Universidade de Lisboa

Faculdade de Motricidade Humana

Partial Scanning Techniques to Assess Body Composition in Broad Individuals using DXA

A Validation Study for Hologic Explorer-W densitometers

Dissertação para obtenção do Grau de Mestre em
Exercício e Saúde

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Abstract

Background/Objectives: Dual-energy X-ray absorptiometry (DXA) is a standard technique for whole-body composition analysis with a known limitation: the table dimensions of DXA scanners prohibit the assessment of broad individuals. Newer DXA instruments have an extended active scan area, but these devices are still unavailable in most research facilities and clinics. To solve this methodological gap in the existing equipment, the aim of this study was to validate and compare partial scanning techniques to assess whole-body composition of broad individuals in Hologic Explorer-W densitometers.

Subjects/Methods: The sample consisted of 198 participants (27.8 ± 10.1 yrs; 61% women), including normal weight, overweight and obese non-athletes and athletes (body mass index, BMI: 17.0 – 40.1 kg/m^2). A single scan was performed in an Hologic Explorer-W fan-beam densitometer in each participant according to standard procedures. The whole-body scan was analyzed to obtain estimates for the reference procedure. The same scan was reanalyzed to obtain estimates for three partial scanning techniques: RSU) the left upper limb is set equal to the right side limb; RSUL) the left upper and lower limbs are set equal to the right side; HS) an half-scan is taken from the right side of the body and the contralateral side is set equal to it. Bone mineral content (BMC), lean soft tissue (LST) and fat mass (FM, %FM) were considered. Multiple regression analysis, mean group comparison, linear regression and agreement analysis, including the inspection of the concordance coefficient of correlation (CCC), were performed for the BMC, LST, FM and %FM compartments.

Results: RSU was the best performing strategy of this study. The estimates for BMC, LST, FM and %FM from RSU were significantly different from those of the reference whole-body scans, though differences were small (0.010 kg, 0.172 kg, -0.026 kg and -0.10% for BMC, LST, FM and %FM, respectively). The alternative procedures explained more than 99% of the variance of the reference scan with low limits of agreement (RSU: -0.010 to 0.031 kg, -0.109 to 0.453 kg, -0.219 to 0.167 kg, and – 0.35 to 0.15% for BMC, LST, FM and %FM, respectively). The CCCs were greater than 0.99 for all compartments.

Conclusions: Regardless of BMI, athletic status and gender, partial scanning techniques are valid and simple solutions to be used in individuals broader than the DXA scan area. RSU is the recommended technique, followed by RSU. However, individual errors for BMC and LST may be higher in athletes engaged in lateral dominant sports practice.

Keywords: DXA; body composition; athletes; obesity; partial scan.

Resumo

Background/Objectivos: A densitometria radiológica de dupla energia (DXA) é uma técnica *standard* para avaliação da composição corporal a nível de corpo inteiro com uma limitação identificada: as dimensões da área de *scan* dos densitómetros impedem a sua utilização em indivíduos largos. Apesar da largura dos equipamentos ter sido estendida em versões recentes, os mesmos ainda não estão disponíveis na maioria dos laboratórios e clínicas. Reconhece-se por isso pertinência no desenvolvimento de abordagens que solucionem esta limitação metodológica nos densitómetros existentes, sendo o objectivo do presente estudo a validação e comparação de técnicas de *scanning* parcial para avaliação da composição corporal de indivíduos largos e em densitómetros QDR Explorer-W.

Sujeitos/Métodos: A amostra consistiu em 198 sujeitos (27.8 ± 10.1 anos; 61% mulheres), incluindo atletas e não-atletas com peso normal, excesso de peso e obesidade (índice de massa corporal, IMC: 17.0 – 40.1 kg/m^2). Cada sujeito foi sujeito a uma avaliação de corpo inteiro num densitómetro Hologic Explorer-W (*fan-beam*) de acordo com procedimentos estandardizados. Os *scans* de corpo inteiro foram analisados para obter as estimativas de referência, e foram novamente analisados para obter estimativas para três técnicas de *scanning* parcial: RSU) assume-se que o membro superior esquerdo é igual ao direito; RSUL) os membros superior e inferior esquerdos são considerados iguais aos direitos; HS) assume-se que a totalidade do lado esquerdo, seccionado pelo plano sagital, é igual ao lado direito. As variáveis de interesse do estudo foram conteúdo mineral ósseo (CMO), massa isenta de gordura e osso (MIGO) e massa gorda (MG, %MG). Para cada uma destas, efectuou-se uma abordagem validativa contemplando comparação de médias de grupos, regressão linear e análise de concordância, incluindo o coeficiente de concordância da correlação (CCC) .

Resultados: RSU foi a técnica com melhor desempenho. As estimativas para CMO, MIGO, MG e %MG por RSU foram significativamente diferentes dos valores de referência, ainda as diferenças tenham sido pequenas (0.010 kg, 0.172 kg, -0.026 kg e -0.10% para CMO, MIGO, MG e %MG respectivamente). As técnicas alternativas explicaram > 99% da variância dos *scans* de referência, com baixos limites de concordância (RSU: -0.010 to 0.031 kg, -0.109 to 0.453 kg, -0.219 to 0.167 kg e – 0.35 to 0.15% para CMO, MIGO, MG e %MG respectivamente). Os CCCs foram > 0.99 para todos os compartimentos e %FM.

Conclusão: Independentemente do IMC, tipo de prática desportiva e género, a técnicas de *scanning* parcial são soluções válidas e simples para avaliar indivíduos largos em densitómetros QDR Explorer-W. RSU é a opção recomendada, seguida por RSUL. No entanto, os erros individuais para BMC e MIGO poderão ser superiores em praticantes de desportos pautados pelo uso preferencial de membros dominantes.

Palavras-Chave: DXA; composição corporal; atletas; obesidade; partial scan.

Acknowledgements

> *It's good to have an end to journey toward; but it is the journey that matters, in the end.*
>
> — **Ernest Hemmingway**

Every life tells a story and the chapters included in this thesis are part of mine. However, this report is far from accurately portraying the valuable experiences and feelings that being a student at the Faculty of Human Kinetics is also about. This journey was as much about memorable people and moments as it was about science-related matters, since the very beginning. My gratitude is extended to the following individuals for their support:

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[1]Professor Analiza Silva's Public Defense / "Provas de Agregação", 2014, Faculty of Human Kinetics, Lisbon University, Portugal.

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List of Acronyms

3C 3-Compartment Model

4C 4-Compartment Model

ADP Air Displacement Plethysmography

BW Body Weight

BMC Bone Mineral Content

BMD Bone Mineral Density

BMI Body Mass Index

CCC Concordance Correlation Coefficient

CV Coefficient of Variation

DA Directional Assymetry

DXA Dual Energy X-ray Absorptiometry

FM Fat mass

GE General Electric

HS Half-Scan

ISAK International Society for the Advancement of Kinanthropometry

ISCD International Society for Clinical Densitometry

LDSP Lateral Dominant Sports Practice

LST Lean Soft Tissue

MRI Magnetic Resonance Imaging

ROI Region Of Interest

RSU Reflexion Scan – Upper Left Limb Excluded

RSUL Reflexion Scan – Upper and Lower Left Limbs Excluded

SD Standard Deviation

SEE Standard Error of Estimation

TEM Technical Error of Measurement

WB Whole Body

Introduction

1

> *An obese man is dead after he refused to have an X-ray taken in a machine for zoo animals because he was too large for one at a Hamburg hospital (. . .) Because [Thomas Lessmann, 230 kilos] couldn't fit into the hospital's X-ray machine, doctors there recommended that he go to the nearby Hagenbeck animal park (. . .) "It sounded as if they wanted to mock us," his wife Petra Lessmann told the paper.*

> — **The Local (2009)**

Because DXA was primarily developed for the diagnosis of osteoporosis, the need for scanning and analyzing broad subjects was not foreseen. Although the scanning width of most DXA machines (60–69cm) is appropriate for the elderly, it makes it impossible to evaluate individuals whose width exceeds the active area of these systems. This size constraint can be problematic when one needs to assess broad individuals that would benefit from accurate body composition analysis, namely the obese or muscular athletes, examples of which are bodybuilders and rugby players (Olds, 2001; van Marken Lichtenbelt et al., 2004). Moreover, in research studies whose purpose is to characterize a population and/or to develop population-specific field body composition equations, the exclusion or improper evaluation of individuals that are too wide for the table of the equipment is unacceptable. The general and athletic population's trend of increasing body proportions exacerbates this problem even further. In order to overcome DXA scan width limits for broad subjects, manufacturers developed a system whose width limits are broader and whose weight limit is higher (GE Helthcare, Inc., 2012). However, in many laboratories and clinics, this equipment is not available. Alternatively, partial scanning techniques have been proposed and validated. Such approaches do not attempt to measure body

region(s) that do not fit into the active scan area of the densitometers; instead, the missing region(s) are set equal to the symmetrical one(s) at the contralateral side. More complex approaches based on partial scan summation —thereby requiring more than one scan— have also been proposed and validated in athletic populations (Misic & Evans, 2006; Nana et al., 2012). However, the need for more than one scan per subject comes at the cost of increased evaluation times and also added complexity for scan analysis. These drawbacks limit the practical interest of summation techniques and make it an unattractive option in comparison to their partial scanning counterparts.

Attempts to measure broad subjects in normal size DXA systems were pioneered by Tataranni & Ravussin (1995). The authors proposed an half-scan strategy that partitioned the body in two halves according to a sagittal line positioned along the middle of the body. In this manner, the whole-body measurements were estimated by doubling the analysis results of just one side of the scan. With the advent of the iDXA densitometer (GE Lunar Medical Systems, Madison, WI), these restraints have been shifted upward, thereby making it possible to perform complete whole-body scans in broader and heavier subjects. However, there are individuals who keep exceeding the table limits of the iDXA and for whom partial scanning techniques are still appealing options; this reason explains why Lunar densitometers include an half-scanning analysis mode in the iDXA devices. In contrast, Hologic devices implement a "reflection" technique that only needs to exclude left limb(s) from the scan (Hangartner et al., 2013; Sherman, 2011). Despite being already available for automated use in commercial DXA analysis programs, validation studies on this topic are scarce. For reflexion and half-scanning, only one abstract and three papers were published that support the use of partial scanning in broad populations. However, some studies would have benefited from higher sample sizes so as to account for the variability of certain populations's physiques, including muscular athletes. Also, by the time results were published, the algorithms implemented for image acquisition and analysis were disclosed and not standardized across manufacturers (Shepherd et al., 2012a). Therefore, the external validity of such studies is not guaranteed for other equipments (and even software versions) than the tested ones.

Original contributions The methodological gaps mentioned motivated us to tackle the issue of individuals that exceed the scan width of Hologic Explorer-W densitometers, while taking the possible implications of gender and sports practice into account. Specifically, this investigation addressed partial scanning based techniques for determining whole-body composition variables. Using a dataset comprising DXA scans from adult athletes and non-athletes of both genders, the estimates derived from partial scans were validated against reference (whole-body) scans. The central theme, present throughout this investigation, was the validation and comparison of two partial scanning approaches: half- and reflexion-scanning. These techniques are similar in the sense that they substantiate its inferences on missing body regions by side-to-side symmetry. However, they are conceptually different in the anatomical regions required for extrapolation to the contralateral side; whereas half-scanning requires extrapolation of body composition data from one "half-side" to another, reflexion-scanning only requires the estimates for the right limb(s) to be "reflected" to the left side.

Published abstract The present investigation resulted in one poster communication: Moço, A., Matias, C. N., Santos, D. A., Sardinha, L. B., & Silva, A. M. (2014). A Partial Scanning Technique for the Assessment of Broad Individuals using DXA. The 12ᵗʰ International Symposion on Body Composition (ISBC), Cascais, Portugal.

Thesis Structure This thesis comprises six chapters, including this introduction. Chapter 2 sets the context for the application of DXA in the field of body composition, introducing the molecular level of analysis. Then, the physical principles and technological advances of the DXA methodology are highlighted, as well as its inherent limitations. A discussion follows on the validity and precision with which DXA systems determine body compartments in different populations. The chapter also justifies the need for partial scanning techniques to evaluate broad individuals in DXA machines, exploring where a contribution to knowledge may be situated. In Chapter 3, the methodological approach to this research project is described and explained. Firstly, considerations are made regarding how DXA scans were collected. Then, this chapter describes the partial scanning techniques that this thesis will address and describes the statistical approach that was applied to its validation. The main research findings are presented in Chapter 4, including validation results. In Chapter 5, results are discussed and limitations are identified. To conclude, Chapter 6 highlights the main findings and recommends further research.

Dual Energy X-Ray Absorptiometry

2

> Users do not care about what is inside the box, as long
> as the box does what they need done.
>
> — **Jef Raskin**
>
> about Human Computer Interfaces

This chapter starts by providing a theoretical background on the field of body composition research, with an emphasis on the molecular model of dual energy X-ray absorptiometry (DXA). Fundamental physical principles and assumptions underpinning the application of DXA for whole-body composition assessment are discussed in Sections 2.2 and 2.3. Since validity and precision are paramount to the clinical value of body composition methods, these concepts are addressed in Sections 2.4 and 2.5. In particular, the focus is on the performance of DXA systems to determine body compartments in different populations. Then, Section 2.6 revises the literature on partial scanning solutions to overcome the width limit of DXA instruments. A brief discussion on morphological asymmetry was also included, hoping that it contributes to a better understanding of the limitations of the partial scanning techniques discussed throughout this thesis. The final portion of this chapter is devoted to the objectives of this investigation.

2.1 Background

The study of human body composition is directed towards the *in vivo* quantification of body components, their connections and also changes in these components as a response to influencing factors such as nutrition, exercise, growth, development, aging and selected diseases. Body composition variables, examples of which are percent fat (%FM), muscle mass and bone content, have been proven effective in diagnosing and/or stratifying the risk of numerous health conditions, including cardiovascular disease, diabetes, certain types of cancers, osteoporosis and osteoarthritis (Sardinha & Teixeira, 2005). The field of body composition can be subdivided into three distinct, interconnected areas: body composition rules, methodology and biological factors that influence body composition

(Heymsfield et al., 1997). The body composition rule area organizes body components into distinct levels of increasing complexity, where the higher levels are obtained by progressive subdivision of body mass into more compartments (Wang et al., 1992). As shown in Table 2.1, five levels were proposed: atomic, molecular, cellular, tissue system and whole-body. Each level refers to specific compartments and measurement techniques, examples of which are also provided in Table 2.1.

Table 2.1: 5-Level Model of body composition research: components and examples of methods used to their determination [APD, Air displacement plethysmography].

LEVELS	COMPONENTS	METHODS
Atomic	O, C, H, other (N, Ca, P, K, Na, Cl)	*In Vivo* Neutron activation analysis
Molecular	Lipids, Water, Proteins, Glycogen, Minerals	Multicompartment models (DXA, ADP, Tracer dilution), DXA
Cellular	Intracellular water Extracellular Fluid and Solids	Bioimpedance analysis Dilution techniques
Tissue system	Adipose tissue, Skeletal muscle Visceral Organs & Residual, Skeleton	Computerized axial tomography, Magnetic Resonance Imaging, Ultrasound
whole-body	Weight, BMI, Skinfolds	Anthropometry

The 5-level model provides the opportunity to clearly define the concept of *body composition steady state*. Although each level and its multiple compartments are distinct, unless the homeostasis of the body is interrupted, the assumption of stability implies that biochemical and physiological connections exist, namely constant (or relatively constant) relations between components across time, within an individual and between different individuals. This concept is important to the application of methods for determining body composition, since an unknown component can be estimated with another component or measured property. An example is the application of DXA in body composition, where the characteristic attenuation of X-rays by different tissues to low and high energy photons make it possible to estimate its composition.

If the needs for reliable, easy-to-get and clinically relevant data were key reasons for research in the field of body composition, the advent of new technologies provided the opportunity to accomplish so. The result is an exponential increase in the amount of publications on body composition over the last 60 years, most of them addressing methodological issues (Sardinha, 2012). Heymsfield et al. (2005) define body composition methodology as "an area of investigation dedicated to the study and application of methods used to quantify body components from atomic to whole-body levels". The

in vivo quantification of body components and their quantitative variation in response to various influencing factors such as nutrition, exercise, growth, aging and selected diseases is an imperative aspect of contemporary practice of health, nutrition, exercise and sports related professionals (Ellis, 2000; Heymsfield et al., 1997; Pietrobelli et al., 2001). Despite having reached a "mature state" (Heymsfield et al., 2005), this field is still not without unmet needs, particularly as it pertains to the assessment of individuals with extreme phenotypes (Santos et al., 2014). The obese and athletes are among such paradigmatic examples that still present technical challenges.

Increasingly obesogenic environments render difficult the adoption of healthy lifestyles, the consequence being the fast growing prevalence of obesity among the world population. Obesity is a serious threat to public health, and is even regarded by the World Health Organization as an epidemic in countries all over the world due to its associations with chronic disease and loss of overall health (Chopra et al., 2002). An implication of this scenario is the fact that the assessment of body composition of the obese has become commonplace in the clinical setting and also in research facilities. On the one hand, simple indicators like Body Mass Index (BMI) and waist circumference have proven to be effective indicators of health with functional implications to metabolic and cardiovascular disease stratification (WHO Working Group, 1986). On the other hand, more powerful imaging-based examinations have led to valuable additional insights for health status assessment, as are the adipose tissue distribution and the diagnosis of states like sarcopenia, osteoporosis or a combination of both (Ergun & Rothney, 2012). The urgency of effective counteracting measures justifies active research on the development of strategies on diet, physical activity, exercise adherence and health. Research on these topics often require samples of obese individuals and the accurate assessment of their body composition is paramount to the quality of the study designs (Silva et al., 2008).

Because sports performance is significantly affected by body composition, and because training induces body composition changes, there is also considerable interest in the evaluation of body composition in sport (Ackland et al., 2012). Applications include the assessment of the progress and effectiveness of nutrition and/or training interventions which aim to assess fat or lean mass changes, and, to a minor extent, the characterization of athletic populations and validation of body composition field methods (Burke & Deakin, 2010; Harley et al., 2011). Although frequently used in general populations, BMI alone or even bioimpedance based methods are not suitable to characterize body composition in

athletes, as their accuracy is compromised by deviations from the reference population and the individual errors may be unacceptably high (Burke & Deakin, 2010). Anthropometry filled this methodological gap and established itself as the most popular field method to assess body composition method in athletes, with 67% of the professionals surveyed by the International Olympic Committee reporting its use (Meyer et al., 2013). However, this approach requires specific population equations that convert skinfold thickness values into whole-body percentage fat mass and these can only be developed based on criterium methods, preferably the four compartment model (4C).

While the 4C model has been recommended for its accuracy and precision, there are drawbacks that prohibit its implementation in clinical settings and even in some research designs. The requirements for more equipment, time and tester expertise make it advisable to look for alternatives that can also deliver "criterium" level performance while allowing routine assessment of body composition. In this regard, imaging techniques are the prime candidates, as they measure several independent compartments accurately in just one evaluation moment. Among them, the only imaging methods that can accurately estimate clinically relevant compartments are DXA, computed tomography (CT) and magnetic resonance imaging (MRI). "However, DXA is low dose in comparison to whole-body CT scanning and inexpensive compared to MRI" (Shepherd, 2014), thereby justifying its relevance in body composition assessment. Time efficiency is an additional advantage of the DXA method that also contributed to its use in research facilities, clinics and even large multicenter studies, including the National Health and Nutrition Examination Survey (Centers for Disease and Control, 2000a).

DXA at the molecular level of analysis   Figure 2.1 depicts the DXA model. According to the conceptual framework of Wang et al. (1992), the DXA methodology is at the molecular level of analysis and provides simultaneous estimates for fat mass (FM), lean soft tissue (LST) and bone mineral content (BMC).
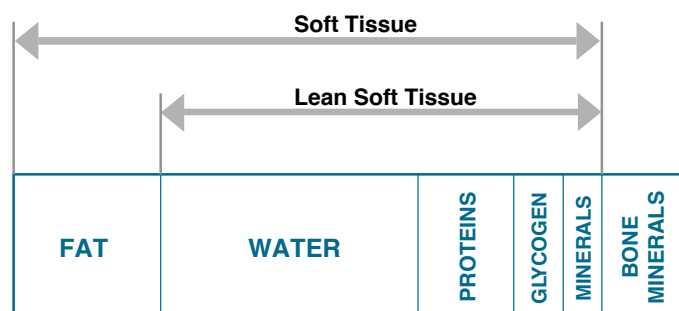
Figure 2.1: DXA three compartment (3C) molecular model comprising fat, lean soft tissue and bone minerals. Adapted from Pietrobelli et al. (1996).

Because water is not explicitly solved for in the DXA 3C model, the equipment assumes that 73.2% of the free-fat mass (FFM) is water. However, concerns emerged as to whether and to what extent variation in soft tissue hydration could cause errors in fat estimates, particularly in altered hydration scenarios, where changes in hydration could be incorrectly confounded with changes in lean tissue (Pietrobelli et al., 1998). In a revision of five studies on changes in water content of FFM and its effect on %FM, including Pietrobelli et al. (1998), Lohman et al. (2000) suggested that a 5% change in the water content of FFM introduces small but systematic errors in whole-body %FM in the order of 1 – 2.5%. Though to a minor extent, fluid balance changes may also impact on the measurements the adipose tissue itself, since it comprises $\approx$ 10 % water (Fuller et al., 1992; Wang & Pierson, 1976). The actual hydration of the FFM in healthy populations is in between $\approx$ 72 – 74.5%, and it may indeed vary slightly from the assumption of 73.2%. However, the impact of any over- or underestimation of these proportions is expected to have a reduced effect in the overall DXA results (Lohman et al., 2000).

2.2 Physical Principles

Although densitometers differ in architecture and specific configurations (see Section 2.3.1 for details on pencil vs. fan beam based architectures), they share the same physical principles. Contrast between tissues in X-ray images arises from differential attenuation of the X-rays as they pass from the source through the body to the detector. Typically, in a DXA scan, the energy source produces photons at "low" and "high" energy levels, which pass through tissues and attenuate at rates related to their elemental composition. The specific energy levels differ between manufacturers; the X-ray tubes of the Hologic Inc. densitometers (Bedford, MA, USA) emit switched pulses at 100 kVp and 140 kVp[1] to generate spectra with maximum photon energies at 45 and 100 keV. GE-Lunar Inc. (GEHealthcare, Madison, WI, USA) and Cooper Surgical (Norland; Trumbull, CT, USA) take a different approach: they use an X-ray source at constant voltage and a K-edge filter to produce stable beams of X-rays at energies 40 keV / 70 keV and 40 keV / 80 keV, respectively (Hull et al., 2009; Lohman & Chen, 2005; Pietrobelli et al., 1996).

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[1]The peak kilovoltage (kVp) of the tube determines the amount of radiation that is delivered by the tube (Bonnick & Lewis, 2006). Currently, the peak X-ray tube voltages used to generate the dual-energy images for the Hologic systems are different between their current fan-beam systems (140 / 100 kVp) and previous pencil-beam models (140 / 70 kVp) (Fan et al., 2010).

X-ray attenuation is often characterized in terms of mass attenuation coefficients, defined as the linear attenuation coefficient divided by the density of the tissues. Owing to its significant amount of highly attenuating minerals (calcium and phosphorous), bone has a higher mass attenuation coefficient than soft tissues, whose main components are oxygen or carbon. The unique elemental profiles of BMC, FM, and LST allow for separate analysis of these components. Bone is readily distinguished from soft tissues (fat and lean tissue), which are mainly organic compounds (Pietrobelli et al., 1996). Fat is described chemically as the lipids in our body and consists mostly of fatty acids and triglycerides (Pietrobelli et al., 1996; Shepherd, 2014). The LST compartment includes body water, protein, soft tissue mineral mass (i. e., non bone mineral mass) and glycogen. The amount of glycogen in the body is neglected in the DXA model, as the overall amount stored in the liver and muscle tissue accounts for just 1 kg of body weight in most individuals (Heymsfield et al., 1997).

Based on the above properties, the DXA methodology assumes that BMC, LST and FM can be distinguished based on its characteristic R-values, defined as the ratio-of-attenuation to low and high incident photons. However, solving for three unknown components with just two known R-values (low and high photon energies sources) is an ill-posed problem. Consequently, DXA cannot estimate soft tissue composition in bone containing pixels as it can only solve for two materials simultaneously (either lean + fat mass or bone + soft tissue mass). This issue is addressed by pre-segmenting the image into bone-containing pixels and pixels containing soft tissues only. This stage of processing is error prone as, for the bone-containing pixels, it is necessary to assume that the soft tissue it contains can be estimated from surrounding pixels. Algorithms can accomplish this task by simply taking the average of the surrounding soft tissue composition or through more complex approaches that also account for adipose and skeletal muscle distribution (Nord & Payne, 1995; Pietrobelli et al., 1996). When there is not adequate soft tissue surrounding bone regions, the accuracy of the estimates may become compromised, particularly at head, hands, feet, and upper torso. In these cases, manufacturers turn to proprietary methods to reference the soft tissue (IAEA, 2010). As an example, Hologic APEX v3.3 for the QDR series assumes that the head contains 17% fat tissue. Since bone is typically contained in 40% or more of the body image pixels and algorithms are not standardized across manufacturers, it is suggested that estimation of soft tissues in high bone-containing

areas may be inaccurate, and when results from different software versions are compared with one another, results may be different (IAEA, 2010). Even still, when the devices are properly calibrated, the validity of DXA is generally acceptable in comparison with the 4C model, in different populations and devices (see Section 2.4 for details).

2.3 Technological advances

In a revision on the evidence regarding the trueness and precision of DXA body composition measurements, Toombs et al. (2012) highlighted that the technological advances resulted in enhanced precision, large availability and low radiation dose, thus turning DXA into a more convenient and useful diagnostic tool for body composition assessment. In this regard, the improvements in beam technologies (Section 2.3.1) and the possibility to assess body composition in broader patients (Section 2.3.2) merit special consideration.

2.3.1 Beam technology

Advances in DXA technology have resulted in the progressively replacement of pencil- by fan-beam densitometers. Pencil-beam systems scan anatomical sites in a rectilinear fashion, whereas fan-beam densitometers use a fan-beam X-ray source and a multiple-element detector array so that the measurement of the whole-body can be made with a motored table and sweeps of a "C"-shaped X-ray arm (Toombs et al., 2012). Not surprisingly, scanning times for fan-beam beam systems are relatively faster that their pencil-beam counterparts ($\approx$ 5min vs. 10–20 min for total body, previously) (Barthe et al., 1997; Tothill et al., 2001). Examples for each category include:

Pencil-Beam
- Hologic QDR-1000/W and QDR-2000-pencil;
- GE Lunar's DPX and DPXL series.

Fan-Beam
- Hologic QDR-2000 fan, QDR-4500 A/W;
- Hologic QDR Explorer, Discovery W/A.

(Narrow) Fan-Beam
- GE Prodigy, PDX-NT and MD+;
- GE iDXA.

Currently, the "Hologic QDR" Series of densitometers employ fan-beam X-ray sources. This family includes the Hologic Explorer[2], which is depicted in Figure 2.2 alongside its main hardware specifications.

Detector System
Multi-element detector array

Bed Table
Scan Region: 196 x 65 cm
Weight Limit: 136 kg

Scanning Method
Linear X-ray fan-beam
(Motorized table and C-arm)

X-ray System
Switched-pulse dual-energy
(100 kVp/140 kVp)
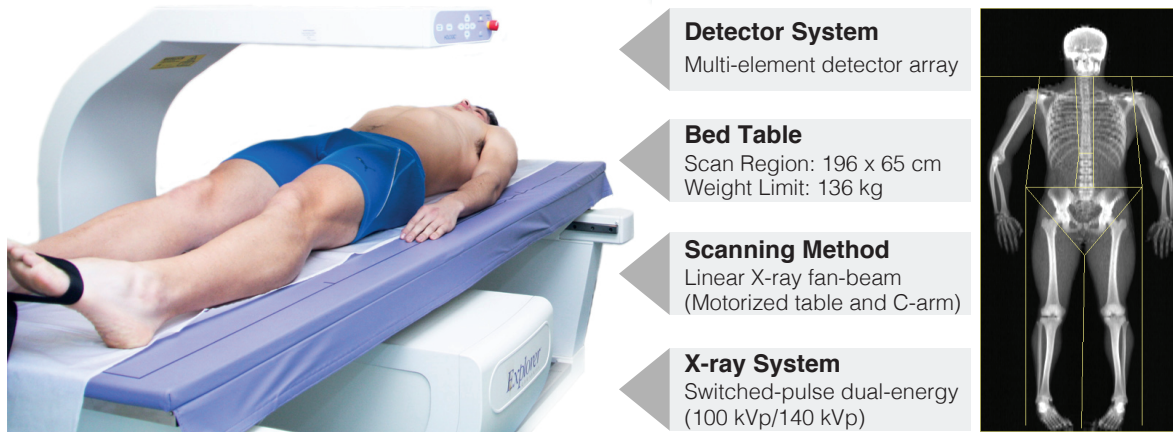
Figure 2.2: Main hardware specifications of Hologic Explorer-W. Explorer-W is a fan-beam DXA system whose width limit is 65 cm (Hologic, Inc., 2014).

