | 38 | 108,68       | 96,98 | 43 | 95,42        | 5,39       |    |                |      | -1,530             | *-2,56             |                        |
| Na-pr        | 89 | 69,15    | 6,49 | 27 | 70,33        | 8,55  | 44 | 71,43        | 4,04       |    |                |      | -0,770             | *-2,13             |                        |
| NB           | 86 | 23,98    | 2,51 | 46 | 23,87        | 1,54  | 44 | 24,75        | 2,20       |    |                |      | 0,280              | -1,72              |                        |
| NH           | 86 | 51,58    | 3,03 | 46 | 52,07        | 2,50  | 44 | 53,75        | 3,56       |    |                |      | -0,932             | °-3,64             |                        |
| Female       |    |          |      |    |              |       |    |              |            |    |                |      |                    |                    |                        |
| CL           | 88 | 172,68   | 6,40 | 46 | 176,78       | 7,77  | 47 | 178,81       | 5,87       | 41 | 179,22         | 6,26 | °-3,27             | °-5,45             | °-5,44                 |
| Ba-br        | 88 | 132,35   | 6,67 | 44 | 129,32       | 4,48  | 47 | 130,64       | 5,30       | 25 | 125,80         | 8,31 | 2,723              | 1,52               | °4,10                  |
| CB           | 88 | 133,70   | 5,79 | 46 | 139,28       | 5,40  | 47 | 137,81       | 4,79       | 40 | 132,18         | 5,35 | °-5,41             | °-4,15             | 1,42                   |
| MaxFrB       | 88 | 118,85   | 5,42 | 45 | 119,04       | 5,66  | 47 | 115,60       | 5,86       | 38 | 109,55         | 6,05 | -0,190             | ° 3,23             | ° 8,53                 |
| MinFrB       | 88 | 93,19    | 4,41 | 46 | 94,33        | 4,61  | 47 | 93,62        | 4,74       | 36 | 92,47          | 4,78 | -1,394             | -0,52              | 0,80                   |
| BizyB        | 87 | 121,93   | 4,60 | 46 | 122,78       | 4,01  | 47 | 122,02       | 3,54       | 18 | 120,61         | 6,17 | -1,061             | -0,12              | 1,04                   |
| FML          | 88 | 34,52    | 2,33 | 46 | 36,02        | 2,50  | 47 | 36,17        | 1,88       |    |                |      | <sup>b</sup> -3,46 | °-4,18             |                        |
| FMB          | 88 | 28,91    | 2,49 | 46 | 30,28        | 1,89  | 47 | 30,55        | 1,89       |    |                |      | <sup>b</sup> -3,26 | °-3,94             |                        |
| MaH          | 88 | 28,51    | 3,56 | 46 | 27,89        | 2,74  | 47 | 30,89        | 3,89       |    |                |      | 1,027              | °-3,59             |                        |
| Ba-na        | 88 | 96,31    | 6,38 | 45 | 96,62        | 4,61  | 47 | 96,32        | 4,10       |    |                |      | -0,289             | -0,01              |                        |
| Ba-pr        | 88 | \$\$,\$4 | 5,57 | 33 | 87,79        | 5,55  | 47 | 90,04        | 5,03       |    |                |      | 0,922              | -1,24              |                        |
| Na-pr        | 88 | 64,13    | 6,31 | 6  | 68,50        | 4,64  | 46 | 66,02        | 5,13       |    |                |      | -1,664             | -1,76              |                        |
| NB           | 85 | 23,15    | 2,09 | 46 | 23,35        | 1,72  | 47 | 22,89        | 2,05       |    |                |      | -0,561             | 0,67               |                        |
| NH           | 85 | 48,14    | 2,98 | 46 | 49,46        | 3,28  | 47 | 49,83        | 2,21       |    |                |      | * -2,33            | ° -3,40            |                        |

Table 7.1.1.4 A t-Test comparison of the Cretans with the Helladic population, White Americans and South Africans. T-Test Values are significant at \* p<0.05; b p<.01; c p<.001

## 7.1.2 Postcranial skeleton

## 7.1.2.1 Discriminant function analysis

## Upper Extremity

A total of 12 measurements were taken from the bones of the upper limb. More specifically, 6 dimensions were measured on the humerus: maximum humeral length (HL), vertical head diameter (HVD), maximum midshaft diameter (HMaxMid), minimum midshaft diameter (HminMid), midshaft circumference (HmidCirc) and biepicondylar breadth (HBB). For the ulna, maximum length (UL), notch height (UNH) and distal breadth (UDB) were measured, while in the case of the radius, maximum length (RL), head diameter (RHD) and distal breadth (RDB) were taken (Table 6.2).

Descriptive statistics of humeral, radial and ulnar measurements and associated univariate F-ratio to measure the differences between the sexes are shown in Table 7.1.2.1 The differences between the means in males and females are significant (p<0.001). The differences between means of right and left lower limb bones were compared and found not to differ significantly between the sexes (p<0.05). Therefore right bones were also used in the analysis.

## Univariate statistics

Table 7.1.2.2 presents the results of the discriminant function analysis for single dimensions. F-ratios, degrees of freedom, cut-off values and classification accuracy for both original and cross-validated data are presented here.

## Humerus

The most effective single dimensions for the humerus, as determined by direct discriminant analysis, was found to be HVD (89.6%), followed by HBB (85.6%) and HML 84.4%. Cross-validation procedure produced results very close to the original classification in all cases.

#### Radius

RL was found to be the best single predictor for sex estimation among all upper limb measurements and the best single variable for the radius with 91% classification accuracy. RHD also performed well for both original (86.1%) and cross-validated data. According to Table 7.1.2.2, a radius with length greater than 223mm will be classified as male, while in the opposite case it will be assigned as female.

## Ulna

UL was found to be the most effective single dimension for the ulna with classification accuracy of 89%, while the two other single dimensions were less effective in sex determination, with classification accuracy not exceeding 79%.
# Multivariate statistics

Tables 7.1.2.3, 7.1.2.4 and 7.1.2.5 demonstrate various discriminant function statistics, where the sex of a skeleton can be determined by measurements of the upper extremity long bones. These functions are constructed so that different preservation conditions can be considered to make identification. Exact F gives an indication of the contribution of each variable entered in the equation to separate the sexes.

|           |    | Males  |       |    | Females |       |                      |
|-----------|----|--------|-------|----|---------|-------|----------------------|
| Variables | N  | Mean   | SD    | N  | Mean    | SD    | <sup>*</sup> F-ratio |
| HML       | 94 | 321.34 | 14.47 | 79 | 294.2   | 13.7  | 158.73               |
| HVD       | 94 | 46.39  | 2.49  | 79 | 41.12   | 2.34  | 203.69               |
| HMaxMid   | 94 | 22.51  | 1.66  | 79 | 20.16   | 1.63  | 88.04                |
| HMinMid   | 94 | 18.43  | 1.57  | 79 | 15.75   | 1.52  | 128.74               |
| HMidCirc  | 94 | 65.89  | 4.86  | 79 | 58.3    | 4.72  | 107.6                |
| HBB       | 94 | 61.7   | 3.85  | 79 | 54.13   | 3.7   | 171.91               |
| UL        | 93 | 258.4  | 19.52 | 78 | 231.9   | 10.87 | 114.49               |
| UNH       | 93 | 23.41  | 2.29  | 78 | 20.72   | 2.46  | 54.55                |
| UDB       | 92 | 20.85  | 2.57  | 77 | 18.39   | 1.72  | 51.1                 |
| RL        | 94 | 238.38 | 11.43 | 79 | 213.2   | 10.74 | 219.92               |
| RHD       | 94 | 22.74  | 1.63  | 79 | 19.86   | 1.17  | 172.34               |
| RDB       | 94 | 30.3   | 2.72  | 79 | 26.58   | 3.09  | 70.9                 |
| FBL       | 94 | 441.10 | 19.98 | 78 | 405.36  | 19.69 | 138.18               |
| FMaxL     | 94 | 443.79 | 20.10 | 78 | 408.17  | 20.14 | 133.67               |
| FHMaxD    | 94 | 47.27  | 2.55  | 78 | 42.42   | 2.29  | 169.64               |
| FSubTap   | 94 | 27.52  | 2.16  | 78 | 24.82   | 2.42  | 59.71                |
| FSubTtr   | 94 | 32.37  | 2.33  | 78 | 30.22   | 2.22  | 37.86                |
| FMidap    | 94 | 29.23  | 2.54  | 79 | 26.29   | 1.83  | 73.64                |
| FMidtr    | 94 | 28.16  | 1.97  | 79 | 26.56   | 2.17  | 25.84                |
| FMidCirc  | 94 | 89.51  | 5.24  | 79 | 82.81   | 4.95  | 73.80                |
| FDB       | 94 | 81.25  | 4.26  | 79 | 74.13   | 3.66  | 136.37               |
| TL        | 93 | 363.02 | 19.41 | 79 | 332.49  | 17.40 | 116.17               |
| NFMax     | 93 | 35.24  | 2.33  | 79 | 30.81   | 2.29  | 157.24               |
| NFMin     | 93 | 24.83  | 2.29  | 79 | 22.26   | 2.08  | 58.59                |
| NFCirc    | 93 | 94.65  | 6.31  | 79 | 83.86   | 5.75  | 135.42               |
| TMinCirc  | 93 | 74.62  | 4.63  | 79 | 68.10   | 4.48  | 87.29                |
| TUB       | 93 | 75.33  | 3.83  | 79 | 68.28   | 3.88  | 143.40               |
| TLB       | 93 | 45.14  | 3.05  | 78 | 40.43   | 2.52  | 118.50               |
| FibL      | 92 | 358.84 | 16.65 | 77 | 329.53  | 15.63 | 137.32               |

Table 7.1.2.1 Descriptive statistics for the measurements of the upper and lower limb.

\* p<0.001

|           |       |                  |       |      | Original |      |       |       | Cr   | oss valida | ted  |       |
|-----------|-------|------------------|-------|------|----------|------|-------|-------|------|------------|------|-------|
| Variables | df    | Cut-off<br>value | Ma    | le   | Fem      | ale  | Total | Ma    | le   | Fem        | ale  | Total |
|           |       | value            | N     | %    | N        | %    | %     | N     | %    | N          | %    | %     |
| HML       | 1.172 | 307.76           | 81/94 | 86.2 | 65/79    | 82.3 | 84.4  | 81/94 | 86.2 | 65/79      | 82.3 | 84.4  |
| HVD       | 1.172 | 43.76            | 84/94 | 89.4 | 71/79    | 89.9 | 89.6  | 84/94 | 89.4 | 71/79      | 89.9 | 89.6  |
| HMaxMid   | 1.172 | 21.33            | 70/94 | 74.5 | 64/79    | 81.0 | 77.5  | 70/94 | 74.5 | 64/79      | 81.0 | 77.5  |
| HMinMid   | 1.172 | 17.09            | 76/94 | 80.9 | 65/79    | 82.3 | 81.5  | 76/94 | 80.9 | 65/79      | 82.3 | 81.5  |
| HMidCirc  | 1.172 | 62.1             | 69/94 | 73.4 | 67/79    | 84.8 | 78.6  | 69/94 | 73.4 | 67/79      | 84.8 | 78.6  |
| HBB       | 1.172 | 57.91            | 78/94 | 83.0 | 70/79    | 88.6 | 85.6  | 78/94 | 83   | 70/79      | 88.6 | 85.6  |
| UL        | 1.170 | 245.1            | 83/93 | 89.2 | 69/78    | 88.5 | 88.9  | 83/93 | 89.2 | 69/78      | 88.5 | 88.9  |
| UNH       | 1.170 | 22.1             | 66/93 | 71.0 | 65/78    | 83.3 | 76.6  | 66/93 | 71   | 65/78      | 83.3 | 76.6  |
| UDB       | 1.168 | 19.3             | 66/92 | 71.7 | 67/77    | 87.0 | 78.7  | 66/92 | 71.7 | 67/77      | 87.0 | 78.7  |
| RL        | 1.172 | 225.8            | 86/94 | 91.5 | 71/79    | 89.9 | 90.8  | 86/94 | 91.5 | 71/79      | 89.9 | 90.8  |
| RHD       | 1.172 | 21.3             | 75/94 | 79.8 | 74/79    | 93.7 | 86.1  | 75/94 | 79.8 | 72/79      | 91.1 | 85.0  |
| RDB       | 1.172 | 28.35            | 80/94 | 85.1 | 59/79    | 74.7 | 80.3  | 80/94 | 85.1 | 59/79      | 74.7 | 80.3  |
| FMaxL     | 1.171 | 425.98           | 80/94 | 85.1 | 58/78    | 74.4 | 80.2  | 78/94 | 83   | 58/78      | 74.4 | 79.1  |
| FeBL      | 1.171 | 423.23           | 81/94 | 86.2 | 60/78    | 76.9 | 82.0  | 81/94 | 86.2 | 60/78      | 76.9 | 82.0  |
| FHMaxD    | 1.171 | 44.85            | 79/94 | 84.0 | 65/78    | 83.3 | 83.7  | 79/94 | 84   | 65/78      | 83.3 | 83.7  |
| FSubTap   | 1.171 | 26.17            | 71/94 | 75.5 | 62/78    | 79.5 | 77.3  | 71/94 | 75.5 | 62/78      | 79.5 | 77.3  |
| FSubTtr   | 1.171 | 31.3             | 61/94 | 64.9 | 55/78    | 70.5 | 67.4  | 61/94 | 64.9 | 55/78      | 70.5 | 67.4  |
| FMidap    | 1.172 | 27.76            | 66/94 | 70.2 | 61/79    | 77.2 | 73.4  | 66/94 | 70.2 | 61/79      | 77.2 | 73.4  |
| FMidtr    | 1.172 | 27.36            | 65/94 | 69.1 | 54/79    | 68.4 | 68.8  | 65/94 | 69.1 | 54/79      | 68.4 | 68.8  |
| FMidCirc  | 1.172 | 86.16            | 68/94 | 72.3 | 62/79    | 78.5 | 75.1  | 68/94 | 72.3 | 62/79      | 78.5 | 75.1  |
| FDB       | 1.172 | 77.69            | 79/94 | 84.0 | 71/79    | 89.9 | 86.5  | 79/94 | 84   | 71/79      | 89.9 | 86.5  |
| TL        | 1.171 | 347.76           | 73/93 | 78.5 | 64/79    | 81.0 | 79.7  | 73/93 | 78.5 | 64/79      | 81.0 | 79.7  |
| NFMax     | 1.171 | 33.02            | 75/93 | 80.6 | 66/79    | 83.5 | 82.0  | 75/93 | 80.6 | 66/79      | 83.5 | 82.0  |
| NFMin     | 1.171 | 23.55            | 65/93 | 69.9 | 57/79    | 72.2 | 70.9  | 65/93 | 69.9 | 57/79      | 72.2 | 70.9  |
| NFCirc    | 1.171 | 89.25            | 74/93 | 79.6 | 67/79    | 84.8 | 82.0  | 74/93 | 79.6 | 67/79      | 84.8 | 82.0  |
| TMinCirc  | 1.171 | 71.36            | 67/93 | 72.0 | 64/79    | 81.0 | 76.2  | 67/93 | 72   | 64/79      | 81.0 | 76.2  |
| TUB       | 1.171 | 71.81            | 76/93 | 81.7 | 65/79    | 82.3 | 82.0  | 76/93 | 81.7 | 64/79      | 81.0 | 81.4  |
| TLB       | 1.170 | 42.78            | 76/92 | 82.6 | 67/77    | 87.0 | 84.6  | 76/92 | 82.6 | 67/77      | 87.0 | 84.6  |
| FibL      | 1.171 | 344.18           | 76/92 | 82.6 | 67/77    | 87.0 | 84.6  | 76/92 | 82.6 | 67/77      | 87.0 | 84.6  |

Table 7.1.2.2 Univariate statistics for the measurements of upper and lower limb bones.

## Humerus

About 91.9% of cases were correctly classified when all humeral measurements were applied jointly, forming function HF1 (Table 7.1.3). Stepwise discriminant function analysis (HF2) selected only 4 dimensions (HML, VHD, HMinMid and HBB), producing an accuracy rate of 91.3%. Assuming that distal epiphysis is missing, a stepwise DFA was performed, forming function HF3. When proximal epiphysis is not present stepwise DFA selects 2 variables forming HF4. HF5 is the result of a direct DFA using HVD and HBB,

|      |           |          |       |                  |       |      | Original |      |       |       | С    | ross validate | ed   |       |
|------|-----------|----------|-------|------------------|-------|------|----------|------|-------|-------|------|---------------|------|-------|
|      | Functions |          |       |                  | Ma    | le   | Fema     | ale  | Total | Mal   | e    | Fema          | ale  | Total |
| Step | HF1       | F-ratios | df    | Raw coefficients | N     | %    | N        | %    | %     | N     | %    | N             | %    | %     |
| 1    | HML       | 158.73   | 1.172 | 0.029458         |       |      |          |      |       |       |      |               |      |       |
|      | HVD       | 203.69   | 1.172 | 0.186518         |       |      |          |      |       |       |      |               |      |       |
|      | HMaxMid   | 88.04    | 1.172 | 0.173062         |       |      |          |      |       |       |      |               |      |       |
|      | HMinMid   | 128.74   | 1.172 | 0.245281         | 85/94 | 90.4 | 74/79    | 93.7 | 91.9  | 85/94 | 90.4 | 72/79         | 91.1 | 90.8  |
|      | HMidCirc  | 107.60   | 1.172 | -0.097004        |       |      |          |      |       |       |      |               |      |       |
|      | HBB       | 171.91   | 1.172 | 0.087156         |       |      |          |      |       |       |      |               |      |       |
|      | Constant  |          |       | -25.640318       |       |      |          |      |       |       |      |               |      |       |
|      | HF2       |          |       |                  |       |      |          |      |       |       |      |               |      |       |
| 1    | HVD       | 203.69   | 1.171 | 0.1883148        |       |      |          |      |       |       |      |               |      |       |
| 2    | HML       | 138.27   | 2.170 | 0.02852316       | 86/04 | 01.5 | 72/70    | 01.5 | 01.2  | 86/04 | 01.5 | 71/70         | 80.0 | 00.8  |
| 3    | HBB       | 101.79   | 3.169 | 0.07824173       | 00/94 | 91.5 | 12/19    | 91.5 | 91.5  | 00/94 | 91.5 | /1//9         | 69.9 | 90.8  |
| 4    | HMinMid   | 78.64    | 4.168 | 0.13910141       |       |      |          |      |       |       |      |               |      |       |
|      | Costant   |          |       | -23.927105       |       |      |          |      |       |       |      |               |      |       |
|      | HF3       |          |       |                  |       |      |          |      |       |       |      |               |      |       |
| 1    | HVD       | 203.69   | 1.171 | 0.307412         | 82/04 | 87.2 | 72/70    | 01.1 | 80.0  | 82/04 | 87.2 | 72/70         | 01.1 | 80.0  |
| 2    | HMinMid   | 123.09   | 2.170 | 0.292621         | 82/94 | 07.2 | 12/19    | 71.1 | 89.0  | 62/94 | 07.2 | 12/19         | 91.1 | 89.0  |
|      | constant  |          |       | -18.45303        |       |      |          |      |       |       |      |               |      |       |
|      | HF4       |          |       |                  |       |      |          |      |       |       |      |               |      |       |
| 1    | HBB       | 171.91   | 1.171 | 0.311035         | 91/04 | 96.7 | 71/70    | 80.0 | 87.0  | 91/04 | 96.7 | 71/70         | 80.0 | 87.0  |
| 2    | HMinMid   | 105.19   | 2.170 | 0.1834           | 01/94 | 80.2 | /1//9    | 89.9 | 07.9  | 01/94 | 80.2 | /1//9         | 89.9 | 07.9  |
|      | Constant  |          |       | -15.937176       |       |      |          |      |       |       |      |               |      |       |
|      | HF5       |          |       |                  |       |      |          |      |       |       |      |               |      |       |
| 1    | HVD       | 203.69   | 1.172 | 0.268776         | 82/04 | 07.0 | 72/70    | 02.4 | 80.5  | 82/04 | 07.0 | 72/70         | 02.4 | 80.5  |
| 2    | HBB       | 171.91   |       | 0.13521          | 82/94 | 87.2 | 13/19    | 92.4 | 89.0  | 82/94 | 81.2 | 13/19         | 92.4 | 89.0  |
|      | Constant  |          |       | -19.591343       |       |      |          |      |       |       |      |               |      |       |

# Table 7.1.2.3 Discriminant functions and classification accuracies for the humerus. The sectioning point is set to zero in all cases.

|      |           |          |       |                     |       |       | Original |       |       |       | С     | ross validat | ed    |       |
|------|-----------|----------|-------|---------------------|-------|-------|----------|-------|-------|-------|-------|--------------|-------|-------|
|      | Functions |          |       |                     | Ma    | le    | Fen      | nale  | Total | Ma    | ale   | Fen          | nale  | Total |
| Step | UF1       | F-ratios | df    | Raw<br>coefficients | N     | %     | N        | %     | %     | N     | %     | N            | %     | %     |
| 1    | UL        | 110.81   | 1.168 | 0.040967            |       |       |          |       |       |       |       |              |       |       |
|      | UNH       | 53.08    | 1.168 | 0.167036            | 81/02 | 88.04 | 69/77    | 99 21 | 00 7  | 80/02 | 86.06 | 69/77        | 99.21 | 97.6  |
|      | UDB       | 51.1     | 1.168 | 0.203402            | 01/92 | 88.04 | 08/77    | 00.51 | 00.2  | 80/92 | 80.90 | 08/77        | 00.51 | 87.0  |
|      | Constant  |          |       | -17.7172            |       |       |          |       |       |       |       |              |       |       |
|      | UF2       |          |       |                     |       |       |          |       |       |       |       |              |       |       |
| 1    | UL        | 110.81   | 1.168 | 0.050112            | 82/02 | 00.22 | 70/77    | 00.01 | 00.5  | 82/02 | 00.22 | 60/77        | 90.61 | 80.0  |
|      | UDB       | 51.1     | 1.168 | 0.222482            | 03/92 | 90.22 | 10/11    | 90.91 | 90.5  | 65/92 | 90.22 | 09/11        | 89.01 | 89.9  |
|      | Constant  |          |       | -16.654983          |       |       |          |       |       |       |       |              |       |       |
|      | RF1       |          |       |                     |       |       |          |       |       |       |       |              |       |       |
|      | RL        | 219.92   | 1.172 | 0.059914            |       |       |          |       |       |       |       |              |       |       |
| 1    | RHD       | 172.34   | 1.172 | 0.314912            | 88/94 | 93.62 | 88/94    | 94.94 | 94.28 | 75/79 | 93.62 | 75/79        | 93.7  | 93.64 |
|      | RDB       | 70.9     | 1.172 | 0.051598            |       |       |          |       |       |       |       |              |       |       |
|      | Constant  |          |       | -21.703419          |       |       |          |       |       |       |       |              |       |       |
|      | RF2       |          |       |                     |       |       |          |       |       |       |       |              |       |       |
| 1    | RL        | 219.92   | 1.172 | 0.061252            | 80/04 | 04.69 | 76/70    | 06.2  | 05.44 | 80/04 | 04.69 | 76/70        | 06.2  | 05.44 |
| 2    | RHD       | 142.54   | 2.171 | 0.361646            | 89/94 | 94.08 | /0//9    | 96.2  | 95.44 | 89/94 | 94.08 | /0//9        | 90.2  | 95.44 |
|      | Constant  |          |       | -0.112053           |       |       |          |       |       |       |       |              |       |       |
|      | RF3       |          |       |                     |       |       |          |       |       |       |       |              |       |       |
| 1    | RDB       | 70.9     | 1.172 | 0.083873            | 77/04 | 01.01 | 70/70    | 00.41 | 06.71 | 77/04 | 01.01 | 70/70        | 01.1  | 06.12 |
|      | RHD       | 172.34   | 1.172 | 0.603633            | 1//94 | 81.91 | 13/19    | 92.41 | 86.71 | 77/94 | 81.91 | 12/19        | 91.1  | 86.13 |
|      | Constant  |          |       | -15.242627          |       |       |          |       |       |       |       |              |       |       |

# Table 7.1.2.4: Discriminant functions and classification accuracies for the ulna and the radius. The sectioning point is set to zero in all cases.

which resulted in 90% correct classification for both the original and cross-validated sample.

# Ulna

Function UF1 demonstrates the result of a direct discriminant function analysis using all ulnar dimensions. Classification accuracy reached 88.2% for the original and 87.6% for the cross-validated sample. The same results were produced using the stepwise procedure. Function 2 (UF2) is the result of a direct discriminant function analysis using UL and UDB, which performed better than UF1, yielding 91% accuracy. UF3 employed UL and UNH, resulting in 90% correct group membership. All functions, the correspondent coefficients, the constants and the accuracies for the original jack-knife procedure are presented in Table 7.1.2.4

#### Radius

Function RF1 demonstrates the result of a direct discriminant function analysis using all three radial dimensions. Stepwise analysis selected RL and RHD to enter into the formula (RF2). According to RF2, the sex can be calculated by multiplying the values of the two dimensions by the corresponding coefficients minus the constant, as can be seen in Table 7.1.2.4. Values greater than zero indicate a male individual, otherwise the sample is a female. RF3 is the result of a direct DFA using RL and RDB, whereas RF4 employs RL and RHD. RF2 exhibits the highest classification accuracy in sex determination using the radius, reaching 95.4% for both original and cross-validation sample.

When all upper limb measurements were subjected to discriminant function analysis, stepwise procedure selected 4 of them (HVD, HDB, RL and UNH). Table 7.1.2.5 illustrates the corresponding unstandardized coefficients and the constant for upper limb function (UL). Classification accuracy reached 95.3%, with better result in males (96.8%) compared to females (93.6%). Cross-validation produced the same accuracy with the original sample for males and slightly lower for females (91%) (Table 7.1.2.6).

#### Lower Extremity

Descriptive statistics of all lower limb measurements and associated univariate F-ratio to measure the differences between the sexes are shown in Table 7.1.2.1. The differences between the means in males and females are significant (p<0.0001). The differences between the means of right and left lower limb bones were compared and found not to differ significantly between the sexes. Therefore right bones were also used in the analysis.

#### Univariate statistics

Single dimensions of the femur, tibia and fibula were submitted to direct analysis and produced demarking values that separated males from females (Table 7.1.2.2).

|      | *Functions | E-ratio | df    | Raw          |
|------|------------|---------|-------|--------------|
| step | ULF        | 1-1410  | u     | coefficients |
| 1    | HVD        | 215.81  | 1.167 | 0.141042     |
| 2    | HEB        | 156.06  | 2.166 | 0.098625     |
| 3    | RL         | 112.74  | 3.165 | 0.046541     |
| 4    | UNH        | 87.537  | 4.164 | 0.090232     |
|      | Constant   |         |       | -24.3845     |
|      | LLF        |         |       |              |
| 1    | FHMaxD     | 163.97  | 1.165 | 0.2258993    |
| 2    | NFMax      | 117.94  | 2.164 | 0.2526981    |
| 3    | FiL        | 89.134  | 3.163 | 0.0259799    |
| 4    | FeMidC     | 69.108  | 4.162 | -0.051879    |
|      | Constant   |         |       | -22.961239   |

Table 7.1.2.5: Results of stepwise discriminant function analysis for upper and lower limb.

### Femur

The most effective single dimensions as determined by direct discriminant analysis were FDB (86.5%) followed by FHMaxD (83.7%) and FBL (82%). According to the results, an individual with FHMaxD smaller than 44.85 mm was assigned as female, while otherwise it was assigned as male. Interestingly, the variables that performed the worst were the 3 measurements taken on the midshaft (FMidap, FMidtr and FMidCirc)(Table 7.1.2.2).

|       |      | Original |      |       |       | Cr   | oss-validate | ed   |       |
|-------|------|----------|------|-------|-------|------|--------------|------|-------|
| Mal   | es   | Fema     | les  | Total | Mal   | es   | Fema         | ıles | Total |
| N     | %    | N        | %    | %     | N     | %    | N            | %    | %     |
| 90/93 | 96.8 | 73/78    | 93.6 | 95.3  | 90/93 | 96.8 | 71/78        | 91   | 94.2  |
| 85/92 | 92.4 | 69/76    | 90.8 | 91.7  | 85/92 | 92.4 | 69/76        | 90.8 | 91.7  |

Table 7.1.2.6 Classification accuracy for upper and lower limb in Cretans

# Tibia

The most effective single dimension for the tibia was TLB (84.6%), followed by TUB (82%) and NFMax (82%). The least discriminating variable was found to be NFmin (71%), followed by TminCirc (76%). Cross-validation was close to the original data in all cases. All demarking points and accuracies for both original and jack-knifed data are presented in Table 7.1.2.2.

<sup>\*</sup> The sectioning point is set to zero

# Fibula

Fi1 is the result of a direct discriminant procedure using FL as a variable to separate the sexes. Any individual with FL greater than 344.2 was assigned as male, otherwise as female. Cross-validation procedure produced results very close to the original classification in all cases. All classification results and leave-one-out classification are presented in Table 7.1.2.2.

## Multivariate statistics

#### Femur

Several discriminant functions were generated using femoral dimensions. F1 demonstrates the result of a direct discriminant function analysis using 8 femoral dimensions (FBL, FHMaxD, FSubTap, FSubTtr, FMidap, FMidt, FMidCirc and FDB). Classification accuracy reached 91% for the original and 89% for the cross-validated sample. Among the two length variables (FBL and FML), FBL was used because it presented a higher F-value (Table 7.1.2.1). Function two (F2) uses the same eight variables as F1 following a stepwise procedure and three dimensions are entered into the equation (FHMaxD, FBL and FDB), resulting in 90.7% accuracy. Assuming different fragmentary patterns, multiple functions were generated, giving an accuracy rate from 76.3% to 87.2 % (F3-F6). F3 assumed that the distal femur was missing, while midshaft and upper epiphyseal parts are present. In that case, six dimensions (FHMaxD, FSubTap, FSubTtr, FMidap, FMidt and FMidCirc) were submitted to stepwise procedure and only two of them (FHMaxD and FMidap) were selected, resulting in 85% correct group membership. F4 assumed that upper epiphyseal part including major trochanter was present. Stepwise procedure selected in that case two out of three dimensions. If only the femoral midshaft was present (F5), classification dropped to 76.3%. (Table 7.1.2.7).

#### Tibia

T1 demonstrated the result of a direct discriminant function analysis using all tibial dimensions. Function two (TF2) used the same eight variables as TF1 following a stepwise procedure and three dimensions were entered into the equation (TL, NFMax and TUB). Different preservation was assumed in functions TF2 and TF4. TF3 assumed that the proximal epiphysis was missing and five dimensions (NFMax, NFMin, NFCirc, TminCirc and TLB) were submitted to stepwise procedure, of which only two of them (NFMax and TLB) were selected. TF4 assumed that the lower epiphyseal part was missing. Stepwise procedure selected in that case two (NFMax and TUB) out of four (NFMax, NFMin, NFCirc and TUB) dimensions (Table 7.1.2.8). 91.2%f the cases were correctly classified when all seven measurements of the tibia were applied jointly.

| Stan | V          | F-     | df    | Raw          | Ma    | le       | Fem   | ale  | Total |           |
|------|------------|--------|-------|--------------|-------|----------|-------|------|-------|-----------|
| Step | v          | ratios | u     | coefficients | N     | %        | N     | %    | %     |           |
|      |            |        |       | F            | 1     |          |       |      |       |           |
| 1    | FBL        | 138.18 | 1.171 | 0.02249      | 84/94 | 89.0     | 73/78 | 93.6 | 91.3  | Original  |
|      | FHMaxD     | 169.64 | 1.171 | 0.18502      | 82/04 | 07.0     | 71/70 | 01.0 | 80.0  | Cross-    |
|      | FSubTap    | 59.71  | 1.171 | 0.00795      | 02/94 | 87.0     | /1//0 | 91.0 | 89.0  | validated |
|      | FSubTtr    | 37.86  | 1.171 | -0.01287     |       |          |       |      |       |           |
|      | FMidap     | 73.64  | 1.172 | 0.1098       |       |          |       |      |       |           |
|      | FMidtr     | 25.84  | 1.172 | 0.00173      |       |          |       |      |       |           |
|      | FMidCirc   | 73.8   | 1.172 | -0.01719     |       |          |       |      |       |           |
|      | FDB        | 136.37 | 1.172 | 0.06827      |       |          |       |      |       |           |
|      | (Constant) |        |       | -24.5392     |       |          |       |      |       |           |
|      |            |        |       | F            | 2     |          |       |      |       |           |
| 1    | FHMaxD     | 169.64 | 1.170 | 0.1985       | 84/94 | 89.0     | 72/78 | 92.3 | 90.7  | Original  |
| 2    | FBL        | 110.71 | 2.169 | 0.02453      | 82/04 | <u> </u> | 71/79 | 01.0 | 80.5  | Cross-    |
| 3    | FDB        | 76.93  | 3.168 | 0.07056      | 03/94 | 88.0     | /1//0 | 91.0 | 09.5  | validated |
|      | (Constant) |        |       | -24.7657     |       |          |       |      |       |           |
|      |            |        |       | F            | 3     |          |       |      |       |           |
| 1    | FHMaxD     | 169.64 | 1.170 | 0.34181      | 82/94 | 87.0     | 68/78 | 87.2 | 87.2  | Original  |
| 2    | FMidap     | 94.87  | 2.169 | 0.15636      | 82/04 | 87.0     | 69/79 | 87.2 | 87.2  | Cross-    |
|      | (Constant) |        |       | -19.671      | 02/94 | 87.0     | 00/70 | 07.2 | 07.2  | validated |
|      |            |        |       | F            | 4     |          |       |      |       |           |
| 1    | FHMaxD     | 169.64 | 1.170 | 0.36337      | 80/94 | 85.0     | 67/78 | 85.9 | 85.5  | Original  |
| 2    | FSubTap    | 88.57  | 2.169 | 0.10424      | 80/04 | 85.0     | 67/78 | 85.0 | 85.5  | Cross-    |
|      | (Constant) |        |       | -19.0239     | 80/94 | 85.0     | 07/78 | 05.9 | 85.5  | validated |
|      |            |        |       | F            | 5     |          |       |      |       |           |
| 1    | FMidap     | 73.8   | 1.171 | 0.24414      | 68/94 | 72.0     | 64/79 | 81.0 | 76.3  | Original  |
| 2    | FMidCirc   | 44.19  | 2.170 | 0.10773      | 68/04 | 72.0     | 64/70 | 81.0 | 76.3  | Cross-    |
|      | (Constant) |        |       | -16.0591     | 00/94 | 12.0     | 04/19 | 01.0 | 70.5  | validated |
|      |            |        |       | F            | 6     |          |       |      |       |           |
| 1    | FMidap     | 136.37 | 1.171 | 0.20046      | 76/94 | 81.0     | 71/79 | 89.9 | 85.0  | Original  |
| 2    | FDB        | 83.38  | 2.170 | 0.19487      | 76/04 | 91.0     | 71/70 | 80.0 | 95.0  | Cross-    |
|      | (Constant) |        |       | -20.7037     | /0/94 | 01.0     | /1//9 | 09.9 | 63.0  | validated |

Table 7.1.2.7 Discriminant functions and classification accuracies for the femur. The sectioning point is set to zero.

