

An assessment of growth and sex from mandibles of cadaver foetuses and newborns

Ву

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Declaration

I declare that the disse	rtation that I am hereby submitting	g to the University of Pretoria
for the MSc degree in	Anatomy is my own work and that	I have never before
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Abstract

The quantification of skeletal data is one way in which to demonstrate variation in human growth. In South Africa, few researchers have assessed patterns of growth in immature mandibles. The purpose of this study was to evaluate growth and sexual dimorphism in the mandible from the period of 31 gestational weeks to 36 months. A total of 74 mandibles were used, skeletal tissues were sourced from the Raymond A. Dart Collection (University of Witwatersrand), and cadaveric remains were obtained from the University of Pretoria and the University of Witwatersrand. The sources of cadaver materials (both bequeathment and unclaimed remains) included local provincial hospitals. The sample was divided into four groups, namely 31 to 40 gestational weeks (group1), 0 to 11 months (group 2), 12 to 24 months (group 3), and 25 to 36 months (group 4). Twenty-one osteological landmarks were digitized using a MicroScribe G2. Ten standard measurements were created and included: the longest length of mandible, mandibular body length and width, mandibular notch width and depth, mental foramen to inferior border of mandible, mandibular basilar widths bigonial and biantegonial, bigonial width of mental foramen and mental angle. Data were analyzed using PAST statistical software and Morphologika2 v2.5. For the linear measurements, no statistically significant difference between either the foetal and up to 12 month groups or the 2 to 3 years groups. However, statistically significant increases with age were noted between 12 and 24 months for nine variables. This can be associated with growth of the mandibular arch, development and eruption of the dentition and development of the masticatory structures. No evidence of sexual dimorphism was observed until age 3, where the mental angle and mandibular notch were significantly larger in females than males. In conclusion, the mandible develops and grows so as to accommodate development of the tongue, mastication and dental eruption. Future research that considers the influence of secular trends on mandibular growth is needed.



Abstrak

Die kwantifisering van skeletale data is 'n betroubare metode om variasie in menslike groei aan te toon. Slegs enkele Suid-Afrikaanse navorsers, het groeipatrone in onvolwasse mandibulae nagevors. Die doel van hierdie studie was om groei en geslagsdimorfisme in die mandibula vanaf 31 gestasie weke tot 36 maande na geboorte te evalueer. 'n Totaal van 74 mandibulae was gebruik. Skeletale weefsel uit die Raymond A. Dart Versameling (Universiteit van die Witwatersrand), en kadaweroorskot van die Universiteite van Pretoria en van die Witwatersrand was verkry. Die oorsprong van kadawermateriale (beide skenkings en onopgeëisde oorskot) het plaaslike provinsiale hospitale ingesluit. Die steekproef was verdeel in vier groepe, naamlik 31 to 40 gestasie weke (groep1), 0 tot 11 maande (groep 2), 12 tot 24 maande (groep 3), en 25 tot 36 maande (groep 4). MicroScribe, G2 is aangewend om 21 standaard antropometriese landmerke te digitiseer. Hieruit is 10 standaard antropometriese afmetings geskep o.a.: langste lengte van mandibula, lengte en breedte van corpus mandibula, afstand tussen foramen mentalis en inferior grens, basale wydte bigoniaal en biantegoniaal, bigoniale wydte van foramen mentalis asook mentale hoek. Inligting is d.m.v. PAST statistiese sagteware en Morphologika2 v2.5 ontleed. Volgens die Kruskal-Wallis-toets was die verskille tussen groepe 1 en 2, asook 3 en 4 statisties onbeduidend. Alle afmetings by groepe 2 en 3 het beduidende toenames getoon, behalwe dié van die afstand tussen foramen mentalis en inferior grens. Die veranderings mag die gevolg wees van die groei van die mandibula en koustrukture. Geslagsdimorfisme was aantoonbaar in groep 4, by die mentale hoeke (p=0.03) asook dimensies van die incisura mandibularis (p=0.0006), waar dié van vroulike individue groter was. Voor geboorte vergroot die arcus mandibularis om die ontwikkelende tong te huisves, terwyl dit na geboorte verander om die koustrukture te huisves. Gevolglik hermodelleer en groei die been as aanpassing vir die kouproses en om strukturele integreteit te behou. Geslagsdimorfisme word ook beïnvloed deur die kouproses. Die meeste veranderinge, veral dié van die koustrukture, was duideliker in vroulike individue. Toekomstige navorsing wat die invloed van sekulêre tendense op die groei van die mandibula oorweeg, is nodig.



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Table of Contents

Abstr	act				III
Abstr	ak				IV
Ackn	owledg	ements	;		V
List o	f Figur	es			IX
List o	f Table	es			XII
1.0.	Introd	duction			1
2.0.	Litera	nture Re	eview		4
	2.1.	Embry	yology of the	e mandible	4
	2.2.	Anato	my of the in	fantile Mandible	9
		2.2.1	Growth of t	he mandible	12
	2.3.	Morph	nological est	imation of sex in juvenile and adult mandibles	14
		2.3.1.	Juveniles (0 to 19 years)	14
			2.3.1.1.	Gonial eversion	14
			2.3.1.2.	Inferior symphyseal border of the mandible	14
		2.3.2.	Adults		15
3.0	Mate	rials and	d Methods		19
	3.1.	Mater	ials		19
	3.2.	Metho	ods		22



		3.2.1. Dissection	22
		3.2.2. Collection of landmark data	24
		3.2.3. Osteometric analysis	26
		3.2.4 Statistical analyses	30
		3.2.5. Geometric morphometric analysis	30
		3.2.6. Osteometric statistical analysis	31
		3.2.7. Inter-observer error	32
4.0.	Resu	lts	33
	4.1.	Osteometry	33
		4.1.1. Comparisons between osteometric variables and age at death	33
		4.1.1.1 Medio-lateral plane	34
		4.1.1.2. Superior-inferior plane	38
		4.1.1.3. Anterior-posterior planes	39
		4.1.2. Comparison between osteometric variables and sex	42
	4.2.	Geometric morphometrics	43
		4.2.1. Age related morphological changes	44
		4.2.1.1. Medio-lateral plane	45
		4.2.1.2. Superior-inferior plane	46
		4.2.1.3. Anterior-posterior plane	46
		4.2.2. Sex related morphological changes	49
	4.3.	Inter-observer error	51



4.4. Summary

5.0.	Discu	ssion	52
	5.1.	Growth of the mandible: a combination of osteometric variables and	d
		geometric morphometric parameters	52
	5.2.	Sexual dimorphism of the mandible: a combination of osteometric	
		variables and geometric morphometric parameters	59
6.0.	Conc	lusion	61
7.0.	D. References		
8.0.	Appe	ndices	
		Appendix A	67
		Appendix B	69
		Appendix C	72
		Appendix D	73

51



List of Figures

Figure 2.1	Cranial bones formed by endochondral ossification (Shading; cranial vault and face) and by intramembranous ossification (Lines; cranial base and mandible)(redrawn from Ten Cate's Oral Histology 7 th ed. pg 50)	5
Figure 2.2	Site of initial osteogenesis related to mandible formation (redrawn from Ten Cate's Oral Histology 7 th ed. pg 50)	
Figure 2.3	Spread of mandibular ossification away from the Meckel's cartilage at the lingula (redrawn from Ten Cate Oral Histology 7 th ed. pg 51)	
Figure 2.4	Schematic diagram of a coronal section through an embryo (redrawn from Ten Cate Oral Histology 7 th ed. pg 51)	
Figure 2.5	The lingual surface of a mandible with an approximate dental age of 1 year, Raymond A. Dart skeletal collection, University of Witwatersrand	
Figure 2.6	Anterior view of a mandible with an approximate dental age of 1 year, Raymond A. Dart skeletal collection, University of Witwatersrand	
Figure 3.1	Planned incision lines A-E for dissection of a neonate (stillborn), lateral view, School of Anatomical Sciences, University of Witwatersrand	
Figure 3.2	Standard measurement position used to digitize mandibles (symphysis menti superior, left mandibular condyle: bottom right corner, right mandibular condyle:	23



centre)

Figure 3.3	An illustration of the 21 homologous (sp) landmarks recorded on each mandible corresponding to the landmarks defined in Table 3.2.	
Figure 3.4	Foetal/infant mandible: anterior	26
Figure 3.5	Foetal/infant mandible: lateral	27
Figure 3.6	Generalized full procrustes analysis (adapted from Evolutionary and Developmental Morphometrics of the Hominoid Cranium, Mitterocker P pg 29)	
Figure 4.1	Mean and standard deviation values for the mental angle of groups 1-4	35
Figure 4.2	The mean and standard deviation for the mandibular basilar width bigonial (pattern bar) and mandibular basilar width bi-antegonial (solid bar) for groups 1-4.	36
Figure 4.3	Mean and standard deviation of the bigonial width of the mental foramen for groups 1-4	37
Figure 4.4	Mean and standard deviation of the distance between the mental foramen to the inferior border of the mandible on the left for groups 1-4	38
Figure 4.5	Mean and standard deviation of the mandibular notch depth (MND) for the left side for groups 1-4	39
Figure 4.6	Mean and standard deviation of the mandibular width on the left side for groups 1-4	40
Figure 4.7	Mean and standard deviation of the mandibular notch width on the left side for groups 1-4	
Figure 4.8	Mean and standard deviation for the mandibular body length on the left side for groups 1-4	41



Figure 4.9	Mean and standard deviation of the longest length of the mandible on the left sides for groups 1-4	41
Figure 4.10	Mean and standard deviation of the mental angle for males (n=43) (pattern bar) and females (n=31) (solid bar) for groups 1-4	42
Figure 4.11	Mean and standard deviation for the mandibular notch depth on the left side for males (n=43) (pattern bar) and females (n=31) (solid bar) for groups 1-4	43
Figure 4.12	The relationship between size and principle component 1 (PC1) from 30 gestational weeks to 36 months postnatal	44
Figure 4.13	Distribution of variance along principle components 1 and 2 comparing groups 1-4	48
Figure 4.14	Distribution of variance along principle components 1 and 2 comparing males (solid lines) and females (broken lines) within groups 1-4	50
Figure 5.1	Mandibles of varying ages from the Raymond Dart Skeletal collection, University of Witwatersrand.	54
Figure 5.2	Lateral view of mandible aged 1 year (LLM: longest length of mandible, MBL: mandibular body length)	55
Figure 5.3	Lateral view of mandible aged 2 years (MND: mandibular notch depth, MNW mandibular notch width)	56
Figure 5.4	Lateral view of mandible aged 3 years (MND: mandibular notch depth, MNW: mandibular notch width)	56



List of Tables

Table 3.1	Sample composition collected from the University	
	of Witwatersrand and University of Pretoria	21
Table 3.2	A description of the homologous landmarks on the	24
	mandible	24



CHAPTER 1

INTRODUCTION

Quantification of skeletal data has been shown to be an effective and reliable method of demonstrating variation in human growth as well as for monitoring and interpreting the growth of various skeletal elements in the living [1-3]. Despite numerous studies conducted by Fazekas, Kosa and Hesby on calculating foetal and post-childhood mandibular growth, less research has focused on morphological changes - associated with growth and development - in the mandible between late term-foetuses to early childhood [4,5].

The purpose of this study was to investigate changes in the size and shape of the mandible from the late foetal to early childhood periods. Age categories included a prenatal group of 30 to 40 gestational weeks, and postnatal groups including 0 to 11 months, 12 to 24 months and 25 to 36 months. Differences between the sexes for the various age at death categories were also examined. Knowledge of normal growth trends may be used to more reliably diagnose abnormal changes in growth and development of the mandible. This osteometric and morphological information may also aid in the estimation of age and sex of unidentified persons.

If undetected, abnormal mandibular growth in a foetus may present with life threatening complications at birth. Four anomalies, which are associated with inadequate mandibular development, include micrognathia, Hallermann-Streiff syndrome, Robinson anomalad and retrusion of the mandible. Micrognathia is often regarded as a common congenital defect [6]. Micrognathia is often identified via sonography, with the aid of lateral views or in some cases with the aid of three-dimensional computerized tomography (3D CT), alternatively known as Cone Beam Computed Tomography [7]. These imaging techniques show the facial profile and the relationship of the mandible to the maxilla. The identification of micrognathia is based on a quantitative assessment of a 'small' jaw and the posterior displacement of the



mandible in relation to other facial features [8]. This is best illustrated by means of a ratio where the mandibular width (MD) is compared to the maxillary width (MX). Micrognathia is diagnosed if the calculated ratio value is less than 0.785 [9]. Micrognathia and cleft palate are often found in association with each other. If diagnosed early and delivery of the infant is successful, then treatment of these conditions may include an immediate tracheotomy and, in later teenage years, distraction osteogenesis (bone stretching) [10].

Similar to micrognathia, Hallermann-Streiff syndrome - a congenital defect - presents with a bird-like appearance of the jaw. This is due to the fact that in profile, the mandible is considerably smaller than the maxilla. Patients presenting with Hallermann-Streiff syndrome exhibit hypoplasia of both the mandible and maxilla, which can be seen with radiographic techniques. It can also be diagnosed by means of the anterior displacement of the TMJ (temporomandibular joint). Other radiographic findings include hypoplasia of both condyles, narrowing of the neck of the mandible on the right hand side, absence of the cortical bone on the left, and anterior displacement of the complete TMJ. Macrodontia, hypoplasia of the maxillary complex, broad mandible, obtuse angle, missing successional teeth, late dental development and atypical mandibular molars are observed as well [11].

Underdevelopment of the mandible can also be associated with Robinson anomalad and mandibular retrusion. The causes of which is due to an interruption in the main growth centre of the mandibular condyles. With regard to retrusion, postnatal changes lead to general bone loss which could be attributed to infection, trauma or even fractures [12]. Correction of these problems involve surgical intervention. Some of the techniques employed, which date as far back as the 1950's, remain in use today. These procedures include sliding osteotomy, osteotomies with bone grafts, flail joints, condylectomy, condylar displacement and, more recently, distraction osteogenesis [12,13].



Another possible use of osteometric dimensions from foetal and infant mandibles is for the estimation of sex from unknown remains. Potential morphological and metric indicators of sex have been investigated using the skull and pelvis of immature skeletons with only moderate success [14,16]. Schutkowski (1987) measured the greater sciatic notch, the ilium and femur of immature remains and was only able to distinguish between the sexes with an accuracy of 70% [17]. Loth and Henneberg (2001) developed a morphological technique, which provided an 81% classification between sexes in South Africans [16]. However the method has not been corroborated in additional studies. According to Franklin et al (2007) the dimorphic features outlined by Loth and Henneberg (2001) could not be identified with an accuracy greater than 59% overall [18]. These researchers suggest that the shape of the border of the mandible was either not sexually dimorphic or varied substantially among black South African population groups. Therefore further clarification with regard to the use of the mandible in the estimation of sex for juvenile remains is needed.



CHAPTER 2

LITERATURE REVIEW

2.1. Embryology of the mandible

A breakdown in the relationships between the building blocks of facial development such as retinoic acid and various growth factors, may contribute to craniofacial anomalies as well as to differences between the sexes. Therefore, facial morphogenesis as the foundation from which morphometric changes in later stages of development can be explained and interpreted is discussed [19].

The structures of the juvenile face and jaws originate from primordial tissue that surrounds the stomodeal depression of the embryo. The development of the facial bones, particularly the mandible, is complex and involves the patterning, fusion, outgrowth and modeling of a variety of tissues for the formation of structures in the upper and lower face. The lower facial structures derive from the first pharyngeal arch, whereas the mesenchymal tissue of the face has been shown to have a neural crest cell origin. Each of the tissue components that make up the structures of the face are regarded as the product of a unique mixture of growth signals and other morphometric determinants. Further evidence suggests that specific sets of molecular signals are the controlling factors in the development of the facial structures. The development of the mandible along the proximodistal and rostrocaudal axes is influenced by these molecular signals [19].