During image acquisition, the X-ray tube emits switched-pulses at two energies (100 kVp/140 kVp). The overall scanning time in enhanced whole-body mode takes approximately 7 min for a radiation dose of 12 μSv, and the resulting (whole-body) images achieve a spatial resolution of 2×2 mm^2. The major outcomes for body composition analysis are BMC, LST and fat mass (absolute value and percentage). As for the regional analysis, relevant regions include limbs, trunk and head, although subregions within the hip and abdomen may also be reported for obesity research (Hologic, Inc., 2014).

Despite superior spatial resolution (0.8–2 mm for fan-beam densitometers vs. 1.5–2.5 mm for pencil-beam densitometers), fan-beam systems may cause *magnification* errors relative to the height of the subject above the scanning table. This effect implies that the higher the body structure of the patient with respect to the radiation source, the smaller the projected area, and it has been shown to affect bone and soft tissue measurements (Blake et al., 1993; Ellis & Shypailo, 1998; Pocock et al., 1997). However, manufacturers were still able to correct for this effect with software upgrades (Griffiths et al., 1997). In the validation study of Visser et al. (1999), FFM was not affected by magnification effects with Hologic QDR 4500 (fan-beam, software version 8.21). GE Healthcare addressed magnification effects with the introduction of narrow fan-beam densitometers, the first of which was

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[2]The software for body composition assessment with whole-body and regional analysis are optional upgrades to the standard QDR Explorer configuration. In this thesis, we use the notation "Hologic Explorer-W" to mean "Body composition with subregional composition analysis" was available in the existing densitometer of the Faculty of Human Kinetics, Lisbon University, Portugal (Hologic, Inc., 2014).

the GE Lunar Prodigy (GE Healthcare, Madison, WI). Narrow fan-beam densitometers scan in a rectilinear fashion with a fan beam that is broader than the original pencil-beam systems but still narrower than the fan-beams.

The most recent improvement in beam technology was led by GE Healthcare with the introduction of the GE Lunar iDXA (GE Helthcare, Inc., 2012). Owing to multiar-ray detectors with higher dimensions, this narrow-angle fan-beam densitometer provides improved spatial resolution (1.05×0.6 mm^2), thereby allowing for superior bone segmen-tation and overall body composition assessment compared to earlier systems.

Considerations on radiation dose Dose is the amount of ionising radiation energy absorbed by the body. The dose required for a DXA whole-body composition examination differs, depending on the manufacturer, model and software configurations. Although the radiation doses incurred during fan-beam DXA imaging are higher than with pencil-beam systems, the overall range is still low ($\approx 0.37 - 4.7 \mu Sv$), though values up to ≈ 28.3 μSv may be required to scan thicker subjects (Shepherd, 2014; Silva et al., 2013). For comparison purposes, the average dose due to natural background radiation is 6.2 μSv (EPA, 2012), thus corroborating the fact that DXA methodology is classified as low dose and minimally invasive. But, although small, dose still makes it unsuitable to pregnant women or for regular monthly examinations (Hangartner et al., 2013; IAEA, 2010).

2.3.2 Assessment of broad individuals

In parallel to the technical developments that have resulted in improved image resolution and faster scan times, there is also an ongoing trend for more robust devices that can accommodate broader and heavier patients. This problem demanded bed tables with extended limits, partial scanning solutions, and also algorithms and/or scanning modes that compensate for the deterioration of the image quality as the patient's thickness increases (with respect to the anterior-posterior axis).

DXA Scan area The DXA table area dimensions prohibits its use in individuals with extreme phenotypes. When an individual's body dimensions exceed these limits, typically in obese patients, the accuracy of the measurements may be compromised. The problem of assessing athletes with larger trunk breadth and musculoskeletal development falls in the same category. Recognizing an emerging need to measure body composition in obese

individuals, manufacturers have been designing DXA tables with extended scan areas and higher weight limits. Table 2.2 lists the weight limits and scan regions of commonly used densitometers for whole-body composition assessment.

Table 2.2: Weight limits and scan regions of densitometers for various manufactures and models. Adapted with permission from Silva et al. (2013).

EQUIPMENT	WEIGHT (kg)	SCAN AREA (cm^2)
GE iDXA	204	197.5×76
GE Lunar Prodigy Advance	159	197.5×60
GE Lunar Prodigy	136	197.5×60
GE Lunar DPX-MD	136	196.8×57.6
Hologic QDR Explorer	136	195×65
Hologic QDR Discovery A/SL/W	159	195×65
Norland XR-46	114	193×64
Norland XR-26	114	193×64

It is apparent from Table 2.2 that Hologic provides only up to 65 cm in the QDR devices. However, at least since the QDR Explorer/Discovery series were created, the analysis results of limbs that could not be scanned could still be automatically copied from the contralateral side(s). GE iDXA provides the highest weight limit and a broader scanning space; the bed table of the iDXA can span 76 cm so that individuals can be positioned off-side to perform a [right-side] half-scan. Is is also apparent from Table 2.2 that the assessment of subjects taller than 193–204 cm can be a problem. To address this methodological gap, summation (head plus subtotal without head or more scans) and patient positioning techniques (knees bended) have been proposed (Evans et al., 2005; Misic & Evans, 2006; Nana et al., 2012; Silva et al., 2004).

Beam hardening As body thickness is increased, DXA may overestimate %FM (Prior et al., 1997; van der Ploeg et al., 2003), while also affecting bone measures (Blake, 1992; Laskey et al., 1992). This phenomena is the ultimate consequence of what is referred to in the literature as "beam hardening effect"; this term is used to denote the artifacts that result from the preferential loss of lower energy photons relative to high-energy photons. As a result of increasing body thickness, the ratio between low and high energy gets distorted, thereby affecting the estimation of the three DXA compartments (Webb, 2003). A practical implication is that body composition results may be systematically different between a thin and an obese person (e.g., lean endurance athlete vs. rugby player) (Prior et al., 1997). Hologic and GE Lunar claimed to have addressed beam-hardening by performing

software upgrades that correct for tissue thickness and/or by providing special scan modes for thick patients (Lohman & Chen, 2005):

GE Healthcare

- Lunar DXP, DPX | v3.4R, v3.6 (v1.3y): Corrections for tissue thickness;

- Lunar Prodigy and PDX-NT and MD+ | v2.16, v3.50: Corrections for total body thickness and increased number of tissue thickness points;

- iDXA | GE Encore 11.10: three scan modes that adjust the X-ray attenuation for the thickness of each patient; automatic half-scanning mode.

Hologic

- QDR-1000 W and QDR-2000 | v5.48–5.54: "Whole-body and enhanced whole-body" and corrections for tissue thickness;

- QDR-4500 A/W | v8.1a–8.26: Corrections for magnification;

- QDR for Windows (Delphi and QDR 4500 A/W) | v11.2: "High-power whole-body released for obesity research".

In the proposed scan modes, the X-ray tube voltage settings remains the same, but the X-ray flux is increased by higher currents or slower scan times. As an example, Hologic currently holds a patent for an invention that optimizes scan parametric values of QDR-4500 systems. These are selected according to the thickness of the patient:

> "An operator initially selects scan parametric values of a fast mode scan which is the recommended default, and then commences the fast mode scan. At the initial portion of the scan, the X-ray thickness of the patient is measured. If the measured X-ray thickness is not greater than a predetermined limit of the fast mode scan, the fast mode scan is continued. If the measured X-ray thickness is greater than the predetermined limit of the fast mode scan, the X-rays are turned off, and the operator is given a choice of continuing with the fast mode scan or restarting with a slower mode scan. If the operator selection is to continue, the fast mode scan is continued. If the operator selects restarting, a slower mode scan is commenced." (Patent US5687211)

By simply detecting when a thresholding value is exceeded, it provides the technologist with an option to do a slow scan instead, thereby preventing beam hardening artifacts to affect the quality of the resulting image (Berger et al., 1997). Since QDR v11.2, Hologic provides a "high-power whole-body" scan mode that should be used if there is a noticeable increase in X-ray noise in the torso region. In this case, the dose is tripled (from 8.5 to 28.3 μSv) (Shepherd, 2014). Recent GE systems will automatically warn the user to the need for the "thick" scan mode if the patient's weight exceeds a particular level and the dose is doubled. The

GE iDXA system has three scan modes that adjust the X-ray attenuation for the thickness of each patient: thin (<13 cm), standard (13-25 cm), and thick (>25 cm). The embedded software (GE Encore 11.10) allows for adjustment of regions of interest including the sagittal line demarcating left from right body sides. In addition, the software automatically detects whether the subject is within the scan space and can be scanned according to standard procedures, or if an half-scanning mode must be activated instead. While more complex approaches are likely be devised in the future, mitigating beam hardening always comes at the cost of more radiation dose to the patient and extended evaluation time.

2.3.3 Lack of standardization

Currently, the three major commercial manufacturers of densitometers are Hologic, GE Medical Systems and Cooper-Surgical, for the QDR series, Lunar & iDXA, and Norland devices, respectively. Some concerns on cross-validation between densitometers of different manufactures arise, as hardware configurations and computer vision algorithms differ across devices (Shepherd et al., 2012a; Toombs et al., 2012). On Section 2.1 it was informed that manufactures apply photoelectric peaks at different energies: this aspect can be outlined as the first evidence for lack of standardization among manufacturers. Different photon energy levels result in different ratio-of-attenuation coefficients (predicted from elemental composition), for the same tissues (Pietrobelli et al., 1996). As a result, the image processing algorithms receive slightly different input images. A second obstacle to standardization is the fact such algorithms are disclosed (Ackland et al., 2012), thus making it likely that images are interpreted differently by different manufacturers and software versions. Although, for bone measures, these aspects can be minimized by cross-calibration with phantoms, soft tissue composition poses an added complexity; at bone containing pixels, soft tissue overlaying bone can only be determined by relying on values of pixels from surrounding tissue. Since bone is contained in 40% or more of the planar whole-body image, the extent of the impact of differences in approaches to tackle missing data can be problematic. Finally, even the recommendations for positioning subjects are slightly different across manufacturers. In the GE iDXA, for example, hands are vertical (midsagittal position) to accommodate broad subjects, whereas the remaining recommend hands prone. With respect to this matter, the International Society for Bone Densitometry (ISBD, http://www.iscd.org/), recommends that the patient's arms are at their sides, palms down, with a separation from the thighs.

However, if patients are large, and as long as there is a space between the patient's arms and sides, the ISBD endorses hands vertically next to the thighs (Hangartner et al., 2013). To the best of my knowledge, the degree to which inconsistencies in different hand positioning practices impact on whole-body composition results has not been throughly investigated.

Although cross-calibration and comparative studies have been undertaken using different devices for the DXA whole-body compartments (Oldroyd et al., 2003; Tothill et al., 2001), until 2010, no attempt was made to standardize BMC (IAEA, 2010). At the time being, only Shepherd et al. (2012b) undertook a standardization study for whole-body composition using GE Lunar and Hologic DXA systems. At the 10th International Symposium on Body Composition, the issue of the lack of standardization was raised by Arthur Stewart during the highlighted session on "The expert view for the future of body composition"[3].

2.3.4 Summary

The DXA methodology has lingering issues that deserve further research and standardization efforts in order to improve accuracy of body composition measurements. Even still, owing to the safety of its procedure for adults, relative inexpensiveness to carry out examinations and capability of providing simultaneous estimates for three clinically relevant body components, DXA has been gaining popularity and is now a standard tool for whole-body composition analysis with recognized relevance for health and sports science (Andreoli et al., 2009; El Maghraoui & Roux, 2008; Kelly et al., 2009; Shepherd, 2014).

Most currently marketed DXA instruments require a low effective radiation dose per scan ($0.37 - 4.7 \mu Sv$), use standardized calibrations and scans are quick ($4 - 17$ min). The ability to analyze scans in an whole-body level or by regions of interest is helpful for studying body fat and lean mass distribution and also regional bone mineral density (e.g. inspection of the android/gynoid regions, bone density at spine) (Ergun & Rothney, 2012). However, caution is advised when using or interpreting results obtained by the DXA methodology, as it has pitfalls and a number of assumptions that raise concerns about the accuracy of this methodology to specific populations, including very lean, obese, elderly populations, and also in altered statuses of hydration (Brownbill & Ilich, 2005; Genton et al., 2006; Scafoglieri et al., 2011). An appreciation of the *validity* of the DXA methodology is, therefore, appropriate and will take place on the next section.

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[3]"The expert view for the future of body composition: from atoms to anthropometry". Contributions by: Timothy G. Lohman, Marinos Elia, Manfred J. Muller, Dale A. Schoeller, Arthur Stewart and Zimian Wang (USA, UK, Germany). International Symposium on Body Composition, Cascais – Portugal, 14th June 2014.

2.4 Validity of DXA

Validity refers to the "degree to which any measurement approach or instrument succeeds in describing or quantifying what it is designed to measure. It reflects systematic or constant errors in measurement" (Weiner, 2007). The most accurate and direct validation technique for DXA is dissection and direct comparison combined with bone ashing (Elowsson et al., 1998). A review of the state of the art of carcass studies related to DXA reveals validation attempts with small to medium size animals mostly based on chemical analysis (Scafoglieri et al., 2011). Although the correlations between the two methods were generally high (Brommage, 2003; Brunton et al., 1993; Lauten et al., 2001; Swennen et al., 2004), skepticism should be exercised when attempting to translate findings on animals to humans. Not only is the distribution of lean and fat mass different but also is anatomy and body size.

2.4.1 Comparison with the criterion technique

Due to the lack of cadaver studies, a four-compartment model (4C) of body composition analysis has been used as the "gold standard" method in validation studies for DXA for body composition (Wang et al., 2010; Withers et al., 1998). The 4C model includes the evaluation of the main free-fat mass (FFM) components (water, bone mineral, protein), thus reducing biological variability. Fat mass is estimated from body volume (generally measured by air displacement plethysmography), while correcting for assumed total-body water and BMC using dilution techniques and DXA, respectively (Wang et al., 2002, 2005). Figure 2.3 illustrates the concept of validating DXA systems against the 4C model.
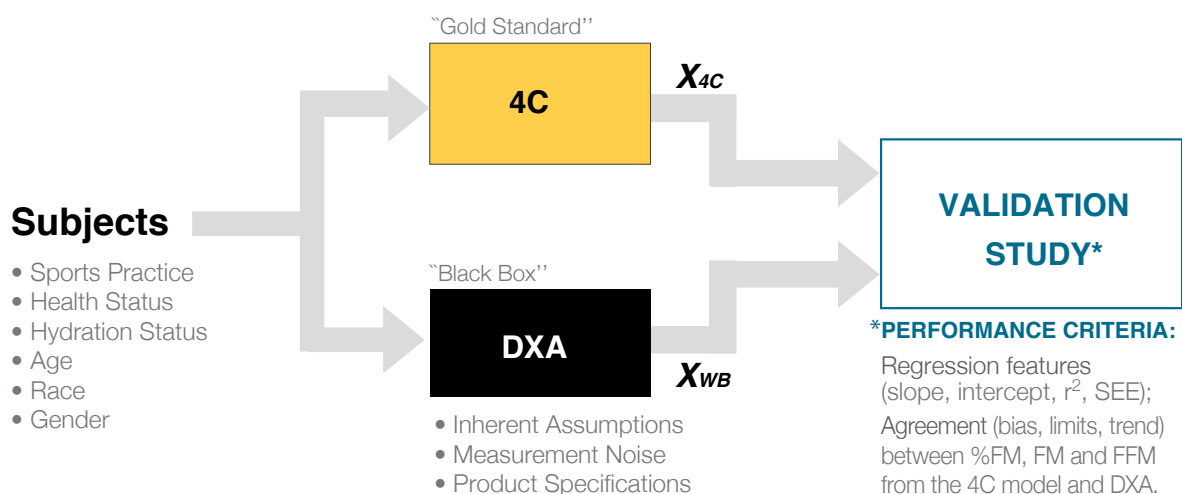

Figure 2.3: Validation of DXA for whole-body composition assessment. The measurements obtained by DXA are compared against the 4C model by statistical procedures that include regression and agreement analysis.

The accuracy is often influenced by features of the subjects (e.g., age, race, gender, health, sports practice, obesity status, biological variation, subject preparation) and by the DXA methodology itself (e.g., inherent assumptions that hold for normal/reference populations). In general, although users may know the physical principals and the general product specifications of a given DXA device, it is not possible to see its inner workings (because it is a closed source program and the hardware details are generally out of the user's expertise). The method to be tested is, therefore, regarded as a "black box" method for which only the overall *performance* needs to be examined (Beizer, 1995). In body composition research, validity testing for DXA is used to check that the body composition results provided by a specific device are as expected, given specific populations. As for the performance criteria, Lohman & Chen (2005) highlighted the inspection of slope, standard error of the estimate (SEE) and the analysis proposed by Bland & Altman (1986): "If two methods have theoretical validity and are properly calibrated to estimate %FM [similar considerations hold for BMC, LST, FM], then the regression line relating the values from the two methods should have a slope equal to 1.0. In addition, a SEE between 2% and 3% and a systematic bias between criterium and DXA methods must be uncorrelated by the mean value" (pp. 71). Widely used performance criteria include mean group comparison, regression features (slope, intercept, coefficient of determination and SEE) and the agreement (bias, limits and trend) between %FM, FM and FFM from the 4C model and DXA (Santos et al., 2010). The following paragraphs provide an overview on linear regression and agreement analysis for validation purposes.

Linear regression analysis Linear regression analysis involves applying the least mean squares algorithm to find the best-fit line to the pairs of points $(x_{DXA,i}, x_{4C,i})$, for $i = 1 \ldots N$ subjects. The outcome is a regression $\hat{x}_{4C,i} = \beta_1 x_{DXA,i} + \beta_0$ such that the errors $\epsilon_i = \hat{x}_{4C,i} - x_{4C,i}$ are minimized in a least mean squares sense, for $i = 1 \ldots N$ subjects. It is desirable that $\beta_1 \approx 1$ and $\beta_0 \approx 0$, meaning that $\hat{x}_{4C,i} \approx x_{DXA,i}$. In this regard, t-tests are useful to confirm that β_1 and β_0 are in fact significantly different from one and zero, respectively. The Pearson's correlation coefficient r and the standard error of the estimate (SEE) are established parameters for evaluating the fit of regression models. For a simple linear regression model, the Pearson's r squared is called the coefficient of determination. It is the fraction of the variation in the values of $x_{4C,i}$ that is explained by least mean squares regression, $\hat{x}_{4C,i}$, on the input values $x_{DXA,i}$, for subjects $i \in 1 \ldots N$. That is, $r^2 \times 100$ (%) is the "amount" by which DXA explains the reference measurements. Magnitude values for r between 0.9 and 1.0 indicate very highly correlated variables, whereas, in the opposite situation, correlation coefficients whose magnitude are less than 0.3 have little or no linear correlation. We can readily see that $0.9 < |r| < 1.0$ corresponds

with $0.81 < r^2 < 1.00$ and $0.0 < |r| < 0.3$ corresponds with $0.0 < r^2 < 0.09$ (Calkins, 2005; Iman, 1994). The concept of SEE is complementary to the Pearson's r. It is the root mean square of the individual errors ϵ_i, $i = 1 \ldots N$, meaning that it can be regarded as a measure of the precision with which the regression coefficients are determined.

Agreement analysis   The agreement between the DXA procedure and the reference 4C model is usually assessed by analyzing the 95% limits of agreement and by plotting the differences against the mean of the methods (Bland & Altman, 1986). The presence of a trend between the differences and the mean of the methods is examined using the coefficient of correlation of the regression on the pairs of points $((x_{DXA,i} + x_{4C,i})/2, x_{DXA,i} - x_{4C,i})$, for $i = 1 \ldots N$ subjects. The statistical significance of the coefficient of correlation (here referred to as trend) is then determined. Some papers report the bias between methods within the framework of the agreement analysis and test if it is significantly different from zero. However, others perform mean group comparison (x_{DXA} versus x_{4C}) instead and only report if the measurements are statistically different (Gately et al., 2003). This is also the case of studies at which validating DXA is an intermediate step, but not the main objective of the study, and the agreement analysis may not be reported (Gallagher et al., 2000).

2.4.2 Validation against the 4C model

Unfortunately, the process of undertaking multi-compartment models is expensive and time consuming (Santos et al., 2010; Wang et al., 2005), the consequence being the relative scarcity of validation studies comparing specific DXA systems and populations with the multi-compartment models (Lohman & Chen, 2005; Toombs et al., 2012). This issue is worsened by the extensive variety of DXA systems currently available, as well as differences in subject phenotypes: features like hardware and software versions, or the obesity and/or athletic statuses of the sample may compromise the extrapolation of validation studies to a particular system and individual. Indeed, an extensive literature review was conducted to select validation studies of DXA against the 4C model, including their detailed validation parameters, but only eighteen studies matched the selection criteria. Full search details are provided below and on the next page.

Protocol, search strategy and selection criteria   The MEDLINE database (PubMed) was searched for English language articles published in peer-reviewed journals, with the last search run on 5 June 2014. The keyword terms included: dual-energy X-ray absorptiometry, DXA, DEXA, 4C, 4-c, four compartment, Hologic, iDXA, Lunar and Norland. The search was solely conducted on abstracts and titles according to Listing 2.1.

```
Search I)
((((((((DXA[Title/Abstract]) OR DEXA[Title/Abstract]) OR dual-energy X-ray
    absorptiometry[Title/Abstract]) OR Lunar[Title/Abstract]) OR iDXA[Title/
    Abstract]) OR Hologic[Title/Abstract]) OR Lunar[Title/Abstract]) OR
    Norland[Title/Abstract]) AND four compartment[Title/Abstract]

Search II)
((((((((DXA[Title/Abstract]) OR DEXA[Title/Abstract]) OR dual-energy X-ray
    absorptiometry[Title/Abstract]) OR Lunar[Title/Abstract]) OR iDXA[Title/
    Abstract]) OR Hologic[Title/Abstract]) OR Lunar[Title/Abstract]) OR
    Norland[Title/Abstract]) AND 4c[Title/Abstract]

Search III)
((((((((DXA[Title/Abstract]) OR DEXA[Title/Abstract]) OR dual-energy X-ray
    absorptiometry[Title/Abstract]) OR Lunar[Title/Abstract]) OR iDXA[Title/
    Abstract]) OR Hologic[Title/Abstract]) OR Lunar[Title/Abstract]) OR
    Norland[Title/Abstract]) AND 4-c[Title/Abstract]
```

The following characteristics and criteria were used:

- Healthy subjects;

- Validation of DXA against the 4C model in assessing FM, %FM, FFM or LST;

- Description of the statistical methods used to validate the procedure.

For the identification of the studies, the process included screening of the identified records, examination of the full text of potentially relevant studies and application of the eligibility criteria to select the included studies. Our search provided a total of 124 citations. Of these, 104 studies were discarded, because, after reviewing the title and abstract, it was apparent that these papers did not meet the criteria or it was impossible to have online access to them. In the end, only a total number of eighteen studies were identified in the review.

The included studies cover detailed validation parameters for %FM (14 studies), FM (5 studies) and FFM (5 studies) and are summarized in Tables 2.3 and 2.4, for %FM and FM/FFM, respectively. Both tables provide information on densitometer specifications, sample size and gender, surrogates of tissue thickness (e.g., %FM, BMI), linear regression parameters (slope; intercept; standard error of the estimate, SEE; coefficient of determination, r^2), limits of agreement at the 95% confidence level and trend.

Our review of validation studies spans two decades; the first study found was Fuller et al. (1992) and the last one was Santos et al. (2010). During this time-frame, the main technological breakthrough was the transition from pencil- to fan-beam densitometers. Because the architecture of these systems is not comparable, it was found appropriate to discuss them in separate.

Table 2.3: Comparison of %FM measured by DXA and the 4C model [cont. on next page].

STUDY	EQUIPMENT (model/software)	SUBJECTS	TISSUE THICKNESS	LINEAR REGRESION ANALYSIS			BLAND-ALTMAN ANALYSIS		
				r²	Slope	Intercept	Bias	95%LoA	Trend
Fuller et al. (1992) %FM	GE Lunar DPX-L sw v1.3z (pencil beam)	16 M; 12 F (18–59 yr)	%FM: F Y 12.0–25.0 F E 19.6–38.0	NR	NR	NR	-1.4^NR	NR	NR
Bergsma-Kadijk et al. (1996) %FM	GE Lunar DPX-L sw v1.3z (pencil beam)	22 F Y (19-27yr) 18 F E (65-78 yr)	BMI: F Y 21.8 ± 1.9 F E 25.4 ± 3.9 %FM: F Y 29.4 ± 3.2 F E 38.8 ± 5.9	NR	NR	NR	F Y: –3.1* F E: –5.3*	NR	NR
Prior et al. (1997) %FM	Hologic QDR1000W sw v5.71 (pencil beam)	91M (21.2±2.1yr); 81F (20.7±2.6yr) (111 A, 61 NA)	M %FM 12.5 ± 5.9 F %FM 22.3 ± 7.6 M BMI 27.0 ± 4.6 F BMI 22.5 ± 3.8	M 0.757*; W 0.88*	M 0.90*; W 0.85*	M 0.75*; W 3.30	-0.4	[-6.1, 5.3]	NR
Withers et al. (1998) %FM	GE Lunar DPX-L sw v1.3z (pencil beam)	24 M; 24 F (18-36 yr)	%FM: M A 12.1 ± 2.8 M NA 21.8 ± 8.2 F A 16.4 ± 2.4 F NA 28.9 ± 4.7	NR	NR	NR	M A: –3.5* M NA: –1.3* F A: –1.3* F NA: –0.4	NR	NR
Gallagher et al. (2000), %FM	Hologic QDR1000W sw v5.71 Lunar DPX sw v3.6 (pencil beam)	Whites: 192 M 225 F African American: 99 M 155 F (20-79 yr)	BMI ≤ 18 to ≥30	M 0.90	0.943	1.60	NR	NR	NR
Deurenberg-Yap et al. (2001) %FM	Hologic QDR4500W sw v8.23a.5 (fan beam)	147 F (36.2 ± 12.0 yr) 144 M (41.9 ± 12.9 yr)	M %FM 26.2 ± 6.5 F %FM 36.2 ± 7.4 M BMI 24.0 ± 3.7 F BMI 23.6 ± 5.2	NR	NR	NR	M -4.2±2.4* to -3.2± 3.0* F -2.5±2.6* to -2.1± 2.6*	NR	M r=-0.56* F r=-0.62*
Wong et al. (2002) %FM	Hologic QDR 2000W sw v5.56 (pencil beam)	141 young females (8-17 yr)	%FM 48-42 BMI 16.1 - 17.3	0.81	1.05	-5.34	3.9**	[-2.7, 10.5]	r=0.13
Gately et al. (2003) %FM	Lunar Prodigy (fan beam)	30 children (14.10 ± 1.83 yr)	%FM 41.23 ± 8.15 BMI 31.56 ± 5.49	0.94	1.06	-4.39	1.9**	[-2.1, 5.9]	NR

Accronims and abbreviations: FM, Fat Mass; FFM, Free Fat Mass; NR, Not reported; M/F, Males/Females; NO/O, Non-Obese/Obese; 95% LoA, 95% Limits of agreement;

Surrogates of Tissue Thickness: Body Mass Index (BMI) [kg/m²], %FM, Antero-Posterior (A-P) chest distance [cm], waist girth [cm].

Significance Levels: ** p < 0.001 , * p < 0.05, NS - Not significant (p ≥ 0.05).

Table 2.3: Comparison of %FM measured by DXA and the 4C model [cont.].

STUDY	EQUIPMENT (model/software)	SUBJECTS	TISSUE THICKNESS	LINEAR REGRESION ANALYSIS				BLAND-ALTMAN ANALYSIS	
				r² (SEE)	Slope	Intercept	Bias	95%LoA	Trend
van der Ploeg et al. (2003) %FM	Lunar DPX-L ssw 1.3z (pencil beam)	118 M (31.1 ± 11.7 yr); 34F (26.1±7.8 yr)	M %FM 22.5 ± 7.7 F %FM 21.5 ± 6.6 M BMI 24.9 ± 3.4 F BMI 20.9 ± 2.1	M 0.952	0.86	4.42	-1.8**	[5.72, 2.12]	r=0.50**
Williams et al. (2006) %FM	Lunar Prodigy sw Encore 2002 (fan beam)	84 adults: • 70 NO M; • 44 NO F; • 14 O F. (18.0-21.3 yr)	%FM: • NO M 15.6±6.3; • NO F 29.9 ± 6.1; • O F 44.4 ± 3.9;	NR	NR	NR	NO M 1.69*; NO F 1.98*; O F 2.28*.	NO M [-2.12, 5.5]; NO F [-1.96, 5.92]; O F [-2.44, 7.00].	NR
Moon et al. (2009) %FM	Lunar Prodigy Advance sw v10.50.086 (fan beam)	29 F Athletes (20 ± 1 yr)	M %FM 24.93 ± 4.63	M 0.86 (1.78)	0.609	7.49	3.71*	[-2.7, 10.1]	r=-0.43*
LaForgia et al. (2009) %FM	Lunar Prodigy sw Encore 2003 7.52.002 (fan beam)	8 M; 6 F (40.0 ± 13.5 yr)	BMI 33.7 ± 3.5 %FM 41.5 ± 6.0 A-P chest 21.3 ± 2.4 WG 106.6 ± 11.3	0.894*	0.83*	7.345	-0.35	NR	r=0.424
Santos et al. (2010) %FM	Hologic QDR 4500A sw v8.21 (fan beam)	24 M (22 ± 3yr)	BMI: 23.6 ± 2.3 %FM: 9.2 ± 4.1	0.61 (2.63)	1.03	-3.24*	2.9	[7.9, 2.2]	-0.26

Accronims and abbreviations: FM, Fat Mass; FFM, Free Fat Mass; NR, Not reported; M/F, Males/Females; NO/O, Non-Obese/Obese; 95% LoA, 95% Limits of agreement;

Surrogates of Tissue Thickness: Body Mass Index (BMI) [kg/m²]; %FM, Antero-Posterior (A-P) chest distance [cm]; waist girth [cm].