Cross-validation gave slightly lower accuracy (88.9%). When stepwise procedure was used, classification accuracy dropped to 90.7%. Assuming different fragmentary patterns, multiple functions were generated giving an accuracy rate from 87.7% to 89.0 % (TF3-TF4).

When all lower limb measurements were subjected to DFA, stepwise procedure selected 4 of them (FHD, FMidCirc, NFMax and FiL) (see Table 7.1.2.5). Classification

accuracy resulted in 91.7% for both original and cross-validated data, with slightly better results for males (92.4%) compared to females (90.8%) (see Table 7.1.2.6).

| Stop | V          | F-     | df    | Raw          | Ma    | le   | Fem   | ale  | Total |           |
|------|------------|--------|-------|--------------|-------|------|-------|------|-------|-----------|
| Step | v          | ratios | u     | coefficients | N     | %    | N     | %    | %     |           |
|      |            |        |       | TI           | 71    |      |       |      |       |           |
| 1    | TL         | 116.17 | 1.17  | 0.0179       | 87/93 | 93.5 | 68/78 | 88.5 | 91.2  | Original  |
|      | NFMax      | 157.24 | 1.17  | 0.2279       | 96/02 | 02.5 | 66/70 | 946  | 000   | Cross-    |
|      | NFMin      | 58.59  | 1.17  | -0.0435      | 80/95 | 92.5 | 00/78 | 84.0 | 00.9  | validated |
|      | NFCirc     | 135.42 | 1.17  | 0.0091       |       |      |       |      |       |           |
|      | TMinCirc   | 87.29  | 1.17  | -0.0096      |       |      |       |      |       |           |
|      | TUB        | 143.4  | 1.17  | 0.0775       |       |      |       |      |       |           |
|      | TLB        | 118.5  | 1.17  | 0.0814       |       |      |       |      |       |           |
|      | (Constant) |        |       | -21.9084     |       |      |       |      |       |           |
|      |            |        |       | TI           | 72    |      |       |      |       |           |
| 1    | TL         | 154.36 | 1.169 | 0.0197       | 87/93 | 93.5 | 69/79 | 87.3 | 90.7  | Original  |
| 2    | NFMax      | 107.31 | 2.168 | 0.239        | 86/02 | 02.5 | 60/70 | 07.2 | 00.1  | Cross-    |
| 3    | TUB        | 78.76  | 3.167 | 0.1001       | 80/95 | 92.5 | 09/19 | 07.5 | 90.1  | validated |
|      | (Constant) |        |       | -21.9317     |       |      |       |      |       |           |
|      |            |        |       | TI           | 73    |      |       |      |       |           |
| 1    | NFMax      |        |       | 0.2972       | 83/93 | 89.2 | 67/79 | 85.9 | 87.7  | Original  |
| 2    | TLB        | 154.36 | 1.169 | 0.1823       | 82/02 | 00 7 | 67/70 | 85.0 | 97.1  | Cross-    |
|      | (Constant) | 99.31  | 2.168 | -17.6135     | 82/93 | 00.2 | 07/79 | 05.9 | 07.1  | validated |
|      |            |        |       | TF           | 54    |      |       |      |       |           |
| 1    | TUB        | 157.24 | 1.169 | 0.1466       | 85/93 | 91.4 | 68/79 | 86.1 | 89    | Original  |
| 2    | NFMax      | 106.66 | 2.168 | 0.2724       | 94/02 | 00.2 | 69/70 | 96.1 | 00 /  | Cross-    |
|      | (Constant) |        |       | -19.5209     | 04/93 | 90.5 | 00/19 | 00.1 | 00.4  | validated |

Table 7.1.2.8 Discriminant functions and classification accuracies for the tibia. The sectioning point is set to zero.

#### 7.1.2.2 Posterior probabilities

Posterior probabilities of each case were also calculated, since they better reflected the affinity of each case to be reassigned to the original group according to the value of the discriminant score. Discriminant scores close to zero (which is set as the sectioning point in all discriminant functions) fall in the area of overlap between the two groups, hence the estimation of sex is likely to be uncertain. Posterior probabilities allow the calculation of the probability of a case to belong to the male or the female group. For sex determination, three thresholds were considered (0.8, 0.9 and 0.95). In order to evaluate the accuracy of the given formulae, posterior probabilities were calculated for all functions which resulted in more than 80% classification accuracy.

#### Univariate statistics

# Humerus

Posterior probabilities for the five single variables of the humerus are presented in Table 7.1.2.9 The most reliable single dimension proved to be HVD, which classified 57% of the specimens at a 0.9 threshold and 40% at a 0.95 threshold. An individual with a discriminant score over 1.0 had 90% probability of belonging to the male group, whereas if the discriminant score fell above 1.38, it had a 95% probability of belonging to the male group. Now the cut-off value for the HVD was 43.76mm. Values over 46.2mm suggested that the specimen had a 90% probability of belonging to the male group, while values over 47.1mm assigned it as male within a 95% confidence interval. The accuracy rate for HVD was 90%.

| PP  | Mal    | es   | Fema    | lles | Total | Male   | es   | Fema   | ıles | Total |
|-----|--------|------|---------|------|-------|--------|------|--------|------|-------|
| (%) | <      | %    | >       | %    | %     | <      | %    | >      | %    | %     |
|     |        |      | HL      |      |       |        |      | RL     |      |       |
| >95 | 330.00 | 30.8 | 286.00  | 27.8 | 29.5  | 241.00 | 38.3 | 211.00 | 43.0 | 40.5  |
| >90 | 325.00 | 40.4 | 291.00  | 48.1 | 43.9  | 237.00 | 57.4 | 215.00 | 62.0 | 58.4  |
| >80 | 318.00 | 56.4 | 296.00  | 62.0 | 66.5  | 233.00 | 74.5 | 218.00 | 72.2 | 73.4  |
| >50 | 307.80 | 86.2 | 307.80  | 82.3 | 84.4  | 225.80 | 91.5 | 225.80 | 89.9 | 90.8  |
|     |        |      | HVD     |      |       |        |      | RVD    |      |       |
| >95 | 47.10  | 43.6 | 40.40   | 35.4 | 39.9  | 32.77  | 33.0 | 23.92  | 26.6 | 30.0  |
| >90 | 46.18  | 54.3 | 41.20   | 59.5 | 56.6  | 31.56  | 45.7 | 24.93  | 49.4 | 47.4  |
| >80 | 45.30  | 69.1 | 42.20   | 72.1 | 70.5  | 30.36  | 58.5 | 26.91  | 69.6 | 63.6  |
| >50 | 43.80  | 89.4 | 43.80   | 89.9 | 89.6  | 28.34  | 79.8 | 28.34  | 93.7 | 86.1  |
|     |        | Max  | MidD    |      |       |        |      | RD     |      |       |
| >95 | 25.00  | 9.6  | 17.70   | 6.3  | 8.1   | 24.10  | 2.1  | 13.70  | 7.6  | 4.6   |
| >90 | 23.90  | 17.0 | 18.80   | 20.2 | 18.5  | 22.90  | 5.3  | 18.54  | 16.5 | 10.4  |
| >80 | 23.00  | 40.4 | 19.70   | 38.0 | 43.9  | 19.70  | 28.7 | 19.74  | 22.8 | 26.0  |
| >50 | 21.30  | 74.5 | 21.30   | 81.0 | 87.3  | 21.30  | 85.1 | 21.30  | 74.7 | 80.3  |
|     |        | ]    | MinMidD |      |       |        |      | UL     |      |       |
| >95 | 19.80  | 20.2 | 0.43    | 27.8 | 23.7  | 276.00 | 7.5  | 215.00 | 3.8  | 5.8   |
| >90 | 19.20  | 33.0 | 0.70    | 35.4 | 34.1  | 267.00 | 18.3 | 223.00 | 23.1 | 20.5  |
| >80 | 18.40  | 55.3 | 1.17    | 51.9 | 53.7  | 259.00 | 35.5 | 231.00 | 56.4 | 45.0  |
| >50 | 17.10  | 80.8 | 17.10   | 82.3 | 81.5  | 245.10 | 89.2 | 245.10 | 88.5 | 88.9  |
|     |        |      | BB      |      |       |        |      |        |      |       |
| >95 | 63.50  | 33.0 | 52.30   | 32.9 | 32.9  |        |      |        |      |       |
| >90 | 62.10  | 45.7 | 53.60   | 44.3 | 50.9  |        |      |        |      |       |
| >80 | 60.70  | 59.6 | 55.20   | 68.3 | 63.6  |        |      |        |      |       |
| >50 | 57.90  | 83.0 | 57.90   | 88.6 | 85.5  |        |      |        |      |       |

Table 7.1.2.9 Posterior probabilities for single dimensions of the upper limb bones

# Radius

All radial measurements gave classification accuracies over 80%. The most effective dimension was proven to be radial length, which classified over 40% of the sample at a 0.95 threshold with 91% accuracy. On the contrary, RD (distal breadth) hardly assigned 5% of the specimens within 95% of confidence interval and 10% within 90% of confidence interval. A radius with total length over 241mm had a 95% probability of being male, whereas a radius with total length less than 211mm was 95% likely to be female (Table 7.1.2.9).

| PP  | Mal     | e     | Fema     | le    | Total | Mal                              | e    | Femal             | e         | Total |  |  |
|-----|---------|-------|----------|-------|-------|----------------------------------|------|-------------------|-----------|-------|--|--|
| (%) | DS      | %     | DS       | %     | %     | DS                               | %    | DS                | %         | %     |  |  |
|     |         | Н     | IF1      |       |       |                                  |      | RF2               |           |       |  |  |
| >95 | >1.0871 | 60.6  | <-1.1068 | 65.8  | 63.0  | >1.1491                          | 53.2 | <-1.2501          | 53.2      | 53.2  |  |  |
| >90 | >0.8455 | 70.2  | <-0.8594 | 81.00 | 72.6  | >0.8767                          | 61.7 | <-0.9579          | 63.3      | 62.4  |  |  |
| >80 | >0.5153 | 79.7  | <-0.5412 | 86.00 | 82.6  | >0.5174                          | 75.5 | <-0.6073          | 77.2      | 76.3  |  |  |
| >50 | >0      | 90.5  | <0       | 93.7  | 91.9  | >0                               | 94.7 | <0                | 94.9      | 95.0  |  |  |
|     |         | Н     | IF2      |       |       |                                  |      | RF3               |           |       |  |  |
| >95 | >1.1335 | 58.5  | <-1.1224 | 63.3  | 60.7  | >1.2390                          | 48.9 | 8.9 <-1.2294 49.4 |           |       |  |  |
| >90 | >0.8230 | 63.8  | <-0.8982 | 79.7  | 71.1  | >0.9458                          | 64.9 | <-0.9312          | 64.6      | 64.7  |  |  |
| >80 | <0.5212 | 67.00 | <-0.6097 | 87.3  | 76.3  | >0.6277                          | 78.7 | <-0.6533          | 72.2      | 75.7  |  |  |
| >50 | >0      | 93.6  | <0       | 89.9  | 91.9  | >0                               | 92.6 | <0                | <0 91.1 9 |       |  |  |
|     |         | Н     | IF3      |       |       |                                  |      | RF4               |           |       |  |  |
| >95 | >1.1062 | 63.8  | <-1.0801 | 67.1  | 65.3  | >1.35                            | 35.1 | .1 <-1.55 30.4    |           |       |  |  |
| >90 | >0.8623 | 70.2  | <-0.8180 | 79.7  | 74.6  | >0.98                            | 45.7 | <-1.18            | 49.4      | 47.4  |  |  |
| >80 | >0.5229 | 77.6  | <-0.6704 | 84.8  | 80.9  | >0.59                            | 62.8 | <-0.82            | 67.1      | 64.7  |  |  |
| >50 | >0      | 91.5  | <0       | 91.1  | 91.3  | >0                               | 81.9 | <0                | 92.4      | 86.7  |  |  |
|     |         | Н     | IF4      |       |       |                                  |      | UF1               |           |       |  |  |
| >95 | >1.7499 | 22.3  | <-1.6600 | 26.5  | 24.3  | >1.4698                          | 26.1 | <-14698           | 28.6      | 27.2  |  |  |
| >90 | >1.2676 | 37.2  | <-1.3079 | 38.00 | 37.5  | >1.1049                          | 41.3 | <-1.0921          | 49.3      | 45    |  |  |
| >80 | >0.8546 | 54.2  | <-0.8036 | 58.2  | 56.1  | >0.6932                          | 57.6 | <-0.7459          | 68.8      | 62.7  |  |  |
| >50 | >0      | 77.6  | <0       | 84.8  | 80.9  | >0                               | 88.0 | <0                | 88.3      | 88.2  |  |  |
|     |         | Н     | IF5      |       |       |                                  |      | UF2               |           |       |  |  |
| >95 | >1.2093 | 51.1  | <-1.2563 | 51.9  | 56.6  | >1.1506                          | 19.6 | <-1.7092          | 16.9      | 18.3  |  |  |
| >90 | >0.9381 | 62.8  | <-0.9478 | 70.9  | 66.5  | >1.1799                          | 34.8 | <-1.1856          | 32.5      | 33.7  |  |  |
| >80 | >0.5902 | 76.6  | <-0.6520 | 79.7  | 78.00 | >0.7681                          | 50.0 | <-0.7573          | 68.8      | 58.6  |  |  |
| >50 | >0      | 87.2  | <0       | 92.4  | 89.6  | >0                               | 90.2 | <0                | 90.9      | 90.5  |  |  |
|     |         | R     | :F1      |       |       |                                  | ULF  |                   |           |       |  |  |
| >95 | >1.1285 | 57.4  | <-1.2599 | 57    | 57.2  | 57.2 >1.021 67.7 <-1.0877 70.5 6 |      |                   |           | 69    |  |  |
| >90 | >0.8588 | 62.8  | <-0.8145 | 68.4  | 65.3  | >0.8419                          | 79.6 | <-0.7590          | 82.1      | 80.7  |  |  |
| >80 | >0.6193 | 76.6  | <-0.5630 | 75.9  | 76.3  | >0.477                           | 87.1 | <-0.5020          | 85.9      | 86.5  |  |  |
| >50 | >0      | 93.6  | <0       | 94.9  | 94.2  | >0                               | 96.8 | <0                | 93.6      | 95.3  |  |  |

Table 7.1.2.10 Posterior probabilities for multivariate functions of the upper limb bones.

# Ulna

Maximum length was the only measurement that performed above 80% for the ulna. However, only 6% of the specimens were classified within a 95% interval of confidence (Table 7.1.2.9). A maximum length under 215 mm classified the specimen as female at a 0.95 threshold, under 223mm at a 0.9 threshold and under 231mm at a 0.8 threshold with 89% accuracy.

# Multivariate statistics

## Humerus

Posterior probabilities were also calculated for the multivariate discriminant functions. The best discriminating function was HF3 (HVD, HML, HBB and HMinMid). At a 0.95 threshold, sex was determined for 65.5% of the sample with 91% accuracy. Discriminant scores over 1.1062 classified males with 95% probability, while values smaller than -1.108 assigned females at the same threshold of 0.95. PP for all multivariate discriminant functions of the humerus and corresponding discriminant scores are presented in Table 7.1.2.10.

#### Radius

The best discriminant function for the radius was RF2 with 95% accuracy. At a 95% threshold, though, RF1 performed better. 57% of the sample was assigned correctly (within 95% intervals of confidence) with 94.2% accuracy. The cut-off discriminant score for males was 1.125 and for females -1.2599. RF4 performed worse, classifying only 33% of the sample at a 0.95 threshold with 86.7% accuracy. All posterior probabilities for RF1-RF4 are presented in Table 7.1.2.10.

# Ulna

Posterior probabilities for the ulna were low for all functions at a 95% threshold. The best function was UF1, which classified about 27% of the specimens with 88.2% accuracy. UF2 exhibited higher accuracies (up to 91%). However, according to this function only 18% of the sample could be classified within 95% of confidence intervals (Table 7.1.2.10).

ULF is the result of a stepwise discriminant function analysis using all measurements of the upper limb bones. This function classified over 80% of the sample at a 0.9 threshold and 69% at a 0.95 threshold with 95.3% accuracy (Table 7.1.2.10). Discriminant scores over 1.021 classified males with 95% probability, while values smaller than -1.0877 assigned females at the same threshold of 0.95.

#### Univariate statistics

# Femur

Femoral dimensions classified the sample up to 47% (FHD) at a 90% threshold and up to 28% (FHD) at a 0.95 threshold, implying that there is a considerable overlap between the two groups. A specimen was considered as male when the FHD measured more than 48.5 mm and female when FHD<41.2mm within 95% of confidence interval with 84% accuracy. Posterior probabilities for all single femoral dimensions that exhibited over 80% accuracy are presented in Table 7.1.2.11.

| PP  | Mal    | es   | Fema   | les  | Total | Mal    | es    | Fema   | les  | Total |
|-----|--------|------|--------|------|-------|--------|-------|--------|------|-------|
| (%) | <      | %    | >      | %    | %     | <      | %     | >      | %    | %     |
|     |        |      | FeBL   |      |       |        |       | TL     |      |       |
| >95 | 456.00 | 26.6 | 390.00 | 26.5 | 25.0  | 381.00 | 18.3  | 314.00 | 12.7 | 15.7  |
| >90 | 450.00 | 36.2 | 399.00 | 42.3 | 38.9  | 373.00 | 34.4  | 323.00 | 27.9 | 31.4  |
| >80 | 439.00 | 55.3 | 407.00 | 60.3 | 57.6  | 364.00 | 48.4  | 332.00 | 50.6 | 49.4  |
| >50 | 423.23 | 86.2 | 423.23 | 76.9 | 82.0  | 347.76 | 78.5  | 347.76 | 81.0 | 79.7  |
|     |        |      | FeL    |      |       |        |       | NFmax  |      |       |
| >95 | 460.00 | 25.5 | 392.00 | 23.1 | 24.4  | 36.60  | 25.8  | 29.30  | 30.4 | 30.2  |
| >90 | 451.00 | 36.2 | 401.00 | 38.5 | 37.2  | 35.90  | 44.1  | 30.30  | 45.6 | 44.8  |
| >80 | 442.00 | 55.3 | 409.00 | 60.3 | 57.6  | 34.70  | 59.1  | 33.20  | 58.2 | 58.7  |
| >50 | 425.98 | 85.1 | 425.98 | 74.4 | 80.2  | 33.02  | 80.7  | 33.02  | 83.5 | 82.0  |
|     |        |      | FHMaxD |      |       |        |       | NFCirc |      |       |
| >95 | 48.50  | 27.7 | 41.20  | 28.2 | 27.9  | 100.00 | 25.8  | 79.00  | 17.7 | 22.1  |
| >90 | 47.80  | 42.6 | 42.10  | 50.0 | 45.9  | 97.00  | 37.6  | 81.00  | 39.2 | 38.4  |
| >80 | 46.60  | 62.8 | 43.10  | 64.1 | 63.4  | 94.00  | 55.9  | 84.00  | 60.8 | 58.1  |
| >50 | 44.85  | 84.0 | 44.85  | 83.3 | 83.7  | 89.25  | 79.6  | 89.25  | 84.8 | 82.0  |
|     |        |      | FBD    |      |       |        |       | UB     |      |       |
| >95 | 84.40  | 25.5 | 70.90  | 21.5 | 23.7  | 78.00  | 21.5  | 65.50  | 25.3 | 23.3  |
| >90 | 82.70  | 38.3 | 72.60  | 38.0 | 38.1  | 76.70  | 34.4  | 67.10  | 39.2 | 26.6  |
| >80 | 80.80  | 58.5 | 74.50  | 57.0 | 57.8  | 75.00  | 52.7  | 68.80  | 58.2 | 55.2  |
| >50 | 77.69  | 84.0 | 77.69  | 90.0 | 86.7  | 71.81  | 81.7  | 71.81  | 82.3 | 82.0  |
|     |        | Fi   | iL     |      |       |        |       | LB     |      |       |
| >95 | 371.00 | 26.1 | 317.00 | 19.5 | 23.1  | 47.80  | 16.13 | 37.50  | 13.2 | 14.0  |
| >90 | 364.00 | 38.1 | 324.00 | 35.1 | 26.7  | 46.60  | 25.81 | 39.00  | 24.4 | 25.2  |
| >80 | 357.00 | 57.6 | 331.00 | 51.9 | 55.0  | 45.10  | 44.09 | 40.40  | 48.7 | 46.2  |
| >50 | 344.18 | 82.6 | 344.18 | 87.0 | 82.8  | 42.78  | 81.72 | 42.78  | 85.9 | 83.6  |

Table 7.1.2.11 Posterior probabilities for single dimensions of the lower limb bones.

Tibia

Posterior probabilities for the measurements taken on the tibia resulted in grouping up to 30% of the specimens at a 0.95 threshold with 82% accuracy (NFMax). Interestingly, LB sexed only 14% of the sample at a 0.95 threshold with slightly higher accuracy compared to NFMax (Table 7.1.1.11).

| PP  | Male    | s    | Female   | es   | Total | Male     | es   | Femal    | es   | Total |
|-----|---------|------|----------|------|-------|----------|------|----------|------|-------|
| (%) | <       | %    | >        | %    | %     | <        | %    | >        | %    | %     |
|     |         |      | F1       |      |       |          |      | TF1      |      |       |
| >95 | >1.2634 | 43.6 | <-1.2334 | 50   | 46.5  | >1.2292  | 49.5 | <-1.2277 | 48.7 | 49.1  |
| >90 | >0.9425 | 58.5 | <-0.9509 | 60.8 | 59.9  | >0.9148  | 63.4 | <-0.9347 | 65.4 | 64.3  |
| >80 | >0.6121 | 74.5 | <-0.6246 | 71.8 | 73.3  | >0.5786  | 76.3 | <-0.6650 | 76.9 | 76.6  |
| >50 | >0      | 89.4 | <0       | 93.6 | 90.8  | >0       | 93.6 | <0       | 88.5 | 91.2  |
|     |         |      | F2       |      |       |          |      | TF2      |      |       |
| >95 | >1.2775 | 45.7 | <-1.2623 | 47.4 | 46.5  | >1.2405  | 47.3 | <-1.2669 | 46.8 | 47.1  |
| >90 | >0.9477 | 58.5 | <-0.9427 | 59   | 58.7  | >0.9396  | 61.3 | <-0.9371 | 65.8 | 63.4  |
| >80 | >0.5945 | 73.4 | <-0.6111 | 74.4 | 73.8  | < 0.6246 | 72.0 | <-0.6303 | 76.0 | 73.8  |
| >50 | >0      | 89.4 | <0       | 92.3 | 90.7  | >0       | 93.6 | <0       | 87.3 | 90.7  |
|     |         |      | F3       |      |       |          |      | TF3      |      |       |
| >95 | >1.4039 | 35.1 | <-1.4112 | 41   | 37.8  | >1.3633  | 38.7 | <-1.3843 | 38.5 | 38.6  |
| >90 | >1.0698 | 48.9 | <-1.0428 | 53.8 | 51.2  | >1.0135  | 51.6 | <-1.0464 | 50.0 | 55.0  |
| >80 | >0.6712 | 67   | <0.6839  | 69.2 | 68    | >0.6462  | 66.7 | <-0.6522 | 69.2 | 67.8  |
| >50 | >0      | 87.2 | <0       | 87.2 | 87.2  | >0       | 89.2 | <0       | 85.9 | 87.7  |
|     |         |      | F4       |      |       |          |      | TF4      |      |       |
| >95 | >1.4455 | 30.9 | <-1.4479 | 33.3 | 32    | >1.3745  | 41.9 | <-1.3396 | 50.6 | 45.9  |
| >90 | >1.083  | 45.7 | <-1.12   | 50   | 47.7  | >1.0106  | 52.7 | <-0.9897 | 60.8 | 56.4  |
| >80 | >0.7027 | 61.7 | <-0.7152 | 65.4 | 63.4  | >0.6673  | 65.6 | <-0.6292 | 69.6 | 67.4  |
| >50 | >0      | 85.1 | <0       | 85.9 | 85.5  | >0       | 91.4 | <0       | 86.1 | 89.0  |
|     |         |      | F5       |      |       |          |      | LLF      |      |       |
| >95 | >2.3285 | 36.2 | <-1.5233 | 32.6 | 34.1  | >1.1682  | 57.6 | <-1.1696 | 55.3 | 56.5  |
| >90 | >1.9572 | 45.7 | <-1.118  | 41.8 | 43.9  | >0.8630  | 68.5 | <-0.8533 | 71.1 | 69.6  |
| >80 | <1.6686 | 63.8 | <-0.7332 | 62   | 63    | >0.5697  | 82.6 | <-0.5350 | 81.6 | 82.1  |
| >50 | >0      | 80.9 | <0       | 89.9 | 85    | >0       | 92.4 | <0       | 90.8 | 91.7  |
|     |         |      | F6       |      |       |          |      |          |      |       |
| >95 | >1.4976 | 34   | <-1.4068 | 37.2 | 35.5  |          |      |          |      |       |
| >90 | >1.0518 | 47.9 | <-1.0791 | 51.3 | 49.4  |          |      |          |      |       |
| >80 | >0.6674 | 66   | <-0.6825 | 67.9 | 66.9  |          |      |          |      |       |
| >50 | >0      | 84   | <0       | 87.2 | 85.5  |          |      |          |      |       |

Table 7.1.2.12 Posterior probabilities for multivariate functions of the lower limb bones.

#### Fibula

Taking into account the demarking point for the maximum length of the fibula, about 23% of the cases were correctly classified within 95% of confidence intervals with 85% accuracy (Table 7.1.2.11). The percentage was slightly higher in males (26.1%) compared to females (20%). A fibula with length greater than 371mm had a 95% probability of being correctly assigned as male, whereas a fibula with length less than 317mm had a 95% probability of being correctly assigned as female.

#### Multivariate statistics

# Femur

Posterior probabilities for the multivariate discriminant functions of the femur are presented in Table 7.1.2.12 The best discriminating functions were F1, which is the result of a direct DFA using all femoral measurements, and F2, which is the result of a stepwise procedure using the same variables. At a 0.95 threshold, sex was determined for 47% of the sample with about 91% accuracy for both F1 and F2. In the case of F1, discriminant scores over 1.2634 classified males with 95% probability, while values smaller than -1.2334 assigned females at a 0.95 threshold. For F2, discriminant scores over 1.2775 classified males with 95% probability, while values smaller than same threshold of 95%.

#### Tibia

The best discriminant functions for the tibia were TF1 and TF2, with about 91% accuracy. At a 95% threshold, though, TF1 performed slightly better, classifying 49% of the sample (within a 95% interval of confidence). For TF1, discriminant scores over 1.2292 classified males with 95% probability, while values smaller than -1.2277 assigned females at the same threshold of 95%.

LLF is the result of a stepwise discriminant function analysis using all measurements of the lower limb bones. This function classified about 70% of the sample at a 0.9 threshold and 57% at a 0.95 threshold with 91.7% accuracy (Table 7.1.2.12). Discriminant scores over 1.1682 classified males with 95% probability, while values smaller than -1.1696 assigned females at the same threshold of 0.95.

# 7.2 Radiometry

# 7.2.1. Estimation of error

Sixty randomly selected specimens (30 males and 30 females) were digitized twice in order to calculate the intra-observer error. The time interval between the two digitalizations was 3 months. A second observer also digitized the same specimens following the instructions as described in "Material and Methods". Then the interlandmark distances were calculated and the differences between the means of each measurement were tested for the same (OB1-A and OB1-B) and two different observers (OB1 and OB2) using the student's T-test. This procedure was followed for both epiphyses of humerus, radius, femur and tibia.

| РН   | OB    | IA   | OB1B  |         | O      | B2    | T-differences between OB1A and |       |  |
|------|-------|------|-------|---------|--------|-------|--------------------------------|-------|--|
|      | Mean  | SD   | Mean  | SD      | Mean   | SD    | OB1B                           | OB2   |  |
|      |       |      |       | Males ( | (N=30) |       |                                |       |  |
| PH1  | 45.02 | 4.20 | 44.96 | 4.39    | 44.74  | 4.354 | 0.42                           | 1.61  |  |
| PH2  | 28.68 | 2.94 | 28.80 | 3.05    | 28.62  | 3.124 | -0.63                          | 0.24  |  |
| PH3  | 49.85 | 4.15 | 49.90 | 4.32    | 49.80  | 4.344 | -0.38                          | 0.21  |  |
| PH4  | 51.41 | 4.51 | 51.33 | 4.68    | 51.44  | 4.588 | 0.79                           | -0.22 |  |
| PH5  | 28.63 | 1.85 | 28.54 | 1.60    | 28.64  | 1.42  | 0.44                           | -0.03 |  |
| PH6  | 6.94  | 0.63 | 7.13  | 0.64    | 7.19   | 0.58  | -1.39                          | -1.75 |  |
| PH7  | 13.04 | 1.94 | 13.17 | 2.23    | 13.30  | 2.153 | -0.93                          | -2.69 |  |
| PH8  | 35.46 | 1.86 | 35.55 | 1.77    | 35.73  | 1.568 | -0.60                          | -1.37 |  |
| PH9  | 40.47 | 3.12 | 40.47 | 3.04    | 40.79  | 2.9   | 0.00                           | -1.34 |  |
| PH10 | 6.82  | 2.04 | 6.79  | 2.23    | 6.82   | 2.269 | 0.27                           | -0.03 |  |
|      |       |      |       | Females | (N=30) |       |                                |       |  |
| PH1  | 44.14 | 2.12 | 44.27 | 1.97    | 44.09  | 2.169 | -0.70                          | -0.48 |  |
| PH2  | 28.12 | 2.09 | 28.39 | 1.79    | 27.95  | 2.194 | -1.14                          | -0.34 |  |
| PH3  | 48.29 | 2.52 | 48.43 | 2.39    | 47.99  | 2.691 | -1.07                          | -1.27 |  |
| PH4  | 48.94 | 3.65 | 49.09 | 3.47    | 49.05  | 3.774 | -0.81                          | 0.80  |  |
| PH5  | 28.34 | 1.30 | 28.40 | 1.31    | 28.46  | 1.799 | -0.21                          | 0.22  |  |
| PH6  | 7.13  | 1.92 | 7.17  | 1.57    | 6.73   | 1.717 | -0.12                          | -1.32 |  |
| PH7  | 13.73 | 1.85 | 13.69 | 1.58    | 13.90  | 1.735 | 0.34                           | 0.78  |  |
| PH8  | 35.13 | 1.98 | 35.22 | 1.75    | 34.85  | 1.778 | -0.23                          | -0.54 |  |
| PH9  | 39.83 | 2.05 | 39.93 | 2.03    | 40.13  | 2.135 | -0.31                          | 0.61  |  |
| PH10 | 7.38  | 2.83 | 7.31  | 2.33    | 8.01   | 2.484 | 0.26                           | *3.05 |  |

Table 7.2.1 T-differences for the measurements of the proximal humerus taken by the same (OB1-A and OB1-B) and by two different observers (OB1-A and OB2).