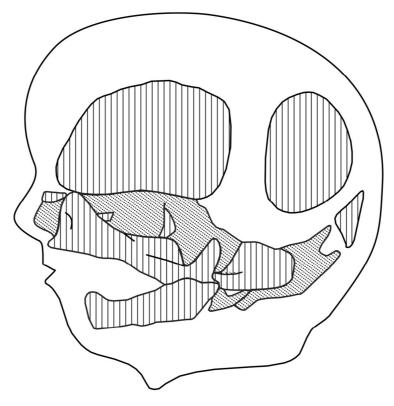


Figure 2.1: Cranial bones formed by endochondral ossification (Shading; cranial vault and face) and by intramembranous ossification (Lines; cranial base and mandible)(redrawn from Ten Cate's Oral Histology 7th ed. pg 50) [20]

Basic structures of the face are established between 4 to 8 weeks of embryonic life. However, certain structures such as the mid-face, remain underdeveloped during embryogenesis and for some time after birth. During gestational weeks 4 and 5, mandibular growth and development is initiated via both endochondral and intramembranous ossification. The cranial bones of the face and cranial vault are formed via intramembranous ossification (see Figure 2.1).

During the 7th week of embryonic life, the primary ossification centres of the mandible begin to develop. These centres are located in the dense fibrous tissue band that surrounds the Meckel's cartilage, and is close to where the future mental foramen is to be located. An illustration of this stage is shown in Figure 2.2, where the mental branch of the trigeminal nerve is near the site of initial ossification. The mental nerve is to emerge from the mental foramen, which is located on the anterior surface of the



developing mandible. Ossification may only commence once the ectomesenchyme of the lower jaw has interacted with the epithelium of the mandibular arch (Figure 2.2) [20-22].

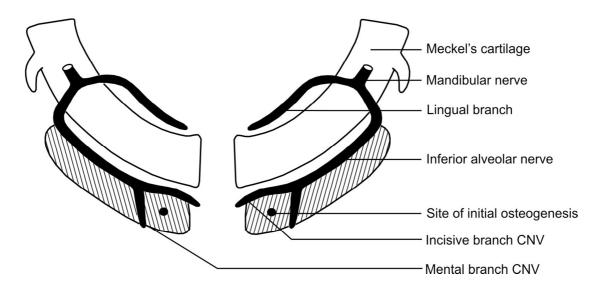


Figure 2.2: Site of initial osteogenesis related to mandible formation (redrawn from Ten Cate's Oral Histology 7th ed. pg 50)[20]

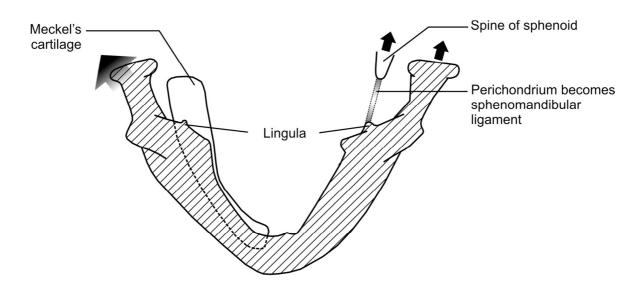


Figure 2.3: Spread of mandibular ossification away from the Meckel's cartilage at the lingula (redrawn from Ten Cate Oral Histology 7th ed. pg 51) [20]



Intramembranous ossification, which commences from the mesenchymal tissue, rapidly progresses both in an anterior and posterior direction on the mandible until it reaches the point where the mandibular nerve splits into its mental and incisive branches (Figures 2.2)[20]. The mesenchymal tissue cells are derived from the midbrain and hindbrain neural crest cells [19].

With growth of the mandible in the anterior, posterior, superior and inferior directions, a bony plate just lateral to the Meckel's cartilage is formed. Meckel's cartilage is a prominent rod that is derived from the neural crest cells of the first pharyngeal arch. This prominent rod serves as the cartilaginous scaffold from which the bone of the mandible is to develop. This is best illustrated in Figures 2.3 and 2.4, where the development of the lateral plate on the Meckels cartilage can be seen as well as the general direction of mandibular growth. The general pattern of intramembranous bone deposition, the relationship between the nerves, Meckels cartilage and the developing tooth germs is indicated in Figure 2.4. The arrowheads are used to illustrate future directions of bone growth in the neural canal and the lateral and medial alveolar plates [20].

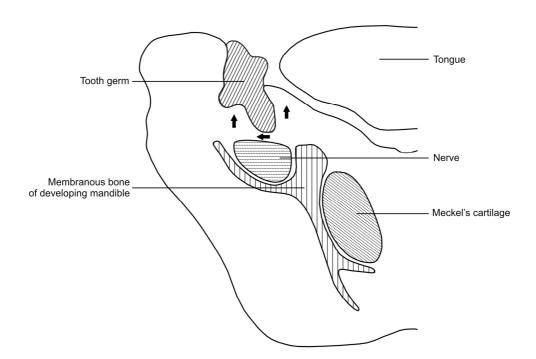


Figure 2.4: Schematic diagram of a coronal section through an embryo (redrawn from Ten Cate Oral Histology 7th ed. pg 51) [20]



The primordial area comprises of unpaired and paired structures, namely the maxillary processes and the mandibular prominences, which are components of the first pharyngeal arches. Secondary structures of the mandible such as the condylar and coronoid processes are formed from the invasion of mesodermal cells from the first two pharyngeal arches. The mandibular prominences enlarge in size until they converge in the midline of the lower jaw to form a point, or dimple, between gestational weeks 4 and 8, after which changes in the proportionality of the mandible continue after birth and into childhood [15,19].

During the 10th week of embryonic development, condensation of mesenchyme at the superior point of the mandible forms the future site of the mandibular condyles. Changes in morphological appearance and size of these condyles occur via appositional as well as interstitial growth [23,24]. In the 14th week, intramembranous bone forms in the temporal region such that at the 22nd gestational week the glenoid fossa and articular disc are created [25]. The mandibular condyles and glenoid fossa undergo morphological changes up to the 33rd gestational week. After birth, the morphology of the mandible continues to change and develop until late adolescence, or eruption of the third molars.

Facial structures, bony or cartilaginous, are influenced with undefined signaling between the pharyngeal endoderm and the neural crest precursors of the facial bones [19]. If development in these areas is abnormal, it will influence mandibular growth as well as maturation of the face and muscular attachments such as the tongue. Imbalances in retinoic acid and other growth related factors may lead to abnormal development.

Retinoic acid has a strong influence in the formation of facial structures in embryonic growth of the mandible. Bone morphogenic proteins (BMP) and retinoic acid determine the initial identity of the facial processes, followed by fibroblastic growth factors 2 and 4, which are responsible for the development of the mandibular



processes [19]. Retinoic acid also controls the development of the maxillary processes and other structures of the mid-face. This is most evident in the control of FGF-8 (fibroblastic growth factor 8) and shh (sonic Hedge Hog), which are responsible for the proliferation in ectomesenchyme of the frontonasal processes and also aids in the apoptosis mediated formation of the lateral region of the mandibular processes [19].

An imbalance of growth factors may lead to hypoplasia of the mandible, which is an anomaly that produces a short, small mandible and often leads to the retrodisplacement of the tongue. A mal-positioned tongue may cause the airways to become partially or completed obstructed, and could possibly lead to respiratory failure. In cases where hypoplasia of the mandible is coupled with a cleft palate, e.g. Pierre-Robin syndrome, the tongue may become impacted within this cleft and may bulge back into the nasopharyngeal cavity to form a plug, which cuts off airflow [26].

2.2. Anatomy of the infantile Mandible

The infantile mandible as in adulthood consists of a body and ramus. The mandibular ramus extends posteriorly in a fairly straight line such that the head of the condyle is at the same level as the superior surface of the body [15]. During post-natal growth, the mandibular condyle shifts downward and forward and away from the cranial base [25]. The articular surface of the mandibular condyle points posteriorly with the mandibular notch found between the condylar processes and the coronoid process.



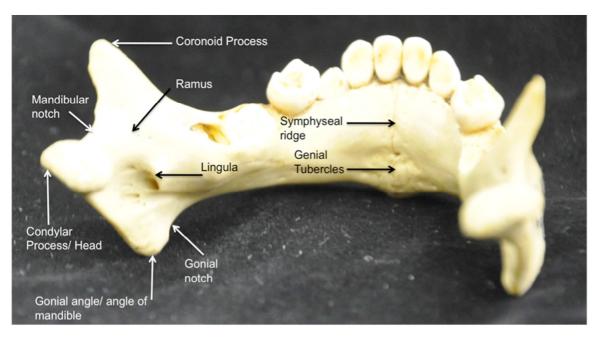


Figure 2.5: The lingual surface of a mandible with an approximate dental age of 1 year, Raymond A. Dart skeletal collection, University of Witwatersrand

In Figure 2.5, the anatomy of the lingual surface of the mandible can be seen. A prominent mandibular foramen, partially enclosed within a well-formed lingula, can be found on the surface of the bone (Figure 2.5). The mandibular foramen contains the mandibular division of the trigeminal nerve, which contributes to the ossification of the mandible (Figure 2.3). The internal surface of the mandibular body plays host to a variety of structures namely, the mylohyoid line on the infero-lateral aspect and the genial tubercles in the midline. The mylohyoid line serves as an attachment point for the mylohyoid muscle, which contributes to the floor of the mouth. The genial tubercles (mental spines) develop from mesenchyme in the midline of the mandible. After the first year of life, the ossicles fuse with the body of the mandible and become the bony genial tubercles [29]. The mental spines serve as attachment points for the genioglossus and geniohyoid muscles.

On occasion, accessory posterior foramina may be observed on the lingual surface of the mandible [30]. No specific role for these foramina has been determined except for additional innervation to the muscles of mastication [30]. Neural bundles originating from the inferior alveolar nerve as well as other superficial nerves of the



region of the neck region traverse through these foramina and are responsible for this innervation.

In Figure 2.6, the anatomy of the buccal surface of the mandible is presented. Anteriorly, the mandibular body has a flattened appearance and becomes rounder just above the base, which is approximately at the level of the crypts for the canines and first molars. The antero-lateral aspect of the mandibular body has various osteological features including: the symphysis menti or mental protuberance, alveolar ridge and mental foramen.

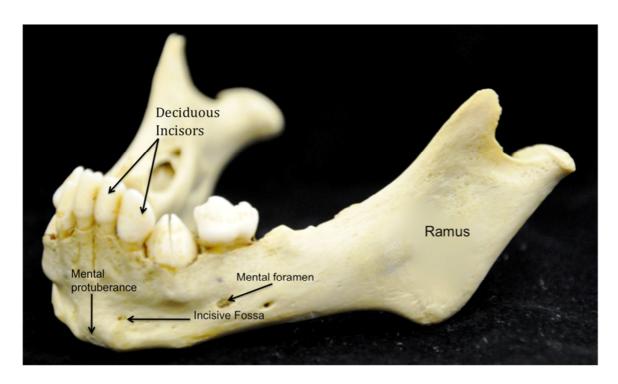


Figure 2.6: Anterior view of a mandible with an approximate dental age of 1 year, Raymond A. Dart skeletal collection, University of Witwatersrand



2.2.1. Growth of the Mandible

To maintain a functional relationship between all components of the mandible, as well as an increase in the length and width of the cranial base throughout growth, the mandibular dimensions develop accordingly.

During the foetal period there is an overall widening and lengthening of the mandibular body, which increases the bigonial width of the mandible [15]. This is accompanied by bone deposition on the posterior border of the ramus and bone reabsorption on the anterior border of the ramus [15]. This leads to the mandibular body forming a straight line between the body and the ramus during this growth period.

The prenatal widening of the mandibular body is further influenced after birth by the fusion of the hemi-mandibles. Scheuer and Black (2000) described the fusion of the hemi-mandibles as starting on the outer inferior surfaces and proceeding towards the inner superior surfaces [15]. They describe the growth of the mandible at the symphysis menti as limited during the foetal period, as the right and left hemi-mandibles only fuse within the first year of life [15]. Becker (1986) suggests a fusion time between 7 and 8 months post-natal. Generally most hemi-mandibles fuse by 6 months [15,21].

During growth, the position and direction of the opening of the mental foramen on the anterior surface of the mandibular body changes. At birth, the mental foramen points anteriorly, but as growth progresses the orientation of the foramen changes to a more posterior position with the neurovascular bundle occupying a posteriorly placed groove. With eruption of the lower incisors, the shape of the menton changes from a ridge to a more triangular shape (Figure 2.6). The mental foramen lies below a line connecting the canine and the first deciduous molar until 36 months.



After birth the mandible presents with the greatest change in shape and size and is in synchronization with the development and eruption of the deciduous dentition, as well as changes in the size and shape of the maxilla [15]. This is necessary for the maintenance of a harmonious occlusal pattern with development. The prenatal bone deposition and reabsorption on the mandibular ramus coupled with the postnatal increase in the body length influences the angle of the mandible. At birth the gonial angle ranges from $135^{0} - 150^{0}$. This angle changes with the eruption of deciduous dentition to 130^{0} - 140^{0} and further to $120^{0} - 130^{0}$ after the eruption of the second molar [15,21]. As the mandibular angle decreases, the height of the ramus increases and results in the condyle being at a higher level than the occlusal plane of the teeth.

Normal mandibular growth is also affected by changes in the tongue as well as tooth development and eruption. Zi-Jun Liu (2008) studied the effects of tongue volume reduction on craniofacial growth with particular emphasis on the orofacial skeleton and dental arches [27]. He stated, "it has been generally accepted that volume increases in soft tissue induces osteogenesis at the growth site of bone". Thus tongue growth and size influence mid-facial growth mechanisms, which determine the growth of surrounding orofacial elements. The study also demonstrated that tongue volume had a measurable effect on jaw growth at a certain point in time [27]. Reducing tongue volume during periods of fast growth should thus lead to consequential alterations in craniofacial skeletal growth and dental arch formation. Although this is not always the case, tongue volume is usually proportional to mandible size.

The influence of tongue volume on the dental arch formation is also accompanied by the development and eruption of the dentition. Aka Sema (2008) described the developmental chronology of the deciduous primary teeth as following a set sequence of events [28]. The formation of hard tissue calcification is described as commencing between 12 and 24 weeks [1,28]. Formation of enamel follows and is usually completed during the first 11 months after birth. Tooth eruption commences 6 to 8 months post birth and may continue up to 36 months [1,15,21,28].



2.3. Morphological estimation of sex in juvenile and adult mandibles

2.3.1. Juveniles (0 to 19 years)

On account of an absence of secondary sex features, and the hormones which accompany them, the estimation of sex in foetuses, infants and early childhood has been shown to be difficult to replicate and generally inaccurate [31]. Morphological features that have been examined in juveniles include gonial eversion and the inferior symphyseal border of the mandible.

2.3.1.1. Gonial eversion

Gonial eversion and/or bilateral eversion may be present at birth. In a study of juveniles (6 months to 6 years), Loth (1995) considered the presence of gonial eversion on the mandible to be a male trait, whereas the absence of gonial eversion was a female trait [31].

The sample was divided into 2 main categories, namely 0-5 years and 6-19 years [31]. In the 0-5 year age group the accuracy of sex estimation was 18% for males and 50% for females, with an overall accuracy of 37%. Similarly, in the 6-19 years age group, accuracy was 21% in males and 46% in females, with an overall accuracy of 29% [31]. Due to poor accuracy, gonial eversion was thus considered to be a poor indicator of sex and may rather be a better reflector of the inherited genetic potential of the individual [31]. Oettlé et al (2009) confirmed this impression by finding that gonial eversion, as an indicator of sex, to also be unreliable [3].