Significance Levels: ** p < 0.001, * p < 0.05, NS - Not significant (p ≥ 0.05).

Table 2.4: Comparison of FM and FFM measured by DXA and the 4C model.

STUDY	EQUIPMENT (model/software)	SUBJECTS	TISSUE THICKNESS	LINEAR REGRESION ANALYSIS				BLAND-ALTMAN ANALYSIS	
				r² (SEE)	Slope	Intercept	Bias	95%LoA	Trend
Goran et al. (1998) FM	Lunar DPX-L sw Lunar v1.3 DPX-L (pencil beam)	82 elderly (69.2±6.8 yr); 41M (68.2±6.6 yr); 41F (70.2±7.0 yr).	%FM: • T: ≈ 30.6%; • M: ≈ 38.1%; • F: ≈ 24.3%.	T: 0.78(3.8); M: 0.73(4.0); F: 0.77(3.6).	T: .83±.05*; M: .81±.08*; F: .76±.07*.	T: 4.0±1.1*; M: 3.4±1.6*; F: 5.9±1.7*.	NS	NR	NS
Salamone et al. (2000) FM	Hologic QDR4500W sw v8.21 (fan beam)	30M (73.9 ± 2.2yr); 30F (73.6 ±2.3yr)	%FM 32.5 ± 8.8 BMI 17.5-38.8	M 0.96** (1.5)	NR	NR	-0.7	[-4.0, 2.6]	r=-0.34*
Schoeller et al. (2005) FM	Hologic QDR4500A (fan beam)	30 M; 28 F (74 ± 2 yr)	BMI 30 ±3	NR	NR	NR	-2.2*	[-5.7, 1.3]	NR
Williams et al. (2006) FM	Lunar Prodigy sw Encore 2002 (fan beam)	84 adults: • 70 NO M; • 44 NO F; • 14 O. (18.0-21.3 yr)	%FM: • NO M 15.6 ± 6.26; • NO F 29.9 ± 6.1; • O F 44.4 ± 3.9;	NR	NR	NR	NO M 1.35*; NO F 1.21*; O F 1.58*.	NO M [-1.47, 4.17]; NO F [-1.08, 3.50]; O F [-1.92, 5.08].	NR
Santos et al. (2010) FM	Hologic QDR 4500A sw v8.21 (fan beam)	24 M (22 ± 3yr)	BMI: 23.6 ± 2.3 %FM: 9.2 ± 4.1	0.67 (1.94)	0.95	-1.57 *	2.1*	[5.8, -1.7]	-0.46
Visser et al. (1999), FFM	Hologic QDR4500A sw v8.21 (fan beam)	30 M (73.9 ± 2.2 yr) 30 F (73.6 ± 2.3 yr)	%FM 32.5 ± 8.8 BMI 27.4 ±4.5	0.98	1	0	1.8	[-1.3 4.9]	r=0.09
Tylavsky et al. (2003) FFM	Hologic QDR4500W sw v8.21 (fan beam)	30 M; 28 F (73.7 ± 2.2 yr)	%FM 25.2 ± 18.9 BMI 27.2 ± 4.5	0.98	0.964	0	2.6**	NR	r=0.16
Schoeller et al. (2005) FFM	Hologic QDR4500A (fan beam)	30 M; 28 F (74 ± 2 yr)	BMI 30 ± 3	0.903*	0.932*	0	3.3*	[-0.2, 6.8]	NR
Williams et al. (2006) FFM	Lunar Prodigy sw Encore 2002 (fan beam)	84 adults: • 70 NO M; • 44 NO F; • 14 O F. (18.0-21.3 yr)	%FM: • NO M 15.6 ± 6.26; • NO F 29.9 ± 6.1; • O F 44.4 ± 3.9;	NR	NR	NR	NO M -0.37; NO F -1.19*; O F -1.97*.	NO M [-3.08, 2.34]; NO F [-3.57 , 1.19]; O F [-5.36, 1.42].	NR
Santos et al. (2010) FFM	Hologic QDR 4500A sw v8.21 (fan beam)	24 M (22 ± 3yr)	BMI: 23.6 ± 2.3 %FM: 9.2 ± 4.1	0.92 (1.88)	1.09	-3.14*	-2.7*	[1.1, -6.4]	-0.42

Accronims and abbreviations: FM, Fat Mass; FFM, Free Fat Mass; NR, Not reported; M/F, Males/Females; NO/O, Non-Obese/Obese; 95% LoA, 95% Limits of agreement;

Surrogates of Tissue Thickness: Body Mass Index (BMI) [kg/m²], %FM, Antero-Posterior (A-P) chest distance [cm], waist girth [cm].

Significance Levels: ** p < 0.001, * p < 0.05, NS - Not significant (p ≥ 0.05).

Pencil-beam densitometers The agreement of pencil-beam systems against the 4C model to determine %FM and/or FM was tested in six studies. For %FM, Bergsma-Kadijk et al. (1996); van der Ploeg et al. (2003); Withers et al. (1998) found Lunar DPX-L [pencil-beam] to underestimate %FM, but, in general, the bias was within –5% to –1%. Two other studies — Fuller et al. (1992) also using Lunar DPX-L and Prior et al. (1997) using Hologic QDR-1000 — reported no significant differences for %FM, and neither did Goran et al. (1998) for FM. As such, pencil-beam DXA systems and the 4C model were in agreement in terms of mean values. The coefficients of determination of pencil-beam systems were in the range 0.73 – 0.88 for FM and 0.757% – 0.952% for %FM, thus indicating strong correlation (Gallagher et al., 2000; Prior et al., 1997; van der Ploeg et al., 2003). Nevertheless, the range of individual differences could be large, particularly in studies comprising >100 subjects; van der Ploeg et al. (2003), for example, observed individual differences in healthy adults that could be as high as –2.6 – 7.3%, but, in a sample of young collegiate athletes and non-athletes, Prior et al. (1997) found that the individual differences could be even higher (–7.5 to 10%). More importantly, the magnitude of the errors appeared to be dependent on the %FM level (van der Ploeg et al., 2003). Not only did DXA underestimate the %FM of leaner individuals, it also overestimated it among those with higher %FM. As an example, in lean to normal weight adults, van der Ploeg et al. (2003) reported 1.8% lower %FM by DXA when compared to the 4C model.

Fan-beam densitometers For fan-beam densitometers, seven studies were reported for %FM. With the exception being made to Bergsma-Kadijk et al. (1996) and LaForgia et al. (2009), it was observed that first generation of fan-beam systems (e.g., Lunar Prodigy, Hologic QDR-2000 fan and QDR-4500W) overestimated %FM by $\approx 2\% - 3\%$ when compared to a 4C model (Gately et al., 2003; Moon et al., 2009; Santos et al., 2010; Williams et al., 2006; Wong et al., 2002). Interestingly, Williams et al. (2006) observed that this overestimation was accentuated in the obese. This research team compared body composition from the Lunar Prodigy to a 4C model and reported a mean overestimation of %FM and FM up to 0.6 % and 0.2kg higher in obese adults than in the non-obese. The differences for the FM and FFM compartments were also higher in the obese group, though the over- and underestimation for FM and FFM was below 1kg. This observation could partly explain the result of Wong et al. (2002), whereby the bias for obese woman reached $\approx 4\%$ in a Hologic QDR-2000 fan-beam system. However, the same does not hold for the studies

conducted by Moon et al. (2009) and Gately et al. (2003). Using the same densitometer as Williams et al. (2006), but now in non-obese athletes, Moon et al. (2009) observed that DXA overestimated %FM by $\approx$ 4%; this bias was even higher that the $\approx$ 2% reported by Gately et al. (2003) for obese children, also in a Lunar Prodigy system (LaForgia et al., 2009). Inline with these efforts, LaForgia et al. (2009) conducted a pilot study in an obese cohort. The author's purpose was to inspect the effect of surrogates of tissue thickness (BMI, %FM and anterio-posterior chest distance) on the validity of DXA against the 4C model. Despite bias was not significant, the correlation between 4C and DXA %FM values was high, meaning that DXA was less robust in the provision of accurate individual values. For this reason, it was suggested that DXA could be unable to accommodate the phenomenon of beam hardening at larger tissue thicknesses (LaForgia et al., 2009). Overestimation results have also been reported for athletic populations. In a sample comprised of elite judo athletes, Santos et al. (2010) verified that, at the athlete's stable period, %FM was overestimated by $\approx$ 3 % when measured by a Hologic QDR-4500 and compared to the 4C model. Likewise, using Lunar Prodigy Advance, Moon et al. (2009) observed a mean underestimation of $\approx$ 4% in female athletes. An underestimation of %FM using fan-beam DXA (Hologic QDR-4500) was only found by Deurenberg-Yap et al. (2001). For the chinese and singaporean ethnic groups, the mean differences in %FM between DXA and the 4C model were between 2.1% and 4.2%, thus suggesting that the higher density of FFM in specific ethnic groups could compromise the accuracy QDR-4500.

With respect to the FM and FFM compartments, Table 2.4 reveals conflicting results. While Williams et al. (2006) and Santos et al. (2010) found DXA to overestimate FM while underestimating FFM, in a comparison of estimates obtained on a Hologic QDR-4500 versus the 4C model, Schoeller et al. (2005) observed that exact opposite. Conversely, significant biases for FM and/or FM by DXA have not been consistently observed by investigators comparing DXA-derived body composition outcomes to those obtained from the 4C model. In samples comprising elderly subjects of genders, Salamone et al. (2000) and Visser et al. (1999) found no significant differences for Hologic QDR-4500A. It is worth point out that both studies were conducted with software version 8.21. As previously mentioned on Section 2.3.1, this version was in the process of corrections for magnification effects. This fact may partly explain the accuracy demonstrated in these two studies, but it is not clear if it was just magnification issues that could have compromised the accuracy of measurements in other studies at which this (or similar)

software version was also available. The inspection of the trends could certainly provide relevant information to clarify this aspect. However, the majority of literature listed in Tables 2.3 and 2.4 did not report trends. And, among those who did, four out of five studies reported a positive trend (two of them reached statistical significance), thus raising suspicions on magnification effects that could have taken place.

Regarding strength of correlation, the coefficients of determination for fan-beam systems were generally in the range 0.90 – 0.98 for FFM and 0.81–0.86 for %FM. However, correlations may be lower in athletic populations (Santos et al., 2010).

As a final note of caution, the reported individual differences could be large (Prior et al., 1997; van der Ploeg et al., 2003). Also, there were discrepancies between body composition measures; some studies underestimated DXA compartments or %FM, whereas others reported the opposite or no differences. The reasons for the conflicting findings are manifold. Firstly, although beam technologies were compared and discussed separately in this review, there have been a mixture of hardware and software versions used in the validation studies, and it is known that some of the errors associated with DXA, such as the magnification effect, are expressed differently with the various technologies (van der Ploeg et al., 2003). Second, these studies were also undertaken with different evaluation protocols (or these were not reported), thus making direct comparison risky (Schoeller et al., 2005). Third, the heterogeneity of the samples varied from study to study in terms of %FM, age, gender, ethnicity and athletic status. The wide variation in the hydration of FFM and %FM among these validation studies (%FM in the range 10-40%) may have partly explained the equivocal results (Deurenberg-Yap et al., 2001; Moon et al., 2009).

2.4.3 Summary

The limited number of validation studies of specific DXA systems (hardware/software) against the 4C model, as well as differences in subject phenotypes, may compromise the extrapolation of validation studies to a particular system and individual. Even still, the differences between DXA estimates of body composition and the criterium 4C model are generally small. It is, therefore, assumed that the validity of DXA measurements for body composition are acceptable, though individual differences may be higher for those with extreme phenotypes, particularly athletes and the obese. As such, DXA can be considered as a valid, practical and safe option to assess body composition.

2.4.4 Accuracy for BMC measurements

Since the 4C model uses the estimates of DXA for the bone compartment, studies against the 4C model leave unanswered the magnitude of errors arising in the estimation of the bone mineral compartment. In case of BMC, it is generally accepted that the "true" measurement is obtained by weighting the ashed weight of bone samples. Comparison of *in situ* bone mineral density (BMD) measurements in cadavers with results of ashing showed differences up to 15%, which may be partly explained by the fat content of bone and bone marrow, or bone segmentation errors during image analysis (Bonnick & Lewis, 2006). Notwithstanding the fact that most validation studies on bone densitometry use phantoms, DXA is well established as an imaging modality for bone mineral content and bone mineral density (Ahmad et al., 2014). The bias that may exist for BMC is minimized if DXA systems are regularly calibrated with phantoms (Hologic, Inc., 2014). Moreover, small accuracy errors are a minor concern provided they remain constant. Often, what is clinically relevant is the precision errors that may compromise the reproducibility of a diagnostic technique or the measurement of changes. Precision is, therefore, an additional key metrics that has to be put into context with the prospective use of the DXA system, be it the assessment of changes during an intervention, screening or other application.

2.5 Precision of DXA

According to Bonnick & Lewis (2006), *precision* is the attribute of a quantitative technique such as DXA imaging that refers to "the ability to reproduce the same numerical result in the setting of no real biologic change when the test is repeatedly performed in an identical fashion (pp. 190)". Like all quantitative tests in medical trials, no DXA system is perfectly reproducible. Not even when whole-body scans are performed in exact accordance with the manufacturer's recommendations (Bonnick & Lewis, 2006). However, it is still possible—and advisable—to assess body composition changes according to conveniently standardized protocols that are congruent with the manufacturer's recommendations. If best practices are followed, then the technique becomes as reproducible as it possibly can be (Nana, 2013). The precision of whole-body composition using DXA assumes great importance when the technique is meant to follow *changes* over time.

Because densitometry is not perfectly reproducible, the results on any given subject are not expected to be identical, even if the composition of the individual has not actually changed. The only way to be confident that a real biologic change has occurred is to verify that the precision error of the DXA system has been exceeded. This means that the precision must be quantified. Precision is usually assessed by performing multiple repeated measurements using the same DXA instrument on the same patients. The precision is generally reported as either as the technical error of measurement (TEM[4]) or as the percentage coefficient of variation (CV[5]). TEM and CV are quality control metrics for expressing the reliability of an observation and account for instrument error, protocol and biological variability of the measured populations, and expertise level of the technologists that performed scan acquisition and analysis (e.g.: positioning of subjects). TEM is the standard deviation between repeated measurements taken independently by one observer. For each DXA compartment, and for the specific case of just one observer, let K be the number of repeated scans of the same subject. If the estimates for whole-body scanning in the $1 \ldots K$ sets are denoted as $x_{WB} \ldots x_{WB}^{(k)} \ldots x_{WB}^{K}$, then the TEM is computed as follows (Bonnick & Lewis, 2006):

$$TEM = \sqrt{\frac{1}{N_{TR}} \sum_{i=1}^{N_{TR}} \frac{1}{K-1} \sum_{k=1}^{K} \left(x_{WB,i}^{(k)} - m_{WB,i} \right)^2} \qquad (2.1)$$

where $m_{WB,i}$ is the average difference between whole-body measurements in the K repeated scans for subject i:

$$m_{WB,i} = \frac{1}{K} \sum_{k=1}^{K} x_{WB,i}^{(k)} \qquad (2.2)$$

Due to radiation dose and time issues, it is generally not possible to perform more than two consecutive DXA scans for each subject. For this reason, short-term precision studies for body composition assessment generally require two consecutive scans (test-retest, TR) per subject with repositioning in between. For statistical validity, the number of required subjects in test-retest studies is thirty, so that the precision study have thirty degrees of

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[4]Note: TEM is also referred to in the literature as root mean-square standard deviation (RMS-SD).

[5]Note: CV is also referred to in the literature as %CV or as root mean-square coefficient of variation (RMS-CV).
Bonnick & Lewis (2006); Sun & Chumlea (2005)

freedom [6] (Bonnick & Lewis, 2006). By substituting $K = 2$ in Equation 2.1, it is possible to arrive at a much simpler expression for TEM:

$$TEM = \sqrt{\frac{1}{N_{TR}} \sum_{i=1}^{N_{TR}} (x_{WB,i} - m_{WB,i})^2 + \left(x'_{WB,i} - m_{WB,i}\right)^2} \qquad (2.3)$$

where $m_{WB,i}$ is the average difference between whole-body measurements in the first and repetition scans for subject i:

$$m_{WB,i} = \frac{x_{WB,i} + x'_{WB,i}}{2} \qquad (2.4)$$

Combining Equations 2.5 and 2.4 results in:

$$TEM = \sqrt{\frac{1}{N_{TR}} \sum_{i=1}^{N_{TR}} \left(x_{WB,i} - \frac{x_{WB,i} + x'_{WB,i}}{2}\right)^2 + \left(x'_{WB,i} - \frac{x_{WB,i} + x'_{WB,i}}{2}\right)^2} \qquad (2.5)$$

which is in fact the same expression as provided by Ulijaszeka & Kerra (1999):

$$TEM = \sqrt{\frac{1}{2 N_{TR}} \sum_{i=1}^{N_{TR}} \left(x'_{WB,i} - x_{WB,i}\right)^2} \qquad (2.6)$$

where the term $\left(x'_{WB,i} - x_{WB,i}\right)^2$ is the squared difference of the test and retest measurements for subject $i = 1 \dots N_{TR}$. TEM is expressed in grams or kilograms. Often, it is useful to normalize TEM by the mean value of the observations, resulting in another metrics called coefficient of variation (CV). CV is expressed in percentage (Sun & Chumlea, 2005). For a test-retest [short-term] precision study with one observer and N_{TR} subjects, CV (%) is computed from TEM as follows:

$$CV = 100 \frac{TEM}{\frac{1}{N_{TR}} \sum_{i=1}^{N_{TR}} m_{WB,i}} \qquad (2.7)$$

The lower the TEM and the CV, the better the precision and the easier it is to detect small changes in measurements. Based on 30 subjects (15 healthy males and females) with

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[6] According to Bonnick & Lewis (2006), "Thirty degrees of freedom ensure that the upper limit for the 95% confidence interval of the precision value is no more than 34% greater than the calculated precision value." (pp. 193)

similar phenotypes as the included in the present study, the TEMs and CVs for whole-body composition assessment in the Exercise and Health Laboratory are:

TEM$_{EHLab}$) 0.02 kg for BMC, 0.36 kg for LST, 0.37 kg for FM, and 0.1 % for FM;

CV$_{EHLab}$) 1.0%for BMC, 0.7% for LST, 2.8% for FM, and 0.6% for %FM.

In contrast to validation studies against the 4C model, precision studies of DXA devices are relatively common. In fact, each laboratory is advised to conduct short-term precision studies in subjects identical to its target populations. A search on titles and abstracts on Pubmed with the keywords DXA, Norland, Hologic, iDXA and Lunar resulted in 62 results, most of them valid. There have also been recent revisions of precision studies, including Hangartner et al. (2013) / International Society for Clinical Densitometry. The precision of DXA technology has been reported for whole-body assessment of the general population, and, to a minor extent, obese and athletic populations. However, results for regional body composition analysis are relatively scarce. Table 2.6 lists CVs for several equipments, for whole-body %FM, FM, LST, BMC and also upper and lower limbs. Overall, Lunar iDXA stands out as the most precise DXA system for whole-body composition analysis (CVs < 1.5% for all compartments and <1% for %FM). In general, DXA devices provide high precision in BMC scans of 1–2%, even in athletic and obese populations (Bilsborough et al., 2014; Carver et al., 2013; Cordero-MacIntyre et al., 2002). We note, that the CV for BMC in the obese (1.1–1.7%) are higher than that of the athletes (0.6%). This results seems to be in agreement with the finding of Knapp et al. (2014) about the deleterious effect of obesity on precision errors in whole-body BMD (the same might hold for BMC). In whole-body scans, CVs for LST and FM are in between 1–4%, and, for %FM, short-term precision in non-athletic populations has been reported as low as 1–2%. It is observed that results for FM and %FM are worse in athletes ($\approx$ 2.5 %). Ackland et al. (2012) provide an insightful explanation with respect to this matter:

> "As training methods have become more sophisticated, each athletic group has become more specialized, modifying its typical physique imperatives away from general morphological norms. (...) Furthermore, athletes are reluctant to interrupt what for many is a full-time occupation for the sake of body composition assessment, thereby making the more involved laboratory techniques less appealing. These factors all conspire against the scientist seeking to make accurate [the same reasoning holds for precision] measurements on athletes, with the inevitable consequence that data may be misleading, misinterpreted or perhaps used inappropriately." (pp. 230)

Table 2.6: Precision of %FM measured by DXA

STUDY	EQUIPMENT (model/software)	SUBJECTS	PRECISION [CV, %]
Hangartner et al. (2013); Toombs et al. (2012) %FM	various	revision of artices	0.6 . . . 1.9%
Hind et al. (2011) %FM	GE Lunar iDXA sw enCORE v11.0 (narrow-fan beam)	52 M & F (34.8 ± 8.4 yr; BMI 16.7–42.7 kg/m^2)	0.9%
Clark et al. (2004); Hind et al. (2011) %FM	2 studies combined	73 M Ath	2.4 – 2.5%
Carver et al. (2013) %FM	GE Lunar iDXA sw enCORE narrow fan-beam	65 O (46 ± 11 yr, BMI 49 ± 6 kg/m^2).	0.8%
Hangartner et al. (2013) FM	various	revision of articles	0.7 . . . 3.4%
Buehring et al. (2014) FM	GE Lunar iDXA sw enCORE vs. 11.0-13.4 narrow fan-beam	30M Ath (20.6 ± 1.3 yr); 30F Ath (19.9 ± 1.3 yr);	M 0.64%; F 1.46%
Clark et al. (2004); Hind et al. (2011) FM	2 studies combined	73 M Ath	2.5%
Cordero-MacIntyre et al. (2002) FM	Hologic QDR-4500A sw v8.21	20 F O (40–70 yr)	1.2%
Carver et al. (2013) FM	GE Lunar iDXA sw enCORE narrow fan-beam	65 O (46 ± 11 yr, BMI 49 ± 6 kg/m^2).	0.9%.
Franck & Munz (2000) FM regional	Hologic QDR-2000 (fan beam)	165 F; 136 M (43–80 yr)	3.4 . . . 10.9%
Cordero-MacIntyre et al. (2002) FM U/L	Hologic QDR-4500A sw v8.21	20 F O (40–70 yr)	11.4 / 3.9%
Lohman et al. (2009) FM upper limbs	Lunar Prodigy	30 M (22–61 yr)	4.1 / 2.7%
Bilsborough et al. (2014) FM U/L	GE Lunar DPX-L sw v1.3z (pencil beam)	22 M Ath (22.5 ± 1.3 yr)	4.3 / 2.3%
Buehring et al. (2014) FM upper limbs	GE Lunar iDXA sw enCORE vs. 11.0-13.4 narrow fan-beam	30M Ath (20.6 ± 1.3 yr); 30F Ath (19.9 ± 1.3 yr);	M 7.1–8.0%; F 3.9–4.3%

Buehring et al. (2014) FM lower limbs	GE Lunar iDXA sw enCORE vs. 11.0-13.4 narrow fan-beam	30M Ath (20.6 ± 1.3 yr); 30F Ath (19.9 ± 1.3 yr);	M 2.2–3.3%; F 1.5–2.1%.
Hangartner et al. (2013); Toombs et al. (2012) LST	various	revision of studies	0.4 . . . 2.2%
Bilsborough et al. (2014); Clark et al. (2004) LST	2 studies combined	75 M Ath	1.0 . . . 1.3%
Cordero-MacIntyre et al. (2002) LST	Hologic QDR-4500A sw v8.21	20 F O (40–70 yr)	1.1%
Carver et al. (2013) LST	GE Lunar iDXA sw enCORE narrow fan-beam	65 O (46 ± 11 yr, BMI 49 ± 6 kg/m^2).	1.1%.
Franck & Munz (2000) LST regional	Hologic QDR-2000 (fan beam)	165 F; 136 M (43–80 yr)	1.2 . . . 10.9%
Cordero-MacIntyre et al. (2002) LST U/L	Hologic QDR-4500A sw v8.21	20 F O (40–70 yr)	4.5 / 2.1%
Lohman et al. (2009) LST U/L	Lunar Prodigy	30 M (22–61 yr)	4.8 / 9.1 %
Bilsborough et al. (2014) LST U/L	GE Lunar DPX-L sw v1.3z (pencil beam)	22 M Ath (22.5 ± 1.3 yr)	2.7 / 1.3%
Bilsborough et al. (2014) BMC	GE Lunar DPX-L sw v1.3z (pencil beam)	22 M Ath (22.5 ± 1.3 yr)	0.6%
Cordero-MacIntyre et al. (2002) BMC	Hologic QDR-4500A sw v8.21	20 F O (40–70 yr)	1.7%
Carver et al. (2013) BMC	GE Lunar iDXA sw enCORE narrow fan-beam	65 O (46 ± 11 yr, BMI 49 ± 6 kg/m^2).	1.1%
Cordero-MacIntyre et al. (2002) BMC U/L	Hologic QDR-4500A sw v8.21	20 F O (40–70 yr)	5.6 / 2.3%

Abbreviations and Acronyms: FM, Fat Mass; LST, lean Soft Tissue; BMC, Bone Mineral Content; U/L, Upper/Lower limbs; M/F, Males/Females; O, Obese; Ath, Athlete; CV, Coefficient of variation (%).

As for regional analysis, CVs are worst than the reported results for whole-body measurements. BMC, LST and FM for limbs are generally in the range of ≈ 2–4%, though values up to 9–11% have also be reported. Table 2.6 have also indicates that the CVs for upper (U) limbs are higher than those of the lower (L) limbs (U: 3–11% vs. L: 2–4%) (Cordero-MacIntyre et al., 2002; Franck & Munz, 2000; Lohman et al., 2009).

Additionally, there are long-term precision studies that consider variations over months, and others that study how well DXA tracks changes in body composition (Bonnick & Lewis, 2006; Nana, 2013). Also, the CV can be used to to determine the so called the least significant change (LSC). The LSC is the minimum change in any of the 3C-DXA compartments that constitutes a real biologic change and is useful to determine the minimum interval between follow-up measurements (Bonnick & Lewis, 2006; Buehring et al., 2014).

2.6 Scanning solutions for broad individuals

As briefly mentioned in Section 2.3.2, some DXA systems restrict the width of individuals to be scanned to 60-67 cm. This methodological problem may be overcome by solutions that scan the body partially—such as half- or reflexion-scanning— or that take the sum of two or more partial scans. As depicted in Figure 2.4, obese individuals and/or athletes undertaking half- and reflexion-scanning only need to perform one whole-body composition evaluation. In contrast, those undertaking summation scans are required to be scanned twice. Partial scanning techniques can only handle width limitations, whereas summation scans can be extended to solve both width and height limitations. Because of its relevance to the scope of the present thesis, this section provides detailed information on validation studies reported in the literature of scanning solutions for broad individuals.