| DH   | ОВ    | 1A   | OB    | 1B      | OE    | 32   | T-differen<br>OB1 | ces between<br>A and | DH OB1A        |       | OB1B |       | OB2  |       | T-differences between<br>OB1A and |       |        |
|------|-------|------|-------|---------|-------|------|-------------------|----------------------|----------------|-------|------|-------|------|-------|-----------------------------------|-------|--------|
|      | Mean  | SD   | Mean  | SD      | Mean  | SD   | OB1B              | OB2                  |                | Mean  | SD   | Mean  | SD   | Mean  | SD                                | OB1B  | OB2    |
|      |       |      |       | Males ( | N=30) |      |                   |                      | Females (N=30) |       |      |       |      |       |                                   |       |        |
| DH1  | 9.58  | 0.65 | 9.53  | 0.94    | 9.19  | 0.85 | 0.23              | 1.79                 | DH1            | 10.73 | 1.58 | 10.48 | 1.83 | 10.85 | 1.69                              | -1.87 | 1.03   |
| DH2  | 14.82 | 1.34 | 15.03 | 0.99    | 15.14 | 0.78 | -0.51             | -0.72                | DH2            | 15.58 | 2.52 | 15.73 | 2.31 | 15.84 | 2.36                              | 0.85  | 1.69   |
| DH3  | 29.05 | 2.52 | 28.79 | 2.07    | 28.91 | 2.06 | 0.61              | 0.41                 | DH3            | 29.03 | 4.09 | 28.73 | 3.86 | 29.11 | 4.13                              | -1.57 | 0.84   |
| DH4  | 41.50 | 2.90 | 41.86 | 3.06    | 41.54 | 2.54 | -1.20             | -0.08                | DH4            | 41.75 | 5.74 | 41.70 | 5.87 | 41.33 | 5.95                              | -0.30 | *-3.04 |
| DH5  | 48.61 | 4.07 | 48.61 | 4.14    | 48.67 | 4.14 | -0.04             | -0.23                | DH5            | 48.03 | 6.90 | 47.99 | 6.87 | 47.76 | 7.02                              | -0.30 | -1.25  |
| DH6  | 16.59 | 0.91 | 16.59 | 1.41    | 16.79 | 1.37 | -0.01             | -0.79                | DH6            | 16.74 | 2.58 | 16.60 | 2.73 | 16.53 | 2.63                              | -0.75 | -0.72  |
| DH7  | 14.00 | 2.04 | 14.26 | 2.01    | 14.38 | 2.24 | -0.70             | -1.13                | DH7            | 14.64 | 2.97 | 14.71 | 2.82 | 15.03 | 2.88                              | 0.48  | 1.74   |
| DH8  | 27.09 | 2.45 | 26.93 | 2.15    | 27.05 | 2.19 | 0.59              | 0.19                 | DH8            | 26.90 | 4.21 | 26.61 | 3.91 | 27.03 | 4.06                              | -1.54 | 0.58   |
| DH9  | 40.57 | 2.81 | 41.00 | 3.06    | 40.58 | 2.51 | -1.27             | -0.04                | DH9            | 40.87 | 5.71 | 40.76 | 5.78 | 40.51 | 5.85                              | -0.57 | -1.47  |
| DH10 | 49.71 | 3.80 | 49.69 | 3.97    | 49.71 | 4.00 | 0.15              | 0.02                 | DH10           | 49.13 | 6.74 | 49.02 | 6.69 | 48.97 | 6.83                              | -0.39 | -0.92  |
| DH11 | 24.89 | 1.23 | 24.82 | 1.31    | 24.68 | 1.49 | 0.56              | 1.18                 | DH11           | 25.58 | 2.42 | 25.15 | 2.87 | 25.22 | 2.78                              | -1.60 | -1.09  |
| DH12 | 14.36 | 1.94 | 13.89 | 1.26    | 13.92 | 1.55 | 1.10              | 1.23                 | DH12           | 13.61 | 1.64 | 13.36 | 1.91 | 13.43 | 1.98                              | 1.65  | -1.04  |
| DH13 | 27.22 | 1.96 | 27.38 | 1.99    | 26.89 | 2.08 | -0.40             | 0.50                 | DH13           | 26.92 | 3.00 | 26.68 | 3.33 | 26.20 | 3.48                              | -0.94 | *-2.88 |
| DH14 | 35.76 | 3.05 | 35.49 | 2.82    | 35.38 | 3.14 | 0.58              | 1.37                 | DH14           | 34.59 | 3.88 | 34.40 | 4.01 | 34.05 | 4.21                              | -0.82 | -2.29  |
| DH15 | 30.37 | 1.86 | 30.51 | 2.00    | 30.72 | 1.76 | -0.69             | -1.74                | DH15           | 31.57 | 4.92 | 31.61 | 4.92 | 31.68 | 4.67                              | 0.36  | 0.33   |
| DH16 | 13.61 | 1.05 | 14.18 | 1.09    | 13.64 | 1.10 | -1.23             | -0.08                | DH16           | 14.09 | 1.52 | 14.25 | 1.94 | 13.61 | 1.85                              | 0.66  | -2.73  |
| DH17 | 24.39 | 1.83 | 24.34 | 2.02    | 24.43 | 2.03 | 0.15              | -0.20                | DH17           | 23.56 | 2.64 | 23.74 | 2.83 | 23.36 | 3.04                              | 0.58  | -0.74  |
| DH18 | 44.00 | 3.48 | 43.74 | 3.20    | 44.02 | 3.19 | 1.03              | -0.12                | DH18           | 44.62 | 6.78 | 44.24 | 6.65 | 44.63 | 6.77                              | -2.09 | 0.05   |
| DH19 | 12.62 | 1.33 | 12.13 | 1.35    | 12.83 | 1.94 | 1.10              | -0.34                | DH19           | 11.04 | 1.28 | 11.17 | 1.39 | 11.28 | 1.53                              | 0.49  | 0.71   |
| DH20 | 55.11 | 4.22 | 55.47 | 4.40    | 55.38 | 3.97 | -1.19             | -0.74                | DH20           | 56.15 | 8.66 | 56.12 | 8.79 | 55.81 | 8.70                              | -0.14 | -1.53  |
| DH21 | 59.85 | 5.44 | 59.95 | 5.58    | 60.07 | 5.52 | -1.33             | -1.79                | DH21           | 60.65 | 9.78 | 60.67 | 9.65 | 60.54 | 9.65                              | 0.17  | -0.92  |

Table 7.2.2 T-differences for the measurements of the distal humerus taken by the same (OB1-A and OB1-B) and by two different observers (OB1-A and OB2)

# 7.2.1.1 Humerus

The differences between the mean measurements for the proximal humerus were found insignificant for the same observer (OB1-A and OB1-B).

| PR   | OBI   | A    | OB1B  |          | OB    | 2    | T-differences<br>between OB1A and |        |  |
|------|-------|------|-------|----------|-------|------|-----------------------------------|--------|--|
|      | Mean  | SD   | Mean  | SD       | Mean  | SD   | OB1B                              | OB2    |  |
|      |       |      | Ν     | Males (N | (=30) |      |                                   |        |  |
| PR1  | 14.23 | 1.68 | 14.22 | 1.73     | 14.07 | 1.87 | 0.06                              | 0.70   |  |
| PR2  | 28.72 | 6.79 | 28.66 | 5.04     | 28.85 | 4.79 | 0.02                              | -0.05  |  |
| PR3  | 31.38 | 6.36 | 30.56 | 3.18     | 30.96 | 2.88 | 0.28                              | 0.15   |  |
| PR4  | 40.13 | 4.26 | 42.75 | 4.76     | 43.45 | 4.08 | -0.92                             | -1.31  |  |
| PR5  | 43.34 | 4.17 | 45.59 | 3.71     | 45.79 | 2.92 | -0.83                             | -1.00  |  |
| PR6  | 12.92 | 2.46 | 14.81 | 3.73     | 15.85 | 3.84 | -0.80                             | -1.51  |  |
| PR7  | 18.91 | 2.23 | 20.17 | 1.14     | 20.29 | 1.56 | -1.07                             | -1.78  |  |
| PR8  | 31.16 | 5.64 | 31.21 | 3.30     | 31.43 | 3.44 | -0.02                             | -0.11  |  |
| PR9  | 27.02 | 6.05 | 26.19 | 4.46     | 26.63 | 4.56 | 0.28                              | 0.15   |  |
| PR10 | 42.36 | 3.32 | 44.64 | 3.86     | 45.32 | 3.49 | -0.85                             | -1.24  |  |
| PR11 | 37.62 | 4.01 | 39.88 | 4.95     | 40.19 | 4.55 | -0.80                             | -0.99  |  |
| PR12 | 20.61 | 2.80 | 22.85 | 1.76     | 23.46 | 1.88 | -1.12                             | -1.66  |  |
| PR13 | 11.99 | 1.95 | 13.51 | 2.98     | 13.96 | 3.13 | -0.84                             | -1.40  |  |
| PR14 | 13.73 | 2.59 | 13.48 | 2.05     | 13.70 | 2.03 | 0.26                              | 0.03   |  |
| PR15 | 11.65 | 3.68 | 14.19 | 2.55     | 14.68 | 2.54 | -2.32                             | *-3.63 |  |
| PR16 | 22.43 | 3.21 | 23.98 | 2.32     | 24.06 | 2.48 | -1.74                             | -2.51  |  |
| PR17 | 16.50 | 7.10 | 15.18 | 1.72     | 14.32 | 2.83 | 0.48                              | 0.81   |  |
| PR18 | 21.09 | 4.85 | 19.75 | 0.34     | 19.60 | 1.47 | 0.62                              | 0.71   |  |
| PR19 | 20.25 | 2.22 | 21.68 | 2.43     | 22.00 | 2.54 | -2.33                             | -3.38  |  |
| PR20 | 12.04 | 2.63 | 15.13 | 1.95     | 14.92 | 1.06 | -2.02                             | -2.71  |  |
| PR21 | 23.10 | 6.34 | 22.11 | 0.80     | 21.85 | 1.62 | 0.31                              | 0.41   |  |
| PR22 | 15.09 | 6.15 | 12.70 | 1.59     | 12.68 | 2.36 | 0.93                              | 1.01   |  |
| PR23 | 22.75 | 2.48 | 22.79 | 2.50     | 22.81 | 2.76 | -0.07                             | -0.12  |  |
| PR24 | 27.62 | 3.65 | 28.69 | 2.19     | 28.33 | 2.54 | -0.53                             | -0.35  |  |
| PR25 | 31.44 | 2.31 | 32.10 | 1.96     | 32.30 | 2.33 | -0.41                             | -0.50  |  |
| PR26 | 34.76 | 4.50 | 36.44 | 1.63     | 35.84 | 2.58 | -0.73                             | -0.43  |  |
| PR27 | 26.03 | 4.03 | 26.84 | 2.20     | 26.73 | 2.82 | -0.45                             | -0.35  |  |
| PR28 | 16.50 | 2.30 | 17.92 | 1.91     | 17.77 | 1.71 | -1.15                             | -1.03  |  |

Table 7.2.3 T-differences for the measurements of the proximal radius for males taken by the same (OB1-A and OB1-B) and by two different observers (OB1-A and OB2).

However, a small inter-observer error was recorded for the variable PH 10 (p<0.05) for females (<u>Table 7.2.1</u>). Similarly, the differences between the mean measurements on the

<sup>\*</sup> p<0.05

distal humerus were found insignificant for the same observer (OB1-A and OB1-B), whereas for two of the variables (DH4 and DH13) there was a significant difference of the means between the first and the second observer (p<0.05) for the female group (<u>Table 7.2.2</u>).

| PR   | OB1   | A    | OB1B  |         | OB     | 2    | T-differences between<br>OB1A and |        |  |
|------|-------|------|-------|---------|--------|------|-----------------------------------|--------|--|
|      | Mean  | SD   | Mean  | SD      | Mean   | SD   | OB1B                              | OB2    |  |
|      |       |      | J     | Females | (N=30) |      |                                   |        |  |
| PR1  | 11.78 | 1.12 | 11.43 | 1.38    | 11.46  | 1.20 | 0.41                              | 0.38   |  |
| PR2  | 25.50 | 1.77 | 23.37 | 2.23    | 23.80  | 2.23 | 1.32                              | 1.21   |  |
| PR3  | 26.18 | 1.60 | 24.47 | 2.13    | 25.03  | 2.09 | 1.13                              | 0.86   |  |
| PR4  | 37.47 | 1.32 | 37.93 | 1.88    | 38.40  | 1.96 | -0.42                             | -0.77  |  |
| PR5  | 37.78 | 1.29 | 37.54 | 0.94    | 38.06  | 1.33 | 0.28                              | -0.27  |  |
| PR6  | 13.01 | 1.51 | 12.50 | 1.72    | 13.07  | 2.02 | 0.37                              | -0.05  |  |
| PR7  | 16.38 | 1.15 | 15.84 | 1.15    | 16.30  | 1.22 | 0.62                              | 0.10   |  |
| PR8  | 27.66 | 1.97 | 26.08 | 2.23    | 26.38  | 2.56 | 0.98                              | 0.83   |  |
| PR9  | 23.06 | 1.24 | 21.62 | 2.18    | 22.15  | 2.25 | 1.04                              | 0.71   |  |
| PR10 | 39.09 | 1.05 | 39.68 | 1.78    | 40.06  | 2.04 | -0.55                             | -0.82  |  |
| PR11 | 33.68 | 0.88 | 33.72 | 1.07    | 34.21  | 1.37 | -0.05                             | -0.55  |  |
| PR12 | 19.34 | 1.69 | 19.26 | 1.87    | 19.55  | 2.18 | 0.06                              | -0.16  |  |
| PR13 | 11.60 | 1.50 | 11.36 | 1.18    | 11.83  | 1.49 | 0.25                              | -0.24  |  |
| PR14 | 11.68 | 1.36 | 11.69 | 1.57    | 11.68  | 1.62 | -0.01                             | 0.01   |  |
| PR15 | 12.00 | 1.53 | 14.57 | 2.65    | 14.63  | 2.02 | -2.34                             | *-2.82 |  |
| PR16 | 18.98 | 0.76 | 20.32 | 1.06    | 20.38  | 1.02 | -1.85                             | -1.91  |  |
| PR17 | 13.35 | 2.23 | 12.07 | 1.78    | 11.85  | 1.46 | 0.74                              | 0.94   |  |
| PR18 | 17.44 | 2.10 | 16.39 | 1.73    | 16.33  | 1.76 | 0.67                              | 0.71   |  |
| PR19 | 18.68 | 0.48 | 20.17 | 1.65    | 20.03  | 1.47 | -2.53                             | *-2.73 |  |
| PR20 | 11.63 | 1.38 | 13.08 | 2.39    | 13.05  | 2.08 | -1.11                             | -1.12  |  |
| PR21 | 18.44 | 2.04 | 18.01 | 2.61    | 17.95  | 2.30 | 0.23                              | 0.27   |  |
| PR22 | 11.50 | 1.76 | 10.29 | 1.36    | 10.35  | 1.40 | 0.97                              | 0.97   |  |
| PR23 | 18.30 | 0.53 | 18.47 | 0.72    | 18.47  | 0.85 | -0.36                             | -0.34  |  |
| PR24 | 25.03 | 2.04 | 26.33 | 1.84    | 26.16  | 1.27 | -1.18                             | -1.15  |  |
| PR25 | 28.09 | 1.77 | 28.87 | 1.40    | 28.81  | 1.04 | -0.88                             | -0.83  |  |
| PR26 | 29.34 | 1.41 | 30.13 | 1.40    | 30.04  | 1.22 | -0.71                             | -0.68  |  |
| PR27 | 22.50 | 1.44 | 22.79 | 1.03    | 22.80  | 0.66 | -0.62                             | -0.61  |  |
| PR28 | 14.32 | 1.73 | 14.71 | 2.33    | 14.68  | 2.25 | -0.24                             | -0.23  |  |

Table 7.2.4 T-differences for the measurements of the proximal radius for females taken by the same (OB1-A and OB1-B) and by two different observers (OB1-A and OB2).

# 7.2.1.2 Radius

The results of the student's T-test for the proximal radius are illustrated in <u>Table 7.2.3</u> (males) and in <u>Table 7.2.4</u> (females).

| DR   | OB1   | A    | OB     | 1B        | OF    | 32    | T-differ<br>between<br>and | ences<br>OB1A<br>1 |
|------|-------|------|--------|-----------|-------|-------|----------------------------|--------------------|
|      | Mean  | SD   | Mean   | SD        | Mean  | SD    | OB1B                       | OB2                |
|      |       |      | 1      | Males (N= | =30)  |       |                            |                    |
| DR1  | 30.30 | 2.63 | 30.423 | 2.189     | 30.37 | 2.32  | -0.58                      | 0.48               |
| DR2  | 32.46 | 3.87 | 32.424 | 3.828     | 32.68 | 3.58  | 0.31                       | 1.63               |
| DR3  | 4.22  | 0.69 | 4.2108 | 0.544     | 4.37  | 0.53  | 0.07                       | 1.29               |
| DR4  | 24.84 | 2.61 | 25.047 | 2.26      | 25.24 | 2.41  | -1.10                      | 1.58               |
| DR5  | 31.21 | 3.14 | 31.291 | 3.318     | 31.35 | 3.07  | -0.38                      | 1.43               |
| DR6  | 9.61  | 1.03 | 9.6188 | 0.717     | 9.54  | 0.33  | -0.03                      | -0.19              |
| DR7  | 26.16 | 2.69 | 26.32  | 2.248     | 26.07 | 2.34  | -0.80                      | -0.48              |
| DR8  | 5.92  | 0.92 | 5.7898 | 0.485     | 5.64  | 0.37  | 0.56                       | -0.96              |
| DR9  | 16.38 | 2.28 | 16.484 | 2.261     | 16.75 | 2.32  | -0.81                      | 0.91               |
| DR10 | 28.72 | 3.70 | 28.68  | 3.608     | 28.74 | 3.41  | 0.54                       | 0.14               |
| DR11 | 9.83  | 1.30 | 9.8114 | 1.081     | 9.61  | 0.89  | 0.13                       | -0.85              |
| DR12 | 7.77  | 3.03 | 7.8404 | 3.378     | 8.32  | 3.02  | -0.33                      | 1.28               |
| DR13 | 20.76 | 2.46 | 20.993 | 2.18      | 21.01 | 2.35  | -1.48                      | 1.24               |
| DR14 | 28.00 | 2.94 | 28.018 | 3.016     | 28.00 | 2.95  | -0.17                      | 0.01               |
| DR15 | 14.11 | 1.90 | 14.298 | 2.007     | 14.33 | 2.08  | -0.80                      | 0.49               |
|      |       |      | F      | emales (N | (=30) |       |                            |                    |
| DR1  | 27.82 | 1.53 | 27.63  | 1.335     | 26.79 | 2.441 | 1.34                       | -0.78              |
| DR2  | 28.9  | 1.60 | 28.81  | 1.273     | 27.83 | 1.692 | 0.50                       | -0.99              |
| DR3  | 3.733 | 0.94 | 3.5568 | 0.861     | 3.64  | 1.314 | 0.86                       | -0.13              |
| DR4  | 22.69 | 2.03 | 22.521 | 1.966     | 21.79 | 2.165 | 0.93                       | -0.67              |
| DR5  | 27.79 | 1.74 | 27.588 | 1.398     | 26.47 | 1.984 | 0.93                       | -1.03              |
| DR6  | 8.086 | 0.83 | 8.3267 | 1.185     | 7.892 | 1.183 | -0.83                      | -0.27              |
| DR7  | 24.16 | 1.24 | 24.13  | 1.118     | 23.49 | 1.747 | 0.24                       | -0.71              |
| DR8  | 5.42  | 1.27 | 5.5563 | 1.304     | 5.378 | 0.511 | -0.73                      | -0.07              |
| DR9  | 14.06 | 0.83 | 14.408 | 1.432     | 14.84 | 1.655 | -1.08                      | 0.83               |
| DR10 | 25.55 | 1.29 | 25.617 | 1.161     | 25.09 | 1.407 | -0.42                      | -0.53              |
| DR11 | 8.364 | 1.23 | 8.2795 | 1.321     | 7.953 | 0.553 | 0.35                       | -0.96              |
| DR12 | 6.526 | 1.00 | 6.7167 | 0.872     | 7.724 | 1.924 | -0.98                      | 1.21               |
| DR13 | 19.08 | 2.08 | 19.074 | 1.702     | 18.62 | 1.524 | 0.02                       | -0.42              |
| DR14 | 24.92 | 1.42 | 24.869 | 1.117     | 24.43 | 1.756 | 0.27                       | -0.47              |
| DR15 | 12.28 | 0.6  | 12.182 | 0.895     | 12.45 | 1.244 | 0.37                       | 0.25               |

Table 7.2.5 T-differences for the measurements of the distal radius taken by the same (OB1-A and OB1-B) and by two different observers (OB1-A and OB2).

The differences between the mean measurements were found insignificant for the same observer (OB1-A and OB1-B). However, a small inter-observer error was recorded

for the variables PR 15 (in both males and females) and PR19 for males. The results of the student's T-test for the distal radius are illustrated in <u>Table 7.2.5</u>. The differences between the mean measurements were found insignificant for the same (OB1-A and OB1-B) and two different observers (OB1-A and OB-2).

| PF   | OB    | A    | OB    | IB       | OE    | 32   | T-differ<br>between O | ences<br>B1A and |
|------|-------|------|-------|----------|-------|------|-----------------------|------------------|
|      | Mean  | SD   | Mean  | SD       | Mean  | SD   | OB1B                  | OB2              |
|      |       |      |       | Males (N | V=30) |      |                       |                  |
| PF1  | 48.96 | 5.39 | 48.74 | 4.38     | 49.04 | 5.76 | -0.22                 | 0.31             |
| PF2  | 60.25 | 3.89 | 59.96 | 3.98     | 59.43 | 4.35 | -0.63                 | -1.74            |
| PF3  | 78.44 | 5.00 | 80.74 | 7.14     | 78.27 | 4.86 | 1.12                  | -1.09            |
| PF4  | 69.02 | 4.35 | 69.78 | 3.81     | 69.20 | 4.08 | 2.14                  | 1.12             |
| PF5  | 79.90 | 5.78 | 80.09 | 5.89     | 79.92 | 5.65 | 0.82                  | 0.18             |
| PF6  | 62.33 | 1.94 | 62.04 | 1.76     | 62.00 | 1.86 | -1.59                 | -1.11            |
| PF7  | 32.72 | 3.56 | 32.81 | 3.23     | 32.85 | 3.22 | 0.50                  | 0.76             |
| PF8  | 16.20 | 2.43 | 16.28 | 2.91     | 15.49 | 1.77 | 0.05                  | -1.24            |
| PF9  | 39.84 | 5.68 | 41.14 | 5.04     | 39.52 | 5.74 | 1.23                  | *-3.80           |
| PF10 | 34.23 | 3.66 | 34.35 | 3.63     | 34.09 | 3.72 | 0.31                  | -1.45            |
| PF11 | 61.20 | 6.15 | 61.20 | 7.13     | 61.00 | 6.49 | -0.01                 | -0.79            |
| PF12 | 67.28 | 5.91 | 66.96 | 7.66     | 67.11 | 6.31 | -0.28                 | -0.61            |
| PF13 | 69.88 | 4.29 | 69.37 | 5.72     | 70.31 | 5.40 | -0.38                 | 0.81             |
| PF14 | 47.53 | 5.65 | 47.67 | 5.81     | 47.15 | 5.89 | 0.75                  | -1.35            |
| PF15 | 44.84 | 4.22 | 44.72 | 4.46     | 44.35 | 4.30 | -0.48                 | -1.45            |
| PF16 | 74.17 | 6.98 | 73.96 | 7.30     | 73.55 | 7.38 | -0.57                 | -1.20            |
| PF17 | 83.22 | 7.56 | 82.99 | 7.70     | 82.41 | 7.61 | -0.59                 | -1.91            |
| PF18 | 84.53 | 4.70 | 84.08 | 4.92     | 84.07 | 4.97 | -0.76                 | -0.65            |
| PF19 | 10.06 | 2.16 | 12.08 | 5.65     | 9.67  | 1.83 | 0.75                  | -1.68            |
| PF20 | 34.31 | 9.02 | 36.44 | 7.81     | 34.12 | 8.89 | 0.94                  | -0.71            |
| PF21 | 59.75 | 8.31 | 62.82 | 6.44     | 59.83 | 7.76 | 1.21                  | 0.19             |
| PF22 | 85.53 | 4.69 | 87.94 | 7.22     | 85.88 | 4.90 | 0.96                  | 0.67             |
| PF23 | 31.11 | 7.72 | 31.39 | 7.91     | 31.13 | 7.75 | 2.57                  | 0.17             |
| PF24 | 51.61 | 7.41 | 52.60 | 7.21     | 52.00 | 7.30 | 2.39                  | 1.46             |
| PF25 | 75.57 | 4.25 | 76.13 | 5.21     | 76.32 | 4.82 | 1.08                  | 1.98             |
| PF26 | 34.05 | 7.65 | 34.75 | 7.78     | 34.53 | 7.50 | 1.51                  | 1.89             |
| PF27 | 73.02 | 6.49 | 72.90 | 5.99     | 73.62 | 6.01 | -0.22                 | 0.97             |
| PF28 | 44.03 | 3.99 | 43.25 | 3.02     | 44.12 | 3.52 | -0.97                 | 0.15             |

Table 7.2.6 T-differences for the measurements of the proximal femur in males taken by the same (OB1-A and OB1-B) and by two different observers (OB1-A and OB2).

# 7.2.1.3 Femur

The results of the student's T-test for the proximal femur are illustrated in <u>Table</u> 7.2.6 (males) and in <u>Table 7.2.7</u> (females). The differences between the mean measurements were found insignificant for the same observer (OB1-A and OB1-B). However, a small inter-observer error was recorded for the variables PF 9 for males and PF8, PF10, PF18 for males and PF27 for females.

| PF   | OB    | IA   | OB    | 1B       | OF    | 32   | T-differ<br>between O | ences<br>B1A and |
|------|-------|------|-------|----------|-------|------|-----------------------|------------------|
|      | Mean  | SD   | Mean  | SD       | Mean  | SD   | OB1B                  | OB2              |
|      |       |      | F     | emales ( | N=30) |      |                       |                  |
| PF1  | 41.92 | 4.03 | 41.78 | 3.88     | 42.48 | 3.87 | -0.90                 | 1.24             |
| PF2  | 50.87 | 6.06 | 50.78 | 6.14     | 50.55 | 6.27 | -0.65                 | -1.54            |
| PF3  | 74.13 | 6.77 | 74.10 | 6.67     | 73.13 | 7.06 | -0.19                 | -1.14            |
| PF4  | 61.57 | 5.38 | 61.26 | 5.21     | 61.60 | 5.54 | -1.55                 | 0.11             |
| PF5  | 74.02 | 6.52 | 73.91 | 6.45     | 73.68 | 6.06 | -1.35                 | -0.71            |
| PF6  | 56.90 | 5.49 | 57.04 | 5.22     | 56.86 | 4.21 | 0.40                  | -0.04            |
| PF7  | 31.69 | 1.19 | 31.65 | 1.24     | 31.84 | 1.32 | -0.51                 | 1.09             |
| PF8  | 14.64 | 3.11 | 14.59 | 3.02     | 13.64 | 2.92 | -0.17                 | *-2.27           |
| PF9  | 38.33 | 3.84 | 38.41 | 3.74     | 37.75 | 3.53 | 0.78                  | -1.01            |
| PF10 | 30.19 | 2.98 | 30.24 | 2.98     | 30.54 | 2.96 | 0.32                  | *2.87            |
| PF11 | 55.45 | 3.57 | 55.24 | 3.09     | 55.81 | 3.44 | -0.94                 | 1.06             |
| PF12 | 61.19 | 2.42 | 61.08 | 2.24     | 61.62 | 2.84 | -0.58                 | 0.88             |
| PF13 | 62.09 | 2.42 | 62.20 | 2.36     | 62.14 | 3.16 | 0.70                  | 0.07             |
| PF14 | 44.17 | 3.07 | 44.09 | 3.09     | 43.91 | 3.00 | -1.07                 | -1.03            |
| PF15 | 40.03 | 3.15 | 40.07 | 3.13     | 39.95 | 2.99 | 0.84                  | -0.27            |
| PF16 | 67.37 | 3.07 | 67.04 | 3.04     | 67.11 | 3.26 | -1.94                 | -0.95            |
| PF17 | 75.56 | 2.35 | 75.38 | 2.62     | 74.91 | 2.10 | -0.95                 | -1.44            |
| PF18 | 74.76 | 4.45 | 74.85 | 4.52     | 73.85 | 4.92 | 0.48                  | *-2.37           |
| PF19 | 13.81 | 4.38 | 14.22 | 4.19     | 12.56 | 2.63 | 2.13                  | -0.95            |
| PF20 | 34.13 | 5.94 | 33.83 | 5.62     | 33.24 | 5.41 | -1.19                 | -0.95            |
| PF21 | 60.85 | 5.33 | 60.63 | 5.30     | 59.32 | 4.95 | -0.53                 | -1.27            |
| PF22 | 82.14 | 5.37 | 82.46 | 5.35     | 79.76 | 6.26 | 1.52                  | -1.80            |
| PF23 | 28.59 | 3.93 | 28.04 | 3.53     | 28.41 | 4.13 | -1.81                 | -0.50            |
| PF24 | 49.00 | 3.68 | 48.36 | 3.90     | 48.61 | 4.16 | -1.47                 | -0.49            |
| PF25 | 68.38 | 3.80 | 68.29 | 3.80     | 67.24 | 4.60 | -0.77                 | -1.66            |
| PF26 | 35.03 | 2.82 | 34.86 | 3.83     | 34.24 | 3.87 | -0.30                 | -1.09            |
| PF27 | 68.02 | 5.60 | 68.42 | 5.91     | 66.19 | 5.94 | 1.47                  | *2.61            |
| PF28 | 38.24 | 5.61 | 38.86 | 5.62     | 36.97 | 5.23 | 1.16                  | -1.01            |

Table 7.2.7 T-differences for the measurements of the proximal femur in females taken by the same (OB1-A and OB1-B) and by two different observers (OB1-A and OB2).

The differences between the mean measurements for the distal femur were found insignificant for the same observer (OB1-A and OB1-B) and two different observers (OB1-A and OB2). T-differences for the distal femur are illustrated in <u>Table 7.2.8</u>.

| DF   | OB    | 1A   | OB    | 1B        | OF     | 32   | T-differences between OB1A and |       |  |
|------|-------|------|-------|-----------|--------|------|--------------------------------|-------|--|
|      | Mean  | SD   | Mean  | SD        | Mean   | SD   | OB1B                           | OB2   |  |
|      |       |      |       | Males (N  | N=30)  |      |                                |       |  |
| DF1  | 85.19 | 4.65 | 85.45 | 4.82      | 85.86  | 4.69 | 2.08                           | -2.32 |  |
| DF2  | 20.98 | 1.36 | 20.85 | 1.98      | 17.90  | 3.46 | -1.40                          | 1.59  |  |
| DF3  | 84.14 | 4.09 | 84.06 | 4.15      | 83.86  | 4.47 | -0.51                          | 0.61  |  |
| DF4  | 52.74 | 1.80 | 52.66 | 1.89      | 51.93  | 2.63 | -0.87                          | 0.93  |  |
| DF5  | 82.71 | 4.51 | 82.43 | 4.06      | 83.46  | 4.33 | 1.86                           | -2.25 |  |
| DF6  | 13.84 | 3.15 | 13.69 | 2.75      | 14.21  | 1.43 | 0.63                           | -0.32 |  |
| DF7  | 42.18 | 3.96 | 42.39 | 3.85      | 42.50  | 3.60 | 0.25                           | -0.59 |  |
| DF8  | 78.30 | 4.27 | 77.81 | 3.76      | 78.51  | 4.60 | 1.16                           | -1.25 |  |
| DF9  | 43.54 | 2.53 | 43.04 | 2.77      | 43.96  | 2.49 | 1.81                           | -1.76 |  |
| DF10 | 35.22 | 3.82 | 35.24 | 3.84      | 35.01  | 3.94 | -1.01                          | 0.95  |  |
|      |       |      | -     | Females ( | (N=30) |      |                                |       |  |
| DF1  | 76.19 | 1.46 | 76.20 | 1.42      | 76.22  | 1.61 | 0.07                           | -0.16 |  |
| DF2  | 19.04 | 2.92 | 18.16 | 4.08      | 18.88  | 3.37 | 1.10                           | 0.35  |  |
| DF3  | 74.37 | 1.64 | 74.18 | 1.71      | 74.27  | 1.86 | 0.47                           | 0.48  |  |
| DF4  | 47.17 | 2.35 | 47.14 | 2.84      | 47.41  | 2.22 | 0.57                           | -1.71 |  |
| DF5  | 72.75 | 1.63 | 72.69 | 1.65      | 73.31  | 1.53 | 1.14                           | -1.39 |  |
| DF6  | 14.11 | 2.99 | 13.98 | 3.40      | 16.24  | 3.11 | 1.22                           | -2.12 |  |
| DF7  | 37.60 | 1.82 | 36.96 | 1.24      | 38.14  | 0.95 | 1.29                           | -0.97 |  |
| DF8  | 67.51 | 1.80 | 67.43 | 1.70      | 67.40  | 1.58 | -0.33                          | 0.94  |  |
| DF9  | 37.73 | 1.63 | 38.16 | 1.81      | 38.15  | 1.24 | -0.05                          | -2.13 |  |
| DF10 | 30.01 | 1.26 | 29.53 | 1.49      | 29.88  | 0.99 | -0.09                          | -0.69 |  |

Table 7.2.8 T-differences for the measurements of the distal femur taken by the same (OB1-A and OB1-B) and by two different observers (OB1-A and OB2).