2.3.1.2. Inferior symphyseal border of the mandible

The inferior symphyseal border morphology was examined in a juvenile and adult sample [31]. Loth (1995) classified males as exhibiting a straight or undercut shape, whereas females had rounded or pointed shapes [31]. The accuracy when using this method in adults ranged from 65% (males 64%, females 66%) in U.S. whites to 75% (males 74%, females 75%) in North African blacks.



When assessing the inferior symphyseal border shape on a juvenile sample, it was noted that the adult chin shape was observed in older juveniles (6 to 19 years), with an accuracy of 67% in males and 73% in females [31]. Younger juveniles (0 to 6 years) demonstrated an accuracy of 81%, with differences in the shape of the inferior symphyseal border being noted as early as seven months [16]. Loth and Henneberg (2001) attributed higher accuracy in earlier stages of development to the accommodation of the developing and erupting dentition [16].

Using geometric morphometrics, Franklin and colleagues (2007) assessed the shape of the inferior mandibular border on white and black sub-adults from South Africa (1 to 17 years) [18]. Males classified with 55% accuracy, while females were 65% [18]. This is at least 30% lower than the qualitative results of Loth and Henneberg (2001). Similarly, Scheuer (2002), using qualitative methodology proposed by Loth and Henneberg (2001), produced classification accuracies similar to those of Franklin and colleagues (2007). Therefore, the inferior symphyseal surface of the mandible is not useful in distinguishing between the sexes in sub-adults.

2.3.2. Adults

More reliable methods of estimating sex has been noted in adults, morphological features include rameal flexion, gonial eversion, the shape of the mandibular symphysis (menton), and the angle of the mandible [3,31-37]. As expected, accuracy and reliability of these methods are affected both by antemortem tooth loss and the advancement of age.

The shape of the mandibular ramus may be useful in sex estimation, but the research on this morphological trait is contradictory. The reason for evaluating rameal shape is because its appearance is dependent on the muscles of mastication and not the size of the individual [31]. Likewise de Villiers (1968) noted that the shape of the ramus is more sexually dimorphic when compared to the body of the



mandible [31,38]. Hunter and Garn (1972) reinforce this idea when they demonstrated that the ramus of the mandible was 14% longer in males [31].

Koski (1996) cautioned against the use of the mandibular ramus flexure as a single indicator of sexual dimorphism in a skeleton [36]. He stated that flexure in the ramus could occur in anyone, with no distinct differences between the sexes. Koski (1996), also mentioned that ancestry had an influence on the occurrence of presence of absence of rameal flexures [36].

Kemkes-Grottenthaler and colleagues (2002) described the loss of teeth and the advancement of age as factors greatly reducing the classification accuracy of rameal flexion. They concluded that asymmetry in growth and tooth eruption had a substantial effect on the usefulness of this method [34]. They suggested that rameal flexure, was not a profound sexually dimorphic trait, but a feature that was influenced by localized functional adaptations [34]. They concluded that the severity of the misclassification of sex encountered using the analysis of the rameal flexure could be due to more complex phenomena and even due to functional influences such as mandibular morphology and masticatory stresses [34]. Kemkes-Grottenthaler (2002) indicated that the use of the rameal flexion as an indicator of sex resulted in only 66% of males and 32% of females being identified with an overall accuracy of only 59% [34].

Gonial eversion - as a trait used to estimate sex - has received far more attention than rameal flexion as an indicator of sex. Using this trait, Loth (1995) found that only 44% of North Africans and 58% of U.S. whites, and 56% of South African groups could be correctly classified based on the presence or absence of an everted gonial region. With tooth loss, accuracy decreased. Kemkes-Grottenthaler and colleagues (2002) found a classification accuracy of 75% in males, versus 45% in females; overall accuracy was 70% [34].



When using geometric morphometrics, Oettlé and colleagues (2007) noted higher classification accuracies in black South African males (74%) and females (71%). However, the authors considered that the degree of overlap between the two groups was too great to accurately ascertain sex with confidence in an individual forensic case [3].

The general consensus for the use of rameal flexure and gonial eversion as indicators of sex is that they should be used in conjunction with other morphological and osteometric methods [31,34]. When all variables are considered namely ramus height, symphysis height, coronoid height, bi-gonial breadth, bi-condylar breadth, symphysis breadth, bi-coronoid breadth, corpus length and maximum mandibular length, the sexes can be sorted with a classification accuracy of 84%.

Similar to juveniles, the shape of the mandibular symphysis has also been used to evaluate sex in adults. De Villiers (1968) assessed the shape of the menton based on criteria proposed by Keiters (1928, 1935) and Schultz (1933) [38]. Using these criteria, the mandibular symphysis could be round, square or pointed [38]. This shape was determined based on the relative development of the mental protuberance and mental tubercles of the trigonum mentale and of the fossa mentalis.

When these morphological indicators were applied to a black South African sample, little sexual dimorphism was observed [38]. Moderate growth of the mental protuberance indicated a general accuracy of 42% between sexes. Moderate growth of the tubercles, was found in 38% of females and only 32% of males. Sexual dimorphism was found to be sizeable when the incidence of slight protuberances (8% males) and marked tubercles (16% males) were investigated [38].

The mandibular angle has also been described as a morphological indicator of sex. At birth the mandibular angle is obtuse, with angles greater than 135°. With the onset of various growth imposed changes i.e. dental eruption, the angle becomes more



acute. These changes are further accentuated with the onset of puberty where hormonal influences play a role and often accelerate the changes in the angle [37].

De Villiers (1968) found that the mandibular angle ranged from 103° - 135° (121° mean) in South African males and 115° - 138° with a mean value of 125° in South African females [38]. Ascadi and Nemeskeri (1970) confirmed this finding by observing that the mandibular angle in females is rounded and exhibits a mean angle of 125° [37].

However Oettlé et al (2009) established that the mandibular angle was influenced by tooth loss [3]. They evaluated changes in the mandibular angle with loss to complete absence of molar teeth. They concluded that the loss of dentition, especially if complete or uneven, influenced the mandibular angle. Due to the large overlap between groups, it was not possible to predict an individual's sex based on the gonial angle [37]. Similarly Ayoub et al (2009) found no sexual dimorphism with the mandibular angle [40].

While the mandible can present with relatively accurate means of sex determination in the adult, it appears to be population specific. The estimation of sex from immature mandibles has been shown to be less accurate than that of the adults. This may be due to hormonal influences present in the adult and juvenile. However it has not yet been accurately ascertained as to whether sexual dimorphism exists in the early years of life (foetal to 3 years). Environmental conditions and genetic potential may also influence the accuracy of sex estimation in the juvenile/adult mandible.



CHAPTER 3

MATERIALS AND METHODS

3.1. Materials

Both cadaveric and skeletal tissues were sourced from the Raymond A. Dart Collection at the University of Witwatersrand, South Africa, while only cadaveric remains were obtained from the Department of Anatomy, University of Pretoria, South Africa [41]. A total of 74 mandibles were used and are presented in Table 3.1. As can be seen, the majority (n=50) were obtained from the School of Anatomical Sciences at the University of Witwatersrand, while 24 were from the University of Pretoria. Sex and ancestry was known.

The skeletal component of the sample (n=22) had an estimated age at death. This was previously determined using the dental eruption and morphological features of each mandible and was used to place the skeletal elements into an age of 0, 1, 2 and 3 years [15]. No skeletal foetal specimens were available.

Age at death for the cadaver specimens was unknown. Standard calculations, which used crown-rump length (CRL) + 6, resulted in the gestational age being reported in weeks and the crown-rump length in centimeters [4,43]. The calculation used was recommended for aging of foetal specimens up to 40 weeks. Eighteen specimens had an estimated age between 30 and 40 gestational weeks. A further 34 specimens had an estimated age between 0 and 3 weeks postnatal, their age was estimated based on the presence or absence of the umbilical cord. In cases where the umbilical cord was absent, the presence of symphyseal cartilage and the degree to which mandibular fusion had progressed was used to estimate the approximate age. The sample was divided into 4 groups. Group 1: 18 specimens (30 to 40 weeks); Group 2: 41 specimens (0 to 11 months), Group 3: 8 specimens (12 to 24 months) and Group 4: 7 specimens (24 to 36 months).



The Raymond A. Dart collection was created in 1921, after Professor Dart had visited an anatomical skeletal collection at an American medical school in St. Louis, Missouri [41]. The collection is primarily comprised of dissection room cadavers from South Africa, to date; approximately 2559 skeletons are available for study [41,42]. The collection from the start of its inception in 1921 has incorporated individuals from various socioeconomic backgrounds.

Subsequently studies analyzing growth should consider the possibility of secular trends influencing the data set [44]. The factors influenced by secular trends in the age range assessed may include the birth weight, head breadth and circumference and the growth rate for many individual body measurements [44]. This may be attributed to changes in diet over time i.e. nutrition of infants in 1921 may not have been to the same degree as 2010. Therefore the growth of the mandible and possible stresses placed on the mandible may have changed considerably over an 80-90 year period. The socioeconomic status of the individual may also have been an influencing factor particularly in cases where medical treatment may have been required and subsequently in cases of higher socioeconomic status the access to advanced medical treatment would have influenced the health and the growth of the individual [41,42]. These factors were all considered in the interpretation of the results obtained in this study.

Skeletal materials within the Raymond A. Dart Collection were obtained under the Anatomy Act (No. 20 of 1959), and are currently sourced under South Africa's National Health Act of 2003 [42]. In the initial years of collection, the main source of human material was from unclaimed, but not unknown bodies, which were received from the historical Transvaal provincial hospitals [42]. After 1958, an increase in bequeathments was observed such that in 1992, the number of willful donations exceeded that of the unclaimed remains. However, in the 21st century, bequeathals have been found to be on the decline [42]. The source of the majority of cadaver material, which is housed in various academic institutions in Gauteng are from the local provincial hospitals and contain both bequeathments and unclaimed remains.



The foetal and neonate cadavers received at the University of Pretoria were obtained in a similar manner to those at the University of the Witwatersrand.

Table 3.1. Sample composition collected from the University of Witwatersrand and University of Pretoria

		ty of Witwatersrand ol of Anatomical Sciences Raymond A. Dart Skeletal Collection	University of Pretoria Department of Anatomy Cadaver Material
Black Males	16	11	10
Black Females	9	7	7
White Males	1	0	5
White Females	1	1	2
Other Males	n/a	2	n/a
Other Females	n/a	4	n/a
Total Males	16	13	15
Total Females	10	12	9

Subsequently due to the various socioeconomic backgrounds incorporated into the different collections used in this study, the assessment of the mandible in terms of ancestral traits was not realistic. The sample's population distribution included Black South Africans (80%), with white South Africans and other (coloured and asian) groups representing 13.5% and 6.7%, respectively. The black sample included members from the Sotho, Malawian, Shangaan, Zulu, Swazi and Xhosa populations,



whereas white South Africans groups are of European descent, which mainly originate from Germany, France, United Kingdom, Portugal and Netherlands [45].

3.2. Methods

3.2.1. Dissection

Soft tissue dissections were performed using a standard dissection kit, which included a scalpel with removable blades, sharp nose forceps and blunt nose forceps. Soft tissue was removed so as to expose the antero-lateral surfaces as well as the inferior border of the mandible.

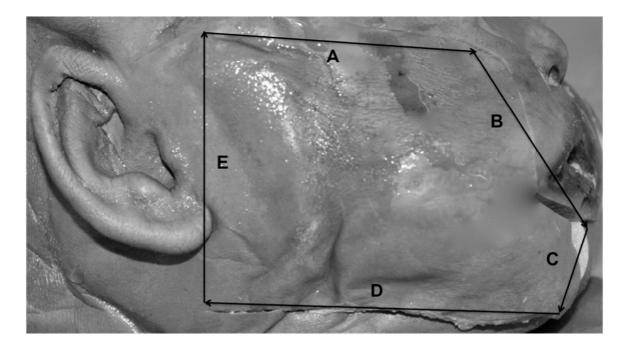


Figure 3.1: Planned incision lines A-E for dissection of a neonate (stillborn), lateral view, School of Anatomical Sciences, University of Witwatersrand

The following incisions were made (Figure 3.1):

Line A. From the zygomatic process of the temporal bone, along the zygomatic arch towards the temporal process of the zygomatic bone.



Line B. From the temporal process of the zygomatic bone towards the superior border of symphysis menti.

Line C. From the superior border of the symphysis menti towards the inferior border of symphysis menti.

Line D. From the inferior border of symphysis menti towards the posterior border of the mandibular angle.

Line E. From the posterior border of mandibular angle towards the zygomatic process of the temporal bone

All musculature and neurovascular structures occurring on the antero-lateral surfaces of the mandible were reflected as one layer. This was done as a means to preserve the overlying soft tissue and to fully expose the bony landmarks.



Figure 3.2: Standard measurement position used to digitize mandibles (symphysis menti superior, left mandibular condyle: bottom right corner, right mandibular condyle: centre)



3.2.2. Collection of landmark data

An Immersion Microscribe G2 with an accuracy of 0.38mm was used to record 21 landmarks on the mandible. These landmarks included both the points necessary to calculate the linear measurements and to define the shape of the mandible. The data was imported into Microsoft Excel™. Each mandible was positioned with the mandibular condyles inferiorly and the symphysis menti superiorly (see Figure 3.2). Landmarks were recorded in a pre-set sequence, as illustrated in Figure 3.3.

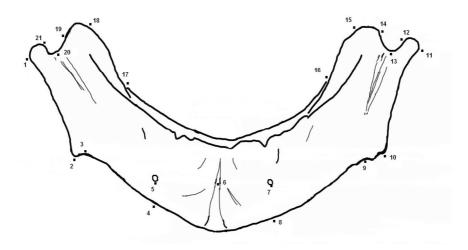


Figure 3.3: An illustration of the 21 homologous (sp) landmarks recorded on each mandible corresponding to the landmarks defined in Table 3.2.

Table 3.2. A description of the homologous landmarks on the mandible

Point	Description
1	Most posterior point on the right condylar process
2	Right gonial angle
3	Gonial notch (right)
4	Inferior border of mandible directly below the right mental foramen
5	Mental foramen (right)



6	Symphysis menti / symphyseal ridge, point horizontally adjacent to the
	mental foramina
7	Mental foramen (left)
8	Inferior border of the mandible directly below the left mental foramen
9	Gonial notch (left)
10	Left gonial angle
11	Most posterior point on the left condylar process
12	Most anterior point on the left condylar process
13	Deepest point on the mandibular notch on the left
14	Most posterior point on the left coronoid process
15	Most anterior point on the left coronoid process
16	Left oblique line termination/point on the oblique line where the body
	and ramus of the mandible meet
17	Right oblique line termination/point on the oblique line where the body
	and ramus of the mandible meet
18	Most anterior point on the right coronoid process
19	Most posterior point on the right coronoid process
20	Deepest point on the mandibular notch on the right
21	Most anterior point on the right condylar process
L	

With each landmark, three coordinates (X, Y, Z) were recorded and represent the horizontal, vertical and depth planes. The 3-dimensional co-ordinates were imported into a Microsoft Excel worksheet and the Pythagorean formula was used to converted the data into linear distances:

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$$= \sqrt{(x_1-x_2)^2 + (y_1-y_2)^2 + (z_1-z_2)^2}$$

where x_1 , y_1 , z_1 and x_2 , y_2 , z_2 refer to the coordinates of the first and second landmarks respectively.

3.2.3. Osteometric analysis

Nine standard anthropometric measurements as adapted from Hesby et al. (2006) and Scheuer & Black (2000), were recorded, as well as the mental angle measurement, which was designed for the purpose of this study [15, 5].