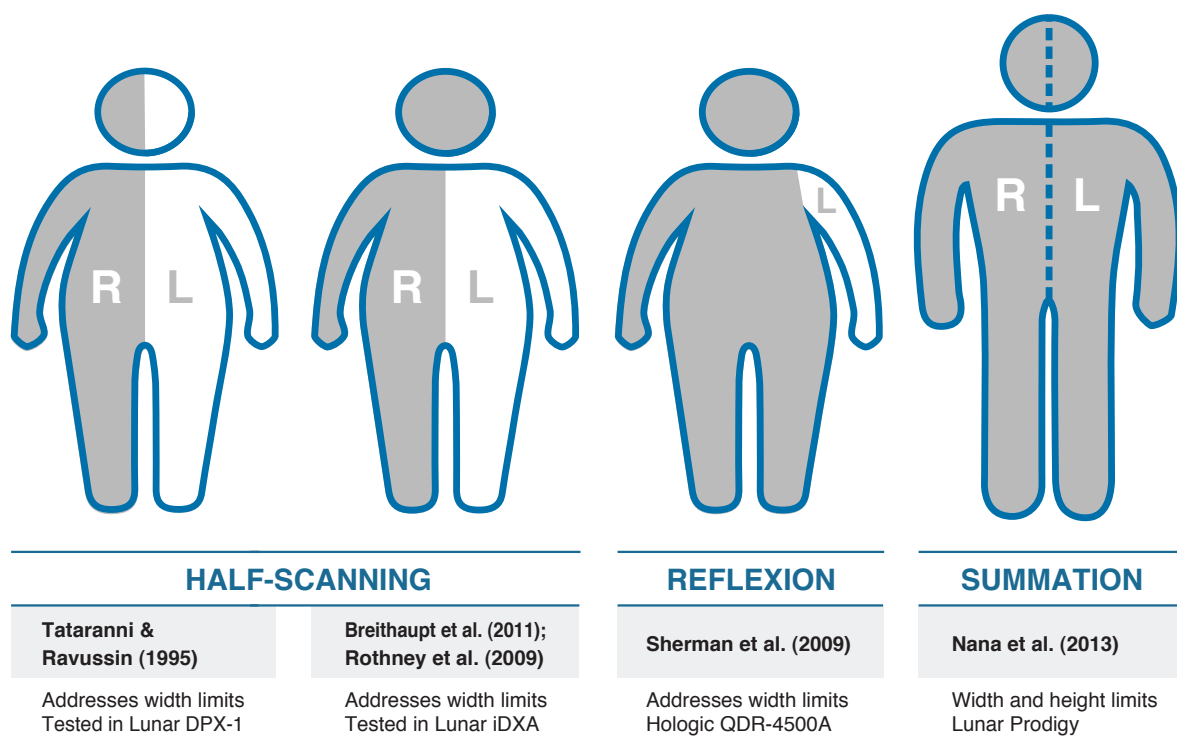| HALF-SCANNING		REFLEXION	SUMMATION
Tataranni & Ravussin (1995)	**Breithaupt et al. (2011); Rothney et al. (2009)**	**Sherman et al. (2009)**	**Nana et al. (2013)**
Addresses width limits Tested in Lunar DPX-1	Addresses width limits Tested in Lunar iDXA	Addresses width limits Hologic QDR-4500A	Width and height limits Lunar Prodigy

Figure 2.4: Scanning solutions for assessing broad individuals in DXA systems.

Partial scanning

The first study that addressed the width limitation of DXA devices was conducted by Tataranni & Ravussin (1995) with the comparison of half-body scans to whole-body composition assessment (see Figure 2.4. The authors assessed 156 subjects that fitted within the DXA scan (BMI 25.8 ± 4.1 kg/m²) table plus 27 subjects (12 females / 15 males; BMI 44.5 ± 4.6 kg/m²; 18–70 yr) wider than the DXA scan area. DXA measurements were performed by using a whole-body scanner (DPX-l; Lunar Radiation Corp, Madison, WI). The outcome variables were FM, LST, BMC, and percent FM. The operator performed a single whole-body or two half-body scans according to his visual judgment of whether or not the patient was fitting within the scanning area (197×58 cm). The subjects scanned once were positioned according to Mazess et al. (1990). For the 27 subjects scanned twice, the central line of the scanning area passed through the midpoint of the left or right clavicula for the left and right half-body scans, respectively. Subjects were scanned from head to toes in both cases, being displaced toward the left side of the table for the scan of the right body side and toward the right side of the table for the scan of the left body side. To adjust for differences in body thickness in the trunk region, scans were performed at three different transverse speeds (slower for thicker individuals). The sagittal line was positioned by the technologist based on anatomical reference points (skull, spine, pelvis, and legs). For the subjects scanned twice, the sagittal line had to be repositioned each time on the two scans. Note the half-scan region "splits" the head in two halves; this option is not elegant nor practical since the head of obese individuals is never out of the scanning area. In a subsequent study, Rothney et al. (2009) did not include the head in the definition of its half-scan region, and neither did Breithaupt et al. (2011).

Rothney et al. (2009) validated the half-scan analysis using a newer DXA equipment by comparing to the standard whole-body scans in a sample of 52 obese adults (37 females) ranging in age from 19 to 63 years. DXA measurements were made using a total-body scanner (Lunar iDXA; GE Healthcare, Madison, WI). As mentioned earlier, the iDXA is a narrow fan-beam DXA instrument with a relatively wider scanning space (66 cm) and an additional 10 cm in one side of the scanning table to allow half-body scans of larger individuals. For the study, all scans were conducted in thick mode (thickness >25 cm), which requires 13 min of scan time with an effective radiation dose of 3 μGy per scan.

Scan analysis was performed using GE Encore 11.10 software. This software allows for adjustment of regions of interest including the sagittal line, which controls the left-right body distribution of tissue. This determination is corroborated by a trained operator. If the subject's body is not contained within the scan space, a half-scan analysis is automatically performed by assuming symmetry of the body. Each whole-body image was analyzed, including the manual placement of an sagittal line. Following this analysis, the authors chose to re-analyze each scan as a right side scan and a left side scan. According to Rothney et al. (2009), this procedure was adopted to eliminate the need of repositioning and rescanning different sides which could introduce estimation errors such as those observed in the previous study.

Breithaupt et al. (2011) determined the validity of a half-body scan methodology for measuring body composition using GE Lunar Prodigy Advance (GE Healthcare, Madison, WI, USA). The sample consisted of 34 obese children (7.7–18.1 yr; 18 girls / 16 boys) that fitted in the active scan area of the scanner. Average scan time was 4.5 min with a radiation dose of approximately 3 μSv. The software (GE encore 11.40) used in the study could detect whether a subject was within the scanning region. If they are not within the active scan region, an automatic half-scan analysis is performed instead. Similarly to Rothney et al. (2009), the determination of the accuracy of the half-scan technique was made in comparison with whole-body scans, so that subjects were only required to perform a single scan.

For reflexion-scanning of the upper limb, Sherman (2011) evaluated 434 subjects (ages 16 – 69 yr) from the NHANES study with repeat whole-body scans. Scans were acquired on Hologic QDR-4500A systems. Linear regression and Bland-Altman analysis were used to compare reflected vs. whole-body scans, and precision was assessed based on TEMs and CVs. Since this study is an abstract, results are not provided.

Summation scanning

Nana et al. (2012) recruited 30 physically active subjects (ages 30 ± 7 yr; 50% females) who would represent the range of physiques found among athletic sub-populations who fit the scanning area. Each subject underwent one whole-body and 4 partial DXA scans in a single testing session under standardized conditions of resting and fasting. Various combinations of the partial scans were summed to estimate total body composition, ho-

wever, in this review, we will only consider the configuration at which the right and left half-scans are summed to obtain whole-body composition estimates of BMC, LST and FM. Body composition was measured using a narrowed fan-beam DXA (Lunar Prodigy, GE Healthcare, Madison, WI) with analysis performed using GE Encore 13.60 software (GE, Madison, WI). All of the scans were undertaken using the standard thickness mode.

Accuracy of the Procedures

Half-scanning Tataranni & Ravussin (1995) reported that whole-body composition can be accurately predicted from the results of half-body DXA scans in a group of 27 obese subjects who did not fit completely in the scanning area. As observed in Table 2.7, parameters of the predictive equations for whole-body composition using DXA half-body scan are available, but SEEs and results of agreement analysis between procedures were not provided by the authors. Data for both the right and left sides of the body were available in 177 subjects who were scanned twice (once each on left and right side). The overall symmetry between the left and right sides were: 0.03 ± 0.08 kg for BMC ($r^2 = 0.89$), -0.04 ± 0.62 kg for FM ($r^2 = 0.99$), and -0.04 ± 0.86 kg for LST ($r^2 = 0.97$). However, in comparison with the group who fitted the scan area, the obese ($n = 21$) had slightly larger differences between sides for FM (0.72 ± 0.11 kg) and LST (0.30 ± 0.11 kg) but similar for BMC (0.03 ± 0.09).

Table 2.7: Review of validation studies of alternative scanning techniques to assess body composition of broad individuals in normal DXA systems, including right half-scanning, reflexion and summation scanning. Adapted with permission from Silva et al. (2013).

VALIDATION STUDIES OF RIGHT HALF SCANS

Components	r^2	SEE	Slope	Intercept	Bias	95% LoA	Trend
Tataranni & Ravussin (1995) in Lunar (DPX-1), N=27 (15M; 12F)							
BMC	0.970	NA	1.87	0.18	NA	NA	NA
LST	0.990	NA	1.88	3.17	NA	NA	NA
FM	0.990	NA	1.93	1.46	NA	NA	NA
%FM	0.990	NA	1.00	-0.26	NA	NA	NA
Rothney et al. (2009) in Lunar (iDXA), N=52 (15M; 37F)							
BMC	0.996	NA	NR	NR	0.03*	-0.04;0.09	NS
LST	0.997	NA	NR	NR	0.05	-1.00;1.10	NS
FM	0.994	NA	NR	NR	0.00	-1.00;1.00	NS
%FM	0.998	NA	NR	NR	0.00	-0.50;0.50	NS

Breithaupt et al. (2011) in Lunar (iDXA), N=34 (16B; 18G)							
BMC	0.996	NR	0.983	0.0515	0.00	-0.10;0.10	NS
LST	0.999	NR	0.999	-0.0574	-0.08*	-0.65;0.85	NS
FM	0.998	NR	1.001	-0.1234	-0.08*	-0.60;0.65	NS
%FM	1.000	NR	0.999	0.0288	0.00	-1.50;1.50	NS

VALIDATION ABSTRACT OF (RIGHT) RSU

Sherman (2011) in Hologic QDR-4500, N=434 (16–69 yr)

"Small significant differences were observed for whole-body bone but no differences on the soft tissue measures. (. . .) [Because of reflected arm values on whole-body scans] (. . .) there may be an impact on the accuracy of bone measures."

VALIDATION STUDY OF SUMMATION SCANS FOR BROAD SUBJECTS

Components	r^2	SEE	Slope	Intercept	Bias	95% LoA	Trend
Nana et al. (2012) in Lunar Prodigy, N=30 Athletes (15M; 15F)							
BMC	NR	NR	NR	NR	-0.2%	NR	NR
LST	NR	NR	NR	NR	0.3%	NR	NR
FM	NR	NR	NR	NR	0.1%	NR	NR

Abbreviations and Acronyms: BMC, bone mineral content; FM, fat mass; LST, lean soft tissue; r^2, squared correlation coefficient; SEE, standard error of estimation; Bias, mean difference between methods (calculated as the alternative minus the reference values); LoA, limits of agreement; NR, Not reported; G/B, Girls/Boys; M/F, Males/Females. **Significance levels:** * Significantly different from 0, $p < 0.05$; NS, Not significant, $p \geq 0.05$.

The results of Rothney et al. (2009) are also displayed in Table 2.7. The group diffe-rences between half-scan simulations and the total-body DXA scan were not significantly different in percent fat, FM, LST, or total body mass. In absolute terms, the BMC was significantly different ($p < 0.001$) between each of the half-body scan estimates and the whole-body, with the right side slightly overestimating BMC (23 ± 31 g). The differences between men and women were similar. The between-individual measures of %FM, FM, LST, and BMC estimated from right side was highly correlated ($r^2 > 0.99$) and closely comparable to the respective measurements from the whole-body. Bland-Altman analysis revealed no significant magnitude bias in prediction of percent fat, FM, LST, or BMC. The 95% confidence intervals were similar between the right and left side methods. The left half-scan was also studied, but it was similar to the right half-scan for all metrics. Rothney et al. (2009) indicated no significant differences between the estimated relative and absolute FM and LST from half-scan compared to whole-body scan results. However, the slight differences between right and left-scan BMC did reach statistical significance. Although the group difference was small, 30 g or 1%, the individual variation ranged from -66 to 95 g (-3.5 to 3.38%) compared to the whole-body BMC values. This is likely

due to lateral BMC differences in right- vs. left-handed subjects. Rothney et al. (2009) referred that handedness was not assessed, but since 90% of the general populations are right-handed, the difference in BMC between right and left scans makes sense. The authors further examined BMC differences in upper and lower limbs between right vs. left sides and found that these were more pronounced for arms (right − left: 5.9 ± 9.17 g) than for legs (2.99 ± 16.1 g). The small differences observed suggest that there is a low risk of introducing systematic bias into a data set by utilizing both whole-body and half-body scans within a single study. No significant trends in between-individual magnitude bias in the differences between half-body and whole-body scans were found. This suggests that the half-body DXA scan results can be comparable to whole-body scans even for larger subjects.

Breithaupt et al. (2011) found no significant differences between half- and full body DXA scans for percent fat, total mass, FM, LST and BMC, as observed in Table 2.7. Small but insignificant differences in absolute values were present within the data between left- and right-side scans. The authors showed that left-side half-body scans were found to overestimate whole-body total mass by 0.14 kg, FM by 0.06 kg, and LST by 0.08 kg whereas right-side half-body scans underestimated whole-body total mass by 0.17 kg, FM by 0.08 kg, and LST by 0.08 kg. No differences were observed between right- and left-side scans for BMC. There was a very strong correlation for percent fat, total mass, FM, lean mass and BMC ($p < 0.01, r^2 = 0.996 − 1.0$) for both half-body side compared to whole-body scans. Lower limits of agreement were observed between half-body estimates and whole-body measurements for all body composition variables.

Reflexion and summation scanning According to Sherman (2011), as a consequence of existing side-to-side differences for upper limbs, small significant differences resulted for whole-body bone but no differences on the soft tissue measures, and it was concluded that the reflected arm values on whole-body scans could impact impact on the accuracy of bone measures. Conceptually, summation scans do not have this issue and Nana et al. (2012) verified that body composition estimates for BMC, LST and FM from summed half-scans of BMC were not substantially different to those of the reference whole-body procedure; i.e. they were less than the respective smallest worthwhile effects[7].

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[7] Nana (2013) defines smallest worthwhile effects as TEM (CV%). These were as follows: BMC 40 g (1.3%);LST 700 g (1.4%); FM: 340 g (2.3%).

2.6.1 Evidence for side-to-side differences

Although side-to-side differences are neglected by partial scanning techniques, this simpli-fication is not supported by publications from researchers in anthropology related fields, physical therapy and neuroscience. This section explores what arguments have been put forward about the influence of handedness, behavioral factors or others (eg.: gender, sports practice, age group) on side-to-side differences in bone measurements at the upper and lower limbs. Unfortunately, the lean soft tissue compartment has not been thoroughly explored in the literature, but since exercise provides weight-bearing stimulus to bone (Layne & Nelson, 1999), muscular and bone development are generally associated (Ireland et al., 2013).

Effect of handedness Hand use patterns are complementary and differentiated, where one hand executes complex tasks while the other performs low frequency tasks, such as supporting an object. Humans are strongly biased towards a specific pattern of hand-use, favoring the right hand (Uomini, 2006). "Right-handers" are thus defined as people who prefer to adopt the precision role with the right hand and the support role with the left hand. On average, the population-level right-handedness appears to remain relatively constant around the 90% mark (Cashmore et al., 2008), although there is some variation in the proportion of left-handers between groups ranging from $\approx$ 3 to 27 % (Llaurens et al., 2009; McManus, 2009). As one would expect from the plasticity of body tissues, bone and skeletal muscle adapt to repeated activity patterns involving mechanical loading (Layne & Nelson, 1999). Accordingly, the differentiated roles of the opposing sides results in side-to-side differences, particularly in the upper limbs (Steele & Mays, 1995).

Bilateral asymmetry in long bones of the human skeleton—particularly from upper limbs—have been extensively studied researchers in archaeology, human origins and lin-guistics. Using measurements of the living and fossil bones from archeological collections, the most commonly compared dimensions have been the weight, lengths and/or breadths of the major long bones of the limbs (humerus, radius, femur, and tibia) (Auerbach & Ruff, 2006; Ruff & Jones, 1981). In order to make inter-individual comparisons regardless of the specific body size of the regions of interest, the quantification of asymmetries usually involves the measurement of asymmetry indexes and statistical procedures (Auerbach &

Ruff, 2006; Carpes et al., 2010; Cuk et al., 2001). One such index—and an extensively used one—is the percentage directional asymmetry ($DA_\%$), which is given by:

$$DA_\% = 100 \left[\frac{R - L}{\frac{R+L}{2}} \right] \qquad (2.8)$$

where R and L are the right and left side measures, respectively. $DA_\%$ also informs on the direction of the asymmetry; negative sign implies left bias and positive implies right side bias. Although derived from a diverse assortment of groups living under a variety of environmental conditions, studies agree that upper limb bones are more bilaterally asymmetric in all dimensions and weight than lower limb bones (Auerbach & Ruff, 2006). Also, right upper limb bones average significantly longer ($\approx$ 1-3%) and heavier ($\approx$ 2-4%) than left upper limb bones (Ruff & Jones, 1981).

Studies on fossil bones can only provide qualitative evidence for side-to-side adaptations (identifiable as directional asymmetry in long bone features) as a consequence of bimanually differentiated and "tool using" activities (Uomini, 2009). But most of these findings are supported by studies using DXA. Akar et al. (2002) investigated whether handedness had an asymmetric effect on distal forearm BMC and BMD, and whether there is an effect of gender on these variables. The mean BMCs and BMDs in bilateral distal forearms were compared in right-handed men and women. If it was the length, rather than the structure of bone, that was different in right vs. left sides, it would be expected that the BMC was higher on the right side. Conversely, differences in terms of BMD would be minimal. Results were consistent with this hypothesis. The right-BMCs were found to be significantly higher than left-BMCs for all regions of the bones studied. Results indicated that the mean BMCs, but not BMDs, were significantly higher in the right than in the left arm at different distal forearm regions, independent of gender. Kontulainen et al. (1999) compared the playing and contralateral arms' BMC of 13 competitive male tennis players ($\approx$ 25 yrs of experience) and 13 controls. It was observed that side-to-side BMC differences were largest in the humeral shaft (25%) and proximal humerus (19%), and the radial shaft and distal radius differences were $13 - 14\%$. In contrast, in controls, the side-to-side BMC differences were small ($< 5\%$).

Crossed symmetry Some studies have reported left-bias in lower limb bone dimensions, which, combined with the contralateral asymmetry in upper limbs, has been termed

"crossed symmetry" (Auerbach & Ruff, 2006). In this case, the lower limb was expressed by the stronger tibia usually on the opposite side of the dominant arm. Thus, right-handers usually have a stronger left leg, left-handers a stronger right. For right-handers, the long bones of the left lower limb (particularly femur) may be slightly longer and heavier (< 1%) than the right lower limb bones (Ruff & Jones, 1981), but, regardless of the hand preference, the supportive limb is associated with greater development (Cuk et al., 2001). Interestingly, more recent populations show a diminishing of the directionality and magnitude of asymmetry, as well as less sexual dimorphism in asymmetry. In this regard, Auerbach & Ruff (2006) suggest that influences from behavioral factors are implicated as the source of these patterns, probably reflecting changes in exogenous factors, such as industrialization and division of labor.

There are descriptive studies using DXA that provide evidence for crossed symmetry in young non-athletic and athletic populations, particularly those engaged in lateral dominant sports practice. Gumustekin et al. (2004) evaluated BMD in a sample comprising 32 right- and 26 left-handed university students. The right and left, total and regional proximal femur BMDs were measured and it was verified that the mean total BMD of the total right-handers and the mean trochanteric BMD of the right-handed males were greater in the left femur. Conversely, left-handers had higher mean intertrochanteric BMD in the right side than in the left side. The results suggest that femur-BMD may be related to hand preference. The results reported by Gumustekin et al. (2004) were not controlled by gender and sports practice, but the study of McClanahan et al. (2002) does not have these limitations; this research team investigated the effects of participation in various sports on side-to-side differences in BMD of the upper and lower limbs. The subjects were 184 collegiate athletes of both genders who participated in baseball, basketball, football, golf, soccer, tennis, cross-country, indoor/outdoor track, and volleyball. Results revealed greater BMD of the right arms compared with the left arms for all teams, with the most pronounced differences observed in men's and women's tennis and men's baseball. Differences in the lower limbs were only observed in lower limb BMD of male football and tennis players, with the non-dominant leg having greater bone mass. The results for football players are puzzling; it would be expected that the dominant lower limb would be stronger because of shooting actions. However, Nazarian et al. (2010) confirmed that it is indeed the non-dominant lower limb of football players that has higher BMD. It was

shown that the non-dominant leg of these atheletes (n=15) had significantly higher BMD than their dominant leg (1.34 vs. 1.29 g/cm^2), whereas differences were not significant at the control group (n=14). The superior BMD for the non-dominant leg was explained by frequent engagement in take off, landing and stance in shooting. In addition to differences at limbs, sports that require lateral dominance combined with increased spinal flexion and rotation may also relate to structural asymmetry of the pelvis (Bussey, 2010).

Cyclic activities   The rationale provided for crossed symmetry as a consequence of ballistic actions does not seem to hold for sedentary individuals who do nothing else but ambulatory functions. In some studies, lower limbs are found to be similar to one another, or the side-to-side differences do not reach statistical significance (Nazarian et al., 2010). Rothney et al. (2009) examined BMC differences for upper and lower limbs between the right vs. left sides in a sample of 52 obese subjects. Notwithstanding the BMC of the right upper limb being higher (right − left: 5.9 ± 9.17 g), there were also small but positive differences for BMC favoring the right lower limb (right-left: 2.99 ± 16.1 g), which were postulated to be due to walking. Likewise, athletes engaged in cyclic activities may also exhibit differences between lower limbs favoring the right side, particularly running and cycling. In a revision of studies on running and cycling performance in healthy subjects, Carpes et al. (2010) verified that evidence supporting symmetry is small. Regarding side preference for the lower limb, literature is consistent in suggesting that only 25–45% of people exhibit right leg preference in lower extremity actions (Cuk et al., 2001). However, studies failed to show a general association between functional asymmetry during running and lateral preference for lower limbs. As for the bilateral assessment of pedaling, cyclists have been shown to present frequent asymmetry. The extent of the asymmetries can vary within subjects and is different for the upper and lower limbs. A finding that is consistent throughout the literature is that the dominant leg, identified as the kicking leg, can generally contribute more to generate propulsion regardless of cadence (Carpes et al., 2010). Based on the current literature, it is not possible to ascertain the extent of differences at the lean soft tissue compartment.

Gender and age factors   Sexual dimorphism in asymmetry is present in some dimensions, especially those of the upper limb, and may implicate differences in behavior and growth. Females have more asymmetric and right-biased upper limb maximum lengths, while males have greater humeral breadths, but the lower limbs demonstrate little sexual

dimorphism (Auerbach & Ruff, 2006). Independent of gender, the pattern of asymmetry decreases with age, primarily due to reduced physical activity and greater losses of cortical bone (Ruff & Jones, 1981).

Summary Individual, lifestyle and sports related factors result in side-to-side adaptations in bone tissue. There is qualitative evidence for side-to-side adaptations at the upper limb as a consequence of handedness. Lateral dominant sports practice (eg.: tennis, basketball, football) induce crossed symmetry, favoring the dominant upper limb and contralateral lower limb. On the other hand, cyclic activities seem to induce superior development at the right lower limb. There are also reasons to suspect that gender and age group impact on the extent of side-to-side differences. Because weight-bearing stimulus develops bone and skeletal muscle (Layne & Nelson, 1999), the qualitative evidence provided for bone measurements may also hold for lean soft tissue at limbs.

2.6.2 Summary

This section pointed out that half-body scanning is a valid approach to assess individuals wider than the DXA table. The outcome measurements for BMC, LST, FM and % FM are closely comparable to whole-body scans using Lunar DPX-1 and the iDXA scanner in children and adults. As for reflexion-scanning of the upper limb and summation scans of two partial scanning, these also seem to work for Hologic QDR-4500A and Lunar Prodigy, and non-athletic and athletic populations, respectively.

2.7 Objectives

This research aims to develop and validate reflexion scanning techniques to evaluate whole-body composition of broad individuals in existing Hologic Explorer-W systems (max. width: 65 cm). Procedure definition encompasses not only options to select and combine body regions for image analysis, but also additional concerns with overall scanning time, safety and comfort for the subject. Factors that can affect morphological asymmetry, including gender, athletic status and being involved in lateral dominant sports are also taken into consideration. A second objective to this study is to compare, at the group and individual level, the performance of reflexion and half-scanning. This investigation is circumscribed to healthy adult populations and to the three DXA compartments: BMC, LST and FM (absolute value and %FM).

Methodology

3

> *"Divide & conquer" (divide et impera)*
>
> — **Julius Ceasar**
> The principle of dividing something large into
> smaller units, so it can be dealt with more easily.

This chapter contains an overall description of the approach taken in this thesis, including subjects, equipment and methods. Section 3.1 describes the sample and controlled variables, while also providing technical specifications for the used DXA system and the protocol for body composition examination. Sections 3.2 and 3.3 describe the partial scanning techniques and statistical analysis that make up the core of this investigation.

3.1 Subjects

This study is ancillary to the "Promoção do Exercício e Saúde na Obesidade" (P.E.S.O., 2005-2007) cohort study and also to the technical study of Santos et al. (2012). PESO, the methodology of which in described in Silva et al. (2008), was a weight-loss randomized controlled trial whose baseline population consisted of obese and overweight women (non-athletes). Participants were followed up for one year and the body composition evaluation moments took place and the beginning and at end of the intervention. Santos et al. (2012) aimed to validate a technique for evaluating tall individuals in Hologic Explorer-W densitometers. For that purpose, the recruited population consisted of non-athletes and athletes engaged in dominant and non-dominant lateral sports practice. The athletic group was comprised of national elite athletes of different sports: triathlon, judo, rowing, track and field athletics, pentathlon, tennis, basketball and wrestling. Both studies collected least one DXA whole-body scan per subject in an Explorer-W densitometer according to the guidelines of Hologic. The Ethics Committee of the Faculty of Human Kinetics approved both studies and all participants gave informed consent. In addition, fifteen additional male rugby players were evaluated and analyzed. Eight additional subjects that approached or exceeded the width limit of the DXA were also recruited to assist in the protocol definition and assessment of the feasibility of the alternative scanning techniques presented in Section 3.2.

3.1.1 Body composition assessment

The protocol for determining body composition in DXA involved preparation/instructions to subjects, anthropometrics and the actual DXA examination for scan acquisition. Overall, each session lasted for approximately 30 minutes.

Preparation Each participant was instructed to fast for, at least, $\geq$ 3h before coming for a morning visit at the study site (University of Lisbon, Faculty of Human Kinetics, Exercise and Health Laboratory, Cruz-Quebrada – Lisbon) and to wear minimal clothing. They were further asked to remove all objects that would interfere with image acquisition, such as jewelry, watches, hair ornaments, glasses, keys and wallets. The following criteria were used for excluding participants: a) Tests taken with radiographic contrast material in the past 72 hours or participation in nuclear medicine studies in the past 3 days; b) Being pregnant; and c) The participant's weight exceeds 136 kg and/or his body dimensions in the supine position are over the DXA table limit (height > 196 cm, width of trunk plus arms >67 cm). All other subjects were asked to undertake a DXA examination.

Anthropometric data Prior to the DXA examination, weight and height were measured with a calibrated scale (BOD POD, Cosmed, Inc., CA, USA) and a stadiometer (Seca, Germany) according to the ISAK (2013) guidelines. Weight and height were measured to the nearest 0.01 kg and 0.1 cm, respectively. These anthropometric variables, the individual's age, gender, and ethnicity were inserted into the interface of the DXA's analysis software and only then the actual DXA examination was initiated.

DXA examination An Hologic Explorer-W (software QDR for windows v13.3 with APEX v3.3 for image analysis, MA, USA) narrow fan-beam system was used in whole-body composition mode. Based on 30 healthy subjects (fifteen of each gender), the CVs in the Exercise and Health Laboratory for BMC, LST, FM and %FM are 1.0%, 0.7%, 2.8% and 0.6%, respectively. The TEMs are 0.02 kg for BMC, 0.36 kg for LST, 0.37 kg for FM, and 0.1% for %FM. The manufacturer's acquisition procedures were followed, implying that participants laid flat in the supine position on the DXA table and each whole-body scan took approximately 7 minutes. After acquisition, BMC, LST, FM and %FM were estimated from the scans using Hologic's proprietary software.

3.1.2 Database construction

DXA scans of 198 eligible subjects were analyzed for this study, including athletes and non-athletes of both genders whose overall phenotypes ranged from thin to obese (WHO Working Group, 1986). The sample was described by age (years), weight (kg), height (cm), gender, BMI (kg/m^2), percent fat mass, athletic status (being a competitive athlete or not) and, for athletes, a lateral dominant sports practice (LDSP) status. Lateral dominant sports are those that induce morphological asymmetry as a consequence of preferential use of dominant limbs for repeated actions, as happens in sports that require throwing and shooting actions. Sports classified as lateral dominant were handball (n=15), rugby (n=15), basketball (n=10), volley (n=6), tennis (n=2), running / barriers (n=2), and judo (n=1). Non-lateral dominant sports are the ones that involve even use of limbs, namely sports that involve cyclic activities like running, cycling, rowing and swimming. Within this category, this study included triathletes (n=19), rowers (n=15), swimmers (n=14) and sprint runners (n=2). The descriptives for athletes vs. non-athletes, males vs. females and whole sample are reported in Table 3.1 as means ± standard deviations.