# 7.2.1.4 Tibia

The differences between the mean measurements of the proximal tibia were found insignificant for the same observer (OB1-A and OB1-B) and two different observers (OB1A and OB2) (<u>Table 7.2.9</u>). The results of the student's T-test for the distal tibia are illustrated in <u>Table 7.2.10</u>. The differences between the mean measurements in females were found insignificant for the same (OB1-A and OB1-B) and two different observers (OB1 and OB2). However, in males, DT7 was found to differ between the first and the second observer (OB1A and OB2) at the level of p < 0.05.

| PT   | PT OB1A | Α    | OB    | 1B       | OB    | 2    | T-diffe<br>between ( | erences<br>DB1A and | OB             | 1A   | OB    | 1B   | OE    | 32   | T-differ<br>between O | rences<br>B1A and |
|------|---------|------|-------|----------|-------|------|----------------------|---------------------|----------------|------|-------|------|-------|------|-----------------------|-------------------|
|      | Mean    | SD   | Mean  | SD       | Mean  | SD   | OB1B                 | OB2                 | Mean           | SD   | Mean  | SD   | Mean  | SD   | OB1B                  | OB2               |
|      |         |      |       | Males (N | I=30) |      |                      |                     | Females (N=30) |      |       |      |       |      |                       |                   |
| PT1  | 32.11   | 1.93 | 32.40 | 2.20     | 32.46 | 2.29 | -1.34                | -1.43               | 30.63          | 1.10 | 30.27 | 0.93 | 31.36 | 0.76 | -0.83                 | 1.93              |
| PT2  | 42.66   | 1.39 | 43.05 | 1.81     | 43.05 | 1.81 | -0.74                | -0.74               | 40.48          | 0.62 | 39.92 | 0.95 | 40.46 | 0.58 | -2.81                 | -0.08             |
| PT3  | 73.07   | 2.44 | 73.29 | 1.82     | 73.20 | 2.00 | -0.22                | -0.16               | 68.75          | 0.68 | 68.62 | 1.28 | 68.99 | 0.74 | -0.39                 | 1.23              |
| PT4  | 76.43   | 3.25 | 76.66 | 3.63     | 76.48 | 3.62 | -0.86                | -0.21               | 71.60          | 0.83 | 71.26 | 0.88 | 71.67 | 0.53 | -1.84                 | 0.31              |
| PT5  | 20.33   | 4.15 | 19.28 | 2.98     | 19.57 | 3.85 | 1.04                 | 1.44                | 18.85          | 2.78 | 18.44 | 2.09 | 17.96 | 2.19 | -1.21                 | -2.12             |
| PT6  | 10.89   | 1.94 | 10.72 | 2.26     | 10.88 | 1.72 | 0.44                 | 0.02                | 10.23          | 1.44 | 9.94  | 1.16 | 9.46  | 1.13 | -0.95                 | -2.40             |
| PT7  | 42.60   | 2.20 | 42.42 | 1.97     | 42.24 | 1.47 | 0.14                 | 0.39                | 40.30          | 0.72 | 40.19 | 0.55 | 39.91 | 0.42 | -0.54                 | -1.66             |
| PT8  | 49.18   | 1.64 | 48.04 | 1.74     | 47.73 | 1.74 | 2.08                 | 3.15                | 46.75          | 1.36 | 46.86 | 1.23 | 46.15 | 1.08 | 0.26                  | -2.04             |
| PT9  | 37.98   | 4.01 | 37.28 | 3.67     | 37.71 | 3.84 | 2.05                 | 2.59                | 36.83          | 2.04 | 35.94 | 1.25 | 36.81 | 1.86 | -1.84                 | -0.06             |
| PT10 | 31.88   | 2.88 | 31.94 | 2.66     | 31.54 | 2.19 | -0.07                | 0.59                | 30.42          | 0.82 | 30.74 | 1.24 | 30.85 | 0.91 | 1.01                  | 4.54              |
| PT11 | 39.55   | 3.00 | 38.38 | 3.38     | 37.80 | 2.88 | 1.40                 | 1.82                | 38.12          | 1.41 | 39.08 | 1.48 | 38.17 | 1.52 | 1.61                  | 0.22              |
| PT12 | 45.96   | 2.94 | 45.57 | 2.43     | 45.97 | 2.68 | 1.50                 | -0.03               | 44.38          | 2.59 | 43.63 | 2.08 | 43.93 | 2.40 | -1.90                 | -1.25             |
| PT13 | 14.82   | 3.71 | 11.54 | 3.04     | 11.29 | 1.69 | 1.31                 | 1.61                | 14.40          | 4.29 | 16.36 | 3.11 | 13.73 | 4.73 | 1.32                  | -2.37             |
| PT14 | 71.10   | 3.07 | 70.98 | 2.59     | 71.24 | 2.57 | 0.12                 | -0.19               | 67.10          | 1.58 | 66.90 | 1.57 | 67.06 | 1.81 | -1.16                 | -0.14             |
| PT15 | 70.40   | 3.32 | 71.41 | 3.68     | 71.61 | 3.61 | -1.54                | -2.54               | 66.12          | 2.18 | 65.19 | 2.17 | 66.26 | 1.99 | -1.28                 | 0.74              |

Table 7.2.9 T-differences for the measurements of the proximal tibia taken by the same (OB1-A and OB1-B) and by two different observers (OB1-A and OB2).

| - | Table 7.2.10 T-differences for the measurements of the proximal tibia taken by the same |
|---|---|
|   | (OB1-A and OB1-B) and by two different observers (OB1-A and OB2).                       |

| DT   | OB    | 1A   | OB    | IB   | OB         | 32   | T-different<br>OB1 | ces between<br>A and |
|------|-------|------|-------|------|------------|------|--------------------|----------------------|
|      | Mean  | SD   | Mean  | SD   | Mean       | SD   | OB1B               | OB2                  |
|      |       |      |       | Ma   | lles (N=30 | ))   |                    |                      |
| DT1  | 12.62 | 2.03 | 12.58 | 1.81 | 12.78      | 2.00 | -0.24              | 0.84                 |
| DT2  | 15.64 | 1.89 | 15.48 | 1.68 | 15.45      | 1.67 | -0.57              | -0.84                |
| DT3  | 52.30 | 3.33 | 52.19 | 3.46 | 52.27      | 3.42 | -0.69              | -0.17                |
| DT4  | 41.45 | 3.24 | 41.29 | 3.11 | 41.28      | 3.06 | -0.76              | -1.10                |
| DT5  | 15.46 | 1.53 | 15.53 | 1.61 | 15.26      | 1.40 | 0.28               | -1.18                |
| DT6  | 48.90 | 3.93 | 48.87 | 4.09 | 48.80      | 3.92 | -0.21              | -0.85                |
| DT7  | 36.69 | 3.90 | 36.54 | 3.86 | 36.39      | 3.70 | -0.96              | *-2.43               |
| DT8  | 36.89 | 2.92 | 36.87 | 2.56 | 37.05      | 2.79 | -0.07              | 0.92                 |
| DT9  | 26.14 | 2.85 | 26.11 | 2.42 | 26.11      | 2.61 | -0.11              | -0.12                |
| DT10 | 13.10 | 2.38 | 13.24 | 1.99 | 13.27      | 2.06 | 0.75               | 1.07                 |
|      |       |      |       |      |            |      |                    |                      |
| DT1  | 12.91 | 1.54 | 12.85 | 1.67 | 12.80      | 1.45 | 0.10               | 0.19                 |
| DT2  | 14.11 | 1.48 | 14.00 | 1.63 | 14.78      | 1.49 | 0.23               | -1.22                |
| DT3  | 47.00 | 2.12 | 47.44 | 2.56 | 47.37      | 2.33 | -0.47              | -0.41                |
| DT4  | 37.96 | 1.91 | 38.43 | 2.35 | 38.70      | 2.24 | -0.46              | -0.76                |
| DT5  | 14.99 | 1.49 | 14.66 | 1.60 | 15.57      | 1.39 | 0.54               | -1.08                |
| DT6  | 43.67 | 2.25 | 43.77 | 2.25 | 43.62      | 2.00 | -0.10              | 0.05                 |
| DT7  | 33.88 | 1.92 | 33.99 | 2.17 | 34.17      | 2.20 | -0.10              | -0.29                |
| DT8  | 33.05 | 1.25 | 33.50 | 1.39 | 32.70      | 1.86 | -0.75              | 0.41                 |
| DT9  | 24.10 | 1.87 | 24.61 | 2.25 | 24.23      | 2.55 | -0.60              | -0.14                |
| DT10 | 9.92  | 2.07 | 10.16 | 2.11 | 9.89       | 1.82 | -0.27              | 0.04                 |

#### 7.2.2 Discriminant function analysis

# 7.2.2.1 Humerus

A total of 101 (53 males and 48 females) adult humerii were X-rayed. Five landmarks were selected on the proximal and seven on the distal humerus (Fig 6.2).

Univariate statistics

#### Proximal humerus

From the five landmarks, 10 distances (PH1-10) were generated (<u>Table 6.3</u>). Descriptive statistics of the 10 dimensions and univariate differences between the sexes are shown in <u>Table 7.2.11</u>. All but PH6 were found significantly different between the sexes at the level of p < 0.001, with the exception of PH6 and PH10, which were found significantly different at the level of p < 0.005.

|                       | F          | roximal    | Humerus      |             |             | Distal Humerus    |            |           |              |             |         |
|-----------------------|------------|------------|--------------|-------------|-------------|-------------------|------------|-----------|--------------|-------------|---------|
| V                     | Mal<br>(N= | les<br>53) | Fema<br>(N=4 | ales<br>48) |             | V                 | Mal<br>(N= | es<br>53) | Fema<br>(N=4 | ules<br>48) |         |
| v                     | Mean       | SD         | Mean         | SD          | F-<br>ratio | v                 | Mean       | SD        | Mean         | SD          | F-ratio |
| PH1                   | 46.82      | 3.73       | 41.52        | 2.00        | 16.94       | DH1               | 9.70       | 1.97      | 8.66         | 0.97        | 11.09   |
| PH2                   | 30.25      | 8.82       | 26.25        | 1.63        | 15.86       | DH2               | 16.31      | 1.67      | 14.22        | 1.07        | 54.58   |
| PH3                   | 51.02      | 3.16       | 45.64        | 2.28        | 16.44       | DH3               | 30.37      | 2.82      | 25.98        | 1.80        | 85.07   |
| PH4                   | 52.82      | 2.95       | 46.69        | 2.46        | 18.33       | DH4               | 41.94      | 2.93      | 36.81        | 1.96        | 104.74  |
| PH5                   | 29.63      | 2.07       | 27.22        | 1.93        | 10.81       | DH5               | 49.33      | 3.47      | 43.41        | 2.60        | 92.59   |
| <sup>a</sup> PH6      | 6.35       | 1.99       | 6.36         | 1.59        | 0.11        | DH6               | 17.12      | 1.91      | 14.75        | 1.05        | 57.81   |
| PH7                   | 13.47      | 1.82       | 12.07        | 2.23        | 8.78        | DH7               | 15.40      | 1.92      | 13.43        | 1.09        | 39.47   |
| PH8                   | 35.76      | 2.79       | 33.33        | 2.42        | 9.42        | DH8               | 28.45      | 2.59      | 24.26        | 1.72        | 89.71   |
| PH9                   | 41.66      | 2.35       | 37.79        | 2.37        | 12.78       | DH9               | 41.01      | 3.11      | 36.07        | 2.01        | 87.68   |
| PH10                  | 7.88       | 2.4        | 6.5          | 2.28        | 7.61        | DH10              | 50.32      | 3.59      | 44.56        | 2.67        | 82.54   |
|                       |            |            |              |             |             | DH11              | 25.37      | 2.85      | 22.17        | 1.40        | 49.58   |
|                       |            |            |              |             |             | DH12              | 14.20      | 1.90      | 11.86        | 1.37        | 49.74   |
|                       |            |            |              |             |             | DH13              | 26.19      | 2.23      | 23.11        | 1.55        | 63.72   |
|                       |            |            |              |             |             | DH14              | 35.02      | 2.74      | 31.18        | 2.19        | 59.82   |
| <sup>a</sup> Not sigr | ificantly  | different  | between t    | he sexes    |             | DH15              | 32.26      | 2.59      | 27.95        | 1.43        | 103.74  |
| <sup>b</sup> p< 0.00  | 5 All the  | rest varia | ables diffe  | r signific  | antly       | <sup>b</sup> DH16 | 12.72      | 1.63      | 11.99        | 1.32        | 6.11    |
| between               | the sexes  | p< 0.001   | _            |             |             | DH17              | 23.66      | 2.10      | 22.05        | 1.99        | 15.57   |
|                       |            |            |              |             |             | DH18              | 45.77      | 3.80      | 39.27        | 2.12        | 109.66  |
|                       |            |            |              |             |             | <sup>b</sup> DH19 | 12.52      | 2.14      | 11.67        | 1.97        | 4.25    |
|                       |            |            |              |             |             | DH20              | 56.11      | 3.92      | 48.90        | 2.36        | 121.89  |
|                       |            |            |              |             |             | DH21              | 61.24      | 4.60      | 53.31        | 3.16        | 99.61   |

Table 7.2.11 Means, standard deviations and F-ratios for all measurements of proximal and distal humerus.

<u>Table 7.2.12</u> presents the results of the discriminant function analysis for single dimensions. F-ratios, cut-off values and classification accuracy for both original and cross-

validated data are presented here. PH1 and PH2 were the best discriminating variables, with 86.1% accuracy for both original and cross-validated data, followed by PH4 (86.1% for the original and 85.1% for the cross-validated data) and PH3 (84.2%). PH2 and PH5 represent the 2 sides of an isosceles triangle, thus they have the same length. PH7-PH10 did not perform that well, with accuracies that did not exceed 80%.

#### Distal humerus

From the seven landmarks, 21 distances were generated (<u>Table 6.3</u>). Descriptive statistics of the 21 dimensions on the distal humerus and univariate differences between the sexes are shown in <u>Table 7.2.11</u>. All variables were found significantly different between the sexes at the level of p<0.001, with the exception of DH1, which was found significantly different at the level of p<0.005, and DH16 and DH19, which were found significantly different at the level of p<0.05.

|       |         | Cut-off          |      |      | Original |         |       | Cross-validated |      |      |      |       |  |
|-------|---------|------------------|------|------|----------|---------|-------|-----------------|------|------|------|-------|--|
| *V    | F-ratio | Cut-off<br>value | Ma   | les  | Fem      | ales    | Total | Ma              | es   | Fema | ales | Total |  |
|       |         |                  | N=53 | %    | N=48     | %       | %     | N=53            | %    | N=48 | %    | %     |  |
|       |         |                  |      |      | Proxima  | l Humer | 18    |                 |      |      |      |       |  |
| PH1   | 121.05  | 44.17            | 43   | 81.1 | 44       | 91.7    | 86.1  | 43              | 81.1 | 44   | 91.7 | 86.1  |  |
| **PH2 | 91.32   | 28.25            | 45   | 84.9 | 42       | 87.5    | 86.1  | 45              | 84.9 | 42   | 87.5 | 86.1  |  |
| PH3   | 94.69   | 48.33            | 42   | 79.2 | 43       | 89.6    | 84.2  | 42              | 79.2 | 43   | 89.6 | 84.2  |  |
| PH4   | 126.71  | 49.8             | 44   | 83.0 | 43       | 89.6    | 86.1  | 43              | 81.1 | 43   | 89.6 | 85.1  |  |
|       |         |                  |      |      | Distal l | Humerus |       |                 |      |      |      |       |  |
| DH2   | 54.58   | 15.26            | 40   | 75.5 | 41       | 85.4    | 80.2  | 40              | 75.5 | 41   | 85.4 | 80.2  |  |
| DH3   | 85.07   | 28.18            | 43   | 81.1 | 43       | 89.6    | 85.1  | 43              | 81.1 | 43   | 89.6 | 85.1  |  |
| DH4   | 104.74  | 39.38            | 44   | 83.0 | 43       | 89.6    | 86.1  | 44              | 83.0 | 43   | 89.6 | 86.1  |  |
| DH5   | 92.59   | 46.37            | 43   | 81.1 | 40       | 83.3    | 82.2  | 43              | 81.1 | 40   | 83.3 | 82.2  |  |
| DH6   | 57.81   | 15.94            | 41   | 77.4 | 43       | 89.6    | 83.2  | 41              | 77.4 | 43   | 89.6 | 83.2  |  |
| DH8   | 89.71   | 26.36            | 45   | 84.9 | 41       | 85.4    | 85.1  | 45              | 84.9 | 40   | 83.3 | 84.2  |  |
| DH9   | 87.68   | 38.54            | 42   | 79.2 | 42       | 87.5    | 83.2  | 42              | 79.2 | 42   | 87.5 | 83.2  |  |
| DH10  | 82.54   | 47.44            | 42   | 79.2 | 41       | 85.4    | 82.2  | 42              | 79.2 | 41   | 85.4 | 82.2  |  |
| DH13  | 63.72   | 24.65            | 41   | 77.4 | 42       | 79.2    | 82.2  | 41              | 77.4 | 42   | 79.2 | 82.2  |  |
| DH15  | 103.74  | 30.11            | 40   | 75.5 | 46       | 95.8    | 85.1  | 40              | 75.5 | 46   | 95.8 | 85.1  |  |
| DH18  | 109.66  | 42.52            | 41   | 77.4 | 46       | 95.8    | 86.1  | 41              | 77.4 | 46   | 95.8 | 86.1  |  |
| DH20  | 121.89  | 52.5             | 43   | 81.1 | 44       | 91.7    | 86.1  | 43              | 81.1 | 44   | 91.7 | 86.1  |  |
| DH21  | 99.61   | 57.27            | 42   | 79.2 | 46       | 95.8    | 84.2  | 42              | 79.2 | 46   | 95.8 | 84.2  |  |

| Table 7.2.12 Univariate statistics for | the measurements on the | e radiographs of the | proximal and distal humerus. |
|--|-------------------------|----------------------|------------------------------|
|  |                         |                      |                              |

<sup>\*</sup> Only variables with classification accuracy>80% are included in the table

<sup>\*\*</sup> PH2 and PH5 have the same values so only PH2 in presented in the table

|      |         | Pro          | oximal Hu | merus |      |      |       |           | Di   | istal Hume  | erus         |      |      |      |      |       |           |
|------|---------|--------------|-----------|-------|------|------|-------|-----------|------|-------------|--------------|------|------|------|------|-------|-----------|
|      | F-ratio | Raw          | Ma        | ıle   | Fem  | ale  | Total |           |      | F-<br>ratio | Raw          | Ma   | ıle  | Fen  | nale | Total |           |
|      |         | coefficients | N=53      | %     | N=48 | %    | %     |           |      |             | coefficients | N=53 | %    | N=48 | %    | %     |           |
|      |         |              | PI        | HF1   |      |      |       |           | DHF1 |             |              |      |      |      |      |       |           |
| PH1  | 46.82   | 0.211340     | 45        | 84.9  | 45   | 93.8 | 89.1  | Original  | DH20 | 56.11       | -0.090012    | 45   | 84.9 | 45   | 93.8 | 89.1  | Original  |
| PH3  | 51.02   | 0.333786     | 44        | 83.0  | 44   | 01.7 | 87.1  | Cross-    | DH18 | 45.77       | 0.050562     | 44   | 83.0 | 13   | 80.6 | 86.1  | Cross-    |
| PH4  | 52.82   | -0.148356    | 44        | 85.0  | 44   | 91.7 | 07.1  | validated | DH4  | 41.94       | 0.962343     | 44   | 85.0 | 45   | 89.0 | 80.1  | validated |
| PH7  | 13.47   | 0.064807     |           |       |      |      |       |           | DH15 | 32.26       | 0.290986     |      |      |      |      |       |           |
| PH8  | 35.76   | -1.006738    |           |       |      |      |       |           | DH21 | 61.24       | 0.039110     |      |      |      |      |       |           |
| PH9  | 41.66   | 0.958213     |           |       |      |      |       |           | DH5  | 49.33       | -0.117982    |      |      |      |      |       |           |
| PH10 | 7.88    | -0.615780    |           |       |      |      |       |           | DH8  | 28.45       | 0.778707     |      |      |      |      |       |           |
| Cor  | nstant  | -17.768589   |           |       |      |      |       |           | DH9  | 41.01       | -0.782126    |      |      |      |      |       |           |
|      |         |              | PI        | HF2   |      |      |       |           | DH3  | 30.37       | -0.713853    |      |      |      |      |       |           |
| PH1  | 46.82   | 0.217321     | 45        | 84.9  | 45   | 93.8 | 89.1  | Original  | DH13 | 26.19       | 0.180293     |      |      |      |      |       |           |
| PH2  | 30.25   | 0.093335     | 44        | 83.0  | 15   | 03.8 | 88.1  | Cross-    | Cons | stant       | -15.559626   |      |      |      |      |       |           |
| PH3  | 51.02   | -0.371965    |           | 05.0  | 45   | 75.0 | 00.1  | validated |      |             |              | Γ    | OHF2 |      |      |       |           |
| PH4  | 52.82   | 0.479580     |           |       |      |      |       |           | DH20 | 56.11       | 0.098027     | 43   | 81.1 | 46   | 95.8 | 88.1  | Original  |
|      |         | -18.121070   |           |       |      |      |       |           | DH18 | 45.77       | 0.046350     | 13   | Q1 1 | 44   | 01.7 | 86.1  | Cross-    |
|      |         |              | PI        | HF3   |      |      |       |           | DH4  | 41.94       | 0.131566     | 43   | 01.1 | 44   | 91.7 | 80.1  | validated |
| PH1  | 46.82   | 0.304936     | 44        | 83.0  | 45   | 93.8 | 88.1  | Original  | DH15 | 32.26       | 0.129463     |      |      |      |      |       |           |
| PH3  | 51.02   | -0.408107    | 12        | 82.0  | 15   | 02.8 | 97.1  | Cross-    | Cons | stant       | -16.196080   |      |      |      |      |       |           |
| PH4  | 52.82   | 0.493376     | 43        | 85.0  | 4.5  | 95.0 | 07.1  | validated |      |             | Γ            | OHF3 |      |      |      |       |           |
| Cor  | nstant  | -18.294018   |           |       |      |      |       |           | DH20 | 56.11       | 0.279626     | 43   | 81.1 | 45   | 93.8 | 87.1  | Original  |
|      |         |              |           |       |      |      |       |           | DH5  | 49.33       | 0.031957     | 12   | Q1 1 | 16   | 05.8 | 96.1  | Cross-    |
|      |         |              |           |       |      |      |       |           | Cons | stant       | -16.163374   | 43   | 01.1 | 40   | 93.0 | 00.1  | validated |

Table 7.2.13 Discriminant functions and classification accuracies for the proximal and distal humerus. Sectioning point is set to zero in all cases

<u>Table 7.2.12</u> presents the results of the discriminant function analysis for single dimensions. F-ratios, cut-off values and classification accuracy for both original and cross-validated data are presented here. DH4, DH18 and DH20 are the single variables that gave the best classification accuracies (86.1%) for both original and cross-validation samples. In total, 13 variables yielded more than 80% correct group assignment. Females tended to be better classified compared to males in almost all cases.

### Multivariate statistics

#### Proximal humerus

Various formulae were produced using direct and stepwise discriminate function analysis of various combinations of the variables for the proximal humerus. When all 10 variables were used with a direct procedure, classification accuracy did not exceed 80%. The same happened when stepwise procedure was applied to the variables. The variables were combined in many different ways, seeking the highest possible classification accuracy for both original and cross-validated data. The best formulae are presented in <u>Table 7.2.13</u>.

PHF1 is the result of direct DFA using PF1, PF3, PF4, PH7, PH8, PH9 and PH10; PHF2 uses PF1-PF4 whereas PHF3 uses 3 measurements (PH1, PH3 and PH4). PHF2 is the formula that separated the sexes with the best accuracy (89.1% for original and 88.1% for cross-validated data). PHF1 and PHF3 performed slightly worse after cross-validation (87.1%) (see <u>Table 7.2.13</u>). It is noteworthy that stepwise procedure using 9 variables that were found significantly different between the sexes selected only one variable: PH4. The cut-off value and classification accuracy for PH4 are presented in <u>Table 7.2.12</u>.

#### Distal humerus

Various formulae were produced using direct and stepwise discriminate function analysis of various combinations of the variables for the distal humerus. When direct DFA was applied to all 21 measurements, 7 of them were rejected automatically due to high covariation with some of the remaining 14 measurements. The combination of the 14 measurements gave a classification accuracy of 89.1% for the original data and 85.1% for the cross-validated sample. Many different combinations of 14, 13, 12 and 11 variables gave exactly the same classification results. <u>Table 7.2.13</u> presents DHF1, which is the one of the formulae with 11 variables and classification accuracy of 89.1% for the original data and 86.1% for the cross-validated sample. Stepwise procedure selected only one variable (DH20) with classification accuracy of 86.1% (see <u>Table 7.2.12</u>). Various combinations of different number of variables were applied and some of the formulae that best separated the sexes are presented in <u>Table 7.2.13</u>. DHF2 is the result of a direct analysis using a

# combination of 4 variables (DH20, DH18, DH4 and DH 15). Classification accuracy for the original sample reached 88.1% and cross-validation performed slightly lower (86.1%).

|                   |          | Pro   | oximal Rad   | lius       |         |   |             | Ι          | Distal Radi   | us          |          |  |  |
|-------------------|----------|-------|--------------|------------|---------|---|-------------|------------|---------------|-------------|----------|--|--|
|                   | Males (1 | N=53) | Fema<br>(N=5 | les<br>50) |         |   | Males (     | N=52)      | Fema<br>(N=4  | ales<br>49) |          |  |  |
| V                 | Mean     | SD    | Mean         | SD         | F-ratio | V   | Mean SD     |            | Mean          | SD          | F-ratio  |  |  |
| <sup>d</sup> PR1  | 13.21    | 1.3   | 12.08        | 3.9        | 4.05    | <sup>a</sup> DR1                              | 31.13       | 2.54       | 28.05         | 1.99        | 45.72    |  |  |
| PR2               | 27.82    | 3.9   | 27.17        | 7.8        | 0.28    | <sup>a</sup> DR2                              | 32.98       | 2.54       | 29.53         | 1.94        | 58.43    |  |  |
| PR3               | 29.58    | 3.9   | 28.3         | 7.9        | 1.10    | DR3   | 4.22        | 1.38       | 4.06          | 1.10        | 0.44     |  |  |
| PR4               | 42.36    | 4.3   | 40.31        | 9.5        | 2.05    | <sup>a</sup> DR4                              | 25.14       | 2.35       | 23.15         | 2.00        | 20.91    |  |  |
| <sup>d</sup> PR5  | 43.68    | 3.9   | 40.42        | 10         | 4.64    | <sup>a</sup> DR5                              | 32.19       | 2.68       | 28.18         | 1.75        | 78.6     |  |  |
| PR6               | 15.07    | 3.3   | 14.73        | 4.9        | 0.17    | <sup>a</sup> DR6                              | 10.25       | 1.6        | 7.89          | 1.22        | 68.65    |  |  |
| PR7               | 19.42    | 2.5   | 18.23        | 5.5        | 2.05    | <sup>a</sup> DR7                              | 27.08       | 1.95       | 24.2          | 1.66        | 63.45    |  |  |
| PR8               | 30.01    | 3.4   | 29.07        | 8.2        | 0.58    | <sup>a</sup> DR8                              | 6.51        | 1.45       | 5.24          | 1.13        | 23.9     |  |  |
| PR9               | 25.6     | 3.8   | 24.82        | 6.6        | 0.54    | <sup>a</sup> DR9                              | 17.66       | 2.11       | 15.37         | 2.20        | 28.59    |  |  |
| PR10              | 43.9     | 3.9   | 41.52        | 9.9        | 2.62    | <sup>a</sup> DR10                             | 29.24       | 2.09       | 26.08         | 1.65        | 70.65    |  |  |
| <sup>d</sup> PR11 | 38.51    | 3.8   | 35.8         | 8.6        | 4.38    | <sup>a</sup> DR11                             | 10.3        | 1.19       | 8.37          | 1.03        | 76.01    |  |  |
| PR12              | 21.86    | 2.6   | 21.14        | 6.8        | 0.51    | DR12  | 8.15        | 1.95       | 8.31          | 2.21        | 0.15     |  |  |
| PR13              | 13.4     | 3     | 13.14        | 4          | 0.14    | <sup>a</sup> DR13                             | 21.12       | 1.82       | 19.38         | 1.75        | 23.96    |  |  |
| PR14              | 13.13    | 1.4   | 12.07        | 4.4        | 2.85    | <sup>a</sup> DR14                             | 29          | 2.14       | 25.45         | 1.53        | 90.51    |  |  |
| <sup>d</sup> PR15 | 14.61    | 3     | 13.17        | 2.8        | 6.36    | <sup>a</sup> DR15                             | 15.26       | 1.64       | 13.34         | 1.76        | 32.04    |  |  |
| <sup>b</sup> PR16 | 22.91    | 2.1   | 20.43        | 5.8        | 8.54    |   |             |            |               |             |          |  |  |
| PR17              | 13.83    | 3.6   | 13.86        | 4.5        | 0.00    |   |             |            |               |             |          |  |  |
| PR18              | 18.84    | 2.7   | 17.71        | 5.5        | 1.81    |   |             |            |               |             |          |  |  |
| <sup>c</sup> PR19 | 21.51    | 1.9   | 19.39        | 5.5        | 6.97    |   |             |            |               |             |          |  |  |
| <sup>b</sup> PR20 | 14.17    | 2.4   | 12.27        | 3.3        | 11.30   | <sup>a</sup> Significa                        | ntly differ | ent at the | level of p<   | 0.001       |          |  |  |
| PR21              | 20.75    | 3.1   | 20.04        | 6.7        | 0.48    | <sup>b</sup> p< 0.005<br><sup>c</sup> p< 0.01 |             |            |               |             |          |  |  |
| PR22              | 12.22    | 3.3   | 11.71        | 3.4        | 0.61    | <sup>d</sup> p< 0.05                          |             |            |               |             |          |  |  |
| <sup>d</sup> PR23 | 21.56    | 1.8   | 19.35        | 6.1        | 6.30    | All the rest                                  | t variables | do not di  | ffer signific | cantly bet  | ween the |  |  |
| PR24              | 27.93    | 4.2   | 26.56        | 5.9        | 1.85    | SEXES   |             |            |               |             |          |  |  |
| <sup>d</sup> PR25 | 31.49    | 3.5   | 29.11        | 7.1        | 4.73    | 3   |             |            |               |             |          |  |  |
| PR26              | 34.01    | 3.7   | 31.59        | 8.9        | 3.31    |   |             |            |               |             |          |  |  |
| <sup>c</sup> PR27 | 25.53    | 3.5   | 23.15        | 5.4        | 7.08    |   |             |            |               |             |          |  |  |
| PR28              | 16.66    | 1.6   | 15.91        | 5.9        | 0.80    |   |             |            |               |             |          |  |  |

Table 7.2.14 Means, standard deviations and F-ratios for all measurements of proximal and distal radius.

DHF3 is the result of a direct DFA using only DH20 and DH5. This formula has the minimum number of variables with the highest possible accuracy for the cross-validated data. All functions, coefficients and constants are presented in <u>Table 7.2.13</u>. Sectioning point is set to zero in all cases.

## 7.2.2.2 Radius

A total of 101 (53 males and 48 females) adult radii were X-rayed. Eight landmarks were selected on the proximal and five on the distal radius (see Fig 6.3).

#### Univariate statistics

#### Proximal radius

Combining the eight selected landmarks on the proximal radius, 28 distances (PR1-PR28) were generated (Table 6.3). Descriptive statistics of the 28 dimensions and univariate differences between the sexes are shown in Table 7.2.14. Seven variables (PR1, PR5, PR11, PR15, PR19, PR23 and PR25) were found significantly different between the sexes at the level of p<0.05 and three variables (PR16, PR20, PR27) were found significantly different at the level of p<0.01. The remaining variables don't exhibit statistically significant differences between the two groups and therefore were excluded from further analysis.

| V    | F-ratio         | Cut-off          | Ma   | les  | Fema   | ales   | Total | Males |      | Females |      | Total |
|------|-----------------|------------------|------|------|--------|--------|-------|-------|------|---------|------|-------|
| v    | r-rauo          | value            | N=53 | %    | N=50   | %      | %     | N=53  | %    | N=50    | %    | %     |
|      | Proximal Radius |                  |      |      |        |        |       |       |      |         |      |       |
| PR16 | 8.54            | 21.67            | 40   | 75.5 | 45     | 90.0   | 82.5  | 40    | 75.5 | 45      | 90.0 | 82.5  |
| PR23 | 6.3             | 20.17            | 41   | 77.4 | 47     | 94.0   | 85.4  | 41    | 77.4 | 47      | 94.0 | 85.4  |
|      |                 |                  |      |      | Distal | Radius |       |       |      |         |      |       |
| v    | F-ratio         | Cut-off<br>value | N=52 | %    | N=49   | %      | %     | N=52  | %    | N=49    | %    | %     |
| DR5  | 78.6            | 30.18            | 39   | 75.0 | 42     | 85.7   | 80.2  | 39    | 75.0 | 42      | 85.7 | 80.2  |
| DR11 | 76.01           | 9.34             | 41   | 78.8 | 40     | 81.6   | 80.2  | 41    | 78.8 | 39      | 79.6 | 79.2  |
| DR14 | 90.51           | 27.22            | 39   | 75.0 | 45     | 91.8   | 83.2  | 39    | 75.0 | 45      | 91.8 | 83.2  |

Table 7.2.15 F-ratios, cut-off values and classification accuracies for the proximal and distal radius.

The 10 single dimensions of the proximal radius that were found to differ significantly between the sexes were submitted to DFA, but only two performed well. More specifically, PR16 and PR23 were the only variables with classification accuracies that exceeded 80%. F-ratios, cut-off values and classification accuracy for both original and cross-validated data are presented in <u>Table 7.2.15</u>. PR23 (85.4%) was the best discriminating variable followed by PR16 (82.5%). Interestingly, PR20, which has the

highest F-ratio (see <u>Table 7.2.14</u>), was not included in the best single variables, yielding only 73% accuracy.

## Distal radius

Combining the five selected landmarks on the distal radius, 15 distances (DR1-PR15) were generated (Table 6.6). Descriptive statistics of the 15 dimensions and univariate differences between the sexes are shown in Table 7.2.14. All variables were found to differ significantly between the sexes at the level of p < 0.0001, except DR3 and DR12, which were excluded from further analysis. The remaining 12 variables were submitted to DFA, but only three of them resulted in classification accuracies higher than 80%. More specifically, DR14 was found to be the most effective single dimension yielding 83.2% accuracy, followed by DR5 and DR11, which both performed the same (80.2%) for the original data. F-ratios, cut-off values and classification accuracy for both original and cross-validated data are presented in Table 7.2.15.

#### Multivariate statistics

#### Proximal radius

Various formulae were produced using direct and stepwise discriminate function analysis of various combinations of the variables for the proximal radius. Only the 10 single dimensions that were found to differ significantly between the sexes (see <u>Table 7.2.14</u>) were employed for this analysis. When all 10 variables were used with a direct procedure, classification accuracy did not exceed 80%. The same happened when stepwise procedure was applied. The variables were combined in many different ways, seeking the highest possible classification accuracy for both original and cross-validated data. The best combination using only two variables did not exceed the accuracies of single variables, nor the cut-off of 80% that was set as a limit in this study; therefore this formula is not presented here.

# Distal radius

Various formulae were produced using direct and stepwise DFA of various different combinations of the 12 variables for the distal radius. When direct DFA was applied to all 12 measurements, 3 of them (DR11, DR13 and DR14) were rejected automatically due to high co-variation with some of the remaining 9 measurements. The combination of the 9 measurements (DRF1) gave a classification accuracy of 88.1% for the original data and 84.2% for the cross-validated sample. When stepwise procedure was applied (DRF2), only two (DR6 and DR14) out of nine variables were selected.

Many different combinations gave similar classification results for the original data, but worse for the cross-validated ones. Some of the best formulae for separating the sexes along with classification results for both original and cross-validated data are presented in <u>Table 7.2.16</u>. Only formulae with accuracies exceeding 80% for cross-validated data are included. Sectioning point is set to zero in all cases. DRF1 is the result of a direct DFA using the three most effective single variables (DR6, DR11 and DR14). Classification accuracy reached 86.1% for original, whereas "leave-one-out" classification was only slightly lower.

| V    | Entin    | Raw          | Ma   | lle  | Fem  | ale  | Total |           |
|------|----------|--------------|------|------|------|------|-------|-----------|
| v    | F-rano   | coefficients | N=52 | %    | N=49 | %    | %     |           |
|      |          |              | DR   | F1   |      |      |       |           |
| DR1  | 45.72    | 2.7045988    | 44   | 84.6 | 45   | 91.8 | 88.1  | Original  |
| DR2  | 58.43    | 0.0192056    | 41   | 79.8 | 44   | 80.8 | 84.2  | Cross-    |
| DR4  | 20.91    | -3.4719622   | 41   | 79.0 |      | 07.0 | 04.2  | validated |
| DR5  | 78.6     | 0.6891741    |      |      |      |      |       |           |
| DR6  | 68.65    | 0.4324208    |      |      |      |      |       |           |
| DR7  | 63.45    | 0.6697782    |      |      |      |      |       |           |
| DR8  | 23.9     | -3.2662015   |      |      |      |      |       |           |
| DR9  | 28.59    | 0.6659321    |      |      |      |      |       |           |
| DR10 | 70.65    | -0.2356804   |      |      |      |      |       |           |
| DR15 | 32.04    | -0.7590823   |      |      |      |      |       |           |
| Co   | nstant   | -13.186245   |      |      |      |      |       |           |
|      |          |              | DR   | F2   |      |      |       |           |
| DR14 | 90.50604 | 0.3693301    | 42   | 80.8 | 43   | 87.8 | 84.2  | Original  |
| DR6  | 57.54669 | 0.3557052    | 42   | 00.0 | 12   | 070  | 84.2  | Cross-    |
| Co   | nstant   | -13.279876   | 42   | 80.8 | 43   | 07.0 | 04.2  | validated |
|      |          |              | DR   | F3   |      |      |       |           |
| DR6  | 68.65    | 0.2419291    | 42   | 80.8 | 45   | 91.8 | 86.2  | Original  |
| DR14 | 90.51    | 0.3293304    | 41   | 78.8 | 44   | 80.8 | 84.2  | Cross-    |
| DR11 | 76.01    | 0.228618     | +1   | /0.0 | 44   | 09.0 | 04.2  | validated |
| Co   | nstant   | -13.29418    |      |      |      |      |       |           |

Table 7.2.16 Discriminant functions and classification accuracies for the distal radius.