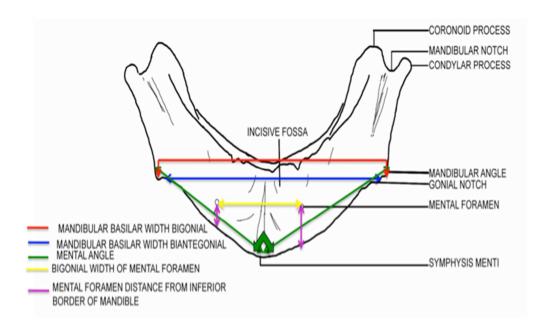
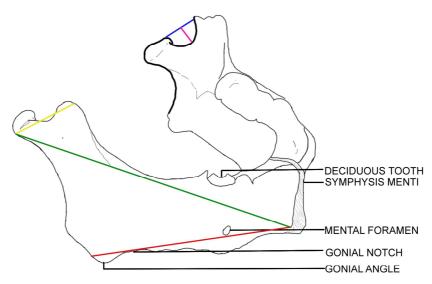


Figure 3.4: Foetal/infant mandible: anterior





- GREATEST DEPTH OF MANDIBULAR NOTCH
- WIDTH OF MANDIBULAR NOTCH
- LONGESTLENGTH OF MANDIBLE
- MANDIBULAR WIDTH
- MANDIBULAR BODY LENGTH

Figure 3.5: Foetal/infant mandible: lateral

1. Mental angle; Figure 3.4 (green)

The angle formed at the mental symphysis between the mandibular rami of both sides of the mandible.

2. Mandibular basilar width bigonial [5]; Figure 3.4 (red)

The width of the mandibular arch measured from the bilateral angles of the mandible (point where the posterior and inferior borders meet).

3. Mandibular basilar width bi-antegonial [5]; Figure 3.4 (blue)

The width of the mandible measured from the bilateral points at the most superior point on the antegonial notch.



4. Bigonial width of mental foramen [38]; Figure 3.4 (yellow)

Distance measured between the two mental foramina on the anterior surface of the mandible.

5. Mental foramen distance from inferior border of the mandible [38]; Figure 3.4 (purple)

Distance measured from the mental foramen to the inferior border of the mandible directly below it.

6. Greatest depth of the mandibular notch [38]; Figure 3.5 (purple)

The line bisecting the width of the mandibular notch at its highest point to the deepest point of the mandibular notch.

7. Width of the mandibular notch [38]; Figure 3.5 (blue)

The measurement taken from the inner surfaces of the mandibular condyle to the coronoid process.

8. Mandibular width [15]; Figure 3.5 (yellow)

The measurement taken from the most posterior tip of the mandibular condyle to the most anterior tip of the coronoid process, i.e. the widest part of the ramus between the coronoid and condylar processes of the mandible

9. Mandibular body length [15]; Figure 3.5 (red)

Distance measured from the mental tubercle of the mandible to the angle of the mandible. In cases where the two halves of the mandible had not fused the mental symphysis was then used as a reference point.



10. Longest length of the mandible [15]; Figure 3.5 (green)

The distance from the mental tubercle to the posterior condylar tip of the mandible.

The measurements used to calculate the mental angle include: the mandibular body length of each side (a+b) as well as the mandibular basilar width bigonial (c). The cosine rule was used for calculations, as the triangle formed by the measurements was not a right-angled triangle; thus, the Pythagorean equation could not be used.

The mental angle was calculated as follows:

$$CosC = (a^2 + b^2 - c^2)/(2ab)$$

a and b represent the left and right mandibular body lengths respectively; c represents the mandibular basilar width bigonial and C represents the calculated mental angle.

The mandibular notch depth was calculated using the Pythagorean equation. The calculation required information obtained from two (2) additional calculations using combinations of landmark points 12, 13, 14, 19, 20 and 21 (see Figure 3.3):

$$A=\sqrt{(C^2-B^2)}$$

A: Mandibular notch depth calculation



B: Calculation no.1

Halfway between the posterior landmark on the coronoid process (19R/14L) and the anterior landmark on the condylar process (21R/12L)

C: Calculation no.2

Distance measured from the anterior landmark of the condylar process (21R/12L) to the greatest depth of the mandibular notch (20R/13L).

3.2.4. Statistical analyses

Statistical analyses were conducted using PAST statistical software.

3.2.5. Geometric morphometric analysis

To analyze the landmarks in terms of shape, data was imported into Morphologika2 v2.5. A generalized full procrustes analysis was performed, which eliminated the influence of size as well as rotation and translation of the mandible. With a generalized full procrustes analysis, the landmarks were superimposed and compared with each other. In doing so, shape variation between the mandibles could be assessed (Figure 3.6). The reflections enabled option was selected, the function of this option is to superimpose landmark configurations of individual mandibles by minimizing the difference between each mandible. If the difference between mandibles can be made smaller by reflecting the sides of the mandible (right becomes left and left becomes right) then the generalized procrustes will do this.

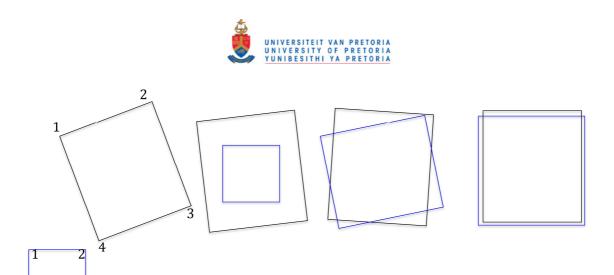


Figure 3.6: Generalized full procrustes analysis (adapted from Evolutionary and Developmental Morphometrics of the Hominoid Cranium, Mitterocker P pg 29)[46]

Centered and

scaled

configurations

Centered, scaled,

and rotated

configurations

Centered

Configurations

In order to perform this analysis, the function required for translation, scaling and rotation, was enabled to remove the information not related to geometric shape (Figure 3.6). Procrustes superimposition renders the data inappropriate for the application of standard linear statistical methods hence the configurations need to be projected orthogonally to the tangent space (see Rohlf, 1999 Journal of Classification 16:197-223). Principal component analysis was used to summarize the data for exploratory analysis (simplifies the data into it's most basic form).

A principle component analysis (PCA) was used to enable the points that had been previously plotted on the tangent space to be adjusted such that variation in shape of the sample could be interpreted. Data were organized and allocated colours and symbols based on the age group and sex and depicted graphically on a PCA graph.

3.2.6. Osteometric statistical analysis

4

Raw

Configurations

In order to assess the relationship between the linear measurements with independent variables such as age at death and sex, the data was evaluated in terms of distribution, i.e. normally distributed or skewed either to the left or to the



right. A Shapiro-Wilk test was performed to assess for normality. Due to the left skewed nature of the data, the Kruskal-Wallis test was used [47].

3.2.7. Inter-observer error

Ten specimens were evaluated by an independent observer and re-evaluated by the primary investigator. Inter and intra-observer errors were tested using the Lin's concordance correlation, which provides information on the level of repeatability of the method and the degree of association between measurements. When a coefficient of 1 was obtained, whether it was a positive or a negative value, it indicated perfect or complete correlation. Values between 0.75 and 0.99 were indicative of a high degree of correlation (negative or positive values applied). When a value range fell within the 0.5 and 0.74 range it indicated a moderate degree of correlation. If a value fell within the 0.25 to 0.49 ranges, the correlation was considered to be low [48].



CHAPTER 4

RESULTS

The size and shape of mandibles from prenatal (30 to 40 gestational weeks) and postnatal (0 to 11 months; 12 to 24 months; 25 to 36 months) groups were recorded so as to establish mean values for normal mandibular growth. These aims were achieved through the analysis of linear measurements as well as geometric morphometrics. Differences between the sexes for the various age at death categories were also examined. The broad application of this data is for the possible detection of growth related abnormalities, such as micrognathia and Hallermann-Streiff, in living foetuses in South Africa.

4.1. Osteometry

In order to assess the relationship of the osteometric variables with age at death and sex, the data was evaluated for normality. The data set was found to be left skewed due to a large number of specimens in the prenatal and one year old categories (Groups 1 and 2, n=59) when compared to the later stages, namely 12 to 36 months (Groups 3 and 4, n=15). On account of this, the Kruskal-Wallis test was used to evaluate statistical significance amongst the four groups.

4.1.1. Comparisons between osteometric variables and age at death

Descriptive statistics for each of the four groups is present in Table 1 (Appendix A pg. 67). As expected, a general increase in size was observed for all osteometric dimensions from Groups 1 to 4. No statistically significant differences were noted between the left and right sides. Therefore, only the mean values of the left side were used.



When the Kruskal-Wallis test was performed, statistically significant differences were noted between the various groups, the values of which are presented in Table 2 (Appendix A pg. 68). As expected, a comparison between group 1 (30 to 40 weeks) and group 4 (24 to 36 months) showed a statistically significant increase for all dimensions. Likewise, statistically significant differences were observed in the linear dimensions between the foetal (group1: 30 to 40 weeks) and across the post-natal groups (groups 2 to 4: 0 to 36 months). These results can be attributed to the large age gap between the groups, which led to a difference in overall size. For the most part, a comparison across the youngest and oldest groups was not useful, as no clear inferences could be made concerning the manner in which the mandible increased in size. Rather, it was necessary to evaluate each group in succession, for example, group 1 to 2, 2 to 3, and 3 to 4. The mandible was divided into three planes: medio-lateral, superior-inferior and anterior-posterior planes for further analysis and interpretation

4.1.1.1. Medio-lateral plane

In the medio-lateral plane, statistically significant increases in size were observed in the mental angle, the mandibular basilar widths bigonial and bi-antegonal, and the bigonial width of the mental foramen for most, if not all, of the groups compared. An increase in these dimensions can be used to suggest that the mandible widens from the late foetal period to 3 years of age.



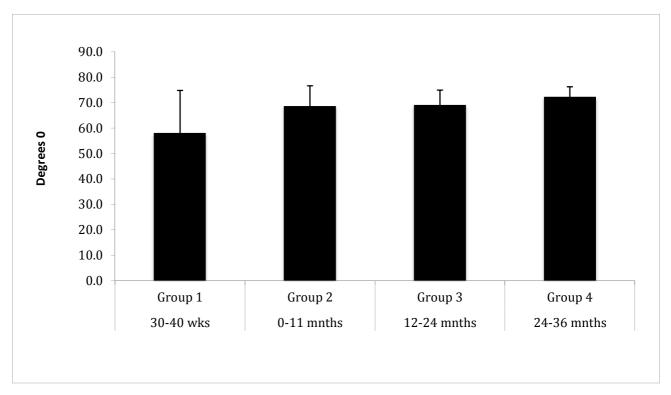


Figure 4.1. Mean and standard deviation values for the mental angle of groups 1-4

In Figure 4.1. an increase in the mental angle (MA) from age groups one to four are shown. In group one, the mean value for the mental angle was 58.16° , whereas in group 2, this increased to 68.56° . This represents an increase of 10.4° (18%) within the first year of life and was statistically significant (p<0.05). After 12 months, the size of the mental angle did not change and ranged from 69° to 72° . The discrepancy between the first year of life and later years may be attributed to the fact that the mandibular symphyses fuse between 4 and 6 months, postnatal.



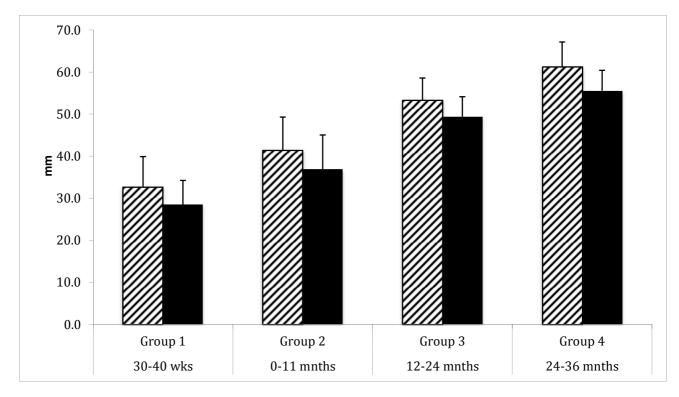


Figure 4.2. The mean and standard deviation for the mandibular basilar width bigonial (pattern bar) and mandibular basilar width bi-antegonial (solid bar) for groups 1-4.

In Figure 4.2, a statistically significant increase in size was illustrated from the late foetal stage to 3 years of age (Figure 4.2). The mandibular basilar width bigonial (MBS_BG) was taken from gonion to gonion; whereas the mandibular basilar width biantegonial (MBS_BAG) was used to determine the distance between the points above the gonial notches. All differences between the groups were statistically significant, with the exception of groups 3 and 4 (12 to 36 months).



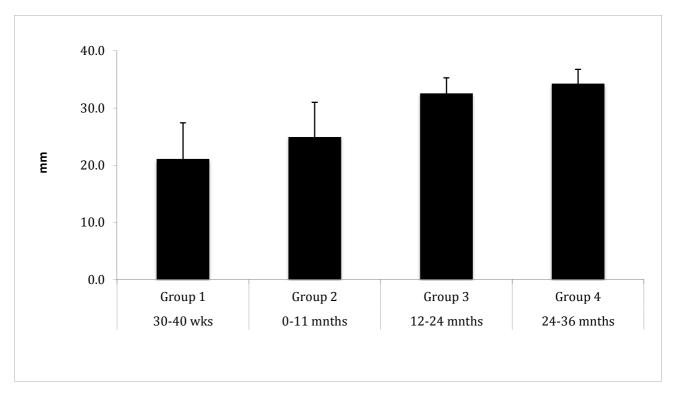


Figure 4.3. Mean and standard deviation of the bigonial width of the mental foramen for groups 1-4

In Figure 4.3, differences in size for the bigonial width of the mental foramina (BW_MF) from group 1 to 4 are presented. A small increase was noted between foetal (21.09 mm) and the early postnatal group (24.90 mm), but this difference was not statistically significant. Likewise, no distinct change was found between the last two postnatal groups.

However, a statistically significant change with age was noted between group 2 (24.90 mm) and group 3 (32.51 mm). Therefore, within ages 1 and 2 (12 to 24 months), the posterior mandible is widening and this may be due to the development of the tongue.



4.1.1.2. Superior-inferior plane

In the superior-inferior plane, an increase in the distance of the mental foramen to the inferior border of the mandible was found along with a statistically significant increase in the depth of the mandibular notch.

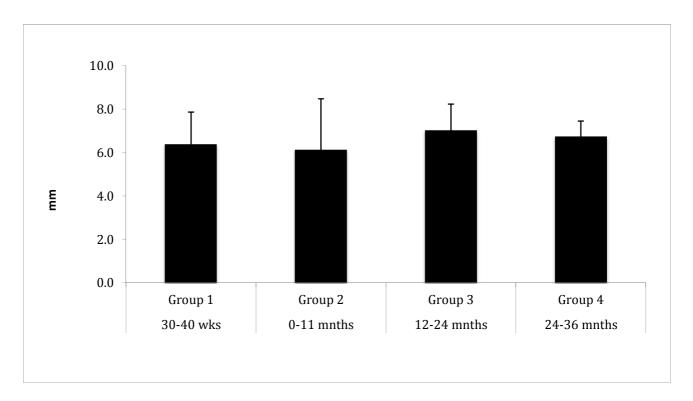


Figure 4.4. Mean and standard deviation of the distance between the mental foramen to the inferior border of the mandible on the left for groups 1-4

For the distance of the mental foramina to the inferior border of the mandible, no increase in the deposition of bone in this region was observed from the prenatal to postnatal period (see Figure 4.4.). This is in contrast to the above-mentioned medio-lateral dimensions from which the mandible was shown to widen with the advancement of age.