Exclusion Criteria Scans were set as invalid as a result of jewelry and other objects not removed by participants; the presence of non-removable objects such as prostheses, pacemakers, breast implants; extremities overlap or are outside the scan area of DXA; and other reasons, including amputees. Different technicians positioned the participants and performed the scans, but it was a single technician that analyzed them according to the operator's manual for the standard whole-body protocol (reference scan) and according to the alternative techniques that are described in the Section 3.2.

Outcomes of Interest Soft tissue and bone measures reported by the software and relevant for this study included BMC (g), LST (g), FM (g) and %FM. By configuring standard the regions of interest (ROI) available in the manufacturer's software, the analysis resulted in whole-body and regional measures. Relevant ROIs are head, trunk and upper / lower limbs. Measures were also obtained for a user-defined (half-scan) rectangular region at the right-hand side of the body, including the right libs and trunk, but not the head (see Figure 6.2 for details).

Table 3.1: Descriptive characteristics of athletes, non-athletes and whole sample.

VARIABLES	ATHLETES			NON-ATHLETES			WHOLE SAMPLE		
	Male	Female	Subtotal	Male	Female	Subtotal	Male	Female	Total
	(n=50)	(n=41)	(n=91)	(n=27)	(n=80)	(n=107)	(n=77)	(n=121)	(n=198)
Age (yrs)	20.4 ± 3.2	21.4 ± 5.9	20.9 ± 4.6§	26.8 ± 7.8	36.0 ± 9.5	33.6 ± 9.9§	22.7 ± 6.1‡	31.0 ± 10.9‡	27.8 ± 10.2
BW (kg)	74.7 ± 9.7	62.1 ± 7.9	69.0 ± 10.9	73.3 ± 9.0	67.4 ± 12.7	68.9 ± 12.1	74.2 ± 9.4‡	65.6 ± 11.6‡	69.0 ± 11.6
Height (cm)	178.5 ± 5.1	168.6 ± 6.3	174.0 ± 7.5§	176.7 ± 5.5	161.4 ± 6.2	165.3 ± 9.0§	177.9 ± 5.3‡	163.8 ± 7.1‡	169.3 ± 9.4
BMI (kg/m²)	23.4 ± 2.6	21.8 ± 2.0	22.7 ± 2.5§	23.4 ± 2.4	25.9 ± 4.8	25.3 ± 4.5§	23.4 ± 2.5	24.5 ± 4.5	24.1 ± 3.9
%FM	15.70 ± 3.84	24.38 ± 5.21	19.78 ± 6.27§	13.72 ± 4.32	31.24 ± 7.92	25.28 ± 10.81§	14.42 ± 4.24‡	29.88 ± 7.93‡	23.87 ± 10.12
LDSP	n=27	n=24	n=51	—	—	—	—	—	n=51

Abbreviations: BW, Body Weight; BMI, Body Mass Index; FM, Fat Mass; LST, Lean Soft Tissue; BMC, Bone Mineral Content; LDSP, Lateral Dominant Sports Practice.

Significant differences: ‡ between genders, § between athletes and non-athletes ($p < 0.05$).

3.2 Partial scanning techniques

Figure 6.2 depicts an whole-body reference scan of a subject alongside the investigated partial scanning techniques (half-scan and two options for reflexion scanning). In all representations, the body is segmented into limbs, trunk and head. For the half-scanning protocol, an additional region of interest consists of an "halved" right-side trunk and limbs. Black and grey pixels represent BMC and soft tissue, respectively, which is further decomposed into LST and FM.
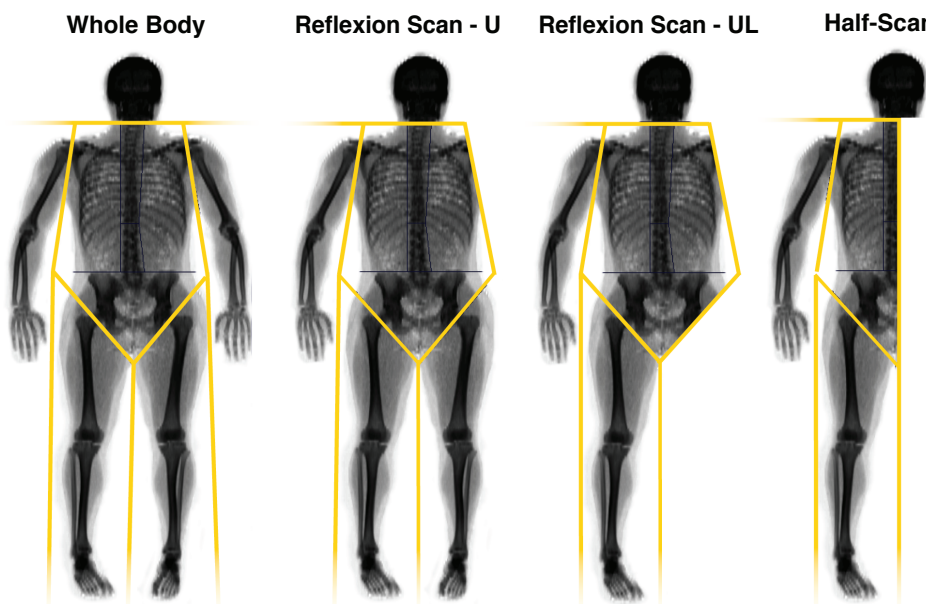

Figure 3.1: Partial scanning techniques for DXA analysis of broad subjects

As specified on Table 3.3, taking reflexion scans that only exclude the upper left limb (RSU) is the most conservative approach, as it only needs to mirror the right upper limb. However, it might still not be possible to scan both lower limbs in practical situations. Thus, an alternative approach that does not require the left leg to fit within the scanning area of the DXA — Reflexion Scan without the upper and lower left limbs (RSUL) — shall also be considered. In contrast to the HS strategy proposed by Tataranni & Ravussin (1995), neither RSU nor RSUL require the trunk to be halved.

Table 3.3: Compared partial scanning techniques for broad subjects.

TECHNIQUES	REQUIRED ANATOMICAL REGIONS	UNKOWN
Half-Scan	Head , Left side of the body	Right side of the body
RSU	Head, Trunk, Right limbs, left lower limb	Upper left limb
RSUL	Head, Trunk, Right limbs	Left limbs

Abbreviations: Reflexion Scanning (RS): RSU – upper left limb removed; RSUL– upper and lower left lims removed.

For RSU and RSUL, insights were drawn from tests with wide individuals who actually exceeded the width limits of the DXA systems, which were explored further by questions in order to establish appropriate positioning and also to determine the maximum subjects' width for which the proposed procedures are recommended. Under the symmetry assumption with respect to the right and left sides of the body, the estimates for the DXA compartments and %FM can be obtained from the scanned anatomic regions according to Equation 3.1:

$$x_{RSU} = Head + Trunk + 2\,Arm_R + Leg_R + Leg_L \tag{3.1a}$$

$$x_{RSUL} = Head + Trunk + 2\,Arm_R + 2\,Leg_R \tag{3.1b}$$

$$x_{HS} = Head + 2\,Half\_Scan_R \tag{3.1c}$$

$$\%FM = 100\,FM/Total\_Weight \tag{3.1d}$$

where the measurements for the DXA outcomes of interest (BMC, LST, FM, %FM) by HS, RSU and RSUL are denoted in generic terms by x_{HS}, x_{RSU} and x_{RSUL}, and subscripts R and L denote right and left hand sides, respectively. In Equation 3.1c, $Half\_Scan_R$ refers to the right-side half of the [subtotal] body, comprising limbs and trunk.

Technical errors   In addition to the same sources of error that affect a standard whole-body scan (e.g., machine's inherent noise, technologist's positioning of subjects), partial scanning techniques are also affected by the propagation of measurement errors that Equations 3.1 entail. In this regard, the errors resulting from delimitation of ROIs during image analysis, namely the segmental lines that demarcate arms/trunk and hips/pelvis, should also be taken into consideration when referring to the reproducibility of total body composition measurements performed on a DXA machine in partial scanning mode. Accordingly, we investigated the short-term precision of total body composition measurements performed in our laboratory using RSU and RSUL. Thirty adults (15 females, 15 males) was scanned twice with repositioning to determine intraobserver[1] technical errors of measurement and coefficients of variation for the estimates of BMC, LST, FM and %FM. For RSU and RSUL, the TEMs and CVs for each compartment were calculated and expressed as $TEM_{x_{RSU}}$ or $TEM_{x_{RSUL}}$, respectively, with $x \in \{BMC, LST, FM, \%FM\}$.

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[1]The technologist was the same for all image analysis procedures undertaken in this investigation.

3.3 Statistical analysis

Statistical analysis was performed using IBM SPSS Statistics version 20.0, 2011 (SPSS Inc., IBM, Chicago, Illinois, USA) and MATLAB R2011a (The Mathworks, MA, USA). Unless stated otherwise, the statistical significance was set at $p < 0.05$.

Characteristics of the sample   The sample was described (means ± standard deviations) in terms of whole-body BMC, LST, FM, and %FM by reference procedures, weight, height and BMI (kg/m^2). Normality was tested using the Shapiro-Wilk or the Kolmogorov-Smirnov tests, depending on whether the subsample sizes were smaller or greater that 30 subjects, respectively. Independent sample t-tests were used for comparisons of subgroups split by gender and athletic status. In addition, for BMD (g/cm^2), BMC, LST and FM, we also performed paired sample t-tests to compare [upper and lower] contralateral limbs with one another of subgroups split by athletic status and lat. dom. sports practice.

Estimates from partial scans   The validation approach of this thesis proceeded with the calculation of estimates for whole-body BMC, FM, LST and %FM by the partial scanning techniques (HS, RSU and RSUL; see Equations 3.1 for details). After this preliminary step was concluded, we sought information on the accuracy with which x_{PS} explained the corresponding whole-body estimates for each compartment, with $PS \in \{HS, RSU, RSUL\}$ and $x \in \{BMC, LST, FM, \%FM\}$.

Confounding factors   In order to examine whether x_{PS} interacted with confounds to explain whole-body estimates (x_{WB}), two-way ANOVA tests were conducted. For each compartment, the interaction of gender, athletic status and lateral dominant sports practice with each main predictors (x_{PS}, estimates for BMC, LST, FM and %FM from the alternative procedure) was tested in separate models:

$$x_{WB} = \beta_0 + \beta_1\, x_{PS} + \beta_2\, gender + \beta_3\, gender\, x_{PS} + \epsilon \tag{3.2a}$$

$$x_{WB} = \beta_0' + \beta_1'\, x_{PS} + \beta_2'\, athletic\_status + \beta_3'\, athletic\_status\, x_{PS} + \epsilon' \tag{3.2b}$$

$$x_{WB} = \beta_0'' + \beta_1''\, x_{PS} + \beta_2''\, LDSP + \beta_3''\, LDSP\, x_{PS} + \epsilon'' \tag{3.2c}$$

If interactions were statistically insignificant or negligible, stepwise multiple regression tests were further undertaken to investigate whether athletic status, gender and regular

practice of lateral dominant sports introduced significant contributions to explain on the outcome variables in HS, RSU and RSUL. The simplified models were:

$$x_{WB} = \beta_0 + \beta_1 \, x_{PS} + \beta_2 \, gender + \epsilon \qquad (3.3a)$$

$$x_{WB} = \beta_0' + \beta_1' \, x_{PS} + \beta_2' \, athletic\_status + \epsilon' \qquad (3.3b)$$

$$x_{WB} = \beta_0'' + \beta_1'' \, x_{PS} + \beta_2'' \, LDSP + \epsilon'' \qquad (3.3c)$$

where the β coefficients were only included if they were significant, $p < 0.05$. Upon the completion of tests for confounding factors, the approach for validating partial scanning techniques against reference whole-body scanning proceeded as depicted in Figure 3.2.
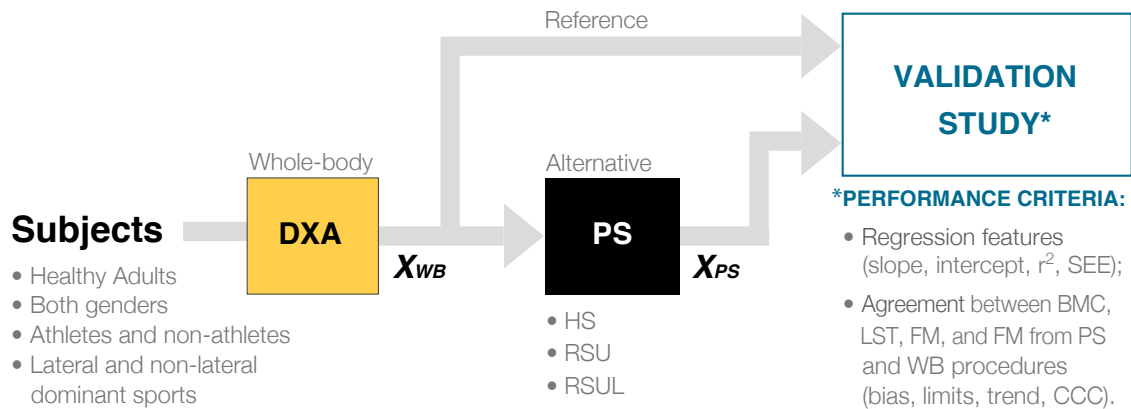

Figure 3.2: Validation of partial scanning for whole-body composition assessment.

The relationship between alternative (half- or reflexion scanning), and reference (whole-body) estimates was examined by linear regression and agreement analysis. Agreement between both methods was assessed by Bland & Altman (1986) analysis and by the inspection of the concordance correlation coefficient.

Linear regression analysis   Linear regression models were then developed for $x \in \{BMC, LST, FM, \%FM\}$ and $PS \in \{HS, RSU, RSUL\}$. As expressed in Equation 3.4, the [reference] whole-body measurements were the dependent variables and the [alternative] partial scan measurements were the dependent ones:

$$x_{WB} = \beta_0 + \beta_1 \, x_{PS} + \epsilon \qquad (3.4)$$

By the least mean squares algorithm, β_0 and β_1 were determined in order to minimize the errors ϵ between x_{WB} and the regression outcome $\hat{x}_{WB} = \beta_0 + \beta_1 \, x_{PS}$. β_0 and β_1 are, hereafter, referred to as intercepts and slopes, respectively. The coefficient of determination r^2 and the standard error of the estimate (SEE) were also determined for all outcome variables.

Bland-Altman analysis   To test of the agreement between methods for whole-body BMC, FM, LST and %FM, the Bland-Altman analysis was conducted. First, the mean difference of alternative (x_{PS}) minus whole-body estimates, defined as bias, was calculated for $PS \in \{HS, RSU, RSUL\}$, and its statistical significance was tested with paired sample t-tests. Then, the magnitude of individual errors of x_{PS} were inspected based on the individual differences $\Delta_{PS,i} = x_{PS,i} - x_{WB,i}$, with i being the subject index of the sample, $i = 1 \ldots N$ and $PS \in HS, RSU, RSUL$ (Bland & Altman, 1986). After the standard deviation of Δ_{PS} was determined, the 95% limits of agreement were determined by $[-1.96\ SD; 1.96\ SD]$. As for possible trends with increasing values, linear regressions were performed on Δ_{PS} (dependent variable) against $(x_{WB} + x_{PS})/2$ (independent variable). The statistical significance of the coefficient of correlation (here referred to as trend) of the linear regression $\hat{\Delta}_{PS} = f[(x_{WB} - x_{PS})/2]$ was also tested.

Concordance analysis   The concordance correlation coefficient (CCC) was also computed to evaluate the extent by which observations for x_{PS} were in agreement with the corresponding reference values x_{WB} (Lin, 1989; McBride, 2005; McGraw & Wong, 1996). The concordance correlation coefficient is a normalized parameter that consists of the product of the Pearson correlation coefficient (r) with a bias correction factor (Cb). In this sense, r's Pearson is the correlation obtained for the regression $\hat{x}_{WB} = \beta_0 + \beta_1\ x_{PS}$ (see defining model in Equation 3.4) and informs on precision, whereas the latter provides insight on how close the points $(x_{PS,i}, x_{WS,i})$, $i = 1 \ldots N$ are to the ideal line $x_{WB} = x_{WB}$ (a 45º line through the origin). The practical interest of CCC stems from the existence of threshold values against which x_{PS} can be compared to infer the strength of agreement of the alternative techniques. As suggested by McBride (2005), a possible descriptive scale comprises values of CCC < 0.90, $0.90 - 0.95$, $0.95 - 0.99$ and > 0.99 that indicate poor, moderate, substantial and almost perfect strength of agreement, respectively.

Summary   The statistical variables that shall be used for validation and comparison of techniques are as follows: mean values for the measurements by the different techniques (mean ± SD), parameters derived from linear regression analysis (r^2, SEE, Intercept, Slope) and agreement analysis (Bias , 95% LoA, Trend, CCC, Pearson correlation coefficient r and the agreement factor Cb).

Results

> *"Bloom, o nosso herói. Eis o que faz primeiro: observa."*

— **Gonçalo M. Tavares**

Viagem à Índia

This chapter was organized with the aim of providing relevant data for comparing and validating the major solution concepts of this thesis. Firstly, we provide information pertaining to the reliability of whole-body and regional body composition assessments reported throughout this investigation. Accordingly, the TEMs and CVs specific to our DXA machine (Hologic Explorer-W) and the conditions of its use are listed in Table 4.1. The reported metrics were computed in thirty subjects with repositioning in between scans, as recommended by Nana (2013) and Bonnick & Lewis (2006).

Table 4.1: TEMs and CVs por whole-body scanning and selected regions of interest.

	TEM (kg)				CV(%)				
PS	**BMC**	**LST**	**FM**	**%FM**	**BMD**	**BMC**	**LST**	**FM**	**%FM**
WB	0.023	0.362	0.370	0.063%	0.85	0.99	0.74	2.78	0.57%
MS_R	0.003	0.072	0.051		2.26	2.12	2.63	7.54	
MS_L	0.003	0.070	0.052		1.71	2.26	2.70	7.51	
MI_R	0.010	0.134	0.111		1.84	2.26	1.56	3.99	
MI_L	0.009	0.156	0.140		2.05	2.13	1.89	5.13	

Acronyms: TEM: Technical Error of the Measurement; CV, Coefficient of Variation; PS, Partial Scanning techniques; ROI, Region of Interest; BMC, Bone Mineral Content; LST, Lean Soft Tissue; FM, Fat Mass.

Section 4.1 provides the participant's characteristics, including comparisons of contralateral limbs. Then, on Section 4.2, partial scanning techniques are examined from a feasibility and practicality perspective. For this purpose, results are provided for preliminary tests that were undertaken on subjects that did not fit into the DXA table, and the TEMs/CVs that inform on the precision of partial scanning techniques for whole-body composition assessment are also provided. Finally, Section 4.3 provides validation results for the half- and reflexion-scanning techniques according to the performance criteria described in Chapter 3, as well as linear regression and Bland-Altman plots.

4.1 Participant's characteristics

Participants' characteristics (N=198) are described in Table 4.2, including body weight, BMC, LST, FM and %FM. The sample is subdivided according to gender and athletic status. Independent sample t-tests revealed significant differences between all characterizing variables except the body weight of athletes vs. non-athletes. Table 4.2 informs that the mean body weight of athletes vs. non-athletes and body weight of males and females are similar. However, the sample has a large variation of %FM ($23.87 \pm 10.12\%$). As could be expected, non-athletes have significantly more adiposity, less lean tissue and bone content than athletes, and the same observations hold for the comparison of females vs. males. Table 4.3 complements the former by providing details on side-to-side differences between limbs. Data on asymmetry were inspected based on BMD and percentage directional asymmetries (DA, %) (see Equation 2.8 on Section 2.6.1 for details). The sample was analyzed collectively and separately, according to athletic status, lateral dominant sports practice and gender[1]. Overall, Table 4.3 indicates that DA is higher for upper limbs and favors the right-hand/dominant side. For a more throughout inspection of side-to-side differences, we shall consider upper and lower limbs separately.

Upper limbs For subgroups (athletes vs. non-athletes; LDSP vs. non-LDSP; males vs. females) and for the whole sample, DAs for LST and BMC in upper limbs are about $\approx$ 4–9% (significant and superior to the corresponding CVs; $CV_{BMC, U}$ and $CV_{LST, U}$ are approximately 2%). Curiously, although the DAs for FM are below the CVs for FM ($\approx$ -5 – -4%; $CV_{FM, U}$: 7.5%), it tends to compensate body composition in the non-dominant limb. DA in the upper limbs is also significant for BMD of the whole sample ($1.3 \pm 4.1\%$, which corresponds to an average difference of just 10 grams favouring the right-hand side) and subgroups. For bone measurements, DAs are most severe for athletes engaged in lateral dominant sports practice For these, the DA for BMD was $3.4\% \pm 4.5\%$ (significant value that corresponds to a bone mineral content of 16 ± 14 g or $8.9 \pm 7.2\%$). Conversely, those classified as non-LDSP had a low and non-significant DA for BMD of upper limb ($DA_{BMD, U}$: 0.50 ± 3.71 % < $CV_{BMD, U}$). For LST , DAs were significant for subgroups and whole sample in the range $\approx 4 - 9\%$, where the smallest and greatest DAs were verified for males and females, respectively. However, the DAs for BMD of handball players alone was 7.1 ± 3.6 (corresponds to a $DA_{BMC, U}$ of 25.2 ± 9.9 g or $13.5 \pm 4.1\%$), and the maximum $DA_{BMD, U}$ was as high as 13.9% (results not reported in Table 4.3).

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[1]Note: the sample was separated according to athletic status, lateral dominant sports practice and gender because of the evidence collected and documented in Section 2.6.1.

Table 4.2: Mean group comparison of athletes, non-athletes and whole sample.

VARIABLES	ATHLETES			NON-ATHLETES			WHOLE SAMPLE		
	Male	Female	Subtotal	Male	Female	Subtotal	Male	Female	Total
	(n=50)	(n=41)	(n=91)	(n=27)	(n=80)	(n=107)	(n=77)	(n=121)	(n=198)
Age (yrs)	20.4 ± 3.2	21.4 ± 5.9	20.9 ± 4.6§	26.8 ± 7.8	36.0 ± 9.5	33.6 ± 9.9§	22.7 ± 6.1‡	31.0 ± 10.9‡	27.8 ± 10.2
BW (kg)	74.7 ± 9.7	62.1 ± 7.9	69.0 ± 10.9	73.3 ± 9.0	67.4 ± 12.7	68.9 ± 12.1	74.2 ± 9.4‡	65.6 ± 11.6‡	69.0 ± 11.6
Height (cm)	178.5 ± 5.1	168.6 ± 6.3	174.0 ± 7.5§	176.7 ± 5.5	161.4 ± 6.2	165.3 ± 9.0§	177.9 ± 5.3‡	163.8 ± 7.1‡	169.3 ± 9.4
BMI (kg/m^2)	23.4 ± 2.6	21.8 ± 2.0	22.7 ± 2.5§	23.4 ± 2.4	25.9 ± 4.8	25.3 ± 4.5§	23.4 ± 2.5	24.5 ± 4.5	24.1 ± 3.9
BMC (kg)	2.89 ± 0.54	2.27 ± 0.33	2.61 ± 0.55§	2.75 ± 0.37	2.11 ± 0.27	2.27 ± 0.41§	2.84 ± 0.49‡	2.16 ± 0.30‡	2.43 ± 0.51
LST (kg)	63.0 ± 7.4	46.6 ± 4.8	55.6 ± 10.4§	60.4 ± 6.9	43.2 ± 6.1	47.5 ± 9.8§	62.1 ± 7.3	44.4‡ ± 5.9‡	51.2 ± 10.8
FM (kg)	10.7 ± 4.0	14.9 ± 4.6	12.6 ± 4.7§	11.8 ± 4.3	23.5 ± 8.3	20.6 ± 9.1§	11.1 ± 4.1‡	20.6 ± 8.3‡	16.9 ± 8.4
%FM	15.70 ± 3.84	24.38 ± 5.21	19.78 ± 6.27§	13.72 ± 4.32	31.24 ± 7.92	25.28 ± 10.81§	14.42 ± 4.24§	29.88 ± 7.93‡	23.87 ± 10.12
LDSP	n=27	n=24	n=51	—	—	—	—	—	n=51

Abbreviations: BW, Body Weight; BMI, Body Mass Index; FM, Fat Mass; LST, Lean Soft Tissue; BMC, Bone Mineral Content; LDSP, Lateral Dominant Sports Practice.

Significant differences: ‡ between genders, § between athletes and non-athletes ($p < 0.05$).

Table 4.3: Side-to-side differences between upper and lower libs for athletes vs. non-athletes, athletes engaged in lateral dominant sports practice vs. others, males vs. females, and whole sample.

DIRECTIONAL ASSIMETRY, Variables %	ATHLETIC STATUS		LATERAL DOM. SPORTS		WHOLE SAMPLE		
	Athletes (n=91)	Non-Athletes (n=107)	LDSP (n=51)	Non-LDSP (n=147)	Male (n=77)	Female (n=121)	Total (n=198)
UPPER L. BMD (%)	1.20 ± 4.70*	1.2845 ± 3.59**	3.39 ± 4.51** (16 ± 14** g)	0.50 ± 3.72 (8 ± 8** g)	1.51 ± 4.72*	1.08 ± 3.72*	1.25 ± 4.13** (10 ± 10** g)
BMC (%)	6.48 ± 6.76**	7.36 ± 5.84**	8.87 ± 7.16**	6.29 ± 5.82**	6.08 ± 7.22**	7.51 ± 5.55**	6.95 ± 6.3**
LST (%)	5.27 ± 4.85**	8.94 ± 6.32**	6.51 ± 5.53**	7.51 ± 6.11**	4.29 ± 4.80**	9.14 ± 5.89**	7.25 ± 6.0**
FM (%)	-4.56 ± 16.07**	-3.67 ± 11.34*	-3.54 ± 13.95	-4.27 ± 13.65**	-3.81 ± 17.11	-4.25 ± 11.07**	-4.08 ± 13.7**
LOWER L. BMD (%)	-0.63 ± 4.16	-0.0771 ± 3.78	-0.85 ± 4.52 (-3 ± 28 g)	-0.15 ± 3.74 (8 ± 20** g)	0.01 ± 4.03	-0.55 ± 3.91	-0.33 ± 3.96 (5 ± 23* g)
BMC (%)	0.19 ± 4.67	2.39 ± 4.84**	-0.59 ± 5.05	2.06 ± 4.64**	0.64 ± 4.85	1.85 ± 4.85**	1.38 ± 4.9**
LST (%)	3.15 ± 3.09**	3.73 ± 3.65**	2.49 ± 3.22**	3.80 ± 3.42**	2.64 ± 2.57**	3.99 ± 3.77**	3.46 ± 3.4*
FM (%)	1.03 ± 7.51	1.78 ± 6.11*	1.70 ± 5.36*	1.34 ± 7.22*	0.59 ± 8.57	1.97 ± 5.31**	1.43 ± 6.8

Abbreviations: FM, Fat Mass; LST, Lean Soft Tissue; BMC, one Mineral Content; LDSP, Lateral Dominant Sports Practice; Upper L, Upper Limb, Lower L, Lower Limb. **Significance levels:** * p < 0.05, ** p < 0.001.
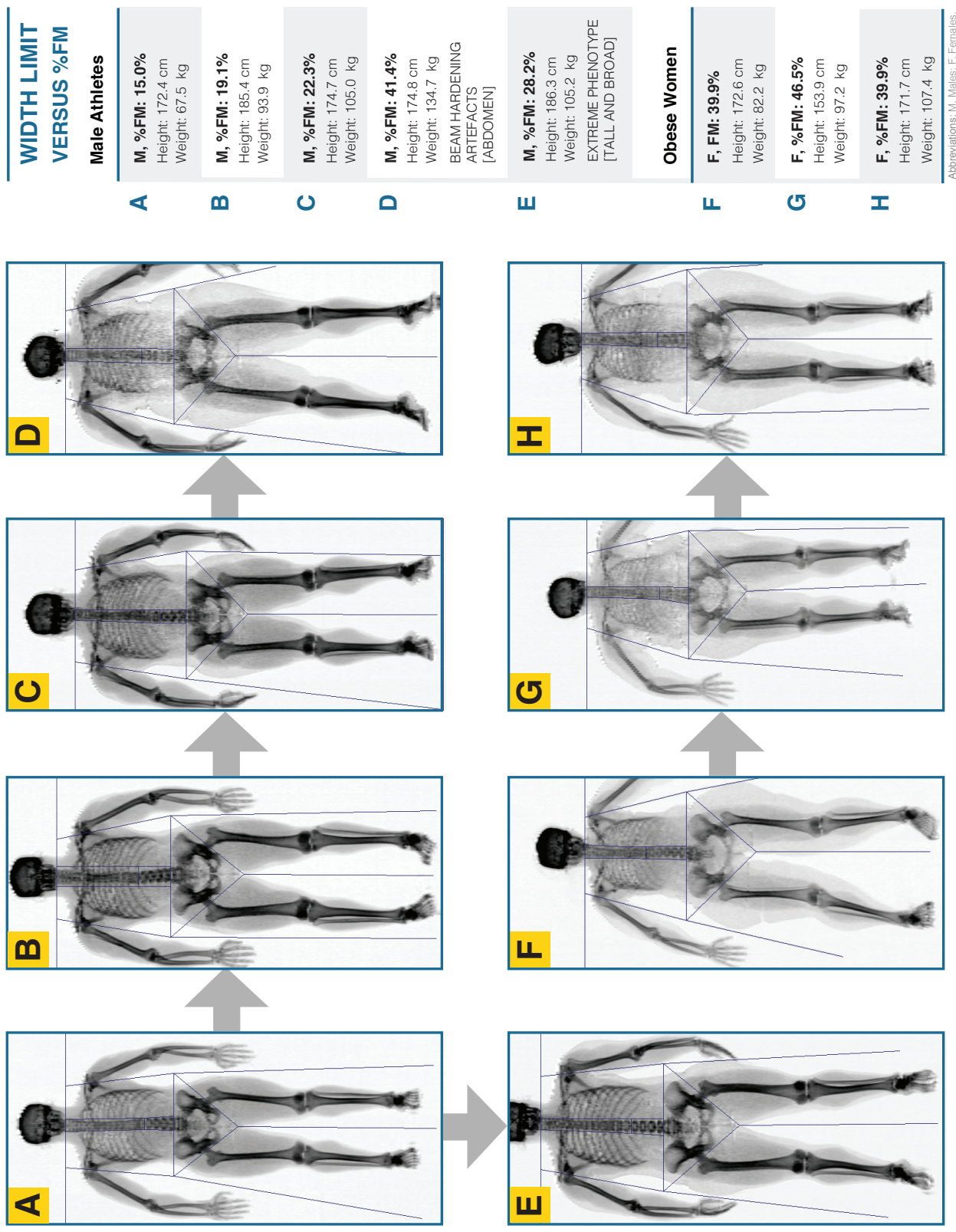
Bias (%), for the upper and lower limbs, is obtained as right side measurements minus the left side ones. It is expressed as a percentage after normalizing the absolute difference by the average value of the right and left limbs, for BMD, BMC, LST and FM

Lower limbs For subgroups and for the whole sample, the DAs for bone measurements and fat mass for lower limbs are less than 4%. Most variables did not reach statistical significance nor exceeded its respective CVs (1.9% for BMD, 2.2% for BMC, and 4.5% for FM). It is interesting to note that the bone measures for athletes in the LDSP group have the lowest (negative sign) results for $DA_{BMD, L}$, although it corresponds to just 3 grams. In handball players, however, $DA_{BMC, U}$ reached -22.4 ± 26.5 g (result not reported in Table 4.3). For LST, the $DA_{LST, L}$ was significant and superior to the CV for LST ($CV_{LST, L}$: 1.9%), but also less than 4% for all subgroups and whole sample.