#### 7.2.2.3Femur

A total of 105 (55 males and 50 females) adult femora were X-rayed. Eight landmarks were selected on the proximal and five on the distal femur (see Fig 6.4).

#### Univariate statistics

#### Proximal femur

Combining the eight selected landmarks on the proximal femur, 28 distances (PF1-PF28) were generated (<u>Table 6.8</u>). Descriptive statistics of the 28 dimensions and univariate differences between the sexes are shown in <u>Table 7.2.17</u>. From the 28 variables, only 3 (PF19, PF20, PF23) were not found to differ significantly between the sexes. Of the remaining 23 variables, PF21 was found significantly different between the two groups at the level of p < 0.05, PF8 at the level of p < 0.01 and all the rest at the level of p < 0.0001.

|                   |           | Proximal   | Femur        |             |         | Distal Femur        |               |            |             |             |         |  |  |  |
|-------------------|-----------|------------|--------------|-------------|---------|---------------------|---------------|------------|-------------|-------------|---------|--|--|--|
| v                 | Ma<br>(N= | les<br>54) | Fema<br>(N=: | ales<br>50) | F-ratio | v                   | Mal<br>(N=5   | es<br>54)  | Fema<br>(N= | ales<br>50) | F-ratio |  |  |  |
|                   | Mean      | SD         | Mean         | SD          |         |                     | Mean          | SD         | Mean        | SD          |         |  |  |  |
| PF1               | 51.44     | 6.73       | 43.66        | 4.81        | 45.60   | DF1                 | 82.82         | 4.63       | 74.52       | 4.00        | 95.00   |  |  |  |
| PF2               | 61.47     | 6.44       | 52.50        | 5.64        | 57.04   | <sup>b</sup> DF2    | 19.35         | 3.97       | 17.18       | 3.32        | 9.01    |  |  |  |
| PF3               | 87.06     | 8.96       | 78.26        | 7.29        | 30.10   | DF3                 | 80.64         | 4.51       | 72.16       | 3.81        | 106.29  |  |  |  |
| PF4               | 71.90     | 7.59       | 61.65        | 5.63        | 60.84   | DF4                 | 49.64         | 3.22       | 44.92       | 2.87        | 62.09   |  |  |  |
| PF5               | 81.96     | 6.15       | 72.93        | 6.10        | 56.79   | DF5                 | 79.59         | 4.41       | 72.06       | 3.56        | 90.98   |  |  |  |
| PF6               | 62.41     | 5.20       | 55.41        | 5.07        | 48.51   | <sup>a</sup> DF6    | 13.96         | 2.21       | 14.25       | 2.56        | 0.37    |  |  |  |
| PF7               | 33.06     | 2.54       | 30.57        | 2.40        | 26.61   | DF7                 | 41.68         | 2.64       | 38.13       | 2.40        | 51.39   |  |  |  |
| <sup>b</sup> PF8  | 15.76     | 3.66       | 14.13        | 2.30        | 7.28    | DF8                 | 74.25         | 4.41       | 66.51       | 3.61        | 94.89   |  |  |  |
| PF9               | 42.41     | 3.86       | 39.40        | 3.00        | 19.52   | DF9                 | 40.41         | 2.71       | 36.71       | 2.14        | 58.96   |  |  |  |
| PF10              | 34.14     | 5.99       | 29.21        | 2.47        | 29.24   | DF10                | 34.17         | 2.54       | 30.18       | 2.05        | 77.18   |  |  |  |
| PF11              | 58.22     | 4.63       | 53.10        | 3.98        | 36.58   |                     |               |            |             |             |         |  |  |  |
| PF12              | 65.57     | 5.18       | 58.91        | 4.06        | 53.10   |                     |               |            |             |             |         |  |  |  |
| PF13              | 70.15     | 6.59       | 61.62        | 4.30        | 60.46   |                     |               |            |             |             |         |  |  |  |
| PF14              | 47.21     | 5.44       | 43.30        | 2.55        | 21.59   |                     |               |            |             |             |         |  |  |  |
| PF15              | 44.23     | 3.16       | 38.84        | 2.73        | 86.59   |                     |               |            |             |             |         |  |  |  |
| PF16              | 70.26     | 6.78       | 64.54        | 4.24        | 26.19   |                     |               |            |             |             |         |  |  |  |
| PF17              | 80.86     | 5.14       | 72.87        | 4.19        | 75.22   | <sup>a</sup> Do not | t differ sig  | nificantly | v between i | the grour   | 15      |  |  |  |
| PF18              | 83.92     | 5.99       | 74.05        | 5.00        | 83.07   | LONO                | t uniter sign | miediti    | between     | une group   |         |  |  |  |
| <sup>a</sup> PF19 | 19.22     | 8.74       | 19.03        | 5.12        | 0.02    | <sup>0</sup> Differ | significant   | ly at the  | level of p  | < 0.01      |         |  |  |  |
| <sup>a</sup> PF20 | 38.71     | 8.59       | 37.28        | 6.81        | 0.88    | <sup>c</sup> Differ | significant   | ly at the  | level of p  | < 0.05      | 4h      |  |  |  |
| °PF21             | 68.13     | 9.80       | 63.93        | 7.03        | 6.25    | sexes at            | the level of  | f p< 0.00  | 01          | ly betwee   | en the  |  |  |  |
| PF22              | 94.02     | 9.91       | 85.43        | 7.50        | 24.72   |                     |               |            |             |             |         |  |  |  |
| <sup>a</sup> PF23 | 27.68     | 4.66       | 26.66        | 3.93        | 1.47    |                     |               |            |             |             |         |  |  |  |
| PF24              | 51.23     | 6.03       | 46.50        | 4.68        | 19.90   |                     |               |            |             |             |         |  |  |  |
| PF25              | 76.37     | 7.89       | 66.62        | 5.47        | 53.09   |                     |               |            |             |             |         |  |  |  |
| PF26              | 36.73     | 5.41       | 33.45        | 4.11        | 12.04   |                     |               |            |             |             |         |  |  |  |
| PF27              | 74.68     | 6.83       | 65.99        | 6.48        | 44.49   |                     |               |            |             |             |         |  |  |  |
| PF28              | 43.08     | 5.75       | 36.97        | 5.72        | 29.70   |                     |               |            |             |             |         |  |  |  |

Table 7.2.17 Descriptive statistics for the measurements of the proximal and the distal femur.

The 25 single dimensions of the proximal femur that were found to differ significantly between the sexes were submitted to DFA and five of them gave accuracies beyond 80%

for both original and cross-validated data. (Table 7.2.18). PF23 (85.7%) is the best discriminating variable followed by PF18 (81.5%).

# Distal femur

The combination of the selected five landmarks on the distal femur resulted in the generation of 10 variables (DF1-DF10) as can be seen in Table 6.8 Descriptive statistics of the 28 dimensions and univariate differences between the sexes are shown in <u>Table 7.2.17</u>. Only one (DF6) out of ten variables was not found to differ significantly between the groups, while the remaining variables were significantly different at the level of p < 0.0001 with the exception of DF2 (p < 0.01). The 9 single dimensions of the distal femur that were found to differ significantly between the sexes were submitted to DFA and five of them gave accuracies beyond 80% for both original and cross-validated data.

|      |         | Cut-                 |      |      | Original |          |       | Cross-validated |      |      |      |       |
|------|---------|----------------------|------|------|----------|----------|-------|-----------------|------|------|------|-------|
| V    | F-ratio | off                  | Ma   | les  | Fema     | ales     | Total | Mal             | es   | Fema | ales | Total |
|      |         | value                | N=55 | %    | N=50     | %        | %     | N=55            | %    | N=50 | %    | %     |
|      |         |                      |      |      | Proxir   | nal Femu | r     |                 |      |      |      |       |
| PF2  | 57.04   | 56.98                | 42   | 76.4 | 44       | 88.0     | 81.9  | 42              | 76.4 | 43   | 86.0 | 81.0  |
| PF10 | 29.24   | 31.68                | 42   | 76.4 | 43       | 86.0     | 81.0  | 42              | 76.4 | 43   | 86.0 | 81.0  |
| PF12 | 53.1    | 62.24                | 43   | 78.2 | 41       | 82.0     | 80.0  | 43              | 78.2 | 41   | 82.0 | 80.0  |
| PF15 | 86.59   | 41.54                | 47   | 85.5 | 43       | 86.0     | 85.7  | 47              | 85.5 | 43   | 86.0 | 85.7  |
| PF18 | 83.07   | 78.98                | 44   | 80.0 | 42       | 84.0     | 91.9  | 44              | 80.0 | 42   | 84.0 | 81.9  |
|      |         |                      |      |      | Dista    | al Femur |       |                 |      |      |      |       |
| v    | F-ratio | Cut-<br>off<br>value | N=54 | %    | N=50     | %        | %     | N=54            | %    | N=50 | %    | %     |
| DF1  | 95      | 78.67                | 44   | 81.5 | 44       | 88.0     | 84.6  | 44              | 81.5 | 42   | 84.0 | 82.7  |
| DF3  | 106.3   | 76.4                 | 45   | 83.3 | 44       | 88.0     | 85.6  | 45              | 83.3 | 43   | 86.0 | 84.6  |
| DF5  | 90.98   | 75.82                | 43   | 79.6 | 41       | 82.0     | 80.8  | 43              | 79.6 | 41   | 82.0 | 80.8  |
| DF8  | 94.89   | 70.38                | 45   | 83.3 | 43       | 86.0     | 84.6  | 43              | 79.6 | 43   | 86.0 | 82.7  |
| DF10 | 77.18   | 32.18                | 45   | 83.3 | 43       | 86.0     | 84.6  | 45              | 83.3 | 43   | 86.0 | 84.6  |

Table 7.2.18 Discriminant functions and classification accuracies for the measurements on the radiographs of the proximal and the distal femur.

More specifically, DF3 and DF10 were found to be the most effective dimensions in sex estimation from the distal femur, with 84.6% correct group membership for the cross-validated data, whereas DF5 did not exceed 80% correct classification. F-ratios, cut-off values and classification accuracies of the single dimensions are presented in <u>Table 7.2.18</u>.

## Multivariate statistics

Proximal femur

Various formulae were produced using direct and stepwise DFA of various different combinations of the 25 variables that were found to differ significantly between the sexes.

|      |          |              | Proxir | nal Femu | ır   |          |       |           |  |  |
|------|----------|--------------|--------|----------|------|----------|-------|-----------|--|--|
| V    | Ensting  | Raw          | Ma     | ıle      | Fem  | ale      | Total |           |  |  |
| v    | r-ratios | coefficients | N=55   | %        | N=50 | %        | %     |           |  |  |
|      |          |              | PFF1   | (stepwis | e)   |          |       |           |  |  |
| PF15 | 86.59    | 0.196622     | 47     | 85.5     | 44   | 88.0     | 86.7  | Original  |  |  |
| PF18 | 52.68    | 0.096889     | 47     | 05 5     | 12   | 96.0     | 057   | Cross-    |  |  |
| Co   | nstant   | -15.82028    | 4/     | 83.3     | 43   | 80.0     | 83.7  | validated |  |  |
|      |          |              | I      | PFF2     |      |          |       |           |  |  |
| PF15 | 86.59    | 0.179154     | 47     | 85.5     | 44   | 88.0     | 86.7  | Original  |  |  |
| PF17 | 75.22    | 0.034372     | 47     | 95 5     | 44   | <u> </u> | 967   | Cross-    |  |  |
| PF18 | 83.07    | 0.079626     | 4/     | 05.5     | 44   | 88.0     | 80.7  | validated |  |  |
| Co   | nstant   | -16.373131   |        |          |      |          |       |           |  |  |
|      |          |              | F      | PFF3     |      |          |       |           |  |  |
| PF2  | 57.04    | 0.129837     | 47     | 85.5     | 48   | 96.0     | 90.5  | Original  |  |  |
| PF10 | 29.24    | -0.141007    | 45     | 01.0     | 45   | 00.0     | 95 7  | Cross-    |  |  |
| PF12 | 53.10    | 0.189398     | 45     | 81.8     | 45   | 90.0     | 85.7  | validated |  |  |
| PF13 | 60.46    | 0.057363     |        |          |      |          |       |           |  |  |
| PF14 | 21.59    | -0.27526     |        |          |      |          |       |           |  |  |
| PF15 | 86.59    | 0.487611     |        |          |      |          |       |           |  |  |
| PF18 | 83.07    | -0.152432    |        |          |      |          |       |           |  |  |
| Co   | nstant   | 14.258185    |        |          |      |          |       |           |  |  |
|      |          |              | Dista  | al Femur |      |          |       |           |  |  |
| V    | E ratios | Raw          | Ma     | ıle      | Fem  | ale      | Total |           |  |  |
| v    | 1-141105 | coefficients | N=54   | %        | N=50 | %        | %     |           |  |  |
|      |          |              | Γ      | OFF1     |      |          |       |           |  |  |
| DF1  | 95.00    | -0.215007    | 46     | 85.2     | 47   | 94.0     | 89.4  | Original  |  |  |
| DF3  | 106.29   | 0.743905     | 45     | 83.3     | 16   | 02.0     | 87.5  | Cross-    |  |  |
| DF4  | 62.09    | -0.293489    | 45     | 85.5     | 40   | 92.0     | 87.5  | validated |  |  |
| DF10 | 77.18    | -0.259801    |        |          |      |          |       |           |  |  |
| Co   | nstant   | -17.685623   |        |          |      |          |       |           |  |  |
|      | DFF2     |              |        |          |      |          |       |           |  |  |
| DF1  | 95.00    | -0.215442    | 47     | 87.0     | 46   | 83.3     | 90.4  | Original  |  |  |
| DF3  | 106.29   | 0.750454     | 15     | 82.2     | 16   | 92.0     | 87.5  | Cross-    |  |  |
| DF4  | 62.09    | -0.294067    | 43     | 05.5     | 40   | 92.0     | 07.5  | validated |  |  |
| DF8  | 94.89    | -0.008329    |        |          |      |          |       |           |  |  |
| DF10 | 77.18    | -0.255968    |        |          |      |          |       |           |  |  |
| Co   | nstant   | -17.661638   |        |          |      |          |       |           |  |  |

Table 7.2.19 Multivariate discriminant functions and classification accuracies for the proximal and distal femur.

When direct DFA was applied to all 25 measurements, 7 of them (PF17, PF18, PF22, PF24, PF25, PF27 and PF28) were rejected automatically due to high co-variation with
some of the remaining 18 measurements. The combination of the remaining 15 measurements gave 92.4% accuracy for the original data and 84.8% for the cross-validated sample. When stepwise procedure was applied (PFF1), only two (PF15 and PF18) out of 25 variables were selected (Table 7.2.19). PFF2 is the result of a direct DFA using 3 variables (PF15, PF17 and PF18) with high F-values and PFF3 is the result of a direct DFA using a combination of 7 variables. The best function was PFF2 with 86.6% correct group membership for both original and cross-validated data. All functions, corresponding coefficients and classification accuracies are shown in Table 7.2.19. The sectioning point is set to zero in all cases.

|                  |          | Proxim | nal Tibia    |             |         |  |             | Distal    | Tibia        |             |         |
|------------------|----------|--------|--------------|-------------|---------|--|-------------|-----------|--------------|-------------|---------|
| v                | Males (I | N=54)  | Fema<br>(N=4 | ules<br>19) |         | v  | Males (     | N=54)     | Fema<br>(N=4 | ales<br>49) |         |
|                  | Mean     | SD     | Mean         | SD          | F-ratio |  | Mean        | SD        | Mean         | SD          | F-ratio |
| PT1              | 32.5     | 2.3    | 29.13        | 1.88        | 65.54   | <sup>b</sup> DT1   | 14.28       | 1.84      | 12.13        | 1.77        | 36.52   |
| PT2              | 42.44    | 2.84   | 38.95        | 2.1         | 49.62   | <sup>b</sup> DT2   | 15.47       | 1.21      | 14.38        | 1.68        | 14.52   |
| PT3              | 73.09    | 4.23   | 66.46        | 3.62        | 72.29   | <sup>b</sup> DT3   | 52.37       | 3.06      | 47.44        | 3.01        | 67.73   |
| PT4              | 77.05    | 4.74   | 69.2         | 3.66        | 87.07   | <sup>b</sup> DT4   | 42.41       | 2.97      | 38.81        | 2.67        | 41.58   |
| <sup>b</sup> PT5 | 19.91    | 2.87   | 18.32        | 2.69        | 8.33    | <sup>b</sup> DT5   | 16.43       | 1.56      | 15.22        | 1.77        | 13.40   |
| <sup>a</sup> PT6 | 10.44    | 1.75   | 10.32        | 1.44        | 0.14    | <sup>b</sup> DT6   | 49.38       | 3.13      | 44.18        | 2.99        | 74.00   |
| PT7              | 42.4     | 2.99   | 39.09        | 2.45        | 37.48   | <sup>b</sup> DT7   | 38.09       | 2.9       | 34.73        | 2.51        | 39.22   |
| PT8              | 49.10    | 3.81   | 44.01        | 2.65        | 60.64   | <sup>b</sup> DT8   | 37.06       | 2.54      | 33.23        | 2.29        | 64.20   |
| PT9              | 38.62    | 2.92   | 34.66        | 2.53        | 53.55   | <sup>b</sup> DT9   | 27.1        | 2.52      | 24.77        | 2.2         | 24.97   |
| PT10             | 32.11    | 2.43   | 28.95        | 2.59        | 40.89   | <sup>b</sup> DT10  | 11.85       | 2.07      | 9.75         | 1.91        | 28.51   |
| PT11             | 39.52    | 3.46   | 34.53        | 2.89        | 62.36   | <sup>a</sup> Do not  | differ sign | ificantly | between      | the group   | ps      |
| PT12             | 45.97    | 3.16   | 41.89        | 2.56        | 51.14   | <sup>b</sup> Differ significantly at the level of $p < 0.005$                                |             |           |              |             |         |
| PT13             | 13.29    | 3.54   | 10.32        | 2.44        | 24.08   |  |             |           |              |             |         |
| PT14             | 71.44    | 4.2    | 64.55        | 3.5         | 80.96   | 6 All the rest variables differ significantly between the sexes at the level of $p < 0.0001$ |             |           |              |             | en the  |
| PT15             | 72.03    | 4.13   | 64.53        | 3.49        | 97.79   |  |             | r (0.00)  |              |             |         |

Table 7.2.20 Means, Standard Deviations and F-ratios for all measurements of proximal and distal tibia.

## Distal femur

Several discriminant functions were produced using different combinations of the nine dimensions of the distal femur that were found to differ significantly between the sexes. Combinations of 7, 6, 5 and 4 variables gave the same results in many cases. Table 7.18 presents the results of one of these functions (DFF1), which employs 4 variables (DF1, DF3, DF4 and DF10). Classification yielded at 89.4%, whereas jack-knife procedure resulted in 87.5% accuracy. DFF2 is the result of a direct DFA using a combination of 5 variables (DF1, DF3, DF4, DF8 and DF10). In that case, group separation reached 91% for the original data, while the cross-validated sample gave 87.5% accuracy. Stepwise

procedure selected only one variable (DF3). All functions and the corresponding accuracies are presented in <u>Table 7.2.19</u>. The sectioning point is set to zero in all cases.

|      |         |                  |      |      | Original |           |       |      | Cr   | oss-valida | ted  |       |
|------|---------|------------------|------|------|----------|-----------|-------|------|------|------------|------|-------|
| V    | F-ratio | Cut-off<br>value | Ma   | ales | Fem      | ales      | Total | Ma   | les  | Fema       | ales | Total |
|      |         |                  | N=53 | %    | N=49     | %         | %     | N=53 | %    | N=49       | %    | %     |
|      |         |                  |      |      | Proxin   | nal Tibia |       |      |      |            |      |       |
| PT3  | 72.29   | 69.78            | 45   | 83.3 | 42       | 85.7      | 84.5  | 45   | 83.3 | 42         | 85.7 | 84.5  |
| PT4  | 87.07   | 73.13            | 44   | 81.5 | 45       | 91.8      | 86.4  | 44   | 81.5 | 44         | 91.8 | 85.4  |
| PT8  | 60.64   | 46.56            | 46   | 85.2 | 41       | 83.7      | 84.5  | 46   | 85.2 | 41         | 83.7 | 84.5  |
| PT11 | 62.36   | 37.02            | 43   | 79.6 | 41       | 83.7      | 81.6  | 43   | 79.6 | 41         | 83.7 | 81.6  |
| PT14 | 80.96   | 67.99            | 46   | 85.2 | 42       | 85.7      | 85.4  | 46   | 85.2 | 42         | 85.7 | 85.4  |
| PT15 | 97.79   | 68.28            | 48   | 88.9 | 43       | 87.8      | 88.3  | 48   | 88.9 | 43         | 87.8 | 88.3  |
|      |         |                  |      |      | Dista    | l Tibia   |       |      |      |            |      |       |
| DT3  | 67.73   | 49.90953         | 44   | 81.5 | 39       | 79.6      | 80.6  | 44   | 81.5 | 39         | 79.6 | 80.6  |

Table 7.2.21 Univariate statistics for the measurements of the proximal and the distal tibia.

## 7.2.1.4 Tibia

A total of 103 (54 males and 49 females) adult femora were X-rayed. Six landmarks were selected on the proximal and five on the distal tibia (see Fig 6.5).

#### Univariate statistics

# Proximal tibia

Combining the six selected landmarks on the proximal tibia, 15 distances (PT1-PT15) were generated (Table 6.10). Descriptive statistics of the 15 dimensions and univariate differences between the sexes are shown in Table 7.2.20. Out of 15 variables, only PT6 was not found to differ significantly between the sexes. The remaining variables were found significantly different between the two groups at the level of p < 0.0001, with the exception of PT5 (p < 0.005). The single variables were than submitted to DFA, resulting in classification accuracies up to 88.3%. More specifically, the most effective single variable for the separation of the two groups was PT15 (88.3%), followed by PT4, PT8 and PT14, which gave 85.4% accuracy (Table 7.2.20).

### Distal Tibia

Combining the six selected landmarks on the proximal tibia, 10 distances (DT1-DT15) were generated (<u>Table 6.10</u>). Descriptive statistics of the 10 dimensions and univariate differences between the sexes are shown in <u>Table 7.2.20</u>. All variables were found to differ significantly between the sexes (p<0.005). However only DT3 resulted in over 80% classification accuracy (<u>Table 7.2.21</u>).

|      |                     |                   | Proxin | nal Tibia   |      |      |       |           |
|------|---------------------|-------------------|--------|-------------|------|------|-------|-----------|
| N    | Endin               | Raw               | Ma     | ıle         | Fem  | ale  | Total |           |
| v    | F-ratios            | coefficients      | N=54   | %           | N=49 | %    | %     |           |
|      |                     |                   | P      | FF1         |      |      |       |           |
| PT15 | 80.96               | 0.27062184        | 48     | 88.9        | 43   | 88.0 | 88.3  | Original  |
| PT14 | 97.79               | -0.0113246        | 40     | 00.0        | 12   | 00 0 | 00 2  | Cross-    |
| Co   | nstant              | -17.707           | 48     | 88.9        | 45   | 88.0 | 88.5  | validated |
|      |                     |                   | P      | FF2         |      |      |       |           |
| PT15 | 80.96               | 0.290229          | 48     | 88.9        | 44   | 90.0 | 89.3  | Original  |
| PT14 | 97.79               | -0.026664         | 16     | 05.2        | 42   | 96.0 | 05 /  | Cross-    |
| PT8  | 60.64               | -0.122884         | 40     | 83.2        | 42   | 80.0 | 63.4  | validated |
| PT11 | 62.36               | 0.124432          |        |             |      |      |       |           |
| Co   | nstant              | -16.888675        |        |             |      |      |       |           |
|      |                     |                   | Dista  | l Tibia     |      |      |       |           |
| V    | Ensting             | Raw               | Ma     | ıle         | Fem  | ale  | Total |           |
| v    | F-ratios            | coefficients      | N=54   | %           | N=49 | %    | %     |           |
|      |                     |                   | D      | FF1         |      |      |       |           |
| DT6  | 36.52               | 0.211350          | 46     | 85.2        | 41   | 83.7 | 84.5  | Original  |
| DT1  | 74.00               | 0.268492          | 16     | 05.2        | 41   | 027  | 015   | Cross-    |
| DT3  | 64.2                | 0.055300          | 40     | 63.2        | 41   | 05.7 | 64.3  | validated |
| Co   | nstant              | -16.191490        |        |             |      |      |       |           |
|      |                     |                   | D      | FF2         |      |      |       |           |
| DT6  | 36.52               | 0.297264          | 46     | 85.2        | 43   | 87.8 | 86.4  | Original  |
| DT1  | 74.00               | 0.283574          | 16     | <b>95</b> 2 | 42   | 957  | 95 /  | Cross-    |
| DT5  | 13.4                | 13.4 -0.144025 46 |        | 03.2        | 42   | 05.7 | 03.4  | validated |
| Co   | Constant -15.370390 |                   |        |             |      |      |       |           |
|      | DTF3                |                   |        |             |      |      |       |           |
| DT6  | 74.00               | 0.260587          | 46     | 85.2        | 41   | 83.7 | 84.5  | Original  |
| DT1  | 48.08               | 0.273534          | 15     | 02.2        | 40   | 016  | 02.5  | Cross-    |
| Co   | nstant              | -15.801350        | 43     | 03.3        | 40   | 01.0 | 82.3  | validated |

Table 7.2.22 Discriminant functions and classification accuracies for the proximal and distal tibia.

## Multivariate statistics

# Proximal tibia

Several discriminant functions were produced using different combinations of the nine dimensions of the proximal tibia. PTF1 is the result of a direct DFA using two variables (PT15 and PT14). Classification accuracy reached 88.3% fot both original and cross-validated data. PTF2 employed four variables and resulted in slightly higher accuracy for the original sample (89.3%). However cross-validation performed worse (85%) compared to PTF1). Both discriminant functions are presented in <u>Table 7.2.22</u>. The sectioning point is set to zero.



Figure 7.2.1: The best single radiometric variables for both epiphyses of the humerus (a, b), radius (c, d), femur (e, f) and tibia (g, h).

# Distal tibia

Various formulae were produced using direct and stepwise DFA of various different combinations of the 10 measurements taken on the radiograph of the distal tibia. Three of them (DTF1, DTF2 and DTF3) are presented in <u>Table 7.2.22</u>. DTF1 is the result of a direct DFA using DT6, DT1 and DT3 which resulted in 84.5% accuracy for both original and cross-validated data. DTF2 performed slightly better (86.5%) for the original sample; however jacknife procedure did not exceed 84.5% accuracy. Again, the sectioning point for all functions is set to zero.

Figure 7.2.1 illustates the best single variables for all the epiphyses examined.

# 7.2.3 Posterior probabilities

7.2.3.1 Humerus

## Univariate statistics

# Proximal humerus

Posterior probabilities for the measurements taken on the radiographs of the proximal humerus resulted in grouping up to 42.6% of the specimens at a 0.95 threshold with 86.1% accuracy. For PH1 values over 45.3mm for males and under 39.6mm for females classify the sample within 95% of confidence intervals (Table 7.2.3.1). Similarly PH4 classified over 73% of the sample at a 0.8 threshold, over 62% at a 0.9 and 42.6% at a 0.95 threshold with 86.1% accuracy. PH2 and PH3 grouped less than 30% of the specimens with 95% probability as it can be seen in Table 7.2.3.1.

# Distal humerus

In the case of distal humerus, 13 variables gave over 80% accuracy and therefore posterior probabilities were calculated for each one of them (Table 7.2.3.1). The most reliable variable was found to be DH20 which classified correctly over 41% of the specimens at a 0.95 threshold with 86.1% accuracy. DH18, DH15, DH21 and DH4 grouped over 30% of the sample at the same threshold with 84-86% accuracy. An individual with PH20>56.21 mm is classified as male within 90% of confidence intervals whereas if PH20> 57.49mm has 95% probability to be a male.

## Multivariate statistics

## Proximal humerus

The best multivariate discriminant functions for the proximal humerus were PHF1, PHF2 and PHF3 with with accuracies that reached 89%. All formulae classified over 70% of the sample with 80% probability. PHF1 performed better classifying over 67% of the specimens at a 0.9 and over 57% of the specimes and a 0.95 threshold with 89.1% accuracy (Table 7.2.3.2).

| PP  | Ma    | les  | Fema  | ales | Total | Ma    | les  | Fem   | ales | Total | Ma    | les  | Fem   | ales | Total | Ma    | les  | Fem   | ales | Total |
|-----|-------|------|-------|------|-------|-------|------|-------|------|-------|-------|------|-------|------|-------|-------|------|-------|------|-------|
| (%) | <     | %    | >     | %    | %     | <     | %    | >     | %    | %     | <     | %    | >     | %    | %     | <     | %    | >     | %    | %     |
|     |       |      | PH1   |      |       |       |      | DH3   |      |       |       |      | DH9   |      |       |       |      | DH20  |      |       |
| >95 | 47.50 | 45.3 | 40.76 | 39.6 | 42.6  | 32.29 | 24.5 | 24.54 | 18.8 | 21.8  | 43.18 | 22.6 | 34.51 | 18.8 | 20.8  | 57.49 | 47.2 | 48.46 | 35.4 | 41.6  |
| >90 | 46.80 | 64.2 | 41.43 | 56.3 | 60.4  | 31.30 | 37.7 | 25.45 | 33.3 | 35.6  | 41.95 | 34.0 | 35.65 | 37.5 | 35.6  | 56.21 | 56.6 | 49.56 | 52.1 | 54.5  |
| >80 | 45.75 | 71.7 | 42.41 | 66.7 | 69.3  | 30.25 | 58.5 | 26.57 | 62.5 | 60.4  | 40.83 | 56.6 | 36.77 | 62.5 | 59.4  | 55.35 | 67.9 | 50.14 | 75.0 | 71.3  |
| >50 | 44.17 | 81.1 | 44.17 | 91.7 | 86.1  | 28.39 | 75.5 | 28.39 | 89.6 | 85.1  | 38.78 | 79.2 | 38.78 | 87.5 | 83.2  | 52.86 | 75.5 | 52.86 | 91.7 | 86.1  |
|     |       |      | PH2   |      |       |       |      | DH4   |      |       |       |      | DH10  |      |       |       |      | DH18  |      |       |
| >95 | 31.59 | 28.3 | 24.95 | 22.9 | 25.7  | 43.36 | 37.7 | 35.98 | 22.9 | 30.7  | 52.95 | 28.3 | 42.54 | 20.8 | 24.8  | 47.26 | 41.5 | 38.38 | 27.1 | 34.7  |
| >90 | 30.87 | 37.7 | 25.77 | 43.8 | 40.6  | 42.43 | 47.2 | 36.92 | 50.0 | 48.5  | 51.60 | 39.6 | 43.68 | 39.6 | 39.6  | 46.16 | 62.3 | 39.51 | 45.8 | 54.5  |
| >80 | 29.95 | 60.4 | 26.66 | 60.4 | 60.4  | 41.38 | 64.2 | 37.88 | 66.7 | 65.3  | 50.17 | 56.6 | 45.27 | 54.2 | 55.4  | 44.99 | 67.9 | 40.67 | 72.9 | 70.3  |
| >50 | 28.25 | 84.9 | 28.25 | 87.5 | 86.1  | 39.63 | 83.0 | 39.63 | 89.6 | 86.1  | 47.73 | 79.2 | 47.73 | 85.4 | 82.2  | 42.84 | 77.4 | 42.84 | 87.5 | 86.1  |
|     |       |      | PH3   |      |       |       |      | DH5   |      |       |       |      | DH13  |      |       |       |      |       |      |       |
| >95 | 52.70 | 30.2 | 43.90 | 25.0 | 27.7  | 51.47 | 28.3 | 41.54 | 25.0 | 26.7  | 28.85 | 15.1 | 20.71 | 4.2  | 9.9   |       |      |       |      |       |
| >90 | 51.52 | 52.8 | 45.18 | 43.8 | 48.5  | 50.41 | 39.6 | 43.07 | 52.1 | 41.6  | 27.50 | 28.3 | 22.08 | 25.0 | 26.7  |       |      |       |      |       |
| >80 | 50.32 | 69.8 | 46.35 | 66.7 | 68.3  | 49.11 | 64.2 | 44.34 | 62.5 | 63.4  | 26.52 | 47.2 | 23.02 | 47.9 | 47.5  |       |      |       |      |       |
| >50 | 48.33 | 79.3 | 48.33 | 93.8 | 84.2  | 46.66 | 75.5 | 46.66 | 83.3 | 82.2  | 24.80 | 77.4 | 24.80 | 87.5 | 82.2  |       |      |       |      |       |
|     |       |      | PH4   |      |       |       |      | DH6   |      |       |       |      | DH15  |      |       |       |      |       |      |       |
| >95 | 53.44 | 45.3 | 46.13 | 39.6 | 42.6  | 19.20 | 15.1 |       | 0.0  | 7.9   | 33.48 | 43.4 | 27.09 | 22.9 | 33.7  |       |      |       |      |       |
| >90 | 52.51 | 66.0 | 47.04 | 58.3 | 62.4  | 18.35 | 30.2 | 13.78 | 18.8 | 24.8  | 32.97 | 49.1 | 27.95 | 50.0 | 49.5  |       |      |       |      |       |
| >80 | 51.57 | 75.5 | 47.99 | 70.8 | 73.3  | 17.51 | 50.9 | 14.51 | 43.8 | 47.5  | 31.84 | 62.3 | 28.86 | 62.5 | 62.4  |       |      |       |      |       |
| >50 | 49.76 | 83.0 | 49.76 | 93.8 | 86.1  | 16.06 | 77.4 | 16.06 | 89.6 | 83.2  | 30.32 | 75.5 | 30.32 | 87.5 | 85.1  |       |      |       |      |       |
|     |       |      | DH2   |      |       |       |      | DH8   |      |       |       |      | DH21  |      |       |       |      |       |      |       |
| >95 | 18.27 | 15.1 | 12.29 | 6.3  | 10.7  | 13.75 | 22.6 | 10.32 | 22.9 | 22.8  | 64.02 | 35.4 | 51.72 | 31.3 | 31.7  |       |      |       |      |       |
| >90 | 17.65 | 24.5 | 13.03 | 14.6 | 19.8  | 13.15 | 39.6 | 10.73 | 39.6 | 39.6  | 62.12 | 43.4 | 52.74 | 43.8 | 43.6  |       |      |       |      |       |
| >80 | 17.01 | 45.3 | 13.96 | 37.5 | 41.6  | 12.78 | 54.7 | 11.21 | 64.6 | 59.4  | 60.81 | 66.0 | 54.88 | 58.3 | 62.4  |       |      |       |      |       |
| >50 | 15.36 | 75.5 | 15.36 | 85.4 | 80.2  | 11.97 | 84.9 | 11.97 | 85.4 | 85.1  | 57.67 | 79.2 | 57.67 | 89.6 | 84.2  |       |      |       |      |       |

Table 7.2.3.1 Posterior probabilities of the measurements taken on the radiographs of the proximal (PH1-PH4) and distal (DH2-DH21) humerus.