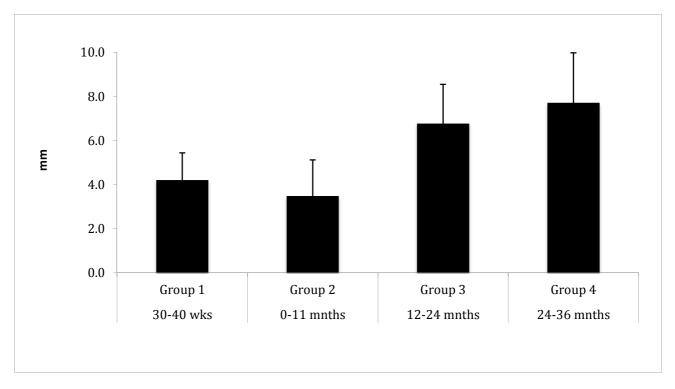


Figure 4.5. Mean and standard deviation of the mandibular notch depth (MND) for the left side for groups 1-4

In Figure 4.5, results for the mandibular notch depth for groups 1 to 4 are shown. A statistically significant difference (p<0.001) was found between groups 2 (3.47mm) and 3 (6.77mm) (Figure 4.5). This increase, between 12 and 36 months of age, may be attributed to the development of the masticatory muscles as well as an increase dependence on mastication.

4.1.1.3. Anterior-posterior planes

In the anterior-posterior planes, statistically significant increases in size were only observed between birth and 2 years of age for the mandibular notch depth and width, the length of the mandibular body, and and the longest length of the mandible (see Figures 4.6 to 4.9). These changes may be attributed to the development and eruption of the decidious dentition as well as an increased reliance on mastication during this period.



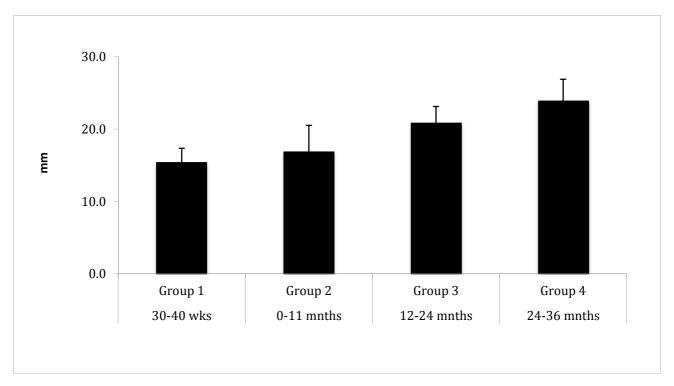


Figure 4.6. Mean and standard deviation of the mandibular width on the left side for groups 1-4

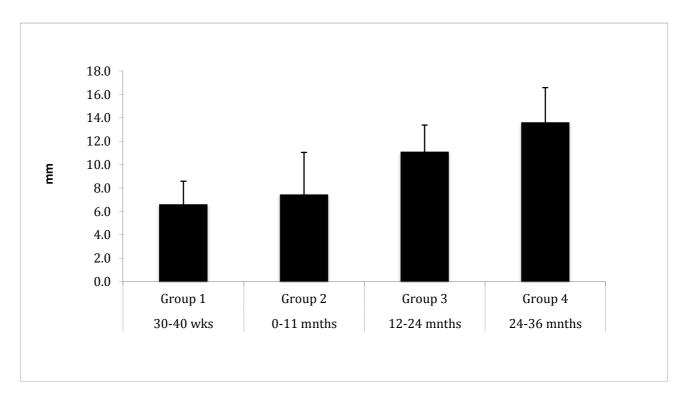


Figure 4.7. Mean and standard deviation of the mandibular notch width on the left side for groups 1-4



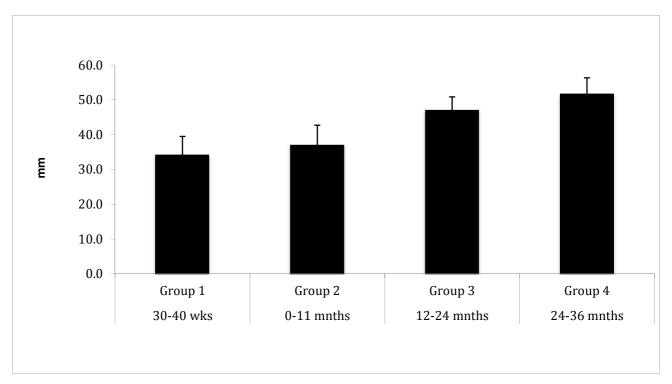


Figure 4.8. Mean and standard deviation for the mandibular body length on the left side for groups 1-4

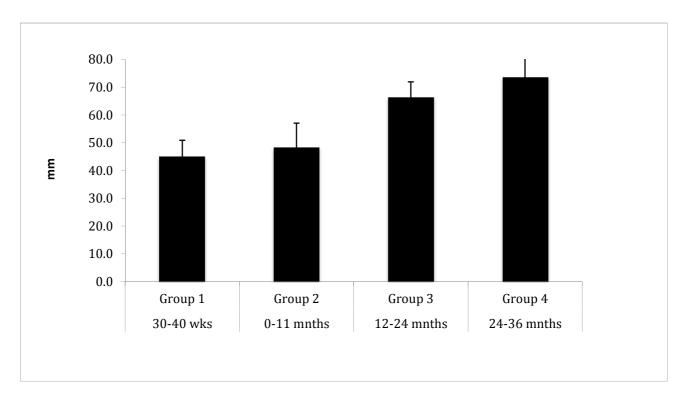


Figure 4.9. Mean and standard deviation of the longest length of the mandible on the left sides for groups 1-4



4.1.2. Comparison between osteometric variables and sex

A Student's t-test and an F-statistic test were used to evaluate statistically significant differences between the sexes in each age group and for each of the ten measurements. The results of these tests can be found in Table 2 (Appendix B pg 71). No statistically significant differences were found between the sexes for the prenatal group (30 to 40 gestational weeks) as well as from birth to 2 years of age (groups 2 and 3). After two years, a statistically significant difference was found between the sexes for the mental angle (see Figure 4.10) and the depth of the mandibular notch (see Figure 4.11). Possible reasons for these differences include unequal age at death distributions within the sample.

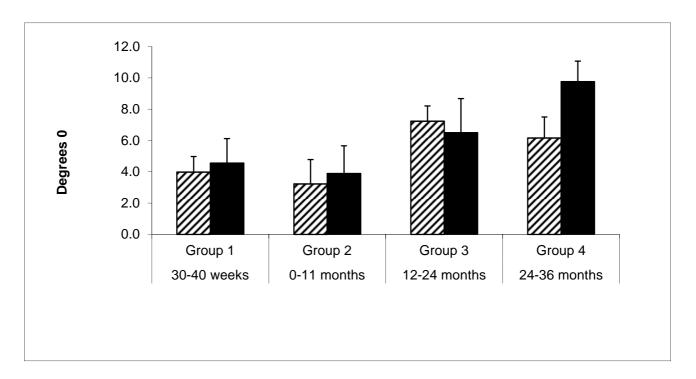


Figure 4.10. Mean and standard deviation of the mental angle for males (n=43) (pattern bar) and females (n=31) (solid bar) for groups 1-4



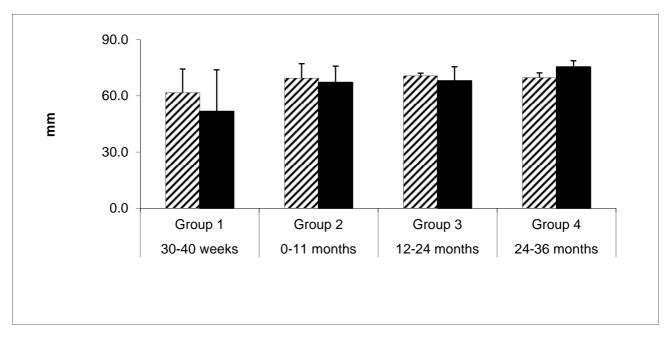


Figure 4.11. Mean and standard deviation for the mandibular notch depth on the left side for males (n=43) (pattern bar) and females (n=31) (solid bar) for groups 1-4

4.2 Geometric morphometrics

Morphological changes associated with an increase in age were assessed using geometric morphometrics. Differences in the morphological appearance of males and females were assessed separately, so as to elucidate shape differences, if any, between the sexes.

Changes in mandibular shape were expressed in terms of their distribution along principle component axes 1 and 2, where the most variation was indicated. The distributions of the various groups were plotted along the PC1 (x-axis) and PC2 axes (y-axis) and visually demonstrated using wireframe images enhanced with thin-plate splines. These visualization techniques (thin-plate splines) were facilitated using Morphologika2 v2.5. Wireframes and thin-plate splines emphasized changes in mandibular morphology through deformations of the mean morphological shape, which was located at the intersection of the PC1 and PC2 axes.



4.2.1. Age related morphological changes

The distribution of variance along the PC1 and PC2 axes when comparing the four groups is shown in Figure 4.13. Principle component 1 (PC1) accounted for the majority of variance found in the sample (scores range from -0.36 to +0.32) as indicated by 86%. PC2 only minimally influenced the variance in the sample (scores range from -0.08 to +0.08) as indicated by 14%.

The linear relationship between PC1 and size is indicated in Figure 4.12, and an increase in length is indicated as PC1 extends from the extreme positive toward the extreme negative specimens. The various measurements assessed and their relationship with both PC1 and PC2 were analyzed from the extreme positive to the extreme negative specimens as indicated in Figures 4.13 and 4.14. There was no relationship found between PC2 and size.

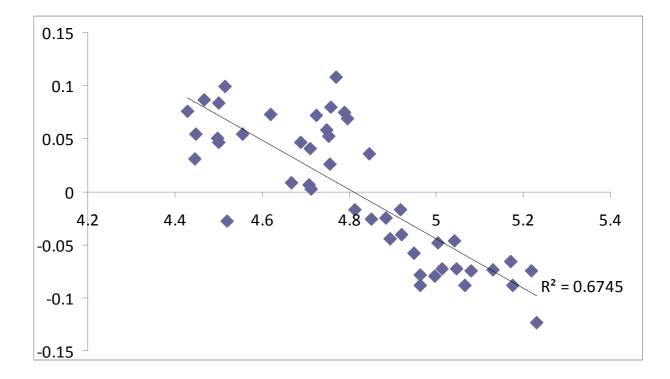


Figure 4.12. The relationship between size and principle component 1 (PC1) from 30 gestational weeks to 36 months postnatal



In both PC1 and PC2, the general trend is that there is a high degree of overlap between groups 1 (blue) and 2 (green) as well as between groups 3 (red) and 4 (black) (Figure 4.13). This is consistent with observations made osteometrically, where no statistically significant changes were observed between consecutive age groups, with the exception of groups 2 and 3, or 0 to 2 years. The statistically significant difference observed between groups 2 and 3 are further shown in the large degree of separation between these groups along the PC1 and PC2 axes. Further detailed morphological observations are discussed according to the mediolateral, superior-inferior and anterior-posterior planes as previously outlined in section 4.1 on Osteometry.

4.2.1.1. Medio-lateral plane

The mental angle was shown to lengthen along PC1 and PC2 axes. This is highlighted in the thin-plate spline and with a notable widening in the wireframes of the arch – which is formed by the mandibular body and the ramus. The observed lateral protrusion (flaring) of the mandibular ramus region, particularly in the area of the gonial angle, corresponds with an increase observed in the mental angle in the osteometric analysis (Figures 4.1 and 4.13). The mandibular basilar widths bigonial and biantegonial also indicated increases along the PC1 and PC2 axes, as a result of the lateral protrusion (flaring) previously observed (Figure 4.2 and Figure 4.13).

The bigonial width of the mental foramen was also observed to increase with age, this was noted with a widening of the thin-plate splines in the anterior region as indicated along PC1 and 2. Morphologically the extreme positive specimen (group 1: 30 to 40 gestational weeks) wireframe indicated a round and smooth appearance. With progression along the PC1 axis, this region progressively narrowed and adopted a very sharp and narrow mental region. Subsequently the extreme negative specimen (group 4: 24 to 36 months) was considerably shaper and narrower in appearance. Therefore this is further indicated by the increase in morphological definition previously observed in the thin-plate splines as well as the increase observed osteometrically (Figures 4.3 and 4.13).



4.2.1.2. Superior-inferior plane

There were no notable morphological changes expressed in the mental foramen to inferior border of the mandible variable in either the thin-plate splines or the wireframes in the specimens along the PC1 and 2 axes. This is consistent with previous observations made in the osteometric analysis for this dimension, where a bone deposition did not appear to change from the youngest to the oldest group (Figures 4.4 and 4.13).

However there was an increase noted in the depth of the mandibular notch from the extreme negative to the extreme positive along the PC1 and PC2 axes. A more defined mandibular notch area on the wireframes morphologically indicated this. The thin-plate splines highlighted the indicated increase in the depth of the mandibular notch. Once again these findings are consistent with those observed, which had been observed osteometrically (Figures 4.5 and 4.13).

4.2.1.3. Anterior-posterior plane

An increase in the rameal/mandibular width from the extreme negative to the extreme positive values along PC1 as well as PC2 axes was indicated in the wireframe projections. A lengthening of the thin-plate splines laterally further emphasized this change (Figures 4.6 and 4.13).

As was indicated previously in the mandibular notch depth, the mandibular notch width was observed to increase from youngest to oldest along both the PC1 and PC2 axes. However morphological changes in the width of the notch area, increased to a greater extent in the wireframes along the PC2 axis. This was in contrast to previous measurements where most observations were more pronounced along the PC1 axis (Figures 4.7 and 4.13).



Furthermore an increased in the length of the lower border of the thin-plate splines between the extreme positive to the extreme negative specimens along both the PC1 and PC2 axes indicated an increase in the mandibular body length. This was also observed in the wireframe images although to a lesser degree. These findings are consistent with previous trends indicated in the osteometric analysis of this dimension (Figures 4.8 and 4.13).

The longest length of the mandible dimension was observed to increase considerably along the PC1 and PC2 axes. This was observed in the lateral bulging of the thin-plate splines noted previously; the bulging indicated a lateral projection of the mandibular ramus away from the central plane of the mandible. This led to an increase in the distance between the anteriorly placed mental region and the posteriorly placed condylar process, both landmarks are used in the measurement of the longest length of the mandible. The increase in the longest length of the mandible measurement was also observed in the lengthening of the wireframes from the extreme positive to the extreme negative along PC1. A similar trend was also observed along the PC2 axis, however to a lesser degree (Figures 4.9 and 4.13).



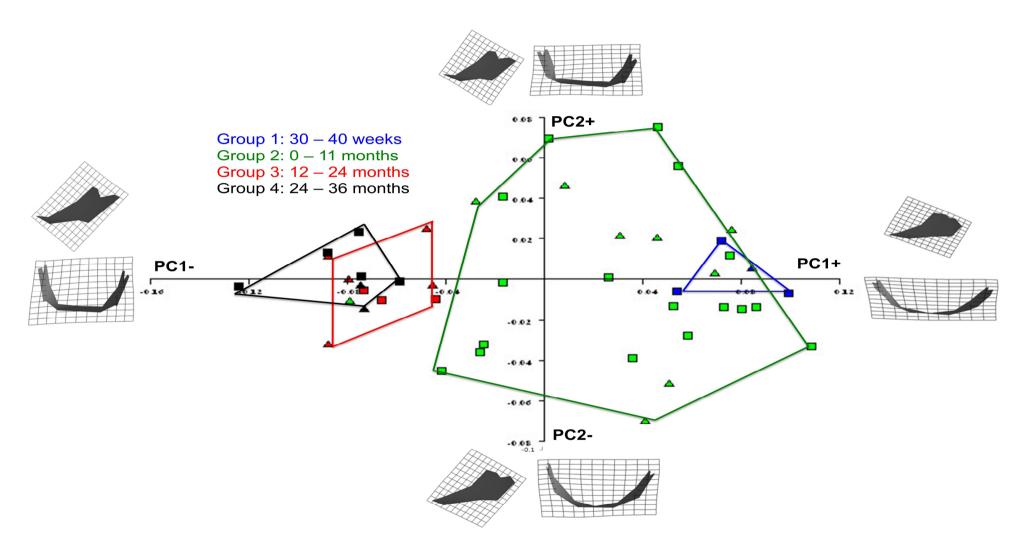


Figure 4.13 Distribution of variance along principle components 1 and 2 comparing groups 1-4



4.2.2. Sex related morphological changes

The distribution of variance along the PC1 and PC2 axes for sex was illustrated in Figure 4.14. In contrast to the group comparisons for age, PC1 indicated a clearer distinction between the male (squares) and female (triangles) specimens within each of the groups assessed. This distinction between the groups was further amplified by the variance indicated in PC2.