4.2 Preliminary tests

Eight additional subjects whose dimensions approached or exceed the limits of the DXA table were recruited for preliminary tests meant to assess pertinence, comfort and safety for the patient, along with feasibility and practicality of partial scanning techniques. Included were 5 male rugby players (see Figure 4.1, case studies A – E, %FM 15.0 ... 41.3 %) and 3 obese females (F: 39.9%FM; G: 46.5%FM and H: 39.9%FM). As the %FM of rugby athletes varies from normal to obese (A – E), the width constraint of the DXA table becomes an issue that eventually compromises proper whole-body scanning. For muscular athletes whose weight ≈ 100 kg (B), regular scanning with palms facing the table may result in inaccurate representation of upper extremities. When the width in excess is minor (1–2 cm), an option to solve these cases is asking the subject to place both hands in midsaggital position (palms facing the hip). While this option seems to prevent hands and forearms from falling outside the active scanning area in some cases, including obese women and tall athletes (E), it seems to remain unable to provide a working solution for wide individuals (C, D, F, G, H). For these, a partial scan that does not include the upper left arm can still be obtained; F, G and H where positioned in such a way that their midsaggittal line was offset from the midline of the table and to their left-hand sides. In this way, the right limbs and trunk could be completely scanned and the software was then allowed to "mirror" the results of the completely imaged side to the contralateral limb(s).

Figure 4.2 depicts zoomed shoulders from scans of different individuals, including obese. It is apparent from these images that the demarcation of arms from trunk is not always clear cut as most images are slightly blurred in this region. Moreover, obesity and high muscularity turn cause overlap between trunk and arms.

WIDTH LIMIT VERSUS %FM

Male Athletes

A M, %FM: **15.0%**
Height: 172.4 cm
Weight: 67.5 kg

B M, %FM: **19.1%**
Height: 185.4 cm
Weight: 93.9 kg

C M, %FM: **22.3%**
Height: 174.7 cm
Weight: 105.0 kg

D M, %FM: **41.4%**
Height: 174.8 cm
Weight: 134.7 kg
BEAM HARDENING ARTEFACTS [ABDOMEN]

E M, %FM: **28.2%**
Height: 186.3 cm
Weight: 105.2 kg
EXTREME PHENOTYPE [TALL AND BROAD]

Obese Women

F F, FM: **39.9%**
Height: 172.6 cm
Weight: 82.2 kg

G F, %FM: **46.5%**
Height: 153.9 cm
Weight: 97.2 kg

H F, %FM: **39.9%**
Height: 171.7 cm
Weight: 107.4 kg

Abbreviations: M, Males; F, Females.

Figure 4.1: Preliminary tests of partial scanning techniques in wide individuals.

In order to minimize technical errors and enhance consistency, the segments demarcating arms from trunk were always placed by the same technologist and intersected the separation between scapula and humerus.

| I | II | III | IV |
| No bone ambiguity, Strength athlete. | Overlap of fat: limbs and trunk. | Blurred image, Non-obese. | Blurred image, Obese. |
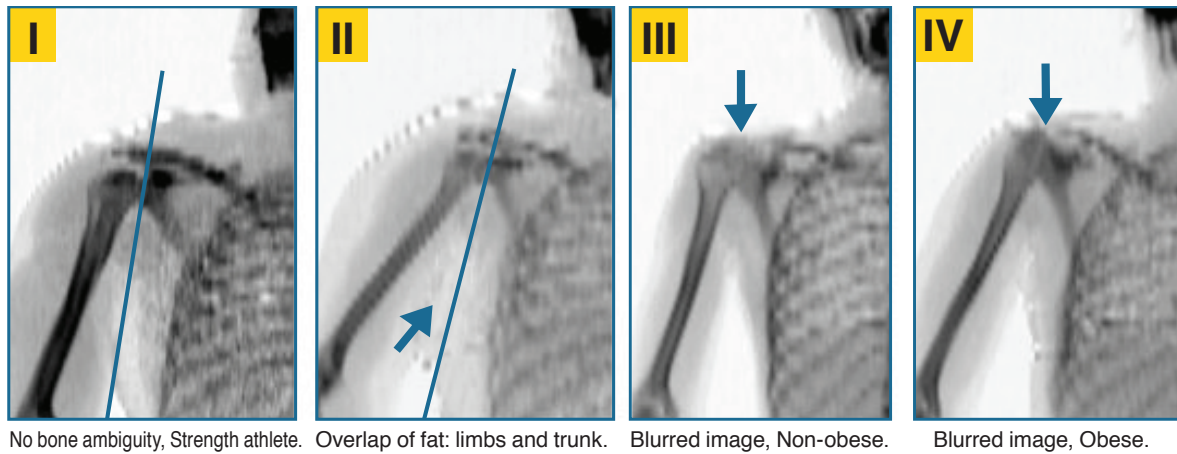
Figure 4.2: Delimitation of upper limbs in obese and non-obese subjects.

Figure 4.3 depicts the analysis software for Hologic Explorer W systems (version 13.0). When the scanning field is exceeded, a window pops up automatically.
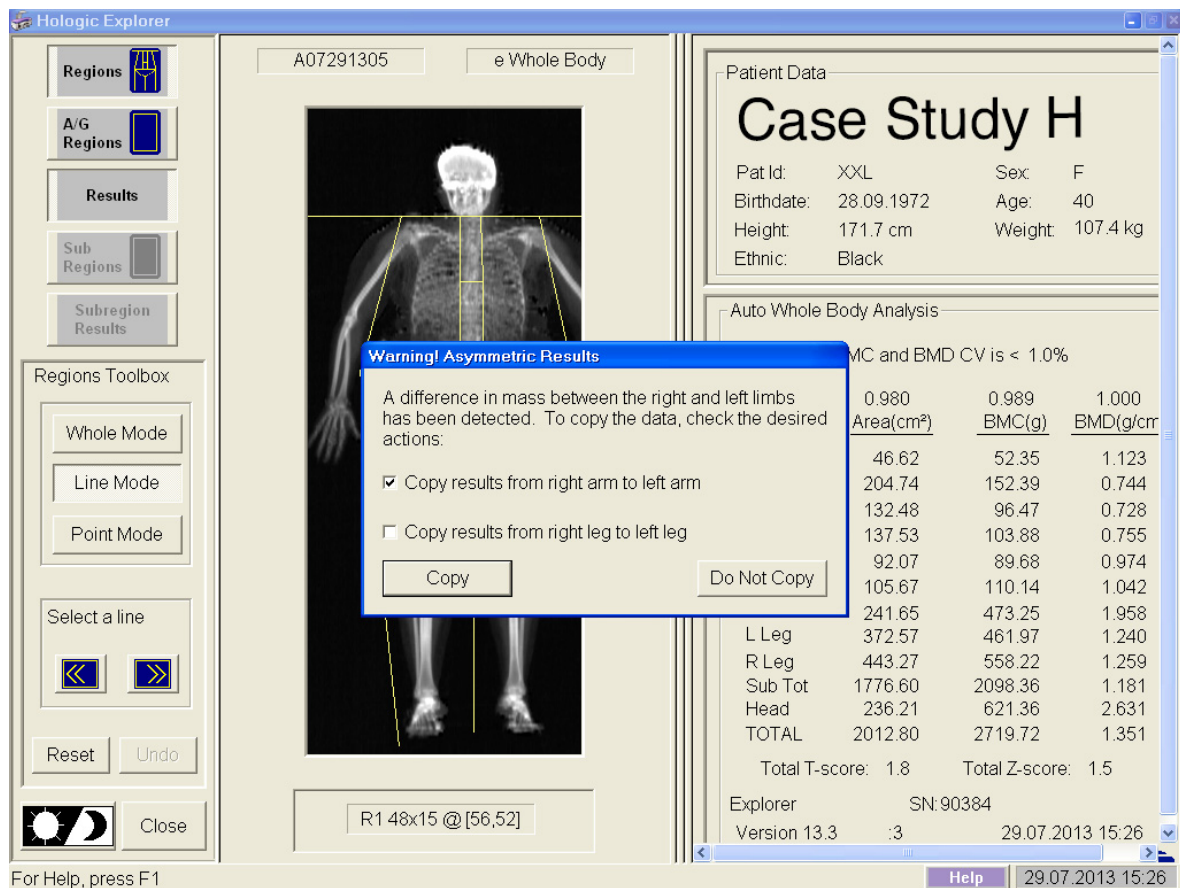
Figure 4.3: Graphic user interface for automatic reflexion-scanning analysis in Hologic Explorer-W densitometers. The user can select RSU or RSUL.

The user can then copy results of the upper, lower or both limbs. In Figure 4.3, Subject H was analyzed without the upper limb only (RSU protocol). Scans A–D did not require reflexion of the left limb nor going for half-scanning. We observed, however, that the weight of the individuals that challenge the limits of the densitometers is lower for women, owing to lesser muscle mass for the same body volume. consequently, it is possible that women weighting $\approx 110 - 136$ kg may still require reflexion of upper and lower limbs. Subjects F–H felt comfortable when the scans were performed with an offset from midline of the table. The maximum offset for which the left arm could still be safe and comfortably accommodated over the side of the table was 10 cm. Subject D was near the weight limit of the densitometer and was the only subject for which beam hardening artifacts compromised the validity of the scan.

TEMs and CVs for reflexion-scanning In order to assist in the interpretation of the precision of DXA results reported for reflexion-scanning techniques, its corresponding TEMs and CVs were investigated in thirty subjects with repositioning in between scans. Accordingly, as previously presented for whole-body and regional measurements, we determined these parameters for Hologic Explorer-W densitometers and for the conditions of this study. Table 4.4 lists these parameters for whole-body versus reflexion-scanning techniques for BMC, LST, FM and %FM.

Table 4.4: TEMs and CVs for whole-body and reflexion-scanning techniques

	TEM (kg)				CV(%)			
PS	**BMC**	**LST**	**FM**	**%FM**	**BMC**	**LST**	**FM**	**%FM**
WB	0.023	0.362	0.370	0.063%	0.99	0.74	2.78	0.57
RSU	0.022	0.387	0.388	0.074%	0.95	0.78	2.93	0.60
RSUL	0.027	0.458	0.401	0.092%	1.15	0.92	3.02	0.61

Acronyms: TEM: technical error of the measurement; CV, coefficient of variation; PS, Partial scanning Techniques; ROI, Region of Interest; BMC, bone mineral content; LST, lean soft tissue; FM, fat mass.

It can be observed from Table 4.4 that the CVs for the studied reflexion-scanning techniques are higher that the CVs for standard whole-body composition assessment. The penalties are about 10% for RSU, but higher for RSUL (range 14-48%). The overall CVs remain in the order or 1% (or less) for BMC and LST, 3% for FM and 0.6% for %FM.

4.3 Validation results

According to the validation approach described in Section 3.3, Equations 3.2 were implemented in SPSS to investigate whether possible confounding factors (gender, athletic status, LDSP) *interacted* with the performance of partial scanning techniques. ANOVA two-way and multiple regressions tests were performed to compute β_3 for each technique and compartment. For HS and RSUL, no significant interactions were found. For RSU, there were significant interaction terms for BMC (*BMC . Sports_Practice*, p=0.003, and *BMC . LDSP*, p=0.019) and %FM (*%FM . Sports_Practice*, p=0.01). The relative contribution of these terms to explain the overall BMC_{WB} and $\%FM_{WB}$ was, nonetheless, negligible in comparison with BMC_{RSU} and $\%FM_{RSU}$. Indeed, $\beta_1 \approx 1$ whereas $|\beta_3| \approx 0.01$ (BMC_{RSU} . *Sports_Practice* : $\beta_3 = 0.009 \pm 0.003$, $(F(194,1) = 9.278, p = 0.003)$; BMC_{RSU} . *LDSP*: $\beta_3 = 0.007 \pm 0.003$, $(F(194,1) = 5.571, p = 0.019)$; $\%FM_{RSU}$. *Sports_Practice*: $\beta_3 = -0.014 \pm 0.005$, $(F(194,1) = 6.716, p = 0.01)$). As such, interaction terms between each main independent predictor (BMC, LST, FM and %FM from the alternative procedures) with gender, LDSP and athletic status were not included in models for any partial scanning technique. Table 4.5 lists results from multiple regressions tests that were conducted to implement and test the models of Equation 3.3, for BMC, LST, FM and %FM from the reference scan. The contribution of fixed factors was either statistically not significant or small; for HS, the contribution of the athletic status accounts for 15 g and 214 goffset in BMC and LST, respectively. In RSU, the influence of factors on the outcome variables was only $\approx 5g$, 42g and $0.3 - 0.4\%$ for BMC, FM and %FM, respectively. Finally, in RSUL, athletic status resulted in a 10g offset for BMC and gender 0.3% for %FM.

Table 4.5: Stepwise multiple regression models for alternative procedures.

PANEL A: MULTIPLE REGRESSION ANALYSIS FOR HS

Model	Summary	Variables	β_i	$S(\beta_i)$	t	*p-value*
BMC (g)	F(2, 195) = 17628.2,	(Constant)	23.289	13.01	1.764	0.079
	p < 0.001,	BMC_HS	0.982	0.006	177.266	<0.001
	R² = 0.994,	Ath. Status	15.194	5.698	2.667	0.008
		Excluded	Gender (*p*=0.57), LDSP (*p*=0.52)			
LST (g)	F(2, 195) = 30846.1,	(Constant)	-256.343	215.591	-1.189	0.236
	p < 0.001,	LST_HS	1.002	0.004	230.484	<0.001

Model	Summary	Variables	β_i	$S(\beta_i)$	t	p-value
	$R^2 = 0.998$,	Ath. Status	214.133	93.165	2.298	0.023
		Excluded	Gender (p=0.36), LDSP (p=0.58)			
FM (g)	$F(1, 196) =$ 109926.3,	(Constant)	459.331	55.685	8.25	<0.001
	$p < 0.001$,	FM_HS	0.999	0.003	331.55	<0.001
	$R^2 = 0.999$	Excluded	Gender (p=0.69), Ath. Status (p=0.77), LDSP (p=0.97)			
%FM	$F(1, 196) =$ 84246.5,	(Constant)	0.773	0.089	8.699	<0.001
	$p < 0.001$,	%FM_HS	1.014	0.003	290.252	<0.001
	$R^2 = 0.998$	Excluded	Gender (p=0.44), Ath. Status (p=0.70), LDSP (p=0.94)			

PANEL B: MULTIPLE REGRESSION ANALYSIS FOR RSU

Model	Summary	Variables	β_i	$S(\beta_i)$	t	p-value
BMC (g)	$F(3, 194) =$ 180217.3,	(Constant)	-4.700	3.618	-1.299	0.195
	$p < 0.001$,	BMC_U	0.998	0.002	651.671	<0.001
	$r^2= 1.000$,	LDSP	-9.946	2.189	-4.543	<0.001
		Athletic Status	5.127	1.800	2.848	0.005
		Excluded	Gender (p=0.51)			
LST (g)	$F(2, 195)$ =1130189.1,	(Constant)	-261.514	49.490	-5.28	<0.001
	$p < 0.001$,	LST_U	1.002	0.001	1063.10	<0.001
	r^2=1.000	Excluded	Gender (p=0.36), Ath. Status (p=0.86), LDSP (p=0.89)			
FM (g)	$F(2, 195) =$ 734757.4,	(Constant)	40.653	15.58	2.609	0.010
	$p < 0.001$,	FM_U	0.998	0.001	1008.92	<0.001
	$r^2 = 1.000$,	Gender	42.263	16.97	2.491	0.014
		Excluded	Ath. Status (p=0.77), LDSP (p=0.96)			
%FM	$F(4, 193) =$ 72370.9,	(Constant)	0.283	0.075	3.789	<0.001
	$p < 0.001$,	%FM_U	1.009	0.003	303.348	<0.001
	$r^2= 0.999$,	Gender	0.270	0.060	4.504	<0.001
		LDSP	0.230	0.058	3.985	<0.001
		Ath. Status	-0.151	0.059	-2.538	0.012
		Excluded	—			

PANEL C: MULTIPLE REGRESSION ANALYSIS FOR RSUL

Model	Summary	Variables	β_i	$S(\beta_i)$	t	p-value
BMC (g)	$F(2, 195) =$ 43100.5,	(Constant)	-2.883	8.536	-.338	0.74
	$p < 0.001$,	BMC_UL	0.993	0.004	277.236	<0.001

	$r^2 = 0.998$,	Athletic Status	10.450	3.655	2.859	0.005
		Excluded	Gender (p=0.57), LDSP (p=0.52)			
LST (g)	$F(1, 196) =$ 212040.6,	(Constant)	-543.524	114.855	-4.732	<0.001
	$p < 0.001$,	LST_UL	1.002	0.002	460.479	<0.001
	$r^2 = 1.000$	Excluded	Gender (p=0.36), Ath. Status(p=0.86), LDSP (p=0.90)			
FM (g)	$F(1, 196) =$ 249760.6,	(Constant)	88.518	37.609	2.354	0.02
	$p < 0.001$,	FM_UL	0.993	0.002	499.761	<0.001
	$r^2 = 0.999$	Excluded	Gender (p=0.69), Ath. Status (p=0.78), LDSP (p=0.97)			
%FM	$F(2, 195) =$ 69124.8,	(Constant)	0.279	0.074	3.786	<0.001
	$p < 0.001$,	FM_UL	1.011	0.004	245.611	<0.001
	$r^2 = 1.000$	Gender	0.265	0.085	3.117	0.002
		Excluded	Ath. Status (p=0.69), LDSP (p=0.94)			

Abbreviations and Acronyms: HS, Half Scan; RSU/RSUL, Partial Scans with Upper/Upper and Lower Left Limbs removed; Ath. Status, Athletic Status; LDSP, Lateral Dominant Sports Practice; BMC, bone mineral content; FM, Fat Mass; LST, Lean Soft Tissue; β_i, Unstandardized correlation coefficients for dependent variables i in main predictors (BMC, LST, FM, %FM_alternative technique) or covariates (Gender, Athletic Status, LDSP); $S(\beta_i)$, Standard Error of β_i; r^2, squared correlation coefficient; F (df1, df2), F value and the degrees of freedom for both the regression (df1) and the residual error (df2). **Significance level** for inclusion of dependent variables in models: $p < 0.05$.

Subjects were grouped together and analyzed collectively. The complete set of validation results is presented in Table 4.6, including mean group comparison of whole-body vs. partial scanning estimates, least-squares linear regression (see Equation 3.4 for details), Bland-Altman and concordance analysis.

Table 4.6: Validation of alternatives to assess wide individuals in DXA.

PANEL A: VALIDATION OF HS

	BMC (kg)	LST (kg)	FM (kg)	%FM
WB, mean ± s	2.43±0.51	51.2±10.8	16.9±8.4	23.9±10.1
HS, mean ± s	2.44±0.51	51.3±10.7	16.5±8.4	23.3±10.1
r^2	0.9943	0.9968	0.9982	0.9981
SEE	0.039	0.616	0.354	0.44
Slope	0.986[*]	1.006[*]	0.9994[*]	0.9966[*]
Intercept	0.019	-0.345	0.459[*]	0.618[*]
CCC	0.99666	0.99834	0.99767	0.9976
r	0.99715	0.99838	0.99911	0.9991
Cb	0.99951	0.99996	0.99856	0.9986

Bias (kg)	0.015 (0.6%)*	0.062 (0.1%)	-0.45 (2.7%)*	-0.54 (2.2%)*
95% LoA	-0.061, 0.091	-1.15, 1.27	-1.14, 0.24	-1.40, 0.32
Trend	0.1450**	-0.1241	-0.0063	0.0565

PANEL B: VALIDATION OF RSU

	BMC (kg)	LST (kg)	FM (kg)	%FM
WB, mean ± s	2.43±0.51	51.2±10.8	16.9±8.4	23.9±10.1
RSU, mean ± s	2.44±0.51	51.4±10.8	16.9±8.4	23.8±10.1
r^2	0.9996	0.9998	0.9999	0.9999
SEE	0.010	0.142	0.098	0.12
Slope	0.995*	1.002*	0.999*	1.003*
Intercept	0.001	-0.262*	0.043*	0.02*
CCC	0.99958	0.99978	0.99993	0.99987
r	0.99980	0.99991	0.99993	0.99993
Cb	0.99978	0.99987	0.99999	0.99995
Bias	0.010 (0.4%)*	0.172 (0.3%)*	-0.026 (-0.2%)*	-0.10 (-0.4%)*
95% LoA	-0.01, 0.03	-0.11, 0.45	-0.22, 0.17	-0.35, 0.15
Trend	0.2149**	-0.1374	0.0822	-0.2667*

PANEL C: VALIDATION OF RSUL

	BMC (kg)	LST (kg)	FM (kg)	%FM
WB, mean ± s	2.43±0.51	51.2±10.8	16.9±8.4	23.9±10.1
RSUL, mean ± s	2.44±0.51	51.7±10.8	17.0±8.4	23.7±10.1
r^2	0.9977	0.9991	0.9992	0.9991
SEE (kg)	0.025	0.328	0.242	0.31
Slope	0.9961*	1.002*	0.993*	1.003*
Intercept	-0.006	-0.544*	0.089*	0.067*
CCC	0.99835	0.99863	0.99958	0.99942
r	0.99882	0.99954	0.99961	0.99953
Cb	0.99953	0.99909	0.99997	0.99989
Bias (kg)	0.016 (0.7%)*	0.459 (0.9%)*	0.034 (0.2%)	-0.143 (-0.6%)*
95% LoA	-0.033, 0.064	-0.18, 1.10	-0.44, 0.51	-0.76, 0.47
Trend	0.0559	-0.0688	0.2375**	-0.1182**

Abbreviations: BMC, bone mineral content; FM, fat mass; LST, lean soft tissue; r^2, coefficient of determination; SEE, standard error of estimation; CCC, concordance correlation coefficient; Bias, mean difference between methods; LoA, limits of agreement; r, Pearson correlation coefficient; Cb, bias correction factor. **Significance levels:** * $p < 0.001$; ** $p < 0.05$.