Distal humerus

The best multivariate discriminant functions for the proximal humerus were DHF1, DHF2 and DHF3 with with accuracies that reached 89%. All formulae classified over 71% of the sample with 80% probability of correct assignment. PHF1 performed better classifying over 58% of the specimens at a 0.9 and over 45.5% of the specimes and a 0.95 threshold with 89.1% accuracy. For this function discriminant scores over 1.2957 classify males and under -1.3454 females at a 0.95 threshold (<u>Table 7.2.3.2</u>).

|           |         | Prox | timal humerus | ;    |       |         | Di   | stal humerus |      |       |
|-----------|---------|------|---------------|------|-------|---------|------|--------------|------|-------|
| PP<br>(%) | Male    | s    | Female        | es   | Total | Male    | s    | Female       | es   | Total |
| (/0)      | <       | %    | >             | %    | %     | <       | %    | >            | %    | %     |
|           |         |      | PHF1          |      |       |         |      | DHF1         |      |       |
| >95       | >1.1903 | 62.3 | <-1.246       | 52.1 | 57.4  | >1.2957 | 47.2 | <-1.3454     | 43.8 | 45.5  |
| >90       | >0.9785 | 67.9 | <-0.9144      | 66.7 | 67.3  | >0.9786 | 62.3 | <-0.9686     | 54.2 | 58.4  |
| >80       | >0.3858 | 73.6 | <-0.8169      | 72.9 | 73.3  | >0.6366 | 71.7 | <-0.6434     | 77.1 | 74.3  |
| >50       | >0      | 84.9 | <0            | 93.8 | 89.1  | >0      | 84.9 | <0           | 93.8 | 89.1  |
|           |         |      | PHF2          |      |       |         |      | DHF2         |      |       |
| >95       | >1.2496 | 58.5 | <-1.2454      | 50.0 | 54.5  | >1.3244 | 49.1 | <-1.3330     | 39.6 | 44.6  |
| >90       | >0.9145 | 67.9 | <-0.9102      | 62.5 | 65.3  | >1.0052 | 58.5 | <-1.0222     | 58.3 | 58.4  |
| >80       | >0.5943 | 75.5 | <-0.6494      | 70.3 | 73.3  | >0.7781 | 67.9 | <-0.6344     | 39.6 | 73.3  |
| >50       | >0      | 84.9 | <0            | 93.8 | 89.1  | >0      | 81.1 | <0           | 95.8 | 88.1  |
|           |         |      | PHF3          |      |       |         |      | DHF3         |      |       |
| >95       | >1.264  | 54.7 | <-1.2911      | 45.8 | 50.5  | >1.3773 | 45.3 | <-1.3342     | 39.6 | 42.6  |
| >90       | >0.9212 | 66.0 | <-0.9599      | 60.4 | 63.4  | >1.3114 | 56.6 | <-0.0283     | 52.1 | 54.5  |
| >80       | >0.5705 | 75.5 | <-0.619       | 70.8 | 77.2  | >0.6552 | 69.8 | <-0.6658     | 72.9 | 71.3  |
| >50       | >0      | 83.0 | <0            | 93.8 | 88.1  | >0      | 75.5 | <0           | 93.8 | 87.1  |

Table 7.2.3.2 Posterior probabilities of the multivariate functions for the proximal and distal humerus.

7.2.3.2 Radius

## Univariate statistics

Proximal radius

Posterior probabilities for the measurements taken on the radiographs of the proximal radius resulted in grouping all the specimens under a 0.8 threshold suggesting that there is a considerable degree of overlap between the two groups.

Distal radius

Posterior probabilities for the measurements taken on the radiographs of the distal radius classified up to 29% of the specimens at a 0.95 threshold. More specifically DR14 classified 43% of the sample at a 0.9 and 29% of the sample at a 0.95 threshold with 83%

accuracy. The cut-off values for this formula at a 0.95 threshold are 30.26mm for males and 16.3mm for females (Table 7.2.3.3).

| PP  | Mal   | es   | Fema  | ales | Total | Males   |      | Femal    | es   | Total |
|-----|-------|------|-------|------|-------|---------|------|----------|------|-------|
| (%) | <     | %    | >     | %    | %     | <       | %    | >        | %    | %     |
|     |       |      | DR5   |      |       |         |      | DRF1     |      |       |
| >95 | 34.00 | 28.9 | 25.93 | 14.3 | 21.8  | >1.2501 | 51.9 | <-1.2378 | 49.0 | 50.5  |
| >90 | 33.14 | 42.3 | 27.27 | 28.6 | 35.6  | >1.0732 | 59.6 | <-0.9202 | 69.4 | 64.4  |
| >80 | 32.46 | 55.8 | 28.37 | 57.1 | 56.4  | >0.5772 | 67.3 | <-0.6349 | 85.7 | 76.2  |
| >50 | 30.18 | 75.0 | 30.18 | 85.7 | 80.2  | >0      | 84.6 | >0       | 91.8 | 88.1  |
|     |       |      | DR11  |      |       |         |      | DRF2     |      |       |
| >95 | 11.35 | 21.2 | 7.40  | 18.4 | 19.8  | >1.4503 | 48.1 | <-1.426  | 28.6 | 38.6  |
| >90 | 10.76 | 32.7 | 7.80  | 36.7 | 34.7  | >1.1898 | 51.9 | <1.0419  | 63.3 | 57.4  |
| >80 | 10.26 | 55.8 | 8.44  | 55.1 | 55.5  | >0.6541 | 65.4 | <-0.6524 | 77.6 | 71.3  |
| >50 | 9.34  | 78.9 | 9.34  | 81.6 | 80.2  | >0      | 80.8 | >0       | 87.8 | 84.2  |
|     |       |      | DR14  |      |       |         |      | DRF3     |      |       |
| >95 | 30.26 | 42.3 | 24.24 | 16.3 | 29.7  | >1.4316 | 46.2 | <-1.3826 | 34.7 | 40.6  |
| >90 | 29.52 | 51.9 | 24.97 | 32.7 | 42.6  | >1.0569 | 59.6 | <-1.0207 | 61.2 | 60.4  |
| >80 | 28.84 | 61.5 | 25.82 | 55.1 | 58.4  | >0.6925 | 69.2 | <-0.7147 | 77.6 | 73.3  |
| >50 | 27.26 | 75.0 | 27.26 | 91.8 | 83.2  | >0      | 80.8 | >0       | 91.8 | 86.1  |

Table 7.2.3.3 Posterior probabilities for univariate and multivariate functions of the distal radius.

#### Multivariate statistics

Proximal radius

Multivariate discriminant functions using different number of variables of the proximal radius did not exceed the cut-off of 80% accuracy that it was set in this study. Therefore, posterior probabilities for the multivariate functions of the proximal radius are not presented here.

# Distal radius

The best multivariate discriminant function for the distal radius (DRF1) classified over 76% of the sample at a 0.8, over 64% at a 0.9 and over 50% at a 0.95 threshold exhibiting 88% correct group membership. For this function discriminant scores over 1.2501 classify males and under -1.2378 classify females at a 0.95 threshold. DRF3 classified over 60% of the sample with 90% probability and over 40% with 95% probability of correct group assignment with 86.1% accuracy. For this function and individual with DS>1.4316 has 95% probability to be a male while if DS<-1.3826 it has 95% probability to be a female. Posterior probabilities for all multiple discriminant functions of the distal radius are shown in Table 7.2.3.3.

|           |       | Pro  | oximal fem | ur   |       |       | Ε    | Distal femu | r    |       |
|-----------|-------|------|------------|------|-------|-------|------|-------------|------|-------|
| PP<br>(%) | Mal   | es   | Fema       | ales | Total | Males |      | Fema        | ales | Total |
|           | <     | %    | >          | %    | %     | <     | %    | >           | %    | %     |
|           |       |      | PF2        |      |       |       |      | DF3         |      |       |
| >95       | 68.84 | 13.3 | 44.19      | 8.0  | 9.5   | 82.53 | 27.3 | 70.20       | 30.0 | 28.9  |
| >90       | 66.46 | 18.2 | 47.83      | 14.0 | 16.2  | 81.09 | 48.2 | 71.59       | 40.0 | 44.2  |
| >80       | 62.92 | 38.2 | 50.99      | 38.0 | 38.1  | 79.31 | 66.7 | 73.41       | 62.0 | 64.4  |
| >50       | 56.98 | 76.4 | 56.98      | 88.0 | 81.9  | 76.40 | 83.3 | 76.40       | 88.0 | 85.6  |
|           |       |      | PF10       |      |       |       |      | DF1         |      |       |
| >95       | -     | 1.8  | -          | -    | 1.0   | 85.85 | 22.2 | 71.98       | 24.0 | 23.1  |
| >90       | 42.59 | 3.6  | -          | -    | 1.9   | 84.02 | 37.0 | 73.34       | 34.0 | 35.6  |
| >80       | 39.43 | 5.4  | 25.61      | 8.0  | 6.7   | 81.97 | 64.8 | 75.52       | 66.0 | 65.4  |
| >50       | 31.91 | 76.4 | 31.91      | 86.0 | 82.9  | 78.67 | 81.5 | 78.67       | 88.0 | 84.6  |
|           |       |      | PF12       |      |       |       |      | DF5         |      |       |
| >95       | 72.59 | 12.7 | 51.76      | 8.0  | 10.5  | 82.17 | 24.1 | 69.30       | 20.0 | 22.1  |
| >90       | 69.82 | 20.0 | 54.70      | 12.0 | 16.2  | 80.66 | 38.9 | 71.02       | 40.0 | 39.4  |
| >80       | 67.39 | 32.7 | 57.86      | 38.0 | 35.2  | 79.04 | 55.6 | 72.82       | 60.0 | 57.7  |
| >50       | 62.56 | 76.4 | 62.56      | 82.0 | 80.0  | 75.82 | 79.6 | 75.82       | 82.0 | 80.8  |
|           |       |      | PF15       |      |       |       |      | DF8         |      |       |
| >95       | 48.22 | 20.0 | 37.95      | 22.0 | 21.0  | 77.09 | 24.1 | 64.09       | 30.0 | 26.9  |
| >90       | 46.74 | 30.9 | 39.20      | 36.0 | 33.3  | 75.05 | 42.6 | 65.64       | 42.0 | 42.3  |
| >80       | 45.34 | 56.4 | 40.51      | 54.0 | 55.2  | 73.50 | 57.4 | 67.32       | 60.0 | 58.7  |
| >50       | 41.80 | 85.5 | 41.80      | 86.0 | 85.7  | 70.38 | 83.3 | 70.38       | 86.0 | 84.6  |
|           |       |      | PF18       |      |       |       |      | DF10        |      |       |
| >95       | 88.43 | 25.5 | 68.58      | 18.0 | 21.9  | 36.21 | 18.5 | 28.05       | 18.0 | 18.3  |
| >90       | 86.02 | 30.9 | 72.15      | 34.0 | 32.4  | 35.15 | 25.9 | 29.08       | 38.0 | 31.7  |
| >80       | 83.37 | 52.7 | 74.50      | 50.0 | 51.4  | 34.06 | 50.0 | 30.21       | 52.0 | 51.0  |
| >50       | 78.98 | 80.0 | 78.98      | 84.0 | 81.9  | 32.18 | 83.3 | 32.18       | 86.0 | 84.6  |

Table 7.2.3.4 Posterior probabilities for the single variables on the radiographs of the proximal and distal femur

## 7.2.3.3.Femur

## Univariate statistics

## Proximal femur

Posterior probabilities for the measurements taken on the radiographs of the proximal femur classified up to 22% of the specimens at a 0.95 threshold with 86.1% accuracy. The best single variable was found to be PF18. Aproximatelly 50% of the sample was correctly classified at a 0.8 threshold and 22% of the specimens at a 0.95 threshold. PF10 classified only one individual (male) with 99.9% probability of correct assignment therefore the cut-off value for the 0.95 threshold could not be estimated. No female was grouped within 90% of confidence intervals therefore no demarking value PF10 in females for is presented in <u>Table 7.2.3.4</u>.

Distal Femur

Single variables for the distal femur performed generally better reaching 29% correct group membership at a 0.95 threshold. Posterior probabilities for the best discriminatory variables (>80% accuracy) are calculated at 0.8, 0.9 and 0.95 thresholds. DF3 is found to be the most reliable single variable classifying 40% of the sample at a 0.9 threshold and 29% at a 0.95 threshold with 85.6% accuracy. DF10 performed slightly lower as it can be seen in Table 7.2.3.4.

## Multivariate statistics

Proximal femur

The best multivariate discriminant function for the proximal femur (PFF3) classified over 66% of the sample at a 0.8, over 52% at a 0.9 and about 40% at a 0.95 threshold exhibiting 89% correct group membership. For this function discriminant scores over 1.3224 classify males and under -1.4862 classify females at a 0.95 threshold. Posterior probabilities for all multiple discriminant functions of the proximal femur with over 80% accuracy are presented in <u>Table 7.2.3.5</u>.

Distal femur

DFF1 and DFF2 performed similarly classifying 32-33% of the specimens at a 0.95 threshold with 89-90% accuracy. Posterior probabilities for these formulae are presented in <u>Table 7.2.3.5</u>.

|           |         | Pro  | oximal femur |      |       |         | D     | istal femur |      |       |
|-----------|---------|------|--------------|------|-------|---------|-------|-------------|------|-------|
| PP<br>(%) | Male    | S    | Femal        | es   | Total | Male    | es    | Female      | es   | Total |
|           | DS      | %    | DS           | %    | %     | DS      | %     | DS          | %    | %     |
|           |         |      | PFF1         |      |       |         |       | DFF1        |      |       |
| >95       | >1.5675 | 30.9 | <-1.4743     | 28.0 | 29.5  | >1.461  | 31.48 | <-1.4782    | 32.0 | 31.7  |
| >90       | >1.1104 | 41.8 | <-1.1323     | 40.0 | 41.0  | >1.0966 | 48.15 | <-1.0555    | 48.0 | 48.1  |
| >80       | >0.7411 | 61.8 | <-0.6982     | 62.0 | 61.9  | >0.6867 | 61.11 | <-0.7155    | 68.0 | 64.4  |
| >50       | >0      | 85.5 | <0           | 88.0 | 86.7  | >0      | 85.19 | >0          | 94.0 | 89.4  |
|           |         |      | PFF2         |      |       |         |       | DFF2        |      |       |
| >95       | >1.1518 | 32.7 | <-1.4684     | 26.0 | 29.5  | >1.4071 | 33.34 | <-1.4828    | 32.0 | 32.7  |
| >90       | >1.0906 | 45.5 | <-1.1798     | 38.0 | 41.9  | >1.0957 | 48.15 | <-1.1184    | 26.0 | 47.1  |
| >80       | >0.7193 | 60.0 | <-0.7122     | 64.0 | 61.9  | >0.6835 | 61.11 | <-0.7235    | 68.0 | 64.4  |
| >50       | >0      | 85.5 | <0           | 88.0 | 86.7  | >0      | 87.04 | >0          | 94.0 | 90.4  |
|           |         |      | PFF3         |      |       |         |       |             |      |       |
| >95       | >1.3224 | 45.5 | <-1.4862     | 34.0 | 40.0  |         |       |             |      |       |
| >90       | >0.9774 | 52.7 | <-0.9706     | 52.0 | 52.4  |         |       |             |      |       |
| >80       | >0.6475 | 69.1 | <-0.6683     | 64.0 | 66.7  |         |       |             |      |       |
| >50       | >0      | 85.5 | <0           | 96.0 | 88.6  |         |       |             |      |       |

Table 7.2.3.5 Posterior probabilities for multiple functions of the proximal and distal femur.

## Univariate statistics

# Proximal tibia

The best single variable for the proximal tibia was found to be PT15 which classified 30% of the sample within 95% of confidence intervals with 88.4% accuracy. According to this function values over 74.14mm for males and under 62.17mm for females classify the sample within 95% of confidence intervals (<u>Table 7.2.3.6</u>). PT8 and PT11 classified more than 555% of the specimens with less than 80% probability of correct group assignment which implies that there is a considerable overlap of these measurements between the sexes.

| PP  | Ma    | les  | Fem   | ales | Total | al Males |      | Fem   | ales  | Total |
|-----|-------|------|-------|------|-------|----------|------|-------|-------|-------|
| (%) | <     | %    | >     | %    | %     | <        | %    | >     | %     | %     |
|     |       |      | PT3   |      |       |          |      | PT14  |       |       |
| >95 | 76.87 | 13.0 | 62.72 | 18.4 | 15.5  | 74.46    | 20.4 | 61.22 | 22.5  | 21.4  |
| >90 | 75.26 | 31.5 | 64.45 | 30.6 | 31.1  | 72.97    | 44.4 | 62.63 | 32.7  | 38.8  |
| >80 | 73.15 | 61.1 | 66.32 | 51.0 | 56.3  | 71.31    | 57.4 | 64.65 | 55.1  | 56.3  |
| >50 | 69.78 | 83.3 | 69.78 | 85.7 | 84.5  | 67.99    | 85.2 | 67.99 | 85.7  | 85.4  |
|     |       |      | PT4   |      |       |          |      | PT15  |       |       |
| >95 | 80.08 | 22.2 | 66.19 | 24.5 | 23.3  | 74.14    | 29.6 | 62.17 | 30.6  | 30.1  |
| >90 | 78.25 | 38.9 | 67.98 | 42.9 | 40.8  | 72.74    | 48.2 | 63.23 | 38.8  | 43.7  |
| >80 | 76.44 | 59.3 | 69.76 | 53.1 | 56.3  | 71.36    | 57.4 | 65.34 | 55.1  | 56.3  |
| >50 | 73.13 | 81.5 | 73.13 | 91.8 | 86.4  | 68.28    | 88.9 | 68.28 | 87.8  | 88.4  |
|     |       |      | PT8   |      |       |          |      | DT3   |       |       |
| >95 | 53.13 | 11.1 | 39.94 | 6.1  | 8.7   | 55.5     | 14.8 | 44.03 | 18.37 | 16.5  |
| >90 | 51.33 | 25.9 | 41.76 | 22.5 | 24.3  | 54.12    | 27.3 | 45.72 | 34.69 | 31.07 |
| >80 | 49.74 | 42.6 | 43.35 | 46.9 | 44.7  | 49.91    | 50   | 46.99 | 44.9  | 47.57 |
| >50 | 46.56 | 88.9 | 46.56 | 83.7 | 84.7  | 49.91    | 81.5 | 49.91 | 79.59 | 80.58 |
|     |       |      | PT11  |      |       |          |      |       |       |       |
| >95 | 44.44 | 9.3  | 30.66 | 8.2  | 8.7   |          |      |       |       |       |
| >90 | 41.65 | 22.2 | 32.32 | 22.5 | 22.3  |          |      |       |       |       |
| >80 | 39.99 | 48.2 | 34.15 | 40.8 | 44.7  |          |      |       |       |       |
| >50 | 37.02 | 79.6 | 37.02 | 83.7 | 81.6  |          |      |       |       |       |

Table 7.2.3.6 Posterior probabilities for the single variables on the radiographs of the proximal and distal tibia.

# Distal tibia

Of the measuremenets on the radiograph of the distal tibia only one exceeded 80% correct group membership. According to DT3, 30% of the specimens are correctly assigned at a 0.9 and 16.5% at a 0.95 threshold with 81% classification accuracy (see <u>Table 7.2.3.5</u>)

| PP  | Male    | s    | Female   | s    | Total | Male    | s    | Female   | es   | Total |
|-----|---------|------|----------|------|-------|---------|------|----------|------|-------|
| (%) | DS      | %    | DS       | %    | %     | DS      | %    | DS       | %    | %     |
|     |         |      | PTF1     |      |       |         |      | DTF2     |      |       |
| >95 | >1.5303 | 29.6 | <-1.5771 | 30.6 | 30.1  | >1.497  | 27.3 | <-1.4552 | 28.6 | 28.2  |
| >90 | >1.1562 | 48.2 | <-1.2998 | 38.8 | 43.7  | >1.1247 | 48.2 | <-1.1253 | 44.9 | 46.6  |
| >80 | >0.8085 | 57.4 | <-0.7635 | 55.1 | 56.3  | >0.7189 | 59.3 | <-0.8132 | 61.2 | 60.2  |
| >50 | >0      | 88.9 | <0       | 87.8 | 88.4  | >0      | 85.6 | <0       | 87.8 | 86.4  |
|     |         |      | PTF2     |      |       |         |      | DTF3     |      |       |
| >95 | >1.5355 | 37.0 | <-1.5247 | 30.6 | 34.0  | >1.6966 | 25.9 | <-1.5837 | 22.5 | 24.3  |
| >90 | >1.1783 | 46.3 | <-1.1137 | 38.9 | 44.7  | >1.1755 | 40.7 | <-1.1411 | 44.9 | 42.7  |
| >80 | >0.8059 | 53.7 | <-0.7264 | 57.1 | 55.3  | >0.7436 | 57.4 | <-0.7204 | 61.2 | 59.2  |
| >50 | >0      | 88.9 | <0       | 89.8 | 89.3  | >0      | 85.6 | <0       | 83.7 | 84.5  |
|     |         |      | DTF1     |      |       |         |      |          |      |       |
| >95 | >1.6933 | 25.9 | <-1.5998 | 22.5 | 24.3  |         |      |          |      |       |
| >90 | >1.1534 | 44.4 | <-1.1334 | 44.9 | 44.7  |         |      |          |      |       |
| >80 | >0.7519 | 57.4 | <-0.7146 | 61.2 | 59.2  |         |      |          |      |       |
| >50 | >0      | 85.6 | <0       | 83.7 | 84.5  |         |      |          |      |       |

Table 7.2.3.7 Posterior probabilities for the multivariate functions of the proximal and distal tibia.

## Multivariate statistics

Proximal tibia

The best multivariate discriminant function for the proximal tibia (PTF2) classified over 55% of the sample at a 0.8, over 44% at a 0.9 and about 34% at a 0.95 threshold exhibiting 89% correct group membership. For this function discriminant scores over 1.5355 classify males and under -1.5247 classify females within 95% of confidence intervals (Table 7.2.3.6).

# Distal tibia

The best multivariate discriminant function for the proximal tibia (DTF2) classified over 60% of the sample at a 0.8, over 46% at a 0.9 and about 28% at a 0.95 threshold exhibiting 86.4% correct group membership. For this function discriminant scores over 1.4970 classify males and under -1.4552 classify females within 95% of confidence intervals (Table 7.2.3.7).

# Chapter 8: Alternative modalities in sex identification

# 8.1 Sex identification and software development using radiographs: An example of the proximal femur.

# Introduction

When a human body is discovered the primary goal in a forensic investigation is the identification of the deceased and the definition of the cause and the manner of death (Di Maio, 2001). The identification is quite an easy procedure in relatively recent deaths, where face and fingerprints are available. Quite often, though, individuals are found disfigured or in a highly decomposed state; without fingerprints, identification becomes more complex and time-consuming. In mass disasters bones are usually commingled, charred and fragmented, thus identification is relayed in few components. The existing skeletal elements are partially exposed due to the remaining soft tissue; hence special techniques, like maceration, are needed in order to carry out the examination. In such cases a forensic anthropologist is considered expert in determining sex from skeletal remains using a variety of techniques in order to make the ultimate decision. In medico-legal routine though such experts are not always available, especially in Greece where there are no forensic anthropologists. Therefore, the necessity of developing new and easily applicable techniques for skeletal identification emerges.

The employment of the radiographic techniques presented in the previous chapters has been proven successful for the identification of sex for the given population. However to design and apply such methods, trained forensic anthropologists are needed. The lack of such expertise in Greece, among other parts of the world, obligates the forensic pathologists to undertake the burden of the identification of unknown skeletal remains. In order to facilitate this procedure and speed the identification project the idea of developing easy and practical software for forensic pathologists emerged. The current study aspires to develop a simplified tool for pathologists for sex identification using radiographs from the proximal epiphysis of the femur. The employment of such software in the medico-legal routine is expected to temporarily substitute the lack of forensic anthropologists in the Greek medico-legal reality.

# Material and Methods

# Material

For this study, a total of 106 (Mean age for men=67.28, Standard Deviation=14.52, N=37; Mean age for women=67.68, Standard Deviation=17.77, N=35) well-preserved adult femora of Cretan origin were examined. Remains were selected from the exhumed skeletons of St. Konstantinos and Pateles Cemeteries, Heraklion, Crete. Of these remains,

70 (36 male and 34 female) randomly selected left femoral heads were used as an original sample while the rest (evaluation sample) were used for cross validation.

# Methods

# Data Acquisition

The bones were radiographed using a digital X-ray machine (TCA 4R PLUS). The camera was placed at a fixed distance of 54 cm from the plane of the radiographic table. The bone was orientated with the anterior surface facing the X-ray table and the epicondyles resting on the horizontal plane. Six landmarks (A-F) were selected in the radiograph and 15 generated distances representing all possible combinations of these landmarks were calculated using specially-designed Java software. The selected landmarks are shown in Fig. 8.1.1 and described as follows:

- A. Point on the shaft under the lower end of the lesser trochanter.
- B. Point on the shaft so that the distance A-B (representing the sub-trochanteric diameter in the radiograph) is vertical to the axis of the shaft.
- C. and D. Points selected on the femoral neck where the curvature changes forming the head so that the distance from C to D is the minimum neck diameter.
- E. and F. Points on the femoral head, so that the distance E-F is the maximum femoral diameter parallel to C-D.

The magnification error was taken into account and corrected, so all distances were calculated in millimeters. Calibration has been accomplished by taking the radiograph of a scale with known length and calculating the correlation between millimeters and pixels in the radiograph. The scaling factor was added to the Java software so that all inter-landmark distances were calculated in millimeters. Landmarks were selected with the objective of being readily distinguishing from a non professional observer and to form variables that are of known significance for sex variation. It must be stressed that even though the variables AB, CD and EF are described as sub-trochanteric transverse diameter, minimum neck and maximum head diameter respectively, they do not represent the homonymous measurements on the actual (dry) bone, because X-ray measurements are two-dimensional and they can not be compared to three dimensional actual bone measurements without some error.

## SIS software

The measuring version of the software (SIS-m) was designed in such a way that the coordinates of the landmarks were recorded and the distances between landmarks were

calculated. Initially the image was loaded onto a JScrollPane to make it possible for the user to click with the mouse and select the landmarks.



Figure 8.1.1: Landmarks selected on the radiograph of the proximal femur.

The measurement can be easily cancelled with a right click and the user can start over. After the landmarks were selected, the user recorded the sex for the given radiograph, which corresponds to the measurements. When the data acquisition was completed, the user obtained, as a text archive, all fifteen measurements for each specimen along with the corresponding sex.

In order to decrease error during the landmarks's selection, a snapping technique was implemented, which is capable of adjusting the selection so as to be tangent to the bone border. That way error due to "wrong landmarking" can be reduced significantly. To increase the effectiveness of the snapping technique, the radiograph was inserted into the system and a blurring algorithm was applied on the image resulting in a second (blurred) image, thus greatly reducing the noise of the first. The data of the latter image were used as the input of the snapping algorithm.

The combination of the 6 landmarks resulted in the generation of 15 variables. *Statistical Analysis* 

All measurements were submitted to analysis of variance ANOVA and discriminant function analysis (DFA). ANOVA tested the significance of sex differences for each variable according to the F-value. Stepwise DFA was used (Method=Wilk's lambda with F=3.84 to enter and F=2.71 to remove) to select the combination of variables that best discriminate sexes. Single variables with high F-ratios were analysed using a direct procedure. A leave one out classification procedure was applied in order to demonstrate the accuracy rate of the original sample and the one created by cross validation. DFA was carried out using SPSS subroutines.

## Estimation of error

Two methods were used in order to quantify the error in the radiometric method.

# Digitizing error

For the quantification of intra-observer variation, standard procedure has been followed (O'Higgins and Jones, 1998; O'Higgins, 1999). Five specimens were randomly selected and each of them was digitized five times. Principal components analysis was carried out, in order to test the relative position of the repeats in respect to each other and to the other individuals. This test evaluated the magnitude of error relative to the differences in size between these 5 specimens and within the sample.

## Measurement error

Twenty specimens (10 males and 10 females) were randomly selected and measured by the same observer over a period of 1 month to estimate the intra-observer error. The same specimens were measured by a second observer and the means were compared with the first measurements of the first observer (inter-observer error) using a student's T-test.

# Results

## Estimation of error

## Digitizing error

The five repeats were submitted to a Principal components analysis, which showed that in all cases the repeats were much closer to themselves than to other individuals or their repeats.

## Measurement error

The results of the student's T-test are illustrated in <u>Table 8.1.4</u>. The differences between the mean measurements were found insignificant for the same (OB1-A and OB1-B) and two different observers (OB1 and OB2).

# Statistical analysis

Descriptive statistics of the 15 femoral dimensions and univariate differences between the sexes are also shown in <u>Table 8.1.1</u>. All but CF were found to be significantly different between the sexes at the level of p < 0.001, apart from DE which was found significantly different at the level of p < 0.05. Function 6 shows the result of a direct DFA

using 14 dimensions (all but CF). Function 7 shows the result of a stepwise DFA using all 15 dimensions. In this analysis 3 dimensions (BD, CE, and DF) were selected. Function 8 was formed using 3 dimensions that were significant in sex determination. The result of a direct DFA using AB (sub-trochanteric transverse diameter), CD (femoral neck diameter) and EF (femoral head diameter) is shown in <u>Table 8.1.2</u>. Dimensions with high F-ratios are noted in <u>Table 8.1.1</u>.

| *Radiometry |       |      |       |       |                    |  |  |  |  |  |
|-------------|-------|------|-------|-------|--------------------|--|--|--|--|--|
|             | М     | ales | Fe    | males |                    |  |  |  |  |  |
|             | N     | =36  | N     | N=34  |                    |  |  |  |  |  |
| Variables   | Means | SD   | Means | SD    | F-values           |  |  |  |  |  |
| AB          | 34.15 | 2.41 | 31.65 | 2.86  | 15.75              |  |  |  |  |  |
| AC          | 74.91 | 7.94 | 63.88 | 6.39  | 40.73              |  |  |  |  |  |
| AD          | 54.44 | 6.73 | 45.19 | 5.41  | 39.93              |  |  |  |  |  |
| AE          | 64.43 | 7.08 | 54.64 | 6.08  | 38.27              |  |  |  |  |  |
| AF          | 86.57 | 8.86 | 75.98 | 8.35  | 26.45              |  |  |  |  |  |
| BC          | 79.79 | 7.53 | 69.84 | 6.06  | 36.83              |  |  |  |  |  |
| BD          | 73.70 | 5.70 | 64.18 | 4.39  | 60.88              |  |  |  |  |  |
| BE          | 87.72 | 6.00 | 77.30 | 5.08  | 61.23              |  |  |  |  |  |
| BF          | 92.59 | 8.53 | 82.93 | 7.50  | 25.19              |  |  |  |  |  |
| CD          | 34.41 | 2.80 | 29.90 | 2.66  | 47.59              |  |  |  |  |  |
| CE          | 45.73 | 2.95 | 39.86 | 2.95  | 69.40              |  |  |  |  |  |
| CF          | 12.92 | 3.67 | 13.18 | 4.43  | <sup>1</sup> 10.08 |  |  |  |  |  |
| DE          | 16.22 | 2.50 | 14.84 | 2.33  | <sup>2</sup> 25.68 |  |  |  |  |  |
| DF          | 40.95 | 2.89 | 37.44 | 3.65  | 20.05              |  |  |  |  |  |
| EF          | 48.57 | 2.95 | 43.42 | 3.10  | 50.63              |  |  |  |  |  |

 Table 8.1.1 Descriptive statistics of femoral dimensions (in mm), standard deviations (SD) and univariate

 F-ratio of the differences between the sexes

Among them CD and EF are projections of minimum neck diameter and maximum head diameter that are expected to differ between sexes, since they reflect the size of the articulation between femur and pelvis. Therefore they are used with direct discriminant function procedure forming Functions 9 and 10 (<u>Table 8.1.2</u>).