A high degree of overlap between groups 1 and 2 as well as between groups 3 and 4 was noted, which is reliable with observations made for the age associated changes. Furthermore there was no overlap found between groups 2 and 3, which remained consistent with the previously noted statistically significant increases in the mandibular dimensions between the two groups. There was no significant difference noted between the sexes for groups 1 to 3 i.e. overlap was found between the sexes. However there was no association observed within group 4 between the sexes, suggesting the possibility of significant morphological differences between the sexes.

The mental region in group 4 females was indicated as being wider and less round in appearance when compared to that of males. The posterior gonial angle region appeared to protrude more laterally in females than it did in males, which when combined with the wider mental region indicate a larger mental angle in females. These observations are consistent with those made in the osteometric assessment where females were indicated to be significantly larger than males within the 24 to 36 month period (Figures 4.10 and 4.14).

As with previous observations in the mandibular notch depth, group 4 females indicated a deeper notch area on the wireframes when compared to males. This was indicated by a greater definition of the notch area on the wireframes in females than what was found for males. When comparing the morphological observations made with previous osteometric observations, the results were similar (Figures 4.11 and 4.14).



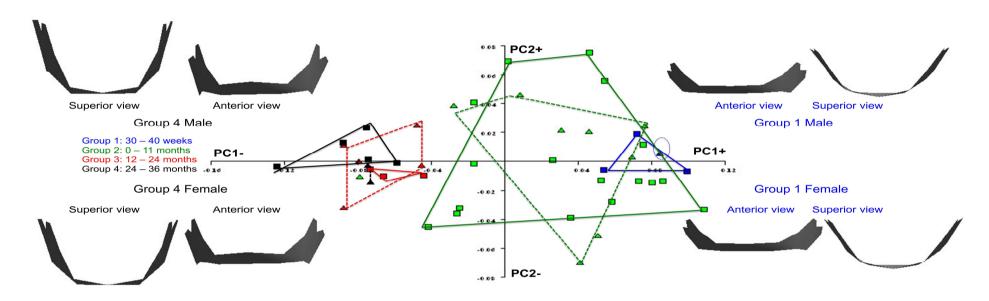


Figure 4.14 Distribution of variance along principle components 1 and 2 comparing males (solid lines) and females (broken lines) within groups1-4



4.3. Inter-observer error

Inter and intra-observer repeatability for each of the measurements evaluated are shown in Table 1 Appendix C (pg 72). In general most of the variables measured obtained values between the 0.79 and 0.99, thus indicating a high level of reliability. The distance between the inferior border of the mandible and the mental foramen was the least repeatable with a value of 0.78 and a range of between 0.41 and 0.93. However, this may be attributed to difficulties in locating the exact point of the inferior border of the mandible.

4.4. Summary

In general, there were no statistically significant increases noted for size and shape from the late foetal period to 11 months. This was also indicated for the comparison between males and females during the same age range. However during the period from birth to 2 years, the mandible was observed to widen (increases in the bigonial and biantegonial widths) and lengthen (increase in the body length and longest length of the mandible) in preparation and accommodation of the dentition. These statistically significant increases were however not observed when comparing males and females during the same period. In contrast to previous osteometric and morphological observations, there were no further statistically significant increases indicated for the period 24 to 36 months. When assessing males and females, statistically significant increases were noted in the mental angle and mandibular notch depth dimensions. These increases may have been an indication of either an increased growth rate in females or an earlier onset of mastication. This would be consistent with the previously observed increase in the mandibular dimensions to accommodate masticatory structures.



CHAPTER 5

DISCUSSION

In South Africa, no osteometric standards exist in which changes in mandibular growth from the late foetal period to three years of age can be evaluated. For clinicians, this is a crucial period in which they can diagnose and treat conditions such as micrognathia and Hallermann-Streiff syndrome. Additionally, foetal and neonatal remains are frequently dumped in the veldt and often discovered in a fragmentary condition. A forensic anthropologist is often requested to provide an assessment of age at death and sex from these remains. Thus the purpose of this study was to document changes in the size and shape of the mandible and to compare it with age at death and sex.

5.1. Growth of the mandible: a combination of osteometric variables and geometric morphometric parameters

From 30 gestational weeks to 36 months, a general increase in size was noted in the mandible, which included: a lengthening of the body, a widening of the basilar arch and a flaring of the ramus (see Figures 4.12 and 4.13, pgs 44,48). These changes may be attributed to growth and development of the tongue, fusion of the symphysis menti, eruption of the deciduous dentition, and influence from the associated muscles of mastication.

An increase in the mandibular body length and a posterior widening of the bigonial and biantegonial dimensions from the late foetal through the first year of life may primarily be associated with development and growth of the tongue. In a study on prenatal growth of the mandible from 5 to 40 gestational weeks, Lee (2001) noted that the tongue commenced its growth in an anterior direction during the 5th gestational week [19,49]. In the 6th gestational week, the genioglossus muscle (intrinsic tongue muscle) attached to the posterior surface of the mandibular symphysis [49]. This attachment to the posterior surface, Lee (2001) suggests, may



have resulted in the tongue movements being responsible for the early induction of mandibular movement and growth [49]. Lee (2001) further stated that as the mandible is supported by the masticatory and tongue muscles from the 8th gestational week, it would be able to control the development of the lower jaw even in the absence of Meckel's cartilage [49]. Thus growth and development of the tongue through associated musculature attachment would have had an influence on the increase in the size and changes in shape of the arch dimensions analyzed in this study; especially in terms of the anterior (mental foramen width) and posterior (bigonial width) dimensions.

When further considering the anterior dimensions, the bigonial width of the mental foramina indicated a general increase across all age groups assessed (30 gestational weeks to 36 months postnatal). This was expected as part of the normal growth process. The bigonial width of the mental foramina also indicated a statistically significant increase over the 0 to 24 month period. These increases may have resulted from influences either on the tongue due to genioglossus muscle attachment or changes in the morphology of the symphysis menti. The morphological changes in the mental symphysis area included both a progressive increase in size as well as shape and could be observed as early as 14 weeks of prenatal development. The morphology of the mental symphysis at 14 week of prenatal development included a limited number of cartilaginous nodules present on the symphyseal surfaces of the hemi-mandibles [15]. These changes persist into the 28th gestational week where mandibular ossicles ossify and consequently fuse with the anterior part of the body during the first year of life [15].

In the transition between the pre-natal and birth periods, alterations in the mandibular arch dimensions were noted (Figure 5.1). Continual growth of the tongue may have been an influencing factor in the increase in the mandibular arch. Further increases in the longest length of the mandible, mandibular width, bigonial width of the mental foramen and mandibular notch dimensions were also observed.



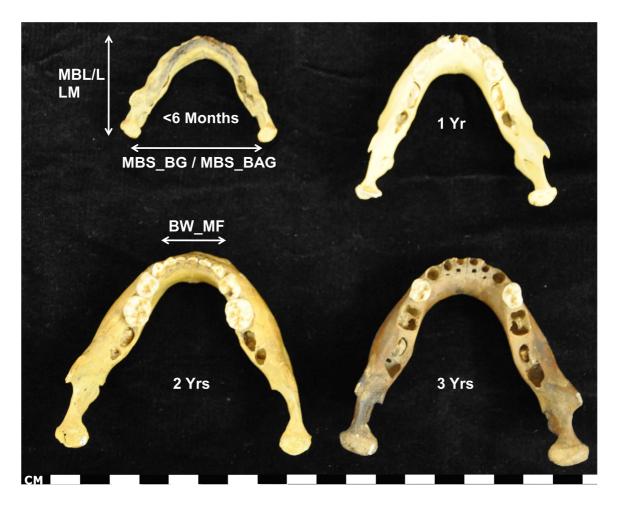


Figure 5.1. Mandibles of varying ages from the Raymond Dart Skeletal collection, University of Witwatersrand.

The progressive increase in the dimensions of the mandibular arch (MBL: mandibular body length, LLM: longest length of the mandible, BW_MF: bigonial width of the mental foramen, MBS_BG: mandibular basilar width bigonial and MBS_BAG: mandibular basilar width biantegonial) from the period of birth to 36 months, is shown in Figure 5.1. When the top left (<6 months) and top right mandibles (1 year) are compared, an increase in the mandibular body length was evident. Increases in the mandibular basilar widths - bigonial and biantegonial - indicate a posterior widening of the mandible with an increase in age.



An increase in the mandibular body length with age can be attributed to continual growth of the tongue as well as development and eruption of the dentition. Eruption of the deciduous dentition would be observable from 6 to 36 months. Scheuer and Black (2000) suggest that the morphology of the developing mandible is attributed to bone deposition occurring posteriorly and bone resorption anteriorly [15]. The resultant effect is the posterior displacement of the ramus and an increase in mandibular body length so as to accommodate the erupting dentition (Figures 5.2 - 5.4). The deciduous central incisors erupt around 6 months of age. The lateral incisors follow at 10 months, first molars at 14 months, canines at 17 months and the second molars erupting between 23 and 24 months, depending on the sex [15].

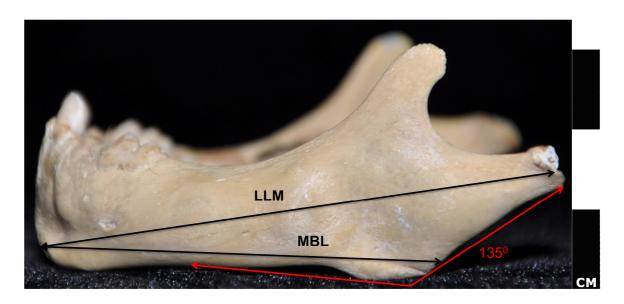


Figure 5.2. Lateral view of mandible aged 1 year (LLM: longest length of mandible, MBL: mandibular body length)

Changes in mandibular body length (MBL) are emphasized through a change in orientation of the mandibular ramus to the mandibular body (Figure 5.2 to 5.4). With age, the mandibular ramus became more upright (see Figure 5.2; 1 year old). This may be the result of the deposition of bone on the superior aspect of the mandibular body along with a progression in dental eruption [15].



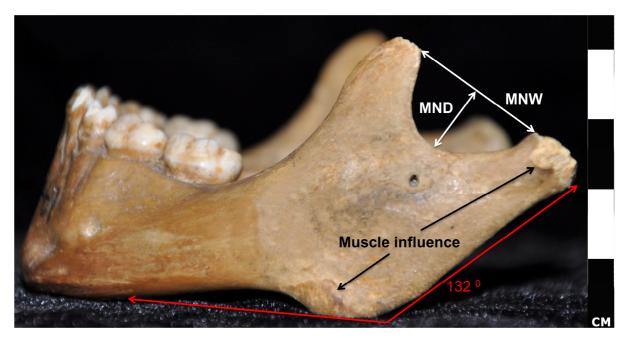


Figure 5.3. Lateral view of mandible aged 2 years (MND: mandibular notch depth, MNW mandibular notch width)

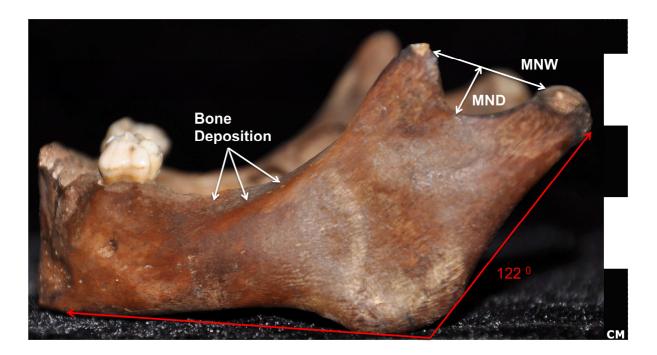


Figure 5.4. Lateral view of mandible aged 3 years (MND: mandibular notch depth, MNW: mandibular notch width)



Dental eruption also accounts for changes expressed in the mandibular angle. Scheuer and Black (2000) noted that the mandibular angle decreased from 150° at birth to 130° after the eruption of the deciduous teeth [15]. The angle of the mandible may have influenced the longest length of the mandible. A change in degrees of the mandibular angle (Figures 5.2 - 5.4) is most evident when comparing mandibles from different age groups. At birth, the angle of the mandible is reported to be obtuse (closer to 180°). As growth progresses, the angle becomes acute (closer to 90°) as seen in Figure 5.4. The changes observed in the mandibular angle in Figures 5.2 to 5.4 indicated a possible relationship with the longest length of the mandible. As the angle of the mandible became more acute, it decreased the distance between the most posterior point of the mandibular condyle and the most anterior point of the symphysis menti (Figures 5.2 - 5.4). Therefore, the previously observed increase in the body length coupled with the changes in the mandibular angle may have also influenced changes in the longest length of the mandible.

The longest length of the mandible may be influenced by the position of the mandibular condyles, as well as the width of the ramus and notch. Both of these dimensions were shown to have statistically significant increases, postnatally (Figures 5.2 - 5.4). These increases along with increases in the mandibular notch depth may also have been due to an increase use in the muscles of mastication.

The muscles of mastication with its attachments to the various mandibular structures had an influence on the morphology and, ultimately functioning, of the mandible. These four muscles include the temporalis, medial pterygoid, lateral pterygoid and the masseter. The temporalis muscle inserts on the apex, anterior and posterior borders as well as the medial surface of the coronoid process of the mandible [22]. The function of the temporalis muscle is to elevate and retract the protruded mandible. This creates pulling stress on the coronoid process, as the muscles attempt to restore the mandible to its resting position. Inferiorly, the temporalis muscle fibers also join with the masseter muscle fibres. The two muscles act together to influence both the morphology and functioning of the mandible, particularly over the coronoid process.



The masseter muscle attaches to the lateral surface of the mandibular ramus, with some attachment to the coronoid process [22]. The function of the masseter muscle is to elevate the mandible during mastication and is most active during the grinding of food. Most of the changes, size and shape, of the ramus of the mandible as well as the mandibular angle were noted after 14 months, which coincided with the eruption of the first molar tooth, and may be associated with an increased dependence on solid foods for nutrition.

The pterygoid muscle complex includes the medial and lateral pterygoid muscles. The lateral pterygoid muscle inserts on the pterygoid fovea of the mandibular condyle [22]. The functions of this muscle include protrusion, depression, lateral exertions as well as retrusion of the mandible. The muscle supports mandibular movement and can assist in extreme mastication [22]. The medial pterygoid muscle inserts on the roughened surface of the medial aspect of the angle of the mandible [22]. The functions of the medial pterygoid muscle are to elevate the mandible and to assist in lateral and protrusive movements [22]. The medial pterygoid and masseter muscles both pass through the mandibular notch area. An increased use of these muscles, due to the eruption of dentition and a greater reliance on mastication, may cause an increase in the mandibular notch width and depth. As previously mentioned, increased stress placed on the muscle attachment sites of the mandible occurs with growth, such that mastication may considerably influence morphological changes in the mandible. Other variables that may contribute include dental eruption and diet.

In summary, the mandible, from foetal to post-natal childhood (3 years), increased in size and changed morphologically. After birth, the mandibular arch dimensions widened so as to accommodate the developing tongue, and eventually, the erupting dentition. This is evident in the 54% increase in the bigonial width of the mental foramen between groups 1 and 2. This is also expressed morphologically by the widening of the mandibular body as well as the changes in the mental angle. With the eruption of the dentition, more emphasis was placed on the muscles of mastication and the consumption of hard foodstuffs; this was morphologically indicated in the mandibular notch depth dimensions, which were more obvious in the 24 to 36 month



age range. The overall increase of 52% in the longest length of the mandible measurement postnatally (groups 2 to 4) when compared to the 7% increase in the pre-natal dimensions emphasizes the mandible accommodating the erupting dentition.