Figures 4.4, 4.5 and 4.6 depict results for the validation of HS, RSU and RSUL. Subfigures (a), (c) and (d) are the linear regression plots for BMC, LST and FM, and (b), (d) and (f) are the corresponding Bland-Altman plots for each compartment. Figures 4.7 (a)–(f) refer to the validation of HS, RSU and RSUL for %FM.
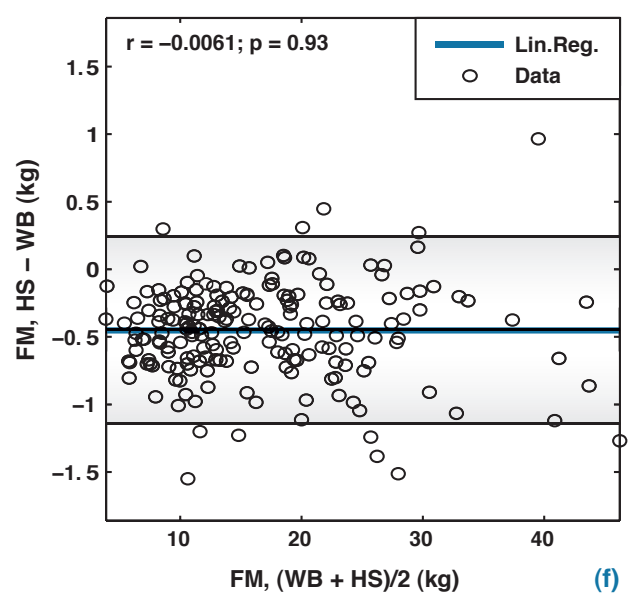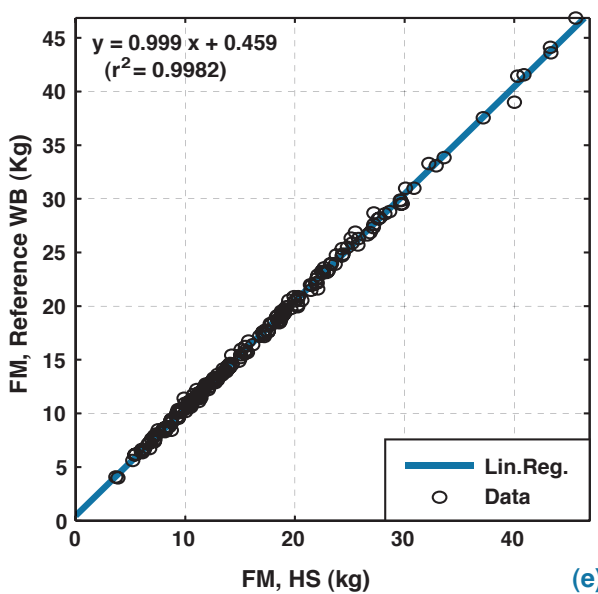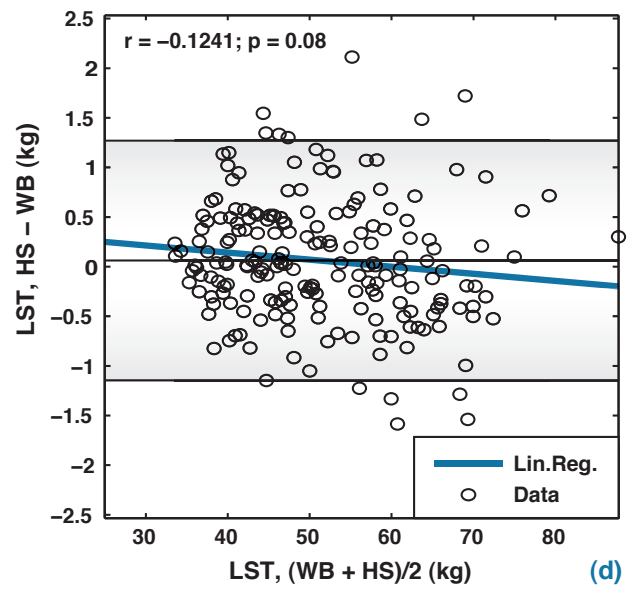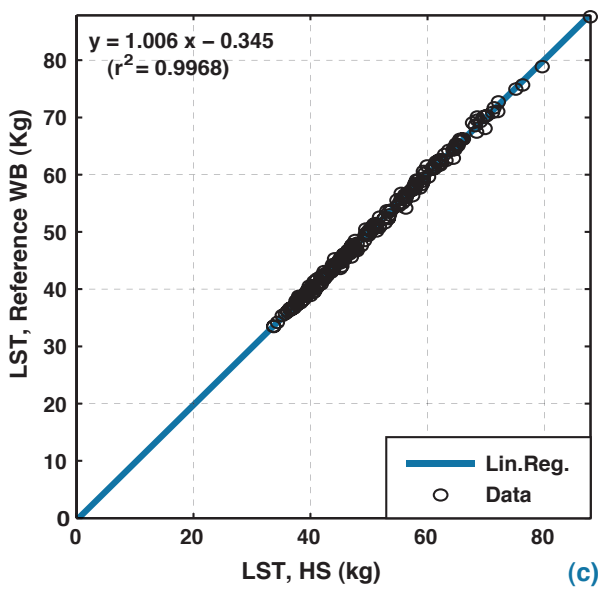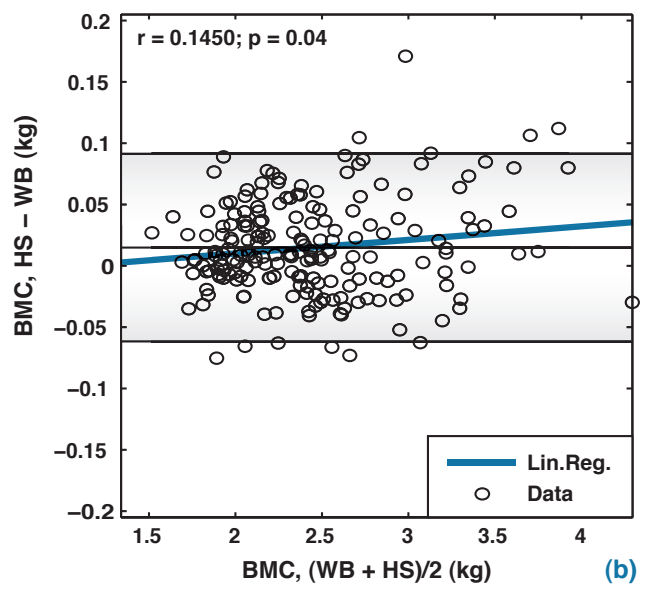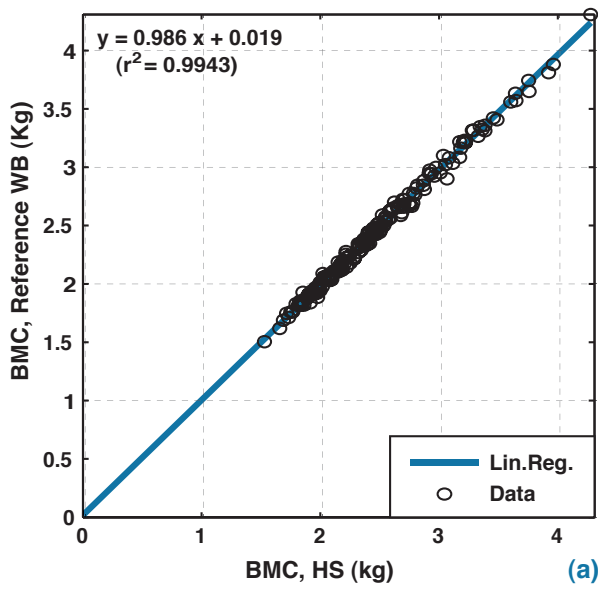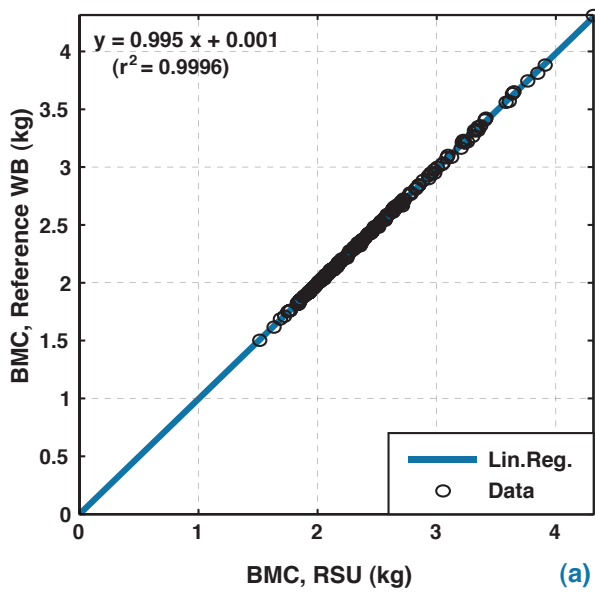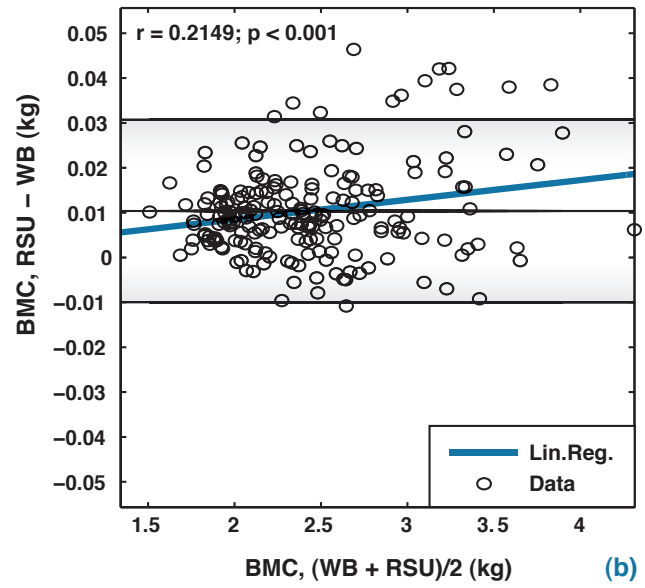
Figure 4.4: Regression and Bland-Altman plots for HS.
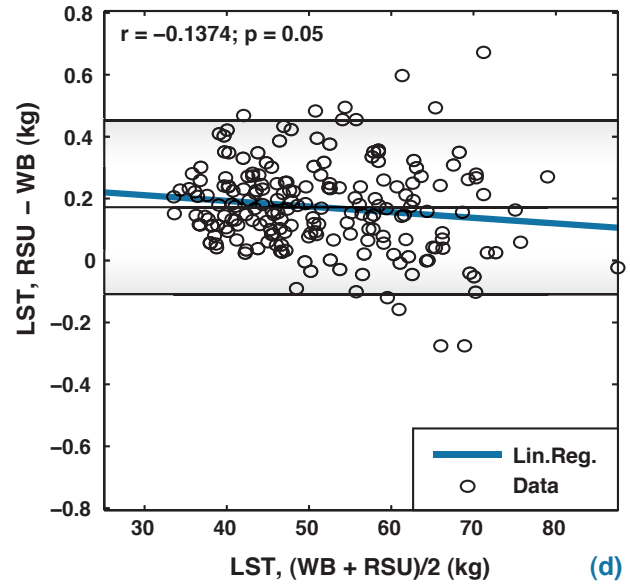
Figure 4.5: Regression and Bland-Altman plots for RSU.
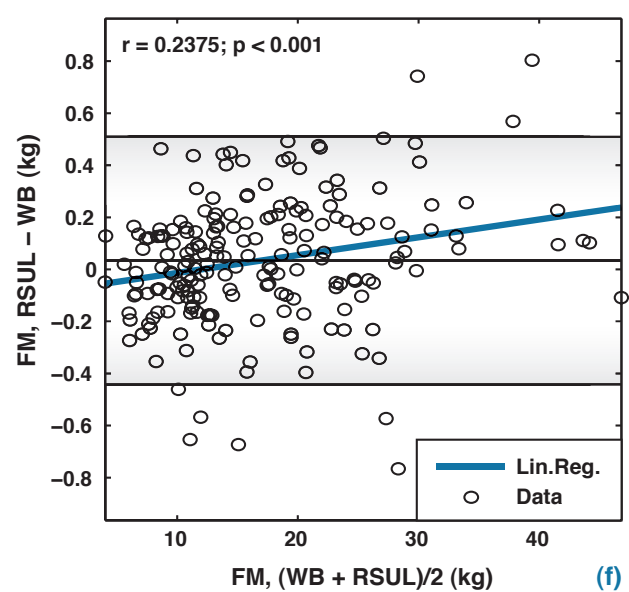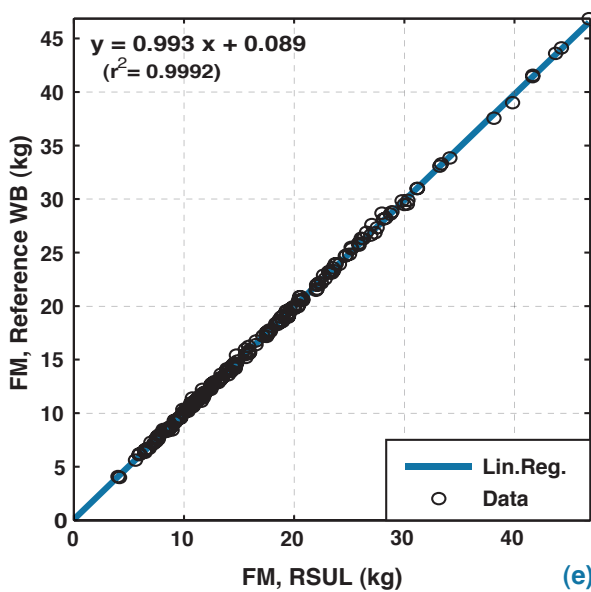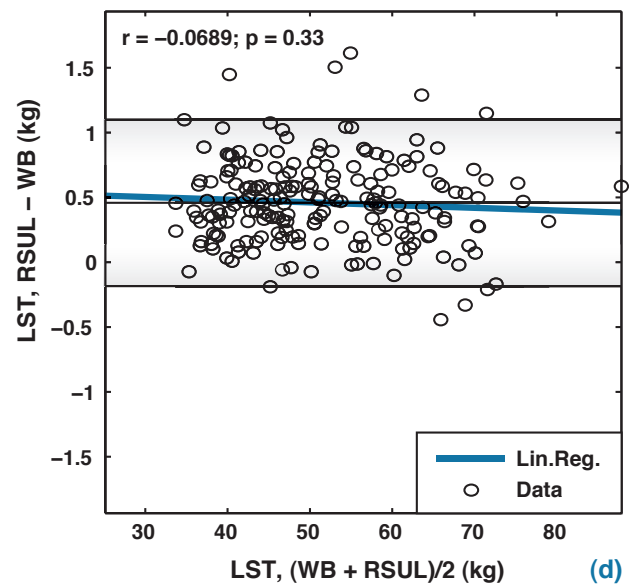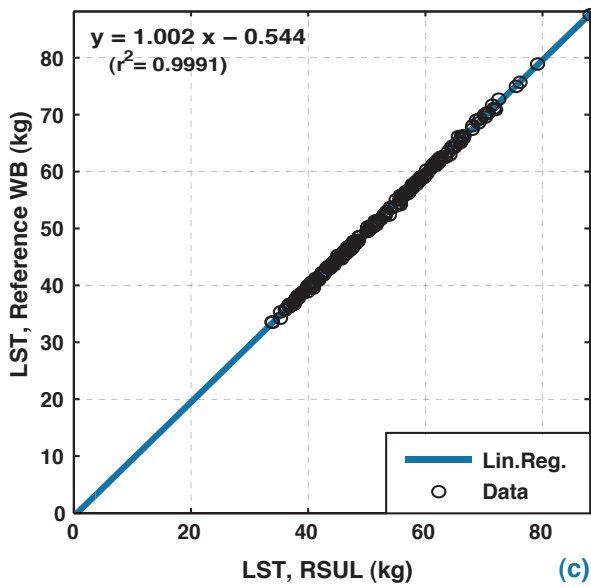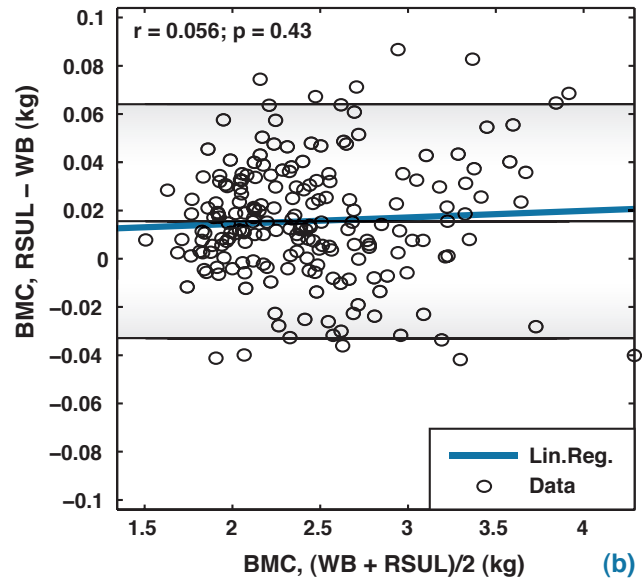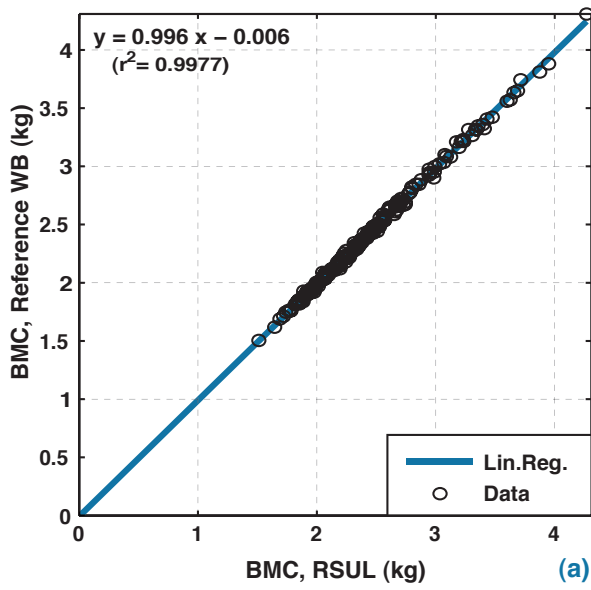
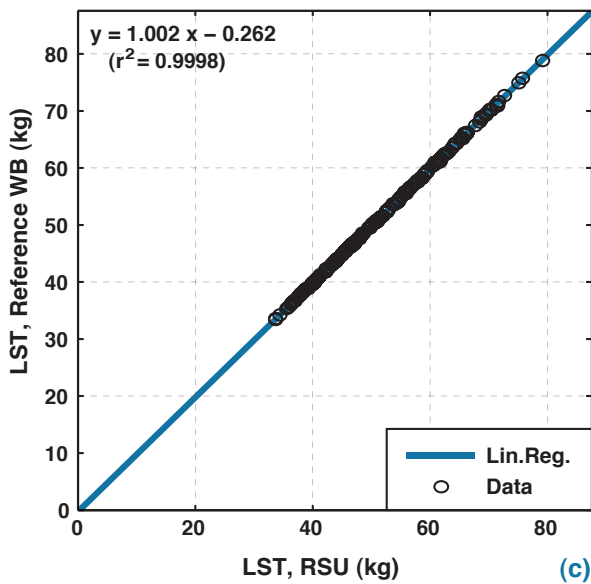**Figure 4.6:** Regression and Bland-Altman plots for RSUL.
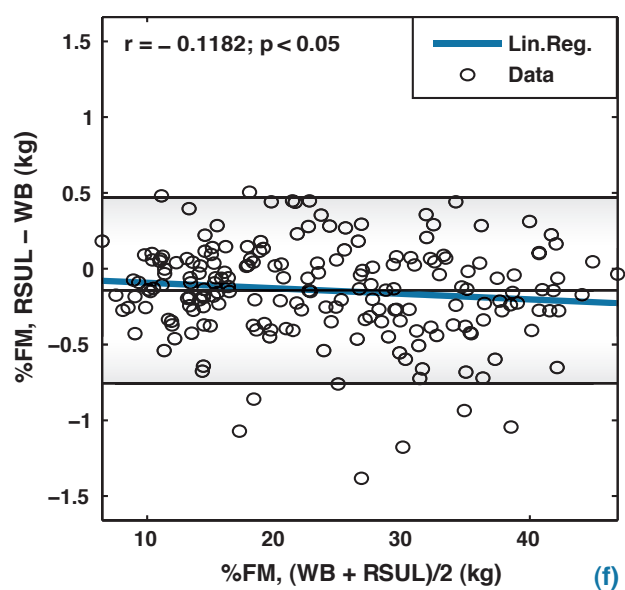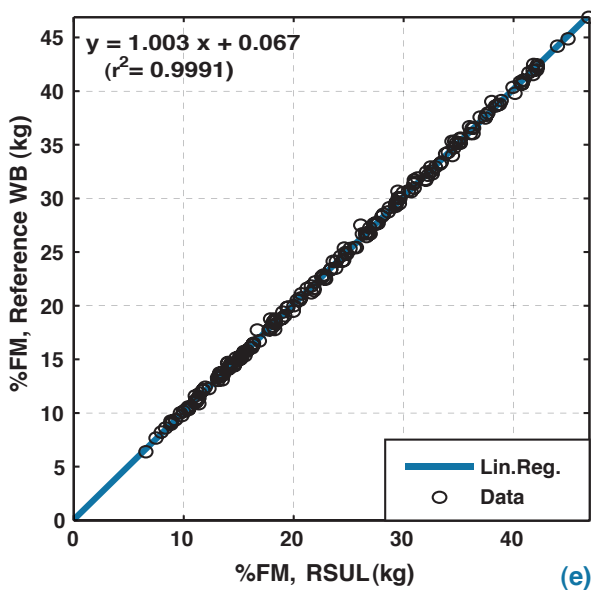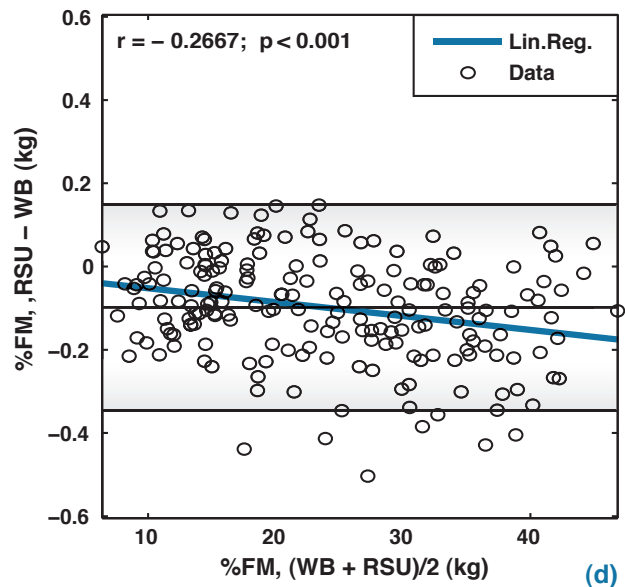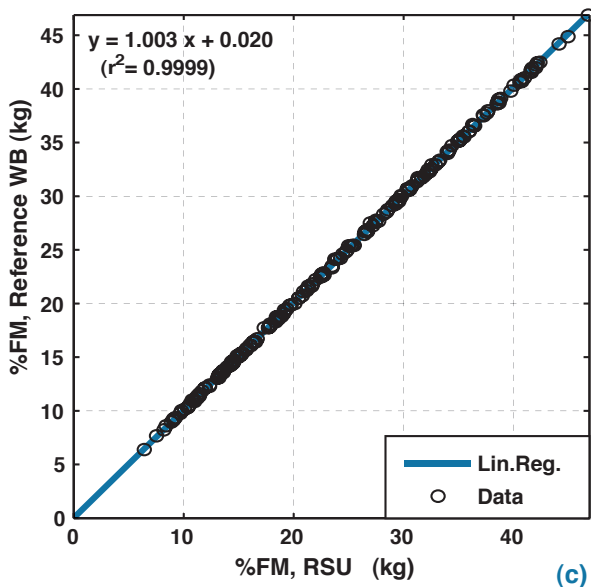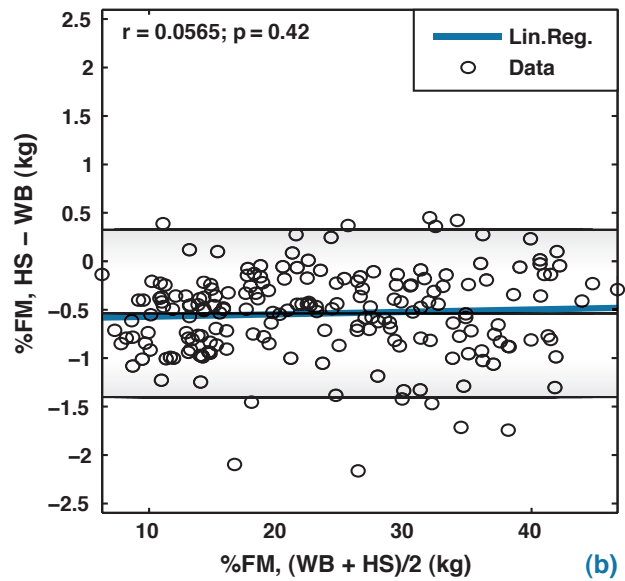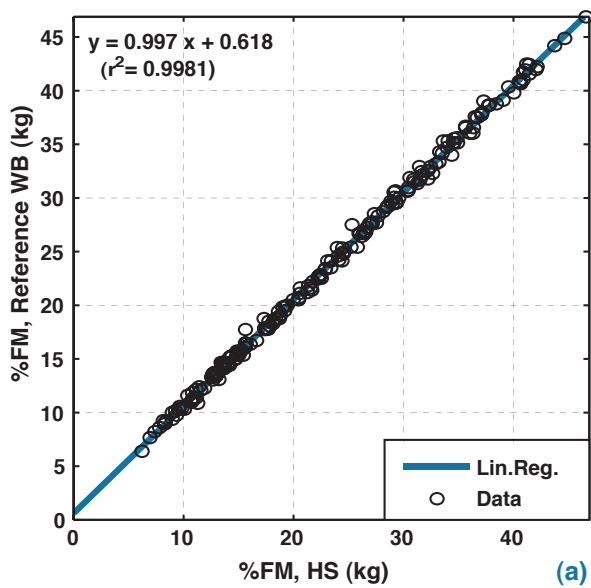
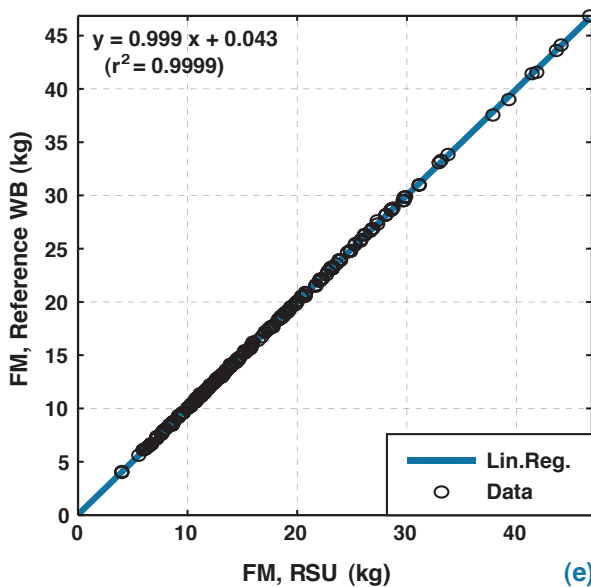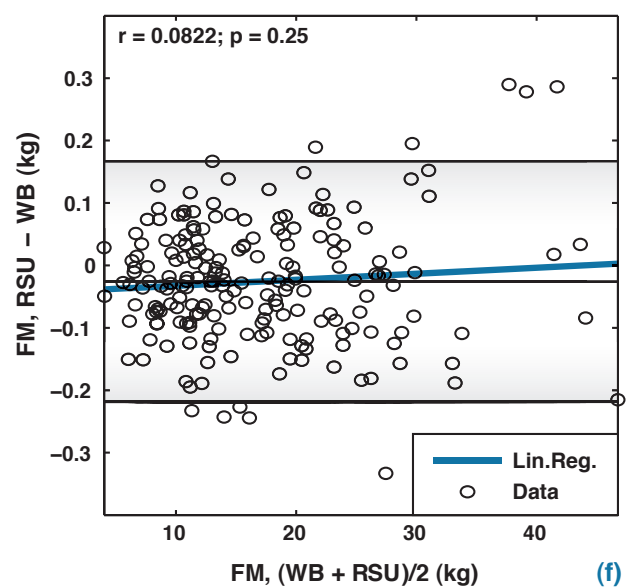**Figure 4.7:** Regression and Bland-Altman plots for HS, RSU and RSUL– % Fat Mass.

The remaining paragraphs of this chapter highlight the information available for linear regression, Bland-Altman and concordance analysis.

Linear regression analysis Subfigures 4.4, 4.5, 4.6 and 4.7 (b, d, f) represent the associations between the reference (dependent variable) and the alternative (independent variable) scans for BMC, LST, FM and %FM. Linear regression analysis showed that the alternative procedures explained more than 99% of the variance of BMC, LST, FM and %FM (Table 4.6). However, RSU was the best performing technique ($r^2 = 0.9996 - 0.9999$) followed by RSU and, lastly, HS ($r^2 = 0.9943 - 0.9982$). The slopes of the regressions for all alternatives were all close to unit (> 0.99), and t-tests confirmed that they were indeed not different to unity. Additional t-tests indicated that intercepts for BMC ($\approx 10 - 15$g) were not significantly different from zero for all techniques. However, the same was not the case for soft tissue variables. In fact, for FM and %FM, the intercepts were significant for all techniques, but whereas these values were below the corresponding TEMs for the reflexion-scanning techniques, for HS the intercepts were still below 0.5 kg / 1%FM. For LST, the intercept values were not significant (for HS), and significant but still less than the TEMs for LST (RSU: -0.263 ± 0.049 kg $<$ TEM$_{\text{LST, RSU}}$=0.387 kg; RSUL: -0.544 ± 0.115 kg $<$ TEM$_{\text{LST, RSUL}}$=0.458 kg). SEEs were lower than 2.1% of the mean measurements for all compartments under HS, RSU, RSUL, and also for %FM (range of absolute values for SEE: BMC $\approx$ 10–40 g, LST $\approx$ 0.1–0.6 kg, FM $\approx$ 0.1–0.5 kg and %FM $\approx$ 0.1 – 0.4%). Specifically, SSEs were about 1.6–1.8% for HS, 0.6–1% for RSU and 0.3–0.5% for RSU, which stood out as the best performing technique.

Bland-Altman analysis As evidenced in Table 4.6, the bias between the alternative techniques and the reference whole-body estimates is either not significant or small in most cases. For BMC, partial scanning techniques significantly overestimated BMC by $\approx$ 10 – 16g, which corresponded to less than 1% of the mean BMC and is less than the TEMs for BMC (TEM$_{\text{RSU, BMC}}$ = 23 g; TEM$_{\text{RSUL, BMC}}$ = 27 g). For LST, HS did not overestimate this component, but RSU and RSUL significantly overestimated whole-body composition measurements by 0.3% (absolute bias: 0.17 kg $<$ TEM$_{\text{LST, RSU}}$) and 0.9% (absolute bias: 0.46 kg = TEM$_{\text{LST, RSUL}}$), respectively. The bias for fat measurements was significantly underestimated by HS by 2–3% (absolute bias: -0.45 ± 0.35 kg, -0.5%). %FM was also significantly underestimated by reflexion-scanning techniques by $\approx$ 0.5%, but these were insignificant or close to its corresponding TEMs for %FM ($\approx$ 1%).

Bland-Altman plots (Subfigures 4.6 and 4.7 (b, d, f) showed that the reflexion-scanning techniques had narrower limits of agreement than half-scanning (HS vs. RS: 0.15kg / 2.5kg / 1.4kg / 1.7% vs. 0.04–0.1kg / 0.6–1.3kg / 0.4–1.0kg / 0.5–1.2% for BMC / LST / FM / %FM). Also, absolute ranges of the 95% limits of agreement were generally higher, but still within the order of magnitude as the corresponding TEMs (TEMs RSU–RSUL: BMC, $\approx$ 0.02–0.03kg; LST, 0.4–0.5kg; FM, 0.40kg; %FM, 0.1%).

There were outliers for all techniques and compartments, and these were not excluded from the data sample. The absolute individual errors are higher among athletes engaged in lateral sports practice, particularly handball. The maximum individual errors registered for handball players were as follows:

- HS:
    - BMC, 0.09 kg;
    - LST, 1.72 kg ;
    - FM, -0.7 kg (%FM, -1.0%);
- RSU:
    - BMC, 0.04 kg;
    - LST, 0.60 kg;
    - FM, 0.15 kg (%FM, -0.3%);
- RSUL:
    - BMC, 0.07 kg;
    - LST, 1.02 kg;
    - FM, 0.45 kg (%FM, +/-0.4%).

As for trends of the differences between the mean of both methods, Bland-Altman analysis reveals significant and positive trends for BMC in HS and RSU, as well as a positive tendency in RSUL estimates. For soft tissue measurements, the trend is not significant for LST, whereas, for fat measurements, the trend is only significant for FM in RSUL (positive trend) and %FM in both reflexion-scanning techniques (negative trend).