 $<sup>^{*1}</sup>$  Not significant,  $^{2}$  significant at p< 0.05, all others significant at p< 0.001

| Radiometry   |      |                   |          |                    |                           |  |  |  |  |  |  |
|--------------|------|-------------------|----------|--------------------|---------------------------|--|--|--|--|--|--|
|              | Step | Variables entered | Exact F  | Degrees of freedom | Coefficient               |  |  |  |  |  |  |
|              | 1    | AB                | 15.75    | 1.69               | 0.472833431               |  |  |  |  |  |  |
|              |      | AC                | 40.73    | 1.69               | -0.60363965               |  |  |  |  |  |  |
|              |      | AD                | 39.93    | 1.69               | -0.26148016               |  |  |  |  |  |  |
|              |      | AE                | 38.27    | 1.69               | 0.745111568               |  |  |  |  |  |  |
| E6. direct   |      | AF                | 26.45    | 1.69               | 0.31053234                |  |  |  |  |  |  |
| FO. difect   |      | BC                | 36.83    | 1.69               | 0.66875721                |  |  |  |  |  |  |
|              |      | BD                | 60.88    | 1.69               | -0.21415491               |  |  |  |  |  |  |
|              |      | BE                | 61.23    | 1.69               | -0.76855869               |  |  |  |  |  |  |
|              |      | BF                | 25.19    | 1.69               | -0.19199587               |  |  |  |  |  |  |
|              |      | Constant          |          |                    | 14.757115                 |  |  |  |  |  |  |
|              |      |                   |          |                    |                           |  |  |  |  |  |  |
|              | 1    | CE                | 69.4     | 1.68               | 0.139011402               |  |  |  |  |  |  |
| F7: stopwise | 2    | BD                | 43.1     | 2.67               | 0.280034272               |  |  |  |  |  |  |
| 17. stepwise | 3    | DF                | 32.52    | 3.66               | -0.16286881               |  |  |  |  |  |  |
|              |      | Constant          |          |                    | -15.252364                |  |  |  |  |  |  |
|              |      |                   |          |                    |                           |  |  |  |  |  |  |
|              | 1    | AB                | 15.74554 | 1.69               | 0.021424536               |  |  |  |  |  |  |
| E8: direct   |      | CD                | 47.59325 | 1.69               | 0.159018649               |  |  |  |  |  |  |
| Fo. unect    |      | EF                | 50.63015 | 1.69               | 0.195662415               |  |  |  |  |  |  |
|              |      | Constant          |          |                    | -14.81799                 |  |  |  |  |  |  |
|              |      |                   |          |                    |                           |  |  |  |  |  |  |
| F9           | 1    | CD                | 47.59325 | 1.69               | F< 32.155 <m< td=""></m<> |  |  |  |  |  |  |
| F10          | 1    | EF                | 50.63015 | 1.69               | F<45.995 <m< td=""></m<>  |  |  |  |  |  |  |

Table 8.1.2 Discriminant function statistics, F-ratios and statistical significance of femoral dimensions.

<u>Table 8.1.3</u> presents the classification accuracy for all functions for both original and cross-validation samples. Functions 6 and 7 present the same correct group membership. Yet F7 has a higher accuracy rate in cross-validated sample and uses a smaller number of variables (3) compared to F6 (9), as is noted in <u>Table 8.1.2</u>.

Therefore F2 was selected as the best function for sex identification in the present study. According to F7 the sex can be calculated by multiplying the values of the three dimensions by the corresponding unstandardized coefficients plus the constant, as can be seen in <u>Table 8.1.2</u>. Values greater than zero indicate a male individual, otherwise a female.

Next, the early measuring form of the software was modified by incorporating the best formula produced by stepwise procedure (F7). The new version of SIS, in addition to the calculation of all "inter-landmark" distances, also calculates the value for the equation compares it with the sectioning point and directly classifies the femur as male or female.

|             |                 | Radiometry |      |         |       |       |
|-------------|-----------------|------------|------|---------|-------|-------|
|             |                 | Males      |      | Females |       | Total |
|             |                 | N          | %    | N       | %     | %     |
| F6 direct   | Original        | 31/36      | 86.1 | 34/34   | 100.0 | 92.9  |
|             | Cross validated | 30/36      | 83.3 | 32/34   | 94.1  | 88.6  |
| F7 stepwise | Original        | 32/36      | 88.9 | 33/34   | 97.1  | 92.9  |
|             | Cross validated | 31/36      | 86.1 | 32/34   | 94.1  | 90.0  |
| F8 direct   | Original        | 32/36      | 88.9 | 28/34   | 82.4  | 85.7  |
|             | Cross validated | 32/36      | 88.9 | 27/34   | 79.4  | 84.3  |
| F9          | Original        | 31/36      | 86.1 | 29/34   | 82.3  | 85.7  |
|             | Cross validated | 31/36      | 86.1 | 29/34   | 82.3  | 85.7  |
| F10         | Original        | 29/36      | 80.6 | 27/34   | 79.4  | 80.0  |
|             | Cross validated | 29/36      | 80.6 | 27/34   | 79.4  | 80.0  |

Table 8.1.3: Classification accuracy of the original and cross validated samples.

Posterior probability of correct group assessment is also calculated. An example of this is presented in Figure 8.1.2. In order to test the software's reliability, a sample of 36 (23 left and 13 right) femoral radiographs was tested by two of the authors independently. Both observers correctly identified sex in 32/36 cases giving an accuracy rate of 91.3% for left femora, 84.6% for right and 88.7% for both groups (Table 8.1.5).

## Discussion

Sexual dimorphism of the femur has been very well studied in many different populations with diverse and interesting results (DiBennardo and Taylor, 1982; Taylor and Dibennardo, 1982; İşcan and Miller-Shaivitz, 1984; İşcan and Miller-Shaivitz, 1986; Wu, 1989; İşcan and Ding, 1995; King et al., 1998; Mall et al., 2000; Asala, 2001; Šlaus et al., 2003; Asala et al., 2004; Purkait and Chandra, 2004; Murphy, 2005; Purkait, 2005).

Since the integrity of the femoral bone in forensic cases can not be assured, different fragmentary models can be assumed. Under that aspect some authors tested the validity of single femoral variables in sex determination (Mall et al., 2000; Purkait and Chandra, 2004), while others created diaphyseal patterns assuming that only one of the

distal ends was preserved (Asala, 2001; Šlaus et al., 2003; Asala et al., 2004; Murphy, 2005; Purkait, 2005).

|                | OB    | OB1-A OB1-B |       | OB-2 |        | *t-Test differences between<br>OB1-A and |       |       |
|----------------|-------|-------------|-------|------|--------|--|-------|-------|
| Males (N=10)   | Mean  | SD          | Mean  | SD   | Mean   | SD                                       | OB1-A | OB2   |
| AB             | 32.99 | 3.30        | 33.52 | 3.67 | 33.50  | 2.98                                     | -2.63 | -1.40 |
| AC             | 68.97 | 3.98        | 68.89 | 3.58 | 69.16  | 3.34                                     | 0.24  | -0.49 |
| AD             | 48.27 | 4.38        | 48.34 | 3.98 | 47.96  | 4.33                                     | -0.23 | 0.68  |
| AE             | 59.32 | 4.66        | 59.51 | 4.01 | 59.74  | 3.59                                     | -0.44 | -0.76 |
| AF             | 78.22 | 4.55        | 78.08 | 4.47 | 79.02  | 4.43                                     | 1.54  | -1.24 |
| BC             | 75.25 | 4.57        | 75.65 | 4.80 | 75.57  | 3.42                                     | -0.94 | -0.47 |
| BD             | 68.99 | 4.63        | 69.58 | 4.74 | 68.98  | 4.61                                     | -1.69 | 0.02  |
| BE             | 83.62 | 5.85        | 84.39 | 5.84 | 84.49  | 4.69                                     | -1.68 | -1.03 |
| BF             | 85.05 | 5.57        | 85.31 | 6.30 | 86.08  | 5.30                                     | -0.62 | -1.67 |
| CD             | 34.11 | 3.80        | 33.91 | 3.90 | 34.23  | 3.85                                     | 2.68  | -0.93 |
| CE             | 44.69 | 4.38        | 44.74 | 4.37 | 45.20  | 3.96                                     | -0.12 | -1.40 |
| CF             | 9.95  | 1.87        | 9.82  | 1.93 | 10.61  | 2.11                                     | 0.41  | -0.83 |
| DE             | 16.22 | 1.47        | 16.43 | 1.38 | 17.09  | 1.10                                     | -0.52 | -1.13 |
| DF             | 39.69 | 4.99        | 39.53 | 5.36 | 40.30  | 5.59                                     | 0.64  | -1.91 |
| EF             | 47.37 | 5.66        | 47.48 | 5.69 | 48.03  | 5.63                                     | -0.36 | -2.60 |
| Females (N=10) |       |             |       |      |        |  |       |       |
| AB             | 30.19 | 2.68        | 30.39 | 2.49 | 30.194 | 2.048                                    | -0.64 | 0.00  |
| AC             | 61.03 | 2.68        | 61.25 | 3.21 | 60.782 | 2.955                                    | -0.76 | 1.16  |
| AD             | 43.64 | 1.61        | 44.62 | 1.48 | 43.552 | 1.424                                    | -1.78 | 0.13  |
| AE             | 53.47 | 1.92        | 54.07 | 1.75 | 53.43  | 2.038                                    | -1.33 | 0.08  |
| AF             | 73.71 | 4.24        | 75.17 | 4.03 | 73.439 | 4.512                                    | -1.15 | 1.37  |
| BC             | 64.23 | 4.14        | 64.90 | 5.67 | 64.885 | 4.808                                    | -0.87 | -1.12 |
| BD             | 60.30 | 3.40        | 61.59 | 4.36 | 60.983 | 3.955                                    | -1.89 | -1.20 |
| BE             | 73.76 | 3.32        | 74.67 | 3.16 | 74.431 | 3.562                                    | -2.67 | -2.00 |
| BF             | 78.04 | 6.07        | 80.16 | 4.95 | 78.746 | 6.401                                    | -1.73 | -1.32 |
| CD             | 29.22 | 1.68        | 29.02 | 1.69 | 29.265 | 1.561                                    | 0.77  | -0.30 |
| CE             | 13.89 | 1.98        | 15.38 | 2.60 | 39.425 | 1.105                                    | -0.06 | -0.61 |
| CF             | 39.22 | 1.39        | 39.24 | 1.50 | 13.919 | 1.839                                    | -0.87 | -0.11 |
| DE             | 14.79 | 1.72        | 14.54 | 2.78 | 14.838 | 1.81                                     | 0.34  | -0.11 |
| DF             | 36.89 | 2.91        | 37.20 | 2.06 | 36.745 | 2.634                                    | -0.34 | 0.33  |
| EF             | 42.64 | 2.39        | 42.76 | 2.56 | 42.625 | 2.728                                    | -0.45 | 0.06  |

Table 8.1.4: A t-Test comparison of the measurements taken by the same observer (OB1-A and OB1-B) and between two different observers (OB1-A and OB2).

<sup>\*</sup> All mean differences were found insignificant

|                            | Males                  |      | Females                |      | Total                  |      |
|----------------------------|------------------------|------|------------------------|------|------------------------|------|
| Evaluation groups          | Correct classification | %    | Correct classification | %    | Correct classification | %    |
| Y1: Left femora<br>(N=23)  | 14/15                  | 93.3 | 7/8                    | 87.5 | 21/23                  | 91.3 |
| Y2: Right femora<br>(N=13) | 6/7                    | 85.7 | 5/6                    | 83.3 | 11/13                  | 84.6 |
| Total (N=36)               | 20/22                  | 90.9 | 12/14                  | 85.7 | 32/36                  | 88.9 |

Table 8.1.5: Classification accuracy of the evaluation samples using SIS software.

Obviously the femur is a very useful bone for sex estimation. Standard osteometric methods performed very well for the given population (see chapter 7). Yet since forensic cases differ significantly, these methods are not always applicable. The need for identification of dismembered semi-decomposed or charred bodies such as the ones recovered in mass disasters or crime scenes led to the development of a radiometric technique for sex estimation based on the proximal epiphysis of the femur.



Figure 8.1.2: An example of sex estimation using SIS-software. The specimen was correctly assigned as female.

The SIS software is a valuable tool for the forensic pathologists that are called to identify semi-decomposed or charred remains in forensic settings. The radiographic examination of the skeleton which constitutes a routine examination in medico-legal practice allows the identification of sex with a simple selection of six landmarks on the femoral head. Since the utility of more long bones for sex estimation from radiographs has been proven in the previous chapter, the early and simple version of SIS software could be further improved in such a way that sex can be identified using landmarks on any radiograph from the long bones that were previously examined. Furthermore the application of all subsets method (the employment of vall possible combination between variables to identify the optimal one) in discriminant function analysis could improve classification results for the femoral head making the software even more accurate for the estimation of sex. A test of this software by other observers and the application in other populations are future goals of the present attempt.

## Conclusions

The current study resulted in the development of a sex estimation method using femoral radiographs that performs equally well as the conventional methods. The radiometric method is presented as an alternative technique, applicable for semidecomposed and charred bodies of crime scenes or mass disasters, when maceration is not an option. The application of metric methods in radiographs and the development of a highly specific program provide a useful tool for sex identification that can be applied in forensic cases. The use of femoral radiographs in sex determination is only one of the various applications that Java technology can have in medico-legal practice. Additional research is needed to improve the SIS software and furthermore to adjust the Java technology in other forms of anthropological radiographic studies. 8.2 The application of geometric-morphometrics in sex identification for forensic purposes: An example of the humerus.

# Introduction

When the entire skeleton is available, sex assessment is considered a relatively easy process (Krogman and İşcan, 1986). However, in forensic investigations that is rarely the case, since the bones are usually recovered in fragmentary state due to the effect of extreme environmental conditions and activities of carnivores and/or other scavengers. Therefore sexual assessment becomes more difficult given that the bones are incomplete and too fragile to be manipulated.

There are mainly two traditional approaches to estimate sex from skeletal remains. Qualitative morphological examination remains the quickest and easiest method and, in experienced hands, results in 95–100% accuracy when the whole skeleton is available (Krogman and İşcan, 1986). Nevertheless, these methods present a certain number of limitations, such as inter- and intra-observer error or classification problems of the qualitative morphological characteristics, which make one sceptical considering their reliability (Pretorius et al., 2006). Morphometric methods, on the other hand, are considered more advantageous in terms of objectivity, repeatability, data evaluation and applicability to both cranial and postcranial skeleton (Krogman and İşcan, 1986; Walrath et al., 2004). However, some characteristics such as the prominence of the glabella or the external occipital protuberance are difficult to assess metrically.

Lately a new technique which combines both morphometric and meristic characteristics is becoming popular. Procrustes-based Geometric morphometrics is a method that provides the means for quantifying shape differences in a 2 or 3 dimensional coordinate system (Kendall, 1981; Bookstein, 1989; Rohlf and Slice, 1990; Bookstein, 1991; Slice, 1993; Bookstein, 1996; O'Higgins, 1997; Adams et al., 2004). As a research tool it has been used to test a variety of hypotheses in a variety of disciplines using various different types of data sets (Richtsmeier, 2002), but it is only recently that it has been introduced in forensic anthropology. More specifically sexual dimorphism has been studied on the greater sciatic notch, mandibular ramus flexure and the orbits (Pretorius et al., 2006), as well as in skulls and mandibles (Rosas and Bastir, 2002; T.J. Buck and Vidarsdottir, 2004; Franklin et al., 2007b; Franklin et al., 2008a; Kimmerle et al., 2008b). It is worth mentioning a recent study on anterior dentition (Kieser et al., 2007) concluding that there are no two individuals with identical tooth morphology, which suggests the potential use of this methodology for positive identification in forensic cases.

The humerus is one of the strongest long bones of the skeleton that even in a fragmented state is likely to be recovered in a forensic case. Several studies using classical osteometric techniques confirm the existence of sexual dimorphism in the humerus (Carretero et al., 1995; İşcan et al., 1998; Steyn and İşcan, 1999; Mall et al., 2001; Sakaue, 2004; Albanese et al., 2005; Frutos, 2005). Scholars agree that a population specific study is required in order to have accurate results in sexing the skeleton for a given population (İşcan and Miller-Shaivitz, 1984; Macho, 1990a).

The objective of this investigation is to discriminate sex from the humerus in a contemporary Greek population, with the application of geometric morphometric techniques on digital radiographs. The study addresses population specific morphological features for identification purposes in forensic investigation and thus provides potentially useful tools for modern medico-legal professionals.

## Material and methods

A total of 97 well preserved, adult humeri of Cretan origin were examined. Remains were selected from the exhumed skeletons of St. Konstantinos and Pateles Cemeteries, Heraklion, Crete. The study population consists of individuals who lived between the end of the 19th century and the beginning of the 20th and buried in Crete. Mean age for males is 68.57 + -13.52 (N=50) and for females 72.98 + -16, 90 (N=47). Of these remains left humeri were radiographed using digital x-ray machine (TCA 4R PLUS). Standard orientation of the bones has been achieved by letting the humerus balance on the horizontal plane, with the anterior surface facing the x-ray camera. The radiographic table has been placed at a distance of 54 cm from the head of the camera.

Within the arbitrary 2-D coordinate system created by this orientation, landmarks were defined as extreme points (Bookstein, 1990; Valeri et al., 1998). The epiphyseal ends were studied separately. In the first analysis 5 landmarks were selected on the radiograph of the proximal humerus as defined in <u>Table 8.2.1</u>. The second analysis included 7 landmarks on the radiograph of the distal epiphysis as described in the same table. <u>Figure 8.2.1a</u> and <u>8.2.1b</u> show the selected landmark on the proximal and distal parts respectively. Landmarks were digitalised using TPSDIG2 software (Rohlf, 1997). Semilandmarks were used to quantify relative height of the caput humeri and slid in order to minimise bending energy following standard methods described elsewhere (Bookstein, 1997; Bookstein et al., 1999; Bastir et al., 2006).

For the quantification of intra-observer variation, standard procedure has been followed (O'Higgins and Jones, 1998; Martinón-Torres et al., 2006). Five specimens were randomly selected and each of them was digitized five times. Principal components analysis was carried out, in order to test the relative position of the repeats in respect to each other and to the other individuals. This test evaluated the magnitude of error precision relative to the differences in shape between these 5 specimens and within the sample.

| Proximal Epiphysis |  |  |  |  |  |  |
|--------------------|--|--|--|--|--|--|
| Lm1                | The projection of the medial and inferior part of the head.  |  |  |  |  |  |
| Lm2                | The projection of the superior part of the anatomical neck   |  |  |  |  |  |
| Lm3                | The sectioning point on the humeral head outline, of the orthogonal projection of the middle point between landmarks 1 and 2 |  |  |  |  |  |
| Lm4                | The maximum curvature point of the greater tubercle  |  |  |  |  |  |
| Lm5                | The most lateral point that defines the maximum distance from landmark 1.  |  |  |  |  |  |
|                    | Distal Epiphysis   |  |  |  |  |  |
| Lm1                | The incision point between the medial epicondilus and medial part of the trochlea.   |  |  |  |  |  |
| Lm2                | The maximum curvature point projected in the distal surface of the medial trochlea.  |  |  |  |  |  |
| Lm3                | The incision point in the distal surface of the troclear groove.   |  |  |  |  |  |
| Lm4                | The maximum curvature point in the distal surface between the capitulum and the trochlea.                                    |  |  |  |  |  |
| Lm5                | The incision point of the capitulum and medial epicondylus.  |  |  |  |  |  |
| Lm6                | The most lateral point of the projection of the lateral epicondilus.   |  |  |  |  |  |
| Lm7                | The most medial point of the projection of medial epicondilus  |  |  |  |  |  |

Table 8.2.1: Definition of landmarks on the proximal and distal humerus

# **GEOMETRIC MORPHOMETRICS**

Generalized Procrustes Superimposition GPA (Rohlf and Slice, 1990; O'Higgins, 1999) and Thin Plate Splines (Bookstein, 1991; Zelditch et al., 2004) are used to obtain Procrustes shape coordinates and shape variables for different statistical analyses. Shape is defined following Kendall [7] as "all the information remaining when location, size and rotational factors are all removed". More technical details about geometric morphometric methodologies can be found in Rohlf and Slice (Rohlf and Slice, 1990), Bookstein (Bookstein, 1991), O'Higgins (O'Higgins, 1997) ,Adams et al. (Adams et al., 2004), O'Higgins and Jones (O'Higgins and Jones, 1998), Zelditch et al. (Zelditch et al., 2004) and Slice (Slice, 2007).

The metrics of the shape space is the Procrustes distance, and is approximately the square root of the summed, squared interlandmark distances of Procrustes registered specimens (Bookstein, 1996). Size is measured as "centroid size" defined as the square root of the summed squared distances between each landmark and the centre of gravity (centroid) of each landmark configuration. It is an individual score obtained as a scaling factor during the partial Procrustes superimposition (Dryden and Mardia, 1998). In the

absence of allometry, centroid size can be considered uncorrelated to shape (Bookstein, 1991; Zelditch et al., 2004).



Figure 8.2.1 a) Landmarks selected on the radiograph of the proximal humerus, b) Landmarks selected on the radiograph of the distal humerus.

The metrics of the shape space is the Procrustes distance, and is approximately the square root of the summed, squared interlandmark distances of Procrustes registered specimens (Bookstein, 1996). Size is measured as "centroid size" defined as the square root of the summed squared distances between each landmark and the centre of gravity (centroid) of each landmark configuration. It is an individual score obtained as a scaling factor during the partial Procrustes superimposition (Dryden and Mardia, 1998). In the absence of allometry, centroid size can be considered uncorrelated to shape (Bookstein, 1991; Zelditch et al., 2004).

# STATISTICAL ANALYSES

Mean comparisons between males and females were carried out using a permutation model of multivariate analysis of variance (MANOVA) of Procrustes shape data (Rohlf and Slice, 1990). In these analyses the *a priori* assigned group membership was permuted by chance (N=1000), and the frequency assessed how often a Procrustes distance equal or larger than the actually observed has been achieved between the permuted group means. This ratio gives a distribution-independent estimate of the significance of the observed mean shape differences between males and females and was performed using Morpheus et al. software (Slice, 1998). More methodological details can also be found in Fontaneto et al. (2004). The associated differences in female and male mean shapes are visualized using thin plate splines transformation grids (Bookstein, 1991) transforming the female mean shape into the male or viceversa. In addition, to aid the

identification of the morphological differences thin-plate splines are used to warp the pixels of the digital x-ray images (Rohlf, 2003).

First, the image of the overall consensus is calculated using images and landmarks of the full sample. Then, consensus image and landmarks are unwarped into an exaggerated shape representing the female and the male mean shape respectively. These "warpings" are calculated using tpsSUPER (Rohlf, 2003). As a result of this, x-ray pictures that visualize shape features of female and male epiphyses are obtained.

Then 3 discriminant function analyses were carried out. One using the PC-scores from Procrustes shape-space, a second using centroid size alone and a third one using PC-scores of GPA residuals plus lnCS for analysis in Procrustes Form space (O'Higgins and Jones, 1998; Mitteroecker et al., 2004; Bastir et al., 2007).

In order to find the optimal combination of variables that best discriminate sexes, the all sub-set method was used. When P predictor variables are available to predict a dependent variable Y by regression, there are altogether  $2^{p}$  different sets of predictor variables that could be formed. That's because each predictor can be included or excluded independently of the others, and there are P such binary choices, making  $2^{p}$  combinations. That includes the "null" regression that contains no predictors, and the full regression containing all P predictors. The optimal combination of P predictor variables can only be found if testing all  $2^{p}$  combinations.

The distributions of females and males in these statistical spaces of reduced dimensionality, as implied by the choice of different PCA axes, are explored via SPSS. Jack-Knife procedures (Zelditch et al., 2004; Rosas et al., 2008) are carried out for cross-validation of the groupings.

# RESULTS

# Digitazing error

The five repeats were submitted to a Principal components analysis, which showed that in all cases (proximal as well as distal epiphyses) the repeats are much closer to themselves than to other individuals or their repeats. The % of variance which is explained by digitizing error was also calculated according to Cardini and Elton (2008). The ratio was proximal humerus (average of 1.9%) and from 3.3 to 8% for the distal humerus (average of 4.8%).



Figure 8.2.2 Plots of the first 2 principal components of PCA in proximal humerus: a) Shape-space b) Form-space and distal humerus c) Shape-space d) Form-space. Note that there is a clear separation of sexes in both proximal (b) and distal (d) end when form variables are used.

# 1. PROXIMAL HUMERUS

# 1a) Shape analysis

The PCA includes 8 principal components that explain 100% of the shape variability in the proximal humerus. The first two principal components of this analysis are plotted in Figure <u>8.2.2a</u>. PC1 (horizontal axis) accounts for 48.6% of the shape variability while PC2 (vertical axis) explains the 23.2 % of the variability.

The MANOVA permutation test showed that the shape differences due to sex dimorphism are statistically significant at the level of p < 0.044.

Table 8.2.2 Classification accuracy using shape, form variables and centroid size for the proximal and the distal humerus.

|                              |                     | Predicted group membership |     |       |       |      |
|------------------------------|---------------------|----------------------------|-----|-------|-------|------|
|                              |                     | M                          | ale | Fem   | Total |      |
| Proximal Hu                  | N                   | %                          | N   | %     | %     |      |
| *Shape variables             | Original<br>group   | 35/50                      | 70  | 37/46 | 80.4  | 75   |
|                              | Cross-<br>validated | 34/50                      | 68  | 36/46 | 78.3  | 72.9 |
| Centroid Size                | Original<br>group   | 42/50                      | 84  | 41/46 | 89.1  | 86.5 |
|                              | Cross-<br>validated | 42/50                      | 84  | 41/46 | 89.1  | 86.5 |
| <sup>†</sup> Form Variables  | Original<br>group   | 44/50                      | 88  | 42/46 | 91.3  | 89.6 |
|                              | Cross-<br>validated | 44/50                      | 88  | 42/46 | 91.3  | 89.6 |
| Distal Hum                   |                     |                            |     |       |       |      |
| <sup>‡</sup> Shape variables | Original<br>group   | 40/50                      | 80  | 32/47 | 68.1  | 74.2 |
|                              | Cross-<br>validated | 38/50                      | 76  | 31/47 | 66    | 71.1 |
| Centroid Size                | Original<br>group   | 40/50                      | 80  | 43/46 | 91.5  | 85.6 |
|                              | Cross-<br>validated | 40/50                      | 80  | 43/46 | 91.5  | 85.6 |
| <sup>§</sup> Form Variables  | Original<br>group   | 44/50                      | 88  | 43/47 | 91.5  | 89.7 |
|                              | Cross-<br>validated | 43/50                      | 86  | 43/47 | 91.5  | 88.7 |

<sup>&</sup>lt;sup>\*</sup> PC 2, 3, 4 and 5

<sup>&</sup>lt;sup>†</sup> PC 1 and 4

<sup>&</sup>lt;sup>‡</sup> PC 1, 6 and 8

<sup>&</sup>lt;sup>§</sup> PC 1, 3, 6 and 9

The MANOVA permutation test showed that the shape differences due to sex dimorphism are statistically significant at the level of p < 0.044.

The first 6 non-zero principal components of form space (accounting for 100% of variance) are used as independent variables in order to identify sex. Several different combinations were calculated according to all-subset method and the best included 4 PCs (2, 3, 4 and 5) following a direct procedure (Wilks's lambda= 0.796, p<0.0001). Classification accuracy was 75% for the original sample while leave-one-out classification yielded at 73%.

Multiple regression of shape using all six PCs revealed that approximately 5% of the total variance is explained by sexual dimorphism.

Figures 8.2.3a and 8.2.3b provide deformation grids for males and females. Observing the two grids one can note that the shape differences are mainly distributed between landmarks 2, 4 and 5. More specifically in females there is an expansion of the grid between landmarks 2 and 4 which corresponds to the relative position of the great tubercle and the projection of the groove of the anatomical neck. Additionally there is a compression between landmarks 4 and 5 which indicates that the most superior point of greater tubercle is relatively closer to the axis defined by landmarks 1 and 5. Furthermore there is an expansion on the grid between landmark 3 and the middle point between landmarks 1 and 2 on females compared to males indicating a relatively more voluminous caput in males.

Figure 8.2.3 provides an average image for females (f), males (h) and the entire group (g) for proximal end of the humerus.

# 1b) Size analysis

In order determinate sex a discriminant function analysis using centroid size is performed (F= 156.183, Wilks's lambda= 0.375). Demarking point is 50.82 therefore values of centroid size greater than that indicate a male individual, while smaller values are assessed as female. Classification accuracy reaches 84% for males and 89.1% for females while cross validation procedure gives exactly the same results (Table 8.2.2).

## 1c) Form (size and shape) analysis

The PCA of form-space extracted 9 principal components that explain 100% of the shape variability. Figure 2b plots the first two principal components of these analyses. More specifically PC1 (horizontal axis) accounts for 64.3% while PC2 (vertical axis) explains 9.1% of the variability; in this subspace, that accounts for most of the variation in the current study. There is a clear separation of the two groups in the direction of the horizontal axis which indicates that sexual dimorphism is mainly contributed to size differences.



Figure 8.2.3 Proximal Humerus: a)Deformation grid of the female configuration, b)Deformation grid of the male configuration c)Deformation grid adjusted to the mean female image e) Deformation grid adjusted to the mean overall image e) Deformation grid adjusted to the mean male image f) Mean female consensus g) Overall consensus h) Mean male consensus. All grids and mean images are exaggerated 5 times in order to visualize better the observed shape differences.

The first 7 non-zero principal components of form space (accounting for 99.9% of variance) are used as independent variables in order to identify sex. Classification accuracy for direct analysis using all seven PCs is 90.6% while leave one out classification yields at 88.5%.Using all subsets method, PC1 and PC4 were selected as the optimal combination of variables giving 89.6% of classification accuracy for both original and cross-validated data (Table 8.2.2).

# 2. DISTAL HUMERUS

# 2a) Shape analysis

The PCA includes 10 principal components that explain 100% of the shape variability. The first two principal components of this analysis are plotted in Figure 8.2.2c.

PC1 (horizontal axis) accounts for 31.78% of the shape variability while PC2 (vertical axis) explains the 17.02% of the variability. Sexual dimorphism is not associated with either of these principal components and thus the two groups cannot be separated visually on scatterplots in this projection of shape space.

After a GPA, data are submitted to a MANOVA permutation test. The shape differences due to sex dimorphism are statistically significant at the level of p < 0.003. This means that of the 1000 permutations only 2 times the Procrustes distance was equal or larger than the observed.

The DFA for the distal humerus yields a significant difference of shape between the sexes using PC 1, 6 and 8 (Wilks's lambda= 0.796, p<0.0001). Ten males and fifteen females were misclassified by the DFA and the classification accuracy for both groups reached 74.2% for original and 71.1% for cross-validated data (<u>Table 8.2.2</u>). Multiple regression of shape using all PCs indicates that approximately 5% of the total variance is explaine<u>d by sexual dimorphism</u>.

<u>Figures 8.2.4a</u> and <u>8.2.4b</u> provide deformation grids for males and females. There is a deformation of the grid of the lateral trochlea which corresponds to the relative expansion of the grid between landmarks 3 and 4 in male configuration.

Additionally a relative compression of the grid between landmarks 4 and 5 is observed, reflecting a relatively smaller capitulum with respect to the trochlea. Furthermore the grid between landmark 6 and 7 is expanded in the male configuration suggesting a relative elongation of the distance between the two most lateral landmarks of the epiphysis in males. As a consequence of these relative changes the female configuration is more square-shaped while the male configuration follows a more rectangular pattern. Figure 8.2.4 also provides an average image with the grid adjusted to the corresponding landmarks for females (c), males (e) and the entire group (d) for the distal end of the humerus.

## 2b) Size analysis

Discriminant function analysis using centroid size is also applied for the distal end (F=126.689, Wilks's lambda=0.428). Demarking point is 55.87 therefore values of centroid size greater than that indicate a male individual, while smaller values are assessed as female. Classification accuracy reaches 80% for males and 91.5% for females while cross validation procedure gives exactly the same results (Table 8.2.2).

#### 2c) Form (size and shape) analysis

The PCA for form-space extracted 13 principal components that explain 100% of the shape variability. Figure 2d plots the first two principal components. PC1 (horizontal axis) accounts for 69% while PC2 (vertical axis) explains only 9.1% of the variability. In this subspace, that accounts for most of the variation in distal humerus. Again there is a clear separation of the two groups in the direction of the horizontal axis which is indicating that sexual dimorphism is mainly contributed to size differences.



Figure 8.2.4 Distal Humerus: a)Deformation grid of the female configuration, b)Deformation grid of the female configuration c)Deformation grid adjusted to the mean female image e) Deformation grid adjusted to the mean overall image e) Deformation grid adjusted to the mean male image f) Mean female consensus g) Overall consensus h) Mean male consensus. All grids and mean images are exaggerated 5 times in order to visualize better the observed shape differences.

The first 11 non-zero principal components of form space (accounting for 99.9% of variance) are used as independent variables in order to identify sex from the distal humerus. Classification accuracy for direct analysis is 89.7% while leave-one-out classification yields at 87.6% All-subsets DFA revealed the 4 PCs (PC 1, 3, 6 and 9) that give the optimal group separation. Classification accuracy yielded at 89.7% for the original and 88.7 for the cross-validated data (Table 8.2.2).

# DISCUSSION

The recovery of fragmentary skeletal remains, in forensic investigations, requires easy and rapid techniques for biological profiling and reconstruction of the scene history. The first and most vital biological characteristic under consideration is sex since it reduces the number of possible matches in the population by fifty percent. Although sex identification can be easily established when a complete skeleton is present, this is rarely the case in forensic investigations where mostly fragmented bony parts are recovered.

According to France (France, 1983) distal measurements are likely to reflect more sexual dimorphism in humerus because they are subjected to greater functional or occupational stress. Scholars agree that epiphyseal structures tend to be more dimorphic than length (Dwight, 1905; Sakaue, 2004). Reviewing the current literature one can note that the best discriminatory measurement varies in different samples. Proximal epiphysis has given better accuracy results in populations from Guatemala (Frutos, 2005), Germany (Mall et al., 2001), China (İşcan et al., 1998) and South Africa (Steyn and İşcan, 1999). On the contrary studies of two different Japanese (İşcan et al., 1998; Sakaue, 2004) and a Thai population (İşcan et al., 1998) concluded that distal part is more effective than the proximal. In all cases though, epiphyseal structures were included in the 3 more effective dimensions. Osteometric data of the Cretan population used in this study conclude that proximal epiphysis is the most dimorphic part with classification accuracy 89.9% while distal epiphysis comes at the third place along with length (85.1%) (Kranioti et al., 2008). However, this is a very small difference which could be reversed by simply adding more specimens.

Apart from the classical osteometric studies, sexual dimorphism of the skeleton was also investigated by means of radiographs and Computed Tomography. Riepert and associates (Riepert et al., 1996) studied sexual dimorphism in radiographs of the calcaneus achieving 80% of correct group membership. Patil and Mody (Patil and Mody, 2005) accomplished sex identification from lateral cephalograms with accuracy of 99%. A recent study on digital radiographs of the femur yielded classification accuracy up to 92.9% (Kranioti et al., 2007). Additionally Harma and Karakas (Harma and Karakas, 2007) predicted sex with 84.6% accuracy by using CT scans of femora derived from hospital patients. It seems that radiography can be quite successful in sex identification apart from its acknowledged value on positive identification and age estimation (K.T. Evans and Knight., 1986; Kahana and Hiss, 1997, 1999; K.M.Stein and Grünberg, 2008). Nevertheless no study to our knowledge deals with digital radiographs of the humerus.