Generally the mandible changed to accommodate three main events. The first change noted was between the prenatal and early postnatal groups (30 gestational weeks to 11 months postnatal). The increase in the mandibular dimensions noted during this period was suggested to occur as a result of the mandible growing in synchronization with the developing tongue. The second major change noted was between 11 and 24 months postnatal, where the mandible adapted its dimensions to accommodate the eruption of the deciduous dentition. The third change was across the entire postnatal group (0 to 36 months), following dental eruption and increase in mastication was noted morphologically.

An increase in mastication had a two-fold effect on changes in the size and shape of the mandible. An increase in muscle mass would necessitate an increase in the dimensions of the mandibular notch [22]. Secondly, an increase in use of these muscles imposed an increase in stress and strain on the muscle attachment sites. This was noted in the posterior flaring of the mandibular angle. Therefore, the bone was remodeled so as to accommodate the imposed stress on this structure as well as to maintain structural integrity of the area from 30 gestational weeks to 36 months.

5.2. Sexual dimorphism of the mandible: a combination of osteometric variables and geometric morphometric parameters

Sexual dimorphism was not observed in this prenatal period studied, which confirmed the previous study of Fazekas and Kosa (1978). The longest lengths of the mandible, mandibular body length and the mandibular width have been previously found not to be sexually dimorphic within 12 to 40 weeks of gestation [4,15].



Postnatally, sexual dimorphism was noted in the mental angle, mandibular notch depth and the mandibular width (or ramus width). A lateral protrusion of the mandibular angle region on the posterior aspect of the mandible was found to be more evident morphologically in female subjects (Figure 4.14). When considering sexual dimorphism found in the current study to previous studies, this was not unexpected as Loth (1995) found gonial eversion as an indicator of sex to be more accurate in females than males in her study of the juvenile mandible (0-19 years)[31]. Loth (1995) also found evidence of shape differences in subjects 6 years or older in an analysis of symphyseal shape as an indicator of sex [31]. However Loth's method of analysis of the symphyseal region has received much criticism [18]. Scheuer (2002) studied Loth's method and found the accuracy to be 20% less in the symphyseal shape of the mandible than was previously recorded [18].

In the current study, sexual dimorphism in the symphyseal region of the mandible showed that the menton of males was smaller and narrower than that of females (see Figure 4.14 pg 50). The mental angle was calculated from a combination of the mandibular body length (MBL) and mandibular basilar width bigonial (MBS_BG). The sexual dimorphism noted previously in the mental angle could have been influenced by changes in the mandibular angle and the symphyseal region (males having a smaller and narrower menton).

Sexual dimorphism was also evident in the width and depth of the mandibular notch as well as the mental angle. Previously, changes in these dimensions were attributed to influences from masticatory structures and eruption of the dentition [22]. Since sex differences have been noted in the development and eruption of teeth, differential onset of eruption and a subsequent increase in mastication during infancy and early childhood may have contributed to differences between the sexes.

Since females demonstrated earlier growth and development in the mandible, it may be possible to suggest that the structures associated with mastication and dental eruption would also be sexual dimorphic. This was shown in this study with the mandibular notch depth indicating sexual dimorphism as well as the mental angle.



CHAPTER 6

CONCLUSION

In the prenatal phase, the mandibular arch increases so as to accommodate the developing tongue. Postnatally, changes in the mandibular arch can be attributed to dental eruption. Increased mastication caused an increase in both the mass of the associated muscles and in the stress and strain on the attachment sites for these muscles was indicated osteometrically by an increase in the mandibular notch dimensions. Therefore, the bone remodeled and grew so as to accommodate these stressors and to maintain structural integrity. Sexual dimorphism was also affected by mastication, where most of the changes noted were in structures related to the process of chewing. These changes were most evident in females, where the mandibular angle was more pronounced.

The analysis of growth in the mandible both prenatally and postnatally provides information regarding the relationship of growth of the mandible with that of the surrounding muscular structures. The current study assessed overall changes in the growth and sexual dimorphism of the mandible. Sexual dimorphism was noted in the postnatal stages. Changes expressed in the general morphology of the mandible both in terms of age and sex provided valuable insight into the complex relationship of growth of the mandible and the surrounding oral cavity tissues. This study further added validity to previous theories about the relationship between the mandible and the structures associated with mastication, by indicating significant changes particularly over periods of increased masticatory activity [4,16,37,38].

The assessment of the shape of the mandible using geometric morphometrics did indicate morphological changes in the mandible. However these changes were only of substantial value in the postnatal groups as those changes observed prenatally were extremely indistinct and could not be used without additional information from the osteometric assessment. Therefore to accurately assess the changes in the growth of the mandible a combination of geometric morphometrics and osteometric analyses is recommended.



CHAPTER 7

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APPENDICES

Appendix A. Table 1: Mean values (mm) and standard deviations for each of the measurements taken for groups 1-4

	Grou	ıp 1	Gro	up 2	Grou	р 3	Grou	ıp 4	
	30-40	wks	0-11 r	nnths	12-24 m	nnths	24-36 n	nnths	
N=	18	8	4	1	8		7		
	Mean	sd	Mean	sd	Mean	sd	Mean	sd	
MA	58.16	16.66	68.56	8.06	69.03	5.89	72.18	4.05	
MBS_BG	32.64	7.24	41.41	7.86	53.32	5.26	61.14	6.00	
MBS_BAG	28.43	5.79	36.90	8.14	49.38	4.73	55.52	4.88	
BW_MF	21.09	6.32	24.90	6.09	32.51	2.78	34.19	2.58	
MFIB_L	6.38	1.48	6.13	2.34	7.02	1.21	6.74	0.71	
MFIB_R	5.86	0.92	6.31	2.16	6.91	1.47	7.42	1.36	
MND_L	4.21	1.23	3.47	1.65	6.77	1.77	7.70	2.28	
MND_R	3.60	0.94	3.39	1.53	6.35	1.69	7.29	2.78	
MNW_L	6.61	1.48	7.44	2.22	11.12	1.85	13.60	2.50	
MNW_R	7.52	2.67	7.92	2.33	11.91	1.75	14.49	3.02	
MW_L	15.38	1.98	16.91	3.59	20.86	2.27	23.90	2.98	
MW_R	17.00	5.45	16.48	3.69	21.55	2.27	24.83	2.51	
MBL_L	34.24	5.23	37.04	5.66	47.07	3.76	51.65	4.65	
MBL_R	30.56	4.20	36.46	5.94	47.23	3.83	52.21	5.16	
LLM_L	45.17	5.73	48.40	8.68	66.47	5.47	73.61	7.26	
LLM_R	41.85	6.26	47.66	9.23	67.37	5.52	74.15	6.88	
	0.00.44	<u> </u>	<u> </u>	L	1400 040	L		1	



Table 2: Kruskall-wallis analysis, statistically significant differences in values for measurements taken for groups 1-4

			GRO	DUPS		
	1-2	1-3	1-4	2-3	2-4	3-4
MA	0.023		0.0209			
MBS_BG	0.004	0.0004	0.0009	0.001	0.0004	
MBS_BAG	0.003	0.0004	0.0009	0.0016	0.0004	
BW_MF		0.0031	0.0057	0.0058	0.0014	
MND_L		0.01348	0.01097	0.00184	0.00134	
MND_R		0.00569	0.00892	0.00225	0.00578	
MNW_L		0.001643	0.0009	0.0034	0.0005	
MNW_R		0.01018	0.0042	0.00183	0.0003	
MW_L		0.0009	0.0009	0.03123	0.00083	
MW_R		0.04051	0.01701	0.002269	0.0003	
MBL_L	0.3634	0.00203	0.0015	0.0008	0.0002	
MBL_R	0.00328	0.0005	0.0009	0.0003	0.0004	
LLM_L		0.0004	0.0009	0.0003	0.0002	
LLM_R		0.0005	0.0009	0.0001	0.0002	



Appendix B. Table 1: Mean values (mm) and standard deviations for each of the measurements taken in the comparison between males and females for groups 1-4

		Gro	up 1	Gro	up 2	Gro	up 3	Gro	up 4	
		30-40) wks	0-11 r	nnths	12-24	mnths	24-36 mnths		
N=		11M / 7F		25M / 16F		3M / 5F		4M / 3F		
		Mean	sd	Mean	sd	Mean	sd	Mean	sd	
MA	M	61.67	12.65	69.50	7.61	70.68	1.40	69.73	2.56	
	F	51.72	22.17	67.13	8.75	68.04	7.52	75.46	3.36	
MBS_BG	М	33.50	5.43	42.92	7.81	55.45	7.73	59.97	6.57	
50_50	F	31.30	9.79	39.15	7.63	52.04	3.61	62.70	6.06	
MBS BAG	М	28.69	3.38	38.16	8.43	50.58	7.64	54.50	5.54	
	F	28.03	8.70	34.93	7.49	48.66	2.88	56.88	4.52	
BW_MF	M	20.08	3.73	25.52	6.83	32.75	4.08	33.93	3.25	
	F	22.52	9.03	23.93	4.74	32.37	2.27	34.53	1.95	
MFIB_L	М	6.43	1.69	6.11	2.58	7.24	0.85	6.40	0.18	
_	F	6.31	1.26	6.16	1.99	6.89	1.47	7.18	0.98	
MFIB_R	М	5.56	0.90	6.52	2.60	7.35	1.96	7.74	1.28	
_	F	6.34	0.79	5.99	1.22	6.64	1.27	6.99	1.62	
MND_L	M	3.99	0.99	3.22	1.56	7.24	0.97	6.16	1.35	
	F	4.55	1.56	3.89	1.77	6.50	2.19	9.76	1.30	
MND_R	M	3.54	0.74	3.20	1.62	7.05	1.26	6.00	2.21	
MND_R	F	3.71	1.25	3.69	1.38	5.93	1.90	9.01	2.83	



Appendix B. Table 1: Mean values (mm) and standard deviations for each of the measurements taken in the comparison between males and females for groups 1-4

		Gro	up 1	Gro	up 2	Gro	up 3	Gro	up 4	
		30-40) wks	0-11 r	nnths	12-24	mnths	24-36 mnths		
N=		11M	/ 7F	25M	/ 16F	3M / 5F		4M / 3F		
		Mean	sd	Mean	sd	Mean	sd	Mean	sd	
MNW_L	М	6.24	1.11	7.68	2.23	11.00	2.73	12.88	3.06	
	F	7.19	1.88	7.06	2.22	11.19	1.49	14.57	1.49	
MNW_R	М	6.62	2.13	8.11	2.51	12.05	2.81	14.63	3.72	
	F	8.93	2.97	7.63	2.06	11.83	1.17	14.31	2.55	
MW_L	М	15.31	2.02	17.41	3.84	21.40	3.52	23.33	3.10	
	F	15.48	2.05	16.09	3.09	20.54	1.57	24.67	3.27	
MW_R	М	17.51	6.94	17.01	3.60	21.85	4.06	24.08	2.69	
	F	16.28	2.47	15.69	3.80	21.38	0.80	25.85	2.31	
MBL_L	М	34.18	5.67	38.04	6.14	48.17	6.06	52.39	5.91	
_	F	34.34	4.89	35.48	4.56	46.41	2.21	50.67	3.17	
MBL_R	М	31.26	4.35	37.33	6.15	47.59	6.38	52.61	6.61	
_	F	29.46	4.03	35.12	5.53	47.02	2.28	51.67	3.68	
LLM_L	М	44.60	4.75	49.20	8.93	67.93	9.06	74.03	9.32	
	F	46.06	7.35	47.08	8.37	65.59	2.94	73.06	5.19	
LLM_R	M	42.06	6.80	48.44	9.56	67.86	9.88	74.39	8.74	
LLIVI_K	F	41.51	5.81	46.49	8.89	67.07	2.04	73.82	5.22	



Table 2. Sex comparison p-values of measurements taken for groups 1-4

		MALES	VS. FEMALES	
	Group 1	Group 2	Group 3	Group 4
	30-41 wks	41-51 wks	52-62 wks	0-11 Months
N=	11M / 7F	25M / 16F	3M / 5F	4M / 3F
MA	0.280	0.352	0.559	0.03
MBS_BG	0.566	0.138	0.448	0.608
MBS_BAG	0.829	0.230	0.626	0.592
BW_MF	0.476	0.426	0.877	0.751
MFIB_L	0.870	0.938	0.768	0.095
MFIB_R	0.073	0.475	0.544	0.534
MND_L	0.361	0.225	0.606	0.006
MND_R	0.735	0.292	0.385	0.164
MNW_L	0.194	0.396	0.886	0.385
MNW_R	0.066	0.551	0.901	0.875
MW_L	0.858	0.252	0.736	0.442
MW_R	0.648	0.250	0.722	0.274
MBL_L	0.947	0.152	0.553	0.716
MBL_R	0.383	0.267	0.834	0.876
LLM_L	0.604	0.466	0.516	0.909
LLM_R	0.857	0.507	0.795	0.948



Appendix C. Mean values (mm) and standard deviations for each of the measurements taken for groups 1-4

	Pc	Min	Max
LLM_L	0.99	0.99	1
LLM_R	0.99	0.95	0.99
MA	0.99	0.99	0.99
MBL_L	0.91	0.70	0.97
MBL_R	0.93	0.77	0.98
MBS_BAG	0.81	0.50	0.93
MBS_BG	0.91	0.85	0.96
BW_MF	0.96	0.86	0.99
MFIB_L	0.79	0.40	0.94
MFIB_R	0.78	0.41	0.93
MNW_L	0.94	0.79	0.98
MNW_R	0.96	0.87	0.99
MW_L	0.99	0.97	0.99
MW_R	0.81	0.46	0.94
MND_L	0.99	0.98	0.99
MND_R	0.85	0.57	0.96



Appendix D. List of individuals and associated measurement values

id	6289W	6312W	3210W	7108W	6311W	6583W	6746W	5935W	5160W	6367W
sex	М	М	F	М	М	М	М	F	F	F
Group	G 1	G 1	G 1	G 1	G 1	G 1	G 1	G 1	G 1	G 1
crl (CM)	25.5	25.5	26	26.5	27	31.5	31.5	32	33	33
AGE	31.5	31.5	32	32.5	33	37.5	37.5	38	39	39
mbs_bg	31.42	30.38	32.68	29.28	29.35	35.62	36.06	24.52	45.23	28.90
mbs_bag	26.71	26.60	29.70	23.12	24.99	30.26	28.58	22.12	42.04	24.17
mfbw	20.06	15.76	21.83	13.60	19.09	25.62	17.17	11.25	39.83	20.18
mf_ibl	3.99	5.31	4.34	4.77	5.96	5.98	7.25	5.65	5.20	7.17
mf_ibr	3.84	4.63	6.74	5.22	5.91	5.23	5.67	5.99	6.19	7.34
mbl_l	28.29	28.66	30.28	31.74	30.64	32.46	38.72	31.21	41.06	35.86
mbl_r	26.82	27.24	29.81	25.81	28.88	33.05	33.82	35.63	30.68	25.05
IIm_I	37.11	39.70	39.96	44.67	41.38	43.21	49.96	41.14	54.25	36.73
IIm_r	35.56	35.31	39.94	34.95	38.91	44.58	40.93	43.91	41.14	47.67
mw_I	13.09	11.76	12.23	17.09	15.98	13.65	15.54	16.27	18.17	14.71
mw_r	12.89	10.77	14.52	10.97	15.99	15.18	27.28	17.89	13.79	14.86
mnw_I	5.57	5.58	4.46	6.64	5.48	5.25	6.45	8.06	7.89	9.96
mnw_r	5.34	4.01	6.57	8.41	6.65	6.30	6.53	8.58	12.40	6.84
mnd_r	2.07	3.10	2.40	4.53	3.62	3.85	3.41	5.53	5.07	3.48
mnd_I	2.33	4.83	2.81	4.27	2.92	4.41	4.28	4.71	3.57	6.23
MA Calc	0.35	0.41	0.41	0.50	0.52	0.41	0.51	0.74	0.23	0.60
MA	69.47	65.78	65.90	60.12	59.00	65.88	59.16	42.40	76.66	53.13



id	6963W	7067W	4238W	6575W	7047W	6833W	7137W	5939W	4533W	6326W
sex	М	F	М	М	М	F	F	М	F	F
Group	G 1	G 1	G 1	G 1	G 1	G 1	G 1	G 1	G 2	G 2
crl (CM)	27	27	28.5	31	31	33.4	33.5	34	35.5	35.5
AGE	33	33	34.5	37	37	39.4	39.5	40	0	0
mbs_bg	35.00	38.91	44.22	35.72	23.73	15.08	33.76	37.67	43.75	52.80
mbs_bag	27.92	30.83	34.23	28.47	32.01	14.72	32.59	32.72	39.28	47.44
mfbw	21.62	23.87	23.64	21.49	-	15.57	25.13	22.78	27.83	31.19
mf_ibl	7.88	7.81	-	9.92	6.86	7.15	6.82	6.41	4.31	6.19
mf_ibr	6.21	4.99	5.99	5.28	5.81	6.08	7.06	7.34	5.66	7.14
mbl_l	32.44	27.61	36.08	35.74	48.42	39.19	35.17	32.78	36.14	39.02
mbl_r	29.81	25.90	41.06	33.61	33.17	25.98	33.15	30.55	40.68	40.70
IIm_I	41.47	43.88	53.45	46.50	45.04	52.80	53.68	48.10	48.38	56.16
IIm_r	39.03	33.58	58.30	46.67	43.00	35.37	48.95	45.43	57.56	53.84
mw_I	13.97	13.71	15.11	17.02	16.73	16.14	17.13	18.50	16.26	19.11
mw_r	17.57	15.16	31.86	13.88	-	16.89	20.85	18.66	20.53	21.25
mnw_I	4.93	5.34	7.53	5.78	6.81	8.10	6.51	8.62	7.08	8.63
mnw_r	5.96	6.41	11.51	4.50	5.27	7.90	13.78	8.39	7.70	9.60
mnd_r	2.78	2.16	4.39	3.22	4.32	3.84	3.49	3.62	5.49	3.88
mnd_I	3.21	2.64	4.79	3.70	3.34	6.28	5.64	5.77	6.09	5.58
MA Calc	0.37	-0.06	0.35	0.47	0.90	0.97	0.51	0.29	0.36	0.12
MA	68.26	-	69.61	61.91	26.22	13.11	59.13	72.90	69.14	82.93



UNIVERSITE IT VAN PRETORIA UNIVERSITY OF PRETORIA YUNIBESITHI YA PRETORIA YUNIBESITHI YA PRETORIA Appendix D. List of individuals and associated measurement values

id	5606T	5936T	6459W	6510W	6502T	3958W	5937T	5945T	6605W	6607W
sex	М	F	F	М	М	F	М	F	М	М
Group	G 2	G 2	G 2	G 2	G 2	G 2	G 2	G 2	G 2	G 2
crl (CM)	36	36	36	36.5	37	37.5	38	38	38	38
AGE	0	0	0	0	0	0	0	0	0	0
mbs_bg	33.18	31.51	34.72	32.15	42.27	28.61	36.58	30.32	46.49	29.03
mbs_bag	29.95	28.25	35.51	24.46	39.57	23.52	31.45	26.06	47.73	19.03
mfbw	16.53	20.60	21.78	27.43	23.63	22.22	16.54	20.19	34.82	9.00
mf_ibl	3.41	5.23	9.09	4.90	7.16	12.02	3.65	3.64	6.77	6.80
mf_ibr	3.82	4.69	8.03	6.91	8.20	5.67	5.22	4.91	9.76	6.83
mbl_l	27.63	31.18	37.75	41.68	31.46	40.00	31.59	26.51	48.98	38.32
mbl_r	27.79	27.52	40.46	36.31	34.21	30.60	31.43	26.24	36.80	25.07
IIm_I	36.10	39.35	52.23	50.34	45.35	60.42	37.57	33.95	61.05	50.61
llm_r	34.87	35.86	53.69	55.24	43.72	51.15	35.95	31.41	48.83	39.19
mw_I	12.61	13.56	19.68	14.07	16.37	21.89	13.98	10.30	18.80	19.03
mw_r	10.13	10.36	16.98	20.53	16.31	22.49	12.33	9.66	18.19	20.08
mnw_I	5.41	6.55	8.01	5.85	6.97	11.45	4.94	2.32	7.76	6.70
mnw_r	3.80	5.57	8.06	8.84	8.76	11.65	5.55	2.98	11.75	6.84
mnd_r	1.20	2.65	4.97	3.28	2.58	5.31	0.77	2.12	4.50	3.42
mnd_I	2.20	3.12	5.94	3.69	3.24	5.80	0.61	1.94	4.44	2.97
MA Calc	0.28	0.43	0.61	0.67	0.17	0.70	0.33	0.34	0.44	0.65
MA	73.55	64.58	52.57	48.08	80.02	45.43	70.96	70.18	63.79	49.25



id	5430T	6275T	6590T	5788T	7103W	6583T	6138T	6051T	6682T	7122W
sex	F	М	F	F	М	М	М	М	F	М
Group	G 2	G 2	G 2	G 2	G 2	G 2	G 2	G 2	G 2	G 2
crl (CM)	40	40	40	41	41	43	43.2	43.8	44	44.5
AGE	0	0	0	0	0	0	0	0	0	0
mbs_bg	35.03	51.67	31.02	33.73	-	44.34	40.49	35.80	37.03	53.56
mbs_bag	28.16	43.25	28.34	29.36	47.01	40.81	34.83	31.31	33.90	47.08
mfbw	21.28	31.34	20.66	18.68	16.32	28.67	26.93	16.01	20.74	30.68
mf_ibl	4.69	5.25	5.80	6.63	16.97	5.99	4.99	6.57	5.51	7.05
mf_ibr	7.11	6.48	7.79	6.12	16.04	4.69	5.11	3.35	5.03	7.57
mbl_l	31.22	42.49	28.48	34.67	43.51	38.32	37.82	30.92	35.21	46.45
mbl_r	31.06	42.39	30.08	31.67	38.75	37.67	38.47	29.38	33.22	44.30
IIm_I	37.87	53.58	40.21	44.15	47.75	48.15	46.97	37.74	44.95	69.07
IIm_r	37.47	51.84	37.70	38.60	-	48.59	43.90	37.15	43.80	65.62
mw_I	12.56	18.10	14.93	15.90	20.16	18.24	14.25	14.13	15.82	27.66
mw_r	12.08	18.16	12.87	12.85	-	19.02	14.73	12.80	15.18	25.71
mnw_I	4.39	8.19	6.50	8.07	10.33	7.87	8.07	6.72	6.68	12.39
mnw_r	5.33	7.20	6.77	7.45	9.17	7.76	4.63	4.73	6.83	10.56
mnd_r	1.24	5.69	3.37	3.79	5.88	2.06	2.04	1.51	2.96	2.64
mnd_I	1.03	4.71	3.27	3.53	6.98	2.25	1.05	2.05	4.13	2.60
MA Calc	0.37	0.26	0.44	0.49	-	0.32	0.44	0.30	0.42	0.30
MA	68.47	75.00	63.91	60.93	-	71.39	64.11	72.79	65.46	72.29



UNIVERSITE IT VAN PRETORIA UNIVERSITY OF PRETORIA YUNIBESITHI YA PRETORIA YUNIBESITHI YA PRETORIA Appendix D. List of individuals and associated measurement values

id	6618T	6939W	5859T	6607T	6152T	6148T	6193T	6285T	6077T	5410T
sex	F	М	F	F	М	М	М	М	М	М
Group	G 2	G 2	G 2	G 2	G 2	G 2	G 2	G 2	G 2	G 2
crl (CM)	46	46	48	48	48.2	48.3	48.3	48.3	52.7	53
AGE	0	0	0	0	0	0	0	0	0	0
mbs_bg	46.83	58.52	41.19	40.42	39.28	46.28	32.69	31.42	44.48	44.62
mbs_bag	40.94	52.79	34.99	34.56	34.22	39.32	26.48	26.71	37.22	38.92
mfbw	27.67	31.75	21.82	17.99	22.68	29.28	19.80	20.06	28.15	24.29
mf_ibl	5.39	5.47	5.70	5.65	6.79	5.00	3.20	3.99	6.17	7.04
mf_ibr	4.98	7.97	5.09	4.42	7.41	3.93	4.07	3.84	7.25	7.59
mbl_l	38.59	46.53	34.52	33.96	30.94	36.81	28.10	28.29	38.45	36.88
mbl_r	40.81	46.73	35.01	34.13	32.82	39.41	29.95	26.82	39.47	40.59
IIm_I	48.33	60.90	43.45	45.91	39.88	46.00	35.46	37.11	49.17	-
IIm_r	49.07	63.27	44.74	44.85	39.43	46.36	36.51	35.56	48.69	48.43
mw_I	16.31	21.72	13.08	16.54	12.62	14.37	13.02	13.09	18.17	17.72
mw_r	16.04	19.91	15.07	13.76	12.29	16.11	12.36	12.89	19.10	16.24
mnw_I	6.63	7.86	5.47	6.61	6.94	6.73	3.66	5.57	8.40	7.38
mnw_r	7.17	11.28	7.53	9.72	5.74	10.10	6.07	5.34	10.23	6.90
mnd_r	3.82	4.47	2.74	3.17	2.64	2.12	0.82	2.07	4.71	3.12
mnd_I	3.98	4.18	1.18	3.88	3.31	1.64	1.47	2.33	4.55	3.64
MA Calc	0.31	0.21	0.30	0.30	0.24	0.26	0.37	0.35	0.35	0.34
MA	72.23	77.73	72.66	72.83	75.99	74.70	68.47	69.47	69.60	70.14



UNIVERSITE IT VAN PRETORIA UNIVERSITY OF PRETORIA YUNIBESITHI YA PRETORIA YUNIBESITHI YA PRETORIA Appendix D. List of individuals and associated measurement values

id	5941T	6589T	535A	614A	3020A	652A	662A	664A	809A
sex	М	М	F	F	М	F	М	М	М
Group	G 2	G 2	G 2	G 2	G 2	G 2	G 2	G 2	G 2
crl (CM)	54	56							
AGE	0	0	0	0	0	0	0	0	0
mbs_bg	45.80	45.65	52.54	43.95	47.69	42.97	45.77	49.54	52.76
mbs_bag	40.40	42.55	49.44	39.49	44.54	39.67	43.16	45.68	45.41
mfbw	25.42	28.66	33.03	27.80	34.63	29.33	32.39	31.78	31.28
mf_ibl	6.25	4.80	7.13	6.32	7.68	5.30	5.84	6.32	4.76
mf_ibr	5.98	6.24	7.75	6.31	7.38	5.10	7.61	5.51	4.37
mbl_l	36.29	39.87	43.55	40.75	43.07	36.12	40.31	43.16	43.02
mbl_r	38.13	41.91	45.73	37.48	44.08	36.49	42.91	43.16	44.62
IIm_I	45.00	54.14	64.34	-	58.42	46.41	58.18	58.56	54.55
IIm_r	47.03	54.78	62.42	56.41	62.28	45.32	57.07	59.05	59.32
mw_I	13.87	17.61	19.90	-	21.52	15.55	21.15	20.47	22.43
mw_r	17.27	19.38	17.28	19.28	16.87	15.28	20.80	17.40	19.51
mnw_I	6.15	5.28	10.50	-	12.20	6.94	11.09	9.49	9.95
mnw_r	7.60	7.64	9.15	9.47	10.87	7.06	7.75	9.96	13.57
mnd_r	2.32	2.68	6.61	4.21	6.32	2.65	4.67	2.78	5.65
mnd_I	3.12	1.69	6.32	-	6.12	2.53	5.25	3.81	2.66
MA Calc	0.24	0.38	0.31	0.37	0.40	0.30	0.40	0.34	0.28
MA	75.92	67.81	72.05	68.21	66.34	72.57	66.66	70.05	73.99



id	3021A	3023A	3190A	3191A	668A	669A	713A	3018A
sex	F	F	М	F	М	М	F	F
Group	G 3	G 3	G 3	G 3	G 3	G 3	G 3	G 3
AGE	1	1	1	1	1	1	1	1
mbs_bg	47.95	53.96	47.09	48.44	56.89	62.36	53.98	55.89
mbs_bag	45.33	50.17	42.89	48.00	50.69	58.17	47.04	52.75
mfbw	29.55	33.83	29.15	33.50	31.90	37.19	30.33	34.63
mf_ibl	8.02	8.26	6.27	7.21	7.62	7.83	6.29	4.65
mf_ibr	7.94	7.19	5.52	7.51	7.12	9.42	5.08	5.51
mbl_l	47.83	47.68	42.03	47.13	48.32	54.16	42.52	46.89
mbl_r	49.68	47.47	40.84	46.33	48.42	53.51	43.56	48.04
IIm_I	63.26	64.75	57.99	67.34	70.08	75.73	62.82	69.80
IIm_r	67.36	66.01	57.06	67.61	70.10	76.43	64.44	69.93
mw_I	17.75	21.27	18.17	21.25	20.88	25.15	20.98	21.45
mw_r	21.29	21.08	17.36	21.13	22.90	25.28	22.75	20.64
mnw_I	9.88	11.72	7.91	13.01	11.96	13.11	11.89	9.44
mnw_r	10.43	12.26	8.83	12.34	13.91	13.42	13.28	10.84
mnd_r	7.10	8.03	5.83	6.68	6.99	8.34	4.14	3.72
mnd_I	7.94	5.00	6.28	8.62	7.20	8.22	7.46	3.44
MA Calc	0.52	0.36	0.35	0.46	0.31	0.33	0.21	0.31
MA	58.87	69.11	69.23	62.43	72.04	70.78	77.66	72.13



Appendix D. List of individuals and associated measurement values

id	667A	656A	1235A	2846A	1469A	1327A	854A
sex	М	F	M	F	F	М	М
Group	G 4	G 4	G 4	G 4	G 4	G 4	G 4
AGE	2	2	2	2	2	3	3
mbs_bg	51.4	56.23	58.65	63.60	68.26	63.2	66.6
mbs_bag	46.5	51.79	54.99	58.44	60.41	58.6	57.9
mfbw	29.5	33.85	33.63	33.02	36.74	35.9	36.8
mf_ibl	6.1	6.86	6.49	6.40	8.28	6.5	6.5
mf_ibr	5.8	5.37	8.50	7.01	8.60	8.4	8.2
mbl_l	44.9	47.51	52.79	50.66	53.84	52.6	59.3
mbl_r	42.8	48.61	54.75	50.66	55.75	56.0	56.9
IIm_I	61.9	68.68	72.37	71.70	78.79	78.1	83.7
IIm_r	62.7	69.39	73.18	72.50	79.57	78.7	83.0
mw_I	21.8	22.87	21.97	22.70	28.44	21.5	28.0
mw_r	22.6	25.92	22.47	23.50	28.12	23.1	28.1
mnw_I	10.9	13.82	11.47	13.59	16.28	11.7	17.4
mnw_r	12.0	13.34	13.41	12.40	17.21	13.0	20.1
mnd_r	3.8	10.37	4.41	5.75	10.90	7.3	8.4
mnd_I	4.6	10.84	7.31	8.31	10.14	7.3	5.4
MA Calc	0.3	0.32	0.41	0.21	0.22	0.3	0.3
MA	71.8	71.60	66.07	77.75	77.03	71.1	69.9