Sexual dimorphism of the humerus has been studied so far in terms of size. One must consider though that sex dimorphism is also expressed in shape and in that concept there is a lack of evidence in this topic (Lague and Jungers, 1999). An exception is considered a shape analysis of the humerus in a Portuguese sample using transformed indices deriving from osteometric data (Carretero et al., 1995). Authors conclude that excluding size (which explains 80% of the observed variability) men tend to have relatively shorter humeri with voluminous epiphyses while women have relatively longer shafts with smaller epiphyses, in the given population. This is consistent with findings of our study (Figure 3).

Another work by Lague and Jungers (Lague and Jungers, 1999), deals with the shape of hominoid distal humerus using geometric morphometrics. Although not the principal goal of this study, sexual dimorphism was mentioned in the results. It was found that the sexes of the American Whites and African Americans showed a mixed pattern of affinities with the males of each group to be closer in shape to the females of the other group. Yet these results were not proven feasible in establishing shape criteria for assessment of sex.

The original concept of the current study is to validate the efficacy of geometric morphometric method in sex identification of humeral radiographs. The existence of sexual dimorphism of the humerus it is well known and mainly attributed to size differences (Dittrick and Suchey, 1986; Carretero et al., 1995; İşcan et al., 1998; Steyn and İşcan, 1999; Mall et al., 2001; Albanese et al., 2005). This is consistent with our results. The existence though of a signal of shape differences is worthy of further investigation.

Observing the plots of the deformation of mean male and mean female proximal radiographs one can note clearly shape differences in the projection of the greater tubercle and the superior border of the anatomical neck. In females the greater tubercle is smoother with its superior border less pronounced. This observation could simply reflect the relatively weaker development of the Supraspinatus muscle and consequently its insertion in females compared to males.

On the distal end, the male configuration is rectangular while the female configuration is squared, probably due to the relatively wider epiphyseal breadth in males. It has also been observed a relative wider lateral trochlea accompanied by a relatively smaller capitulum in males in respect to females (Figure 3). These observations could be related to shape differences of the elbow articulation, but in order to confirm this interpretation, a further investigation is required.

Taking into account factors such as occupational stress and pathology, which could not be entirely controlled in this study, additional research of humeral shape is needed.
Furthermore, the sample consists of individuals with high mean ages, thus age-related factors, may affect differences in shape. Caution must be taken when anatomical interpretation is attempted.

Shape differences between males and females give slightly better classification results in proximal (75%) compared to the distal humerus (73%) which is opposite to France (France, 1983). Nonetheless these differences are too small to lead to any definite conclusion. As anticipated, classification accuracy improves when both size and shape are applied jointly. In a recent study of sexual dimorphism in American skulls, authors concluded that the combination of size and shape has better accuracy results than shape itself and classical osteometrical techniques on the same population (Kimmerle et al., 2008b). In our study the combination of form variables performed well with classification accuracies reaching 90% for both epiphyses. Whether this is statistically better than simply using centroid size needs to be tested in a proper statistical approach.

The analysis of humeral radiographs by geometric-morphometric techniques offers an alternative way to identify sex of unknown skeletal remains. Size differences between sexes are long acknowledged and confirmed by the results of this study. Thus the novelty deriving of this investigation is the existence of shape differences between sexes as they are reflected in the radiographs of the humeral epiphyses. The combination of shape and size characteristics seems to overcome the results based on the analysis of each one of them independently. However, this is a method which requires a background in a complex statistical theory hence its "superiority" compared to classical osteometric studies or the use of centroid size alone cannot be supported by the findings of this study, without further meta-statistical analysis. Nonetheless, the current method can be applied successfully to approximate sex for forensic purposes and could also be applicable in archaeological context.

### CONCLUSION

From the forensic standpoint, the usefulness of this study rests on the estimation of sex from radiographs of fragmentary humeri. The use of radiographs instead of the actual bone allows the identification of semi-decomposed bodies without the need of special preparation (ex. Maceration), thus facilitating the whole medico-legal investigation. The application of Geometric Morphometrics in humeral radiographs has proven to be successful, since it reveals shape differences that could not be assessed with conventional techniques and allows the combination of size and shape for the identification of sex.

# 8.2.3 Estimation of sex from the upper limb with the aid of ROC-analysis

# Introduction

Pelvis and skull are traditionally considered as the most dimorphic elements of the skeleton; hence many studies on the past are focused on producing sex estimation methods on these bones. Lately though, several postcranial elements have proved to be very effective sex predictors when metric methods are employed. Special attention was given by several scholars to the sexual dimorphism of the long bones of the upper limb (Holmann and Bennett, 1982; Mall et al., 2001; Sakaue, 2006; Celbis and Agritmis, 2007, Frutos, 2005; Carretero et al., 1995; Dittrick and Suchey, 1986; Albanese, 2005).

Despite the large amount on osteometric studies worldwide, there is a lack of such data in Greece. The few published studies deal cranial (Kranioti et al., 2008) and pelvic morphology (Papaloucas et al., 2008; Steyn and Iscan, 2008). However, no data for long bones are so far available. The aim of this work is to provide criteria for sex estimation from measurements of the long bones of the upper limp using Receiver Operator Characteristics (ROC) Analysis.

### Materials and Methods

The skeletal material for this study is selected from the cemeteries of St. Konstantinos and Pateles, Heraklion, Crete. A total of 173 well preserved skeletons of Cretan origin were used. A total of 12 measurements are taken: Maximum Humeral Length (HL), Vertical Head Diameter (HVD), Maximum Midshaft Diameter (HMaxMid), Minimum Midshaft Diameter (HminMid), Midshaft Circumference (HmidCirc) and Epicondylar Breadth (HEB) in humerus, Maximum Length (UL), Notch Height (UNH) and Distal Breadth (UDB) in ulna and Maximum Length (RL), Head Diameter (RHD) and Distal Breadth (RDB) in radius.

## Receiver Operator Characteristics (ROC) Analysis

ROC curves are employed in the evaluation of several variables as effective factors on sex estimation. The diagnostic value of the single variables was evaluated using the UAC. The cut-off values and the diagnostic characteristics of each variable (Sensitivity, Specificity, Positive and Negative predictive values) are calculated. The sensitivity of a diagnostic test is the proportion of specimens for whom the outcome is positive that are correctly identified by the test. The specificity is the proportion of specimens for whom the outcome is negative that are correctly identified by the test.

The correlation of normally distributed the variables was tested with the method Pearson correlation coefficient. The level of statistical significance is set to p<0.05 (a-

error). Means, standard deviations and F-ratios for all single dimensions were calculated by performing ANOVA with SPSS 13.0.

### Results

# Univariate statistics

Descriptive statistics of humeral, radial and ulnar measurements and associated univariate F-ratio to measure the differences between the sexes are shown in Table 7.1.2.2. The differences between the means in males and females are significant (p<0.0001) for all variables.

The results of the ROC analysis are shown in <u>Table 8.3.1</u>. Sensitivity, Specificity, Positive and Negative predictive values, AUC as well as the cut-off values for each measurement are presented. All measurements are found statistically significant at the level of 0.0001. According to the results each value equal or greater than the cut-off value for each measurement classifies the specimen as male while in the opposite case as a female. For instance an individual with radial length of 226 mm will be assigned as a male.

|          | Cut-off | Se   | Sp   | *AUC | PV   |      | Males | Females | Total |
|----------|---------|------|------|------|------|------|-------|---------|-------|
|          | value   |      |      |      | (+)  | (-)  | %     | %       | %     |
| HL       | 309.0   | 0.80 | 0.90 | 0.92 | 0.90 | 0.79 | 81.91 | 86.08   | 83.82 |
| HVD      | 43.3    | 0.90 | 0.89 | 0.93 | 0.91 | 0.91 | 92.55 | 87.34   | 90.17 |
| HMaxMid  | 21.2    | 0.77 | 0.80 | 0.85 | 0.82 | 0.74 | 78.72 | 77.22   | 78.03 |
| HMinMid  | 17.1    | 0.80 | 0.86 | 0.89 | 0.87 | 0.78 | 80.85 | 82.28   | 81.50 |
| HMidCire | 60.0    | 0.85 | 0.77 | 0.88 | 0.82 | 0.81 | 92.55 | 68.35   | 81.50 |
| HBB      | 57.1    | 0.90 | 0.84 | 0.93 | 0.88 | 0.87 | 89.36 | 82.28   | 86.13 |
| RL       | 224.0   | 0.96 | 0.87 | 0.95 | 0.90 | 0.95 | 96.81 | 84.81   | 91.33 |
| RHD      | 21.0    | 0.84 | 0.90 | 0.93 | 0.91 | 0.83 | 86.17 | 86.08   | 86.13 |
| RDB      | 28.5    | 0.84 | 0.77 | 0.87 | 0.81 | 0.80 | 85.11 | 74.68   | 80.35 |
| UL       | 241.0   | 0.96 | 0.86 | 0.94 | 0.89 | 0.94 | 95.70 | 83.33   | 89.02 |
| UNH      | 20.8    | 0.90 | 0.68 | 0.83 | 0.77 | 0.86 | 91.40 | 60.26   | 76.30 |
| UBD      | 19.6    | 0.72 | 0.87 | 0.85 | 0.87 | 0.72 | 72.04 | 84.62   | 76.88 |

Table 8.3.1: Cut-off values, Sensitivity, Specificity, area under the curve (AUC), predictive values and accuracies for all single variables

Figure 8.3.1 illustrates the ROC curves and the cut-off values for all humeral measurements and Figure 8.3.2 for radial and ulnar measurements. For UL the cut-off value is set in 241 mm with Se=0.96, Sp=0.86 and AUC=0.935.

The best discriminatory variables was found to be RL (91.3%) followed by HVD (90.2%) and UL (89%).UNH, UDB and HMaxMid did not performed well with less than 80% of correct group assignment.

## **Multivariate statistics**

ROC analysis was also performed using combinations of single variables for each bone separately. The variables were selected according to the AUC value. In the case of the humerus 3 variables (HL, HVD and HEB) were used. According to this combination 91% of the females and 79% of the males were correctly classified. For the radius all three dimensions were combined. According to the ROC analysis, if RL>224mm, RHD>21mm and RDB>28.5mm the individual is assigned as male. Classification accuracy was 99% (78/79) for females and 75% (74/95) for males.For the ulna all three dimensions were used. According to the ROC analysis, if UL>241mm, UNH>20mm and UDB>19.6mm the individual is assigned as male. Classification accuracy is 96.2% (75/78) for females and 63% (58/92) for males. The average accuracy does not exceed 80%.



Figure 8.3.1: ROC curves and cut-off values for the single variables of the humerus.

## Discussion

ROC curves were developed in the 1950's as a by-product of research into making sense of radio signals contaminated by noise (Green and Swets, 1966). More recently it

became clear that they are remarkably useful in medical decision-making (Fawcett, 2006). Despite the fact that traditional osteometric methods use discriminant function analysis for the study of sexual dimorphism, herein ROC curves are employed in the evaluation of several measurements on the long bones of the upper extremity as effective markers for sex identification.



Figure 8.3.2: ROC curves and cut-off values for the single variables of radius and ulna.

According to these data radial length (91%) is the most discriminatory variable for the upper limb measurements, followed by head vertical diameter of the humerus (90%) and ulnar length (89%).Multivariate methods usually perform better than single dimensions in discriminant function analysis. Interestingly for the radius that was not the case.

The results of this study indicate that ROC-analysis is an efficient method to study sexual dimorphism. From forensic standpoint the standards that are produced here can be useful for sex identification in forensic cases that unidentified skeletal remains of the upper extremity are recovered.

# **Chapter 9: Discussion**

Accurate determination of sex from the human skeleton is of great importance in anthropologic and forensic investigations. While the overlap in size of male and female range is still the most important aspect of sexual dimorphism, the accuracy depends on factors causing variation in sex. The greater the sexual dimorphism, the higher the classification accuracy from skeletal remains (Mays and Cox, 2000). It must be stressed that a population specific study is required in order to have accurate results in sexing a skeleton deriving from that population (Krogman and İşcan, 1986; Macho, 1990b). A recent study evaluating standard methods used for North American Whites concluded that they can be only partially applied to modern Greeks (Eliopoulos et al., 2004). Furthermore, the unique biological characteristics of Cretans, formed due to geographical isolation, raise the need for the development of population specific standards.

#### 9.1 Osteometric methods

## 9.1.1. Cranial skeleton

Despite the fact that sex assessment using craniofacial characteristics is commonly done worldwide, a lack of such investigation is noted in the Balkan countries. Among the few published studies, morphological sex determination of crania deriving from a mass murder grave in Serbia should be mentioned (Durić et al., 2005). This sample, consisting of individuals of Albanian descent killed in the recent Kosovo war, was sexed with an accuracy rate that hardly reached 71%. These results are relatively poor compared to the ones mentioned in the literature (Novotny et al., 1993; Walrath et al., 2004) There is beyond any doubt inter-population variation seriously affecting cranial sex accuracy (Novotny et al., 1993). But even in morphological studies exhibiting higher accuracies, a significant amount of intraobserver error is noted, deriving naturally from the subjective nature of the study (Walrath et al., 2004).

Metric studies are considered more advantageous due to the higher objectivity in evaluating data compared to morphological observations. With that in mind, the current work has focused on the development of population specific craniometric standards for a contemporary Cretan population. Although the mandible is considered the most dimorphic part of the skull (Acsádi and Nemeskéri, 1970; Durić et al., 2005), it was excluded from the current study because of a large number of edentulous individuals with excessive alveolar resorption, which has altered the mandibular dimensions.

The accuracies obtained in this study are either similar or even higher when compared to other groups (Giles and Elliot, 1963; Steyn and İşcan, 1998; Durić et al., 2005). A comparison of the modern Cretans is made with American and South African Whites (Caucasoids) of approximately the same period. It is possible to note that Cretans are closer in size to American Whites in most dimensions and furthest from African Whites. African Whites demonstrate a significantly larger cranial length (over 7 mm for males and over 6mm for females), while means for maximum frontal breadth are greater in Cretans for both sexes. Mean values for cranial length are greater in White (Terry) Americans as well, but all other dimensions are very close to contemporary Cretans.

A significant remark comparing all mentioned studies is that cranial length is included in the cranial function only in the present sample, suggesting a higher discriminatory value of this variable in Cretans as compared to other populations. Similar observations are made in the postcranial skeleton as well.

Sexual dimorphism in Cretans is well reflected in cranial dimensions, thus providing a very high accuracy rate of correct classification. From the forensic perspective, this information is essential for the identification of skeletal remains. Further research may provide additional standards for Cretans and Greeks and hopefully will be applicable to other Mediterranean and Balkan populations.

#### 9.1.2. Postcranial skeleton

Osteometric studies of long bones have established their importance in sex and stature estimation when skeletonized bodies or body parts are recovered without any identification. Given that osteometric methods for sex identification are populationspecific, many researchers from around the world have conducted studies on long bones, establishing specific standards of group assessment for several different populations (Singh and Singh, 1972; İşcan et al., 1998; Steyn and İşcan, 1999; Rogers 1999; Frutos, 2005; Carretero et al., 1995; Dittrick and Suchey, 1986; Albanese, 2005; Berrizbeitia 1989; Celbis and Agritmis 2005; Barrier and L Abbe, 2008; Steel, 1972; Singh et al., 1976, Introna et al., 1993; Purkait, 2001; Grant and Jantz, 2003; Matzon et al., 2006; Barrier and L Abbe, 2008; İşcan et al 1998, İşcan and Shihai 1994, Albanese 2003, Seidemann 1998, Mall et al., 2000, Asala 2000, Asala et all 2004; Hanihara, 1958; Holland, 1991; Kieser et al., 1992; İşcan and Milner Savitz 1984b, İşcan et al., 1994; Bruzek, 1995; Steyn and İşcan, 1997; Gonzalez-Reimers et al., 1999; Sakaue, 2004; Slaus and Tomicic, 2006; Sacragi et al., 1994). Among them some data, though limited, derive from Balkan populations (Jantz et al., 2008b; Kimmerle et al., 2008a). However, no information on the osteometric characteristics of the long bones in Greeks has been so far reported.

The current study addresses population specific standards for the Cretan population for single and combined measurements of the six most important long bones that are usually recovered in forensic settings. All long bones performed well and have proven to be effective for the identification of sex. The best single dimension for the humerus was HVD (89.6%), for the radius RL (90.8%), and for the ulna UL (88.9%). For the lower limb classification was slightly lower, with the best single dimension for the femur being FHMaxD (83.7%) and for the tibia TLB (84.6%). Regarding the fibula, only the maximum length was measured, resulting in 84.6% accuracy. When all the measurements of the upper limb bones were combined, better classification results were obtained compared to the lower limb bones.

An interesting point to note is that most of the earlier studies suggest that epiphyseal breadth and circumferential measurements are better sex discriminators than length (France, 1983; Wu, 1989; Işcan et al., 1994), while in the present study length has exhibited high F-ratios in all upper limb long bones, but it performed worse in the case of femur and tibia. More specifically, maximum length was found to be the most discriminatory single variable for the radius (90.8%) and the ulna (88.9%) and the third best variable for the humerus (84.4%). It is noteworthy that in the best combination of variables for each bone in DFA, the maximum length was included in all cases. The same observation was made in the study of the Chinese (İşcan et al., 1998) and German (Mall et al., 2001) populations, while in the Guatemalan sample, a high eigenvalue of length among the other dimensions was observed, which indicates that this is a valuable discriminating factor despite its low percentage of accuracy (Frutos, 2005). A similar result was produced when stepwise discriminant analysis was applied to cranial data of the same population; length enters the equation, indicating that it constitutes a highly discriminatory variable for sex allocations in Cretans.

It is commonly known that the overall reliability of sex estimation depends on the chosen method and the population taken into account (Krogman and İşcan, 1986; Macho, 1990a). The computation of posterior probabilities for all functions allows the observer to evaluate each method for the particular case taken into account. The determination of sex using posterior probabilities and a threshold of 0.95 is highly reliable and therefore it was considered in the present study. Posterior probabilities at 0.8 and 0.9 thresholds were also calculated. According to this principle, the percentage of correctly assigned specimens based on formulae with over 80% accuracy was calculated. For the single variables, RL classified over 40% of the sample with 91% accuracy at a 95% threshold and 58% at a 90% threshold, which indicates that it is a highly dimorphic and reliable variable in the given population. HVD performed equally well by correctly assigning sex to 40% of the specimens with 90% accuracy. The single variables of the lower limb performed considerably worse, since the highest percentage of correct classification at a 95% threshold did not exceed 30% (NFmax). When multivariate statistics were applied, the

reliability of the method rose considerably. Discriminant functions for the humerus correctly assigned sex up to 65% at a 95% threshold and up to 75% at a 90% threshold, with accuracies that reached 92%. Discriminant functions for the radius correctly classified up to 57% of the sample with up to 94% accuracy. Lower limb bones performed worse, with posterior probabilities that did not exceed 50% at a 95% interval of confidence. These results indicate that upper limb bones produce more reliable formulae compared to lower limb bones in the Cretan population. However, the method of choice is always driven by the particular case under investigation, which makes these data highly valuable when a skeleton of Cretan origin is considered.

Naturally, questions concerning the applicability of this method to other Greek and Balkan populations arise. As for the Balkans, recent analysis of sexual dimorphism of the femur revealed size differences of the femoral head and the total length among three groups (Croatians, Bosnians and Kosovars) (Jantz et al., 2008a), suggesting that a population-specific methodology is required for each region (Jantz et al., 2008a; Ubelaker, 2008). Furthermore, studies on craniofacial variation reveal significant differences even between populations which share common Slav ancestry, such as Bosnians and Croatians (Ross, 2004). A small sample of Greeks (N=14) that was included in the later study was found to be the furthest removed from the rest of the Balkan groups and closer to the American Whites (Ross, 2004).

The few published data on modern Greeks are restricted to a few studies on pelvis morphology (Papaloucas et al., 2008a; Steyn and İşcan, 2008). Papaloucas and collaborators (2008a) measured four dimensions on the pelvis and femur of a sample from Athens. They found slightly higher mean values for the acetabular diameter for both males and females, as compared to Steyn and İşcan (2008) on Cretans. Femoral head diameter in the Athens collection was found to be higher in males (mean: 48.5+/- 2.3 mm) and lower in females (mean: 41.6+/-1.9 mm) as compared to the Cretans (males: 47.3+/-2.6 mm, N=94, females 42.4+/-2.3 mm, N=78). This indicates a larger amount of overlaping in Cretans compared to the population from Athens. It must be emphasized, however, that Papaloucas and co-workers (2008a) measured right femora and pelvises, while data for Cretans are obtained from the left side. Nonetheless, the means on the two dimensions that we were able to compare do not differ tremendously between the two populations, implying that standards on Cretans could be applicable to other Greeks. On the other hand, a recent work (Papaloucas et al., 2008b) on the bilateral asymmetry of the humeral length in the Athens collection provided mean values for males and females (males: 342.2+/-6.3 mm, N=100, females 314.1+/-3.2 mm, N=100) that exceeded the values obtained here for Cretans (males: 321.3+/-14.5 mm, N=94, females 294.2+/-13.7 mm,

N=79) by about 2cm. Obviously, more comparative data are needed to test the applicability of the osteometric data provided here to other Greek populations.

Lately there has been a great deal of discussion on secular changes (Jantz and Jantz, 1999; Jantz and Meadows Jantz, 2000; Sparks and Jantz, 2000). Studies in the U.S. detected secular changes on long bones in a time interval of 170 years (Jantz and Jantz, 1999). It is noteworthy that secular trends in Americans are found to be more pronounced in lower limbs compared to upper limbs, and in distal bones as compared to proximal ones (Jantz and Jantz, 1999). Consequently, the humerus exhibits higher resistance on short-time secular changes than the femur for instance. Notwithstanding the lack of similar studies on modern Greeks, the osteometric data derived from 20<sup>th</sup> century Cretans are expected to be applicable to the current population of Crete. Additional research is obviously needed to define the biological characteristics of other Greek sub-groups from the mainland and the islands. Comparative data can provide the scientific proof of whether the metric standards produced in this study can be reliable for the rest of Greece.

The recovery of fragmentary and pathological skeletal remains, in forensic investigations, requires easy and rapid techniques for biological profiling and reconstruction of the scene history. Simple measurements performed during autopsy can provide an immediate and accurate prediction of sex, thus contributing significantly to positive identification in forensic cases. There is no doubt that population differences affect the sexual dimorphism reflected in the dimensions of the long bones. Hence, a specific standard for sex estimation in a modern Cretan population is addressed here. The results of this study demonstrate that long bones are effective for the identification of sex for forensic purposes, since even in a fragmentary state they can give high classification accuracy. Naturally, additional research is required to test the applicability of this technique in other Greeks and Balkan populations.

### 9.2 Radiometric methods

## 9.2.1 Postcranial skeleton

The identification of a deceased person in forensic investigations is quite an easy procedure in relatively recent deaths, where face and fingerprints are available. As demonstrated earlier, postcranial measurements can provide highly accurate sex estimation even for fragmented bones. Not infrequently, however, individuals are recovered in forensic settings disfigured and highly decomposed, without fingerprints or even mummified, and in such cases identification becomes more complex and time-consuming. In mass disasters the pathologist is called in to deal with commingled, charred and fragmented body parts of different individuals, thus making identification more complicated. The existing skeletal elements are partially exposed due to the remaining soft tissue; hence special techniques, like maceration, are needed in order to carry out the examination. In such cases, trained forensic anthropologists are needed. The lack of such expertise in Greece, among other parts of the world, obligates the forensic pathologists to undertake the burden of the identification of unknown skeletal remains. In order to facilitate this procedure and speed up the identification process, the idea of developing an easy and rapid method for the identification of sex emerged.

Radiological identification was first introduced in 1926 by Culbert and Law and since then it has been used extensively in diagnosing skeletal pathology and trauma (Krogman and İşcan, 1986; Kahana and Hiss, 1997, 1999), as well as in the detection of foreign bodies (Brogdon, 1998; Kahana and Hiss, 1999; Brogdon, 2006; Stein and Gruenberg, 2008) and securing evidence for court. In several occasions, classical radiographic methods have been used in skeletal identification (Riepert et al., 1996; Kahana et al., 1997; Kahana and Hiss, 2002; Sağir, 2006; Mahfouz et al., 2007; Petrovecki et al., 2007). Riepert and colleagues (1996) developed a sex estimation method based on radiographs of the calcaneus and reported 84.4% accuracy. Recently, CT scans have been employed in sex assessment of the femur, yielding 84.6% correct group membership (Harma and Karakas, 2007). Mahfouz and associates (2007) predicted sex with up to 93% accuracy by using linear measurements taken from CT scans of patellas.

The method proposed here is based on linear measurements taken on radiographs of long bones. The radiographic machine that was used is an accurate and flexible device used routinely in our department for diagnostic and scientific purposes. This equipment has been widely accepted in everyday medical practice for its sensitivity and accuracy with expanding applications in radiology, cardiology, paediatrics, traumatology, operation rooms, intensive care units, pneumonology, forensic pathology and currently anthropology. The radiographs taken are stored in digital form and can be transferred to a computer for further evaluation and thus can be kept as evidence in case that they are needed in court. The advantage of such a machine lies in the rapid diagnosis and the fact that there are no additional costs for consumables (i.e. films etc).

Since the integrity of the recovered bones in forensic settings cannot be assured, this study considers fragmentary models. Four long bones were employed and each epiphysis was radiographed separately. A certain number of landmarks were selected in each radiograph and all inter-landmark distances were calculated. The landmarks were selected with the objective of being easily distinguished even by an inexperienced observer. The generated distances are the variables used to discriminate males and females with the aid of discriminant function analysis. The identification of sex using linear measurements on radiographs of the long bones has proven feasible according to these data. Of the tested epiphyses, all 8 were proven to be effective for sex identification with accuracies rating from 84% to 92%. In almost every case, both epiphyses performed equally well for both original and crossvalidated data. The radius exhibits a different pattern, with the distal epiphysis (92%) performing considerably better compared to the proximal one (84%). The femur performed better than the tibia for both epiphyses, and the humerus better than the radius for the proximal epiphysis and worse for the distal epiphysis. The most effective bone for sex estimation using the radiometric method is the radius (distal epiphysis), followed by femur (90% for both epiphyses), humerus (89% for both epiphyses) and tibia (proximal epiphysis-88%).

Of the single dimensions, a linear measurement on the proximal tibia performed equally well with the best multivariate discriminant formula. This variable, PT15, corresponds to the projection of the upper epiphysis breadth of the tibia and yielded 88% correct group membership. According to discriminant function analysis each individual with PT15>68.28mm is classified as male, otherwise as female. Interestingly, the upper epiphysis breadth (TUB) as analysed on the osteometric method hardly reached 81% correct group membership with a 71.8 mm cut-off value. The different cut-off value can be attributed to the fact that PR15 is a projection of TUB and not the same variable (tibiae were radiographed with the anterior surface facing the X-ray table, and the distal epiphysis perpendicular to the axis of the camera). Another reason is the different sample size for the osteometric (N=172) and radiometric (N=102) method, which could also be responsible for the distinct classification results. A future comparative study of the two methods employing the same sample is necessary in order to conclude whether the radiographic method is better in the case of single dimensions of the tibia.

The analysis of the femur resulted in 90% correct group membership for both epiphyses, which is higher than the results obtained in Harma and Karakas' study (2007) of the femur (85%). The different classification results can be attributed to several reasons. Firstly, they are related to the different variables employed in each study (Harma and Karakas measured total length and head vertical diameter, while in the present study length was not calculated). Also, PF14 corresponds to the maximum head diameter of the femur on the radiograph and thus it cannot be compared to the HVD measured by Harma and Karakas (2007). However, the results of the osteometric study indicate that the HVD is highly dimorphic in the Cretan population (90% accuracy) contrary to the Anatolians (77%). PF14, on the other hand, did not exceed 80% accuracy in Cretans.

As in any osteometric study, the standards provided here should be treated as specific for the Cretan population and caution should be taken when applying the formulae to other Greek or Balkan populations. Osteometric data for the femur and the humerus that are provided by the literature are contradictory concerning the existing variability between the population of Crete and a mixed population from Athens. However, to test the applicability of the formulae produced in this study to other populations, for both osteometric and radiometric data, several comparative samples are needed, which would certainly be a subject for future studies.

Posterior probabilities for univariate and multivariate discriminant functions of the radiometric variables were also calculated. As in the case of the osteometric variables, determination of sex using posterior probabilities and a threshold of 0.8, 0.9 and 0.95 was considered. According to this principle, the percentage of correctly assigned specimens based on formulae with over 80% accuracy was calculated. As a general observation, multivariate discriminant functions classified the sample with higher reliability than single dimensions independently of the accuracy percentage.

For the humerus, single variables of the proximal epiphysis performed similarly with the ones for the distal epiphysis in classifying the sample at a 0.95 threshold. Multivariate discriminant functions for the proximal humerus, though, seem to be more reliable compared to the distal one, correctly sexing up to 57% of the specimens at a 0.95 threshold (Table 7.2.3.2). In the case of the radius, results for the lower epiphysis were more reliable (compared to the upper epiphysis) for both uni- and multivariate analysis, achieving a classification of more than half of the sample within 95% confidence intervals (DRF1). The variables of the proximal epiphysis seem to overlap considerably, and thus should be considered with caution. Similarly, the lower end of the femur achieved better separation of the groups at a 0.95 threshold for both single and multifactorial analyses. The employment of posterior probabilities in this study allows the evaluation of the method in every case independently, thus facilitating the observer in selecting the method of choice for sex estimation according to the available bones and the population under study.

The lack of forensic anthropologists in Greece and other places around the world calls for the development of rapid and easy techniques that can be applied by pathologists in order to reconstruct a biological profile, thus assisting in positive identification. The radiometric method has proven to be applicable in sex estimation of unknown semi-decomposed, charred or mummified remains, such as the ones recovered in mass disasters or forensic cases. As a further step, a diagnostic tool was created (as shown in chapter 8) based on radiographs of the proximal femur, in order to identify sex. The SIS (Sex Identification Software) is programmed in Java and is based on the selection of landmarks

on a radiograph. The radiometric standards for the femur produced in this study were incorporated in the Java program in such a way that when the observer selects the 6 predefined landmarks on the radiograph, SIS calculates all inter-landmark distances and applies the measurements to the best formula produced by DFA, calculates the discriminant score for this formula and gives the sex along with the posterior probability of the specimen being correctly sexed. This tool is a preliminary demonstration of a more complete tool, which will contain all radiometric standards that are produced by this study, not only for the femur but also for the humerus, radius and tibia, in the Cretan population. With the new software, the observer will be able to choose first the bone under examination, secondly the landmarks on the radiograph and thirdly the formula based on which the sex will be estimated. As a result, the software will provide the sex estimation along with the posterior probability. If, for example, a specimen is assigned as male with posterior probability of 60%, obviously the method is not reliable for the particular case and the observer (the pathologist in forensic cases) should seek a different method to assess sex. On the contrary, a case assigned, for example, as male with 95% probability is highly reliable and the method should be used in the particular case. The availability of such a tool in the medicolegal routine is highly valuable for quick sex assessment by both experienced anthropologist and forensic pathologists.

Another point to highlight is the employment of radiographs in a geometricmorphometric study in which a different methodology was applied in order to separate sexes bases on shape differences. For this study, both epiphyses of the humerus were used and the same landmarks were chosen on the radiographs. Instead of calculating the interlandmark distances, this method is based on shape, size and form variables. On the distal end, the male configuration is rectangular, while the female configuration is squared, probably due to the relatively wider epiphyseal breadth in males. A relative wider lateral trochlea accompanied by a relatively smaller capitulum has also been observed in males in respect to females. The analysis of humeral radiographs by geometric-morphometric techniques offers an alternative way to identify the sex of unknown skeletal remains. Size differences between sexes have long been acknowledged and confirmed by the results of this study. Thus the novelty deriving from this investigation is the existence of shape differences between sexes as they are reflected in the radiographs of the humeral epiphyses. The combination of shape and size characteristics seems to outperform the results based on the analysis of each one of them independently. However, this is a method which requires a background in a complex statistical theory. Thus its "superiority" as compared to classical osteometric studies or to the use of centroid size alone, cannot be supported by the findings of this study, without further meta-statistical analysis. Nonetheless, the current

method can be applied successfully to approximate sex for forensic purposes and could also be applicable in an archaeological context.

The current method yielded comparable classification results with classical osteometric methods applied on the same population. Whether this method is actually better or not should be tested with meta-statistical approaches, which exceeds the purpose of the current study. The important point to be made is that radiometric techniques are applicable in forensic cases for identification purposes and their employment can be advantageous when a rapid examination is required and maceration is not an option. The present study does not aim to propose a method that would replace the osteometric techniques, but instead to offer an alternative method applicable in certain circumstances in which osteometry cannot be applied, acknowledging that the method of choice in forensic anthropology is always case driven.

# 9.3 Conclusions

1. Sex estimation with the aid of linear measurements taken on digital radiographs of 4 long bones (humerus, radius, femur and tibia) is possible, with accuracies up to 95%.

2. Among the 4 bones studied here, both epiphyses are found equally effective in sex estimation, with the exception of the radius, for which the lower epiphysis gave higher accuracies.

3. The radiometric method is advantageous in cases of disfigured, semidecomposed, charred or mummified bodies, or body parts recovered in forensic cases of mass disasters.

4. The application of Geometric Morphometrics in humeral radiographs has proven to be successful, since it reveals shape differences that could not be assessed with conventional techniques and allows the combination of size and shape for the identification of sex.

5. The osteometric method developed here provides standards for sex estimation of cranial and postcranial skeletons of Cretans, a population that has not been represented so far to the known databases.

6.Measurements on the postcranial skeleton are more accurate for sex allocations in the Cretan population as compared to cranial measurements.

7. Posterior probabilities at the threshold of 95% provide a useful tool to the anthropologist and/or the pathologist in order to select the most reliable method according to the particular case under examination.

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The purpose of the study is to develop a sex determination technique using osteometric data from remains exhumed from two contemporary Cretan cemeteries in Heraklion, Crete. The study population consists mostly of Cretans or individuals that lived in Crete for more than three generations that lived and died between the end of the 19th century and the beggining of the 20th. A total of 200 skeletons became available for investigation. A number of people who may have migrated from Turkey, islands and mainland Greece are excluded from the study. All individuals with obvious bone pathology have been also removed from the sample. Age and cause of death has been obtained from the Heraklion City Hall census archives, for only part of the skeletal material, while sex can be obvious from the names written on the boxes that contained the remains. The mean age for males is 68, 57 + -13.52 (N=61) and for females 72, 98 + -16, 90 (N=58).