Concordance Analysis Panel A of Table 4.6 evidences that the concordance correlation coefficient (CCC) values were higher than 0.99 for BMC, LST, FM and %FM for all techniques, and the same holds for the bias term Cb and Pearson's r, which inform (favorably) on accuracy and precision, respectively.

Discussion

<div style="text-align: right">5</div>

> *Only professional mathematicians learn anything from proofs. Other people learn from explanations. (. . .) Experienced parents realize that when a child says "Why?" it just wants more conversation.*

— **Ralph P. Boas**

1912–1992

Due to the need of evaluating athletes and obese patients whose dimensions exceed the width of the DXA scan area, the ability to circumvent this limitation has become a very useful capability for DXA systems (Sherman, 2011). Partial scanning techniques like "Half" and "Reflection" scanning are being used by manufacturers to estimate whole-body values even when body region(s) fall outside the scan window. Morphological symmetry is an implicit assumption to this family of solutions, as missing body regions must be estimated from those that were actually scanned. In this study, we sought to compare the aforementioned techniques to one another in terms of accuracy of estimates for BMC, LST, FM and %FM. We also asked what effect gender and sports practice, particularly lateral dominant sports, had on validity. Since the quality of partial scanning inferences seems to be scanner dependent (Hangartner et al., 2013), we narrowed the scope of this project to Hologic QDR Explorer-W densitometers (software QDR APEX vs. 13, Hologic Inc).

For validation purposes, an whole-body scan was the reference criteria and three partial scanning options were tested: the half-scan, which measures half of the body and assumes the opposite side is equal, and two more conservative alternatives at which only left limb(s) are left out of the scan (RSU, reflection scan – upper left limb removed and RSUL, reflection scan – upper and lower limbs). In the RSU technique, only the left upper limb is excluded from the scan, whereas in RSUL both the left upper and lower limbs are excluded and assumed to be equal to the contralateral ones.

A partial scan can be performed in less than 10 minutes and the Hologic and Lunar software versions already include an automatic mode for reflexion or half-scans, respectively. As such, the time required to evaluate a subject and to analyze its scan by either RSU, RSUL or HS is similar to the standard procedures. Using whole-body scans as the reference criteria, results demonstrated that all partial scanning techniques are valid and accurate options to evaluate wide individuals.

Using a diverse sample of athletes and non-athletes of both genders, and including a broad range of phenotypes, results indicated that RSU was the best performing technique in terms of overall accuracy of estimates for whole-body DXA compartments and %FM. Specifically:

a) The RSU estimates differed slightly from those of the reference procedure (bias: 0.010± 0.010 for BMC, 0.17 ± 0.14 kg for LST, -0.03 ± 0.10 kg for FM, -0.10 ± 0.12 for %FM). However, the differences were within the technical errors of measurement (TEM) of the equipment for each compartment (0.02 kg, 0.39 kg, 0.39 kg and 0.1% for BMC, LST, FM and %FM, respectively).

b) RSU explained more than 99% of the variability in body composition assessed by the reference scan, thereby suggesting a very strong correlation(Iman, 1994), and the standard errors of estimation were low;

c) The Person's correlation coefficient r, agreement factor Cb and concordance correlation coefficient ($CCC > 0.99$) indicate excellent accuracy, very high correlation and an almost perfect strength of agreement (McBride, 2005);

d) Bland-Altman analysis revealed 95% limits of agreement ranging from -0.01to 0.03 kg for BMC, -0.11 to 0.45 kg for LST, -0.22 to 0.17 kg for FM, -0.35 to 0.15% for %FM. The limits of agreement were close to or within the technical errors of measurement (TEM) for the aforementioned compartments. Trends were significant for BMC (positive trend) and %FM (negative trend), therefore meaning that the more mass individuals have, the greater the overestimation or underestimation errors are expected to be for BMC and %FM, respectively.

f) Individual errors can be significant and largely exceed TEMs for BMC and LST, particularly in lateral dominant sports. Handball players fall in this category.

As for the comparison of techniques, we observed that reflexion-scans are superior to the conventional half-scanning approach in terms of accuracy of estimates for whole-body DXA compartments and %FM. The relative performance of the techniques was, in descending order: RSU, RSUL and HS. This result is not surprising since the more area is used from the DXA scan, the more accurate the estimates for the DXA compartments are likely to be, since less data needs to be extrapolated from contralateral regions. However, techniques that require more scanning information are likely to be applicable to a narrower range of physiques, which is also a critical aspect that deserves consideration. Preliminary tests performed in three individuals with extreme phenotypes (see Figure 4.1 on page 60) shown that those who are near the weight limit of the DXA table ($\approx$ 140kg) can be properly assessed by RSU and that RSUL might be used less frequently than RSU.

There are two last points that favor HS in comparison to either RSU or RSUL, which are its inferior radiation and time requirements. These aspects are relevant to the assessment of the obese patients as, for these, the radiation required to prevent beam hardening might be threefold the standard dose for an whole-body composition asessment (Shepherd, 2014). In this regard, if DXA scans are only performed occasionally throughout the year, the radiation levels are still safe for the patient, but there will be less evaluations that can performed in an annual basis for the same accumulated dose. Also, the diminished evaluation time required by HS reduces in one half the penalty time required to overcome beam hardening in obese/thick patients. Time efficiency is pivotal in settings where multiple evaluations need to be scheduled for one morning and where delays compromise the number of subjects being scanned. Nevertheless, since HS is not implemented in the existing Hologic's Explorer-W software, it is not possible to reap the time and radiation benefits of scanning just one side of the body, and the evaluation time is actually larger since there is an additional half-scanning region of interest that needs to be created by the technologist during scan analysis. Thus, reflexion-scanning techniques are definitely preferable to half-scanning.

Insights from the literature   To our knowledge, eight previous studies have discussed, validated and/or reviewed procedures to assess whole-body BMC, LST and FM or BMC in individuals broader than the DXA scan area (Breithaupt et al., 2011; Brownbill & Ilich, 2005; Misic & Evans, 2006; Nana et al., 2012; Rothney et al., 2009; Sherman, 2011; Silva et al., 2013; Tataranni & Ravussin, 1995). However, among these, only four actually attempted to

validate HS or RSU with specific devices and populations (Breithaupt et al., 2011; Rothney et al., 2009; Sherman, 2011; Tataranni & Ravussin, 1995).

For RSU, Table 2.7 informs that Sherman (2011) reported small significant differences for whole-body bone but no differences on soft tissue measures. In contrast, our study indicated that reflexion-scanning protocols resulted in significant differences for bone, lean and fat measurements. For HS, Table 2.7 summarizes the results of such validation studies in [right-side] HS. A general appreciation of the results indicates that the coefficients of determination are all > 0.99, which is in agreement with our results. The reported magnitudes for side-to-side differences in HS are in the order of $\approx 10 - 30$ g for BMC and $\approx 1 kg$ for LST and FM. These results are higher than the ones obtained in this study, which is not surprising if we take into account the higher levels of adiposity of the subjects reported by Breithaupt et al. (2011); Rothney et al. (2009); Tataranni & Ravussin (1995). However, if we attend to percent bias, results are similar. Even still, HS in our sample performed relatively poorly in terms of bias for fat measurements. This shortcoming did not compromise the overall validity of the proposed half-scanning based solutions and the same holds for the current investigation, although individual differences were exacerbated in lateral dominant sports. It should also be recalled that the participants recruited for the present study were mostly sports science students and physically active obese women. Hence it seems reasonable to assume that the sample we considered is more active than sedentary obese adults and children, and this aspect may impact on side-to-side differences induced by lifestyle.

Considerations on side-to-side differences   Across all partial scanning results there was a systematic bias between dominant and non-dominant upper limbs, as indicated by the positive bias and negative intercept values for LST and BMC. This result in not surprising given the information presented on Section 2.6.1 about studies documenting and providing rationale for side-to-side differences in limbs. In short, it is known that humans display a species wide lateralized hand preference with 90% of individuals in all populations being right-handed for most manual actions (Auerbach & Ruff, 2006). Thus, the increased muscular development and bone content of right upper limbs that characterize the crossed symmetry pattern is a plausible consequence of the mechanical loading placed by throwing and grabbing actions as those involved in lateral dominant sports. Likewise, it is known that cyclic activities are associated with higher power

production by dominant limbs; thus, in endurance athletes and also in non-athletes, side differences stem from the duality function-shape adaptations.

The extent to which side-to-side differences are explained by daily living tasks or sports practice is not possible to ascertain based on the current literature (Auerbach & Ruff, 2006; McClanahan et al., 2002; Steele & Mays, 1995). Anthropological studies on fossil bones can only provide qualitative evidence for side-to-side adaptations (identifiable as laterality differences in the BMC and LST compartments) as a consequence of handedness (Uomini, 2009). Likewise, to the best of my knowledge, the influence of sports or lifestyle activities on side-to-side differences in body composition has not been thoroughly investigated and reported in the literature using DXA, although some descriptive studies exist for specific sports (Ireland et al., 2013; McClanahan et al., 2002).

The data collected in this study shows that athletes have higher directional bias (DA) towards the right-hand side. Nor surprisingly, these are the same for which bias between partial scanning and whole-body scanning techniques indicated superior errors in either mean group comparison or Bland-Altman analysis. This observation is clear among athletes engaged in lateral dominant sports, specially handball players. For these, the crossed symmetry pattern dominates. Handball involves jumping with the non-dominant leg to perform throwing actions with the dominant hand. The repetition of this movement might explain the outliers found for BMC and LST—favoring the right-hand side upper limb—, as well as superior BMD for the non-dominant leg. As a consequence, the right upper limb is stronger (in terms of lean tissue and bone measures) than the contralateral side, and the bias for BMC and LST is significant and positive for RSU. In this study, the DAs for BMD were 3.4 ± 4.5% in LDSP and just 0.50 ± 3.7% (non-significant) in non-LDSP. Sherman (2011) reported results for RSU using scans representative of the general american population (NHANES study). Since these are non-athletes, it was expected that DAs would also be non-significant. However, the authors observed that the DA for BMD of upper limbs was 3.4% (significant), a result that is similar to the LDSP group from our study; this finding raises suspicion on the accuracy of bone measures under reflexion-scanning protocols and also population variability. For non-athletes and endurance sports athletes, partial scanning estimates also overestimated BMC and LST. Rothney et al. (2012), in the obese, observed absolute side-to-side differences for BMC at the upper and lower limbs that favored the right-hand side, and these differences where higher for upper limbs.

Its causes was attributed to handedness and walking. The findings of Rothney et al. (2012) seem to be in agreement with ours for non-athletes and non-LDSP, where the right limbs were also stronger. An opposite trend was also verified for the fat mass compartment in the non-dominant upper arms for RSU and left lateral side for RSUL. This phenomena remains to be explained.

Through questioning to coaches during the examination sessions or posteriorly, it was known that the evaluated athletes do complementary work at the gym at a weekly basis (2-5 times/week), the exceptions being made to basketball and handball athletes. Also, handball athletes only train 2–3 times/week, which means that the individual errors may be smaller in athletes classified in the LDSP group, but who perform weight training regularly. Conversely, errors may be even higher in high-level and professional athletes who train daily but do not compensate induced side-to-side asymmetries in limbs (sources: Dr. Anna Volossovich, July 2013; Dr. Fernando Gomes, July 2014; Francisco Assis, June 2013, Faculty of Human Kinetics, Portugal).

Should this discussion discourage the application of partial scanning techniques because of its reliance on symmetry assumptions? Probably not for most individuals. For most non-athletes, daily-living activities or occasional sports practice are not expected to compromise the validity of partial scanning, as these are only likely to induce negligible side-to-side differences. The same rationale may not hold for all athletes, particularly those engaged in DLSP. But even in such sports, strength training for injury prevention and/or muscular development is currently among the training paradigms (Bompa & Haff, 2009), which helps the symmetry assumption hold. However, it is recommendable that those suspicious of side-to-side imbalances take summation scans instead. Examples include individuals recovering from an injury requiring immobilization of limbs or athletes engaged in LDSP who do not perform preventive training for sports induced imbalances.

Considerations on reproductibility   Reflexion and half-scanning protocols require partitioning the body according to different regions of interest. As such, in addition to side-to-side differences between limbs, whole-body estimates obtained by partial scanning are also affected by the skill of the technologist in delimiting regions of interest (head, trunk and limbs) during scan analysis. This issue seems more relevant for the reflexion algorithms implemented in Hologic QDR Explorer-W densitometers than by their GE Lunar iDXA counterparts; the later includes a visible midsagittal line printed the DXA table

to allow correct positioning and performs automatic selection of the right half-scan region, thereby reducing intra- and inter-observer error sources (GE Helthcare, Inc., 2012).

Even when systematic errors are minimized by the adoption of standardized procedures, there remains an error margin in reflexion techniques due to need to separate pelvis from hips and trunk from arms. Here a question arises as to whether the overall precision of the reflexion techniques *reported in this research* was significantly affected by the intervention of the technologist that analyzed scans. The investigation of the susceptibility of RSU and RSUL to random mismatches in scan analysis recommends inspecting the corresponding TEMs and CVs (%).

Recall from Section 2.5 that TEMs or CVs are precision metrics that take into account equipment, protocol, characteristics of the measured populations and expertise level of the technologist(s) that perform scan acquisition and analysis. Table 4.4 lists results for the TEMs for reflexion techniques[1]. In order to compute them, 30 subjects were evaluated twice with repositioning in between. The technologist was the same for all analysis procedures required for this investigation. For each technique and DXA compartment, the TEMs and CVs for standard whole-body scanning, RSU and RSUL were computed. At the end, it was verified that the error metrics for RSU were only slightly worse than for whole-body scanning. The CVs (%) for BMC / LST / FM / %FM for WB are 0.99 / 0.74 / 2.78 / 0.57, whereas for RSU these are up to $\approx$ 7% higher. Similarly, for RSUL, the penalty is $\approx$ 8 − 25%. As such, it is reasonable to assume that, for all compartments, $CV_{WB} \approx CV_{RSU}$. The feasibility of this equivalence adds simplicity to RSU, since, for experienced technologists, precision studies are not required. The adoption of standardized procedures for placement of markers and prior training are recommended practices to reduce intra and inter-observer errors due to mismatches (Hangartner et al., 2013) and might explain this good result. Indeed, for this study, the same technologist analyzed almost 50 scans for training prior to analyzing the 198 scans reported in this investigation. All scans were obtained and analyzed according to the guidelines of the manufacturer (Hologic, Inc.), which are in agreement with those of the Exercise and Health Laboratory / Faculty of Human Kinetics, Lisbon. It is therefore recommended that similar practices are undertaken by those wishing to replicate the reflexion techniques discussed in this thesis.

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[1]Due to time constraints, it was not possible to perform the same study for HS.

Limitations

Despite the encouraging findings reported so far, this study is not without limitations.

Up to 10 cm   Firstly, the proposed partial scanning techniques are only applicable to individuals who exceed the width limit of the DXA table by up to 10 cm (evaluated in the coronal plane). However, tests on subjects that actually do not fit in the DXA table revealed that those in excess of 10 cm of the DXA scanning table are unlikely to be within weight limit of the system (136Kg).

Standartization   Second, the lack of standardization among different manufacturers and models of DXA systems restricts the practical interest of our results to laboratories whose DXA systems are of the same hardware and software as the ones we used in this investigation (Hologic Explorer-W System, fan-beam mode, software APEX for Windows version 13.3; Hologic,Waltham, MA). For the time being, unless conversion equations are available to calibrate variables from one instrument to another (e.g.: Shepherd et al. (2012b)) it cannot be guaranteed that the results here reported hold under different software versions, scan modes or among instruments from different manufacturers.

Generalizability   Third, our sample comprised mostly young healthy adults that were normal or overweight (World Health Organization, 2013). Consequently, the generalizability of the findings is threatened by the fact that the selected sample does not include subjects who actually do not fit within the DXA size limits.That implies imprecision due to the assumption of allometric scaling and beam hardening distortion that affects the accuracy of the DXA system for thick subjects. Another limitation of this study concerns the method used as a reference, i.e., an WB performed in the same equipment as the partial scanning strategies. In order to overcome some of the aforementioned limitations, validation studies at which the reference results are collected in a different DXA system whose width is large enough to accommodate wide individuals to whom the proposed partial techniques are meant are, therefore, worthy of future investigation.

Specific sports   Fourth, although the selected sample size (N=198) is in agreement with the literature, whereby the number of subjects measured in validation studies of partial scans for broad subjects ranged from 30 to 183 (Nana et al., 2012; Santos et al., 2012; Tataranni & Ravussin, 1995), the sample size was not large enough to allow for individual

sports analysis. Instead, we could only investigate the dihcotomic classification according to the statuses of athletic practice and lateral dominant sports practice. The possibility that different sports may induce morphological asymmetries in varying degrees of severity, was, therefore, not accounted for in this study (Bussey, 2010; Ireland et al., 2013; McClanahan et al., 2002). Thus, it may be prudent at this time to exercise caution when using DXA among athletes engaged in a specific lateral dominant sport (e.g.: tennis, handball), as the individuals errors might exceed the accuracy and precision errors reported for partial scanning strategies.

Characterization of the sample   Fifth, it was based on a post-hoc analysis of an RCT and a technical study not specifically designed to study the validity of half- and reflexion-scans. Although the evaluation procedures and equipment are the same as in this study, handedness information, record of previous injuries and detailed sports practice, were not collected, preventing us from drawing firm conclusions about the extent of side-to-side bias in specific athletic populations or individuals. 85% of individuals in all populations are right-handed for most manual actions, whereas only 5% and left-handed Uomini (2009). As such, although it was advantageous to have handedness available, not controlling for this variable might have a minor impact on the overall results.

Body dissatisfaction   Furthermore, partial scanning images might not be well perceived by patients. Many women (and arguably many men as well) feel dissatisfied with their *body image*. According to Carraça (2012), this concept refers to "how someone personally experiences his or her own embodiment. More than a mental representation of the body, it reflects one's personal relationship with its encompassing perceptions, beliefs, thoughts, feelings, and behaviors" (pp. 17). Body dissatisfaction results in pressure to achieve thin ideals and might be more pronounced for overweight and obese women. Predictably, poor body image is often associated with diminished quality of life in obese individuals and may constitute an obstacle to successful weight management. In fact, a prior study conducted with participants from the PESO Trial revealed that body image change mediates changes in weight (Palmeira et al., 2009). Concerns might emerge at this point: how will partial scan images be perceived by weight sensitive individuals? Do they contribute to worsen body image? It is imperative that the patient does not derive the interpretation that partial scans are an evidence that he or she was too fat for the machine. Stated another way, the patient must not get the implicit messages that the machine was designed for "normal"

individuals, and that excludes him or her. These might be damaging to their health and well-being by invoking feelings of shame and inadequacy, particularly in weight sensitive patients. Moreover, if body composition assessments are performed during obesity treatments that proactively address body image investment features as part of their protocols, as recommended by Carraça et al. (2011a,b); Palmeira et al. (2009), then a misinterpretation of the DXA report may jeopardize the interventions. To better deal with this issue, the technologist should be careful when delivering DXA reports to obese patients or print them without image.

Conclusion

6

Considering the need for accurate body composition assessment of individuals who are wider than the DXA scan area, three solutions requiring a single partial scan were compared and validated: HS, left hand side of the body removed; RSU, upper left limb removed; RSUL, upper and lower left limbs removed. The scope of this investigation included BMC, LST, FM and %FM. The exploration of factors that could affect morphological asymmetry, including gender, athletic status and being involved in lateral dominant sports practice were also taken into consideration.

Overall, findings demonstrate that the reflexion-scanning approach is accurate, minimally invasive, and therefore is suitable for the obese and/or athletes engaged in sports at which superior width dimensions translate to performance gains.

RSU is superior to RSUL, and both reflexion techniques outperform half-scanning. RSU is the recommended option for practitioners working with Hologic Explorer-W densitometers, although RSUL might also be worthy in extremely broad individuals. Either reflexion-scanning protocol can compensate a width deficit of approximately 10 cm (evaluated in the coronal plane).

Although differences due to gender and sports practice were found non-significant or small, caution should be exercised when evaluating athletes suspicious of abnormal side-to-side differences as individual errors are likely to be exacerbated.

Future work

This section recommends future research to assess the validity and reliability of partial scanning solutions to assess body compartments, percent fat and bone mineral density. Also included are: approaches to avoid partial scanning (based on the positioning of hands); improve the validity of reflexion-scanning (based on corrective equations) and also summation scanning for individuals at which side-to-side differences are expected to be higher or at which tracking regional body composition is a requirement.

Validation against a 4C model   The results reported in this investigation used an whole-body scan as a reference. Future research on partial scanning solutions should be directed towards the overall accuracy of these techniques, using the 4C model as a reference. Figure 6.1 illustrates the proposed concept, whereby the measurements obtained by partial scanning techniques (reflexion and half-scanning) are compared against the 4C model by statistical procedures that include regression and agreement analysis.
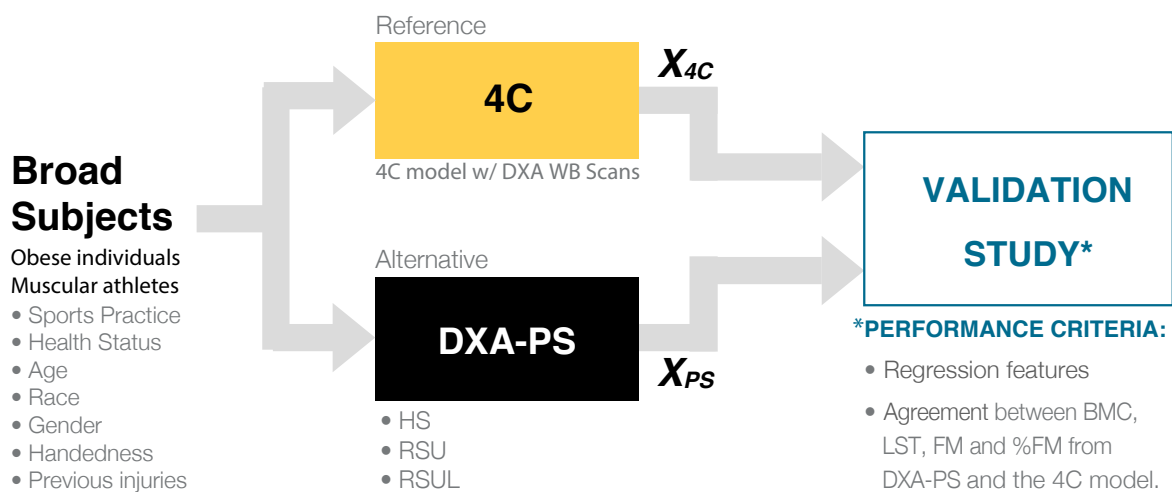
Figure 6.1: Validation of DXA-PS techniques for whole-body composition assessment.

The sample would consist of individuals whose width actually exceeds the limits of Hologic Explorer-W (65 cm), while remaining below its weight limits (136 kg). Accordingly, the 4C model and the DXA-PS methods would require different densitometers: one at which the obese and/or muscular athletes can be assessed using standard whole-body scans and the actual Explorer-W at which the partial scanning techniques are to be tested. Since it is unlikely that Hologic, Inc. would produce an adapted Explorer-W with extended width limits for the sake of one validation study, a GE Lunar iDXA (76 cm)

would be required for this purpose. Due to variations between devices, if cross-calibration equations did not exist for BMC, LST and FM in these systems, this study would have to be preceded by a calibration study similar to those of Oldroyd et al. (2003); Shepherd et al. (2012b) (an additional sample, with individuals that can fit in both scanners, would be required for this preliminary study).

Reliabilty Studies on partial scanning did not inspect the short-term precision of partial scanning solutions, in obese and non-obese populations. Future studies should address this topic for bone and lean tissue measurements, including whoie-body BMC, BMD, LST, FM and %FM. Reliability is important for applications at which the main purpose of DXA is to assess changes in body composition over time (from one semester to several years), and possibly with different technologists.

Bone mineral density Despite the focus of validation studies of partial scans is typically narrowed to %FM or the DXA compartments, the clinical relevance of bone mineral density for risk-stratification of sarcopenic and obese patients also recommends its inclusion in the set of variables to be validated (Gallagher et al., 2000; Petak et al., 2013). Misic & Evans (2006) was the only study published so far that reported validation results for summation scans to obtain estimates for whole-body BMD and BMC. Their summation protocol for wide individuals required the summation of three scans (subtotal without upper limbs plus two subregions for upper limbs) and was accurate; mean values were close to the whole-body reference values, correlations were strong and individual errors were within the coefficient of variation for BMD ($\approx$ 0.9%), as indicated by Bland-Atman analysis. Unfortunately, the proposed summation technique is plagued by added complexity for the technician and a threefold increase in the overall time required to complete a body composition evaluation. In contrast to summation, Sherman (2011) reported results for the more practical RSU in BMD. In a large sample from the NHANES Study comprising four hundred and thirty four scans of adults acquired on Hologic QDR-4500A systems (Centers for Disease and Control, 2000b), the bone tissue of the right arm was found to be slightly denser (3.4%) than the left one. Although this bias had no impact on whole-body precision, it could still affect the accuracy of bone measures. It is unclear whether these results hold for different systems and specific populations. More research is needed to validate partial scanning approaches for BMD, and preferably taking into account lifestyle or sports related factors that may impact on increased side-to-side differences.

When partial scanning can be avoided   In situations where the body in excess of the DXA table is small $(1-3\,\mathrm{cm})$, it is possible to avoid the need for partial scanning by positing subjects with palms facing the hip. Figure 6.2 illustrates the proposed concept. Region R1, in Scan 1, is the right hand-side hand positioned according to the recommendations of the manufacturer. R2, in scan 2, is the same hand, but with palms facing the hip. Region L depicts the left hand in standard position.
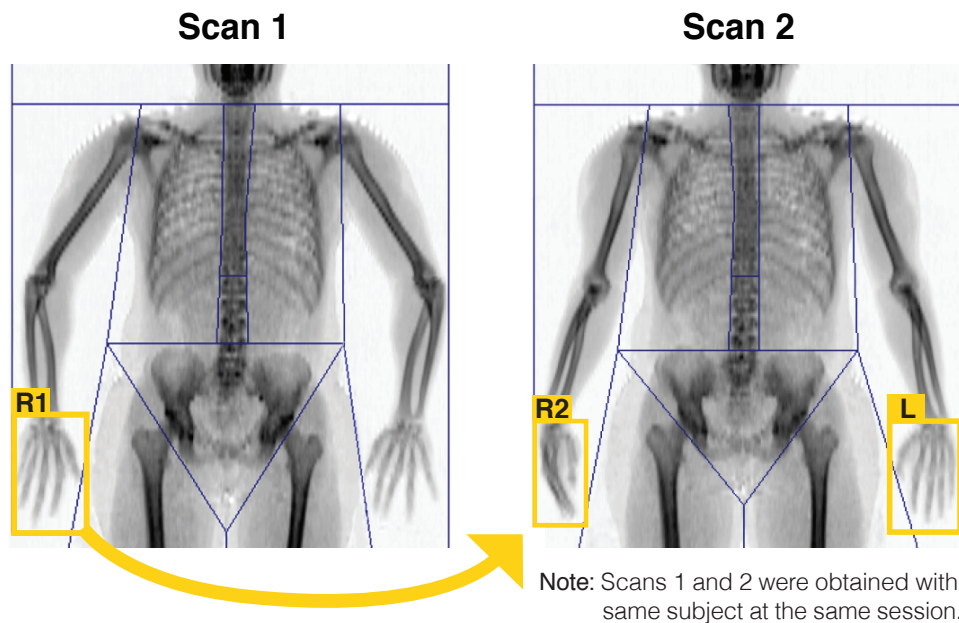
Figure 6.2: Alternative scanning protocol with palms facing the hip.

Future research on this topic should also address whether positioning aids could be used to to standardize the subject hand's positioning at every scanning time point. Figure 6.3 depicts a prototype (custom-made foam block) purposely designed and recommended by Nana (2013) therefore improving consistency in positioning subjects.

Despite its apparent simplicity and inexpensiveness, this solution is useful to standardize the subject's positioning at every scanning time, thereby contributing to minimize TEMs/CVs Nana (2013). Foam blocks were effective in combination with the summation techniques proposed and validated by Nana et al. (2012). However, caution should be exercised when translating these findings to densitometers that are not Lunar iDXA, since placing hands in midsaggital position is the standard/recommended positioning of General Electric for this specific equipment and not other manufacturers. More tests are needed to ensure that image processing algorithms of Hologic and Norland, for example,

Figure 6.3: Positioning aid to be used in scanning protocols with palms facing the hip. Copied with permission from Nana (2013).

can robustly handle this approach without compromising the validity of whole-body and even regional body composition measurements.

Automated summation scan   Partial scanning solutions may compromise the accuracy of measurements of whole-body composition and do not allow regional distribution analysis (at least of the upper limb). These drawbacks make the concept of summation scanning appealing, particularly in athletic populations for whom individuals errors could be higher. Also, following changes in lean tissue is useful for training monitoring and performance optimization. Accordingly, as future work, I propose two versions of summation scanning at which two partial scans are summed. These are based on the best performing technique of this study, RSU, and that of Santos et al. (2012), that addresses the complementary problem of assessing taller individuals.

In version A of Figure 6.4, the upper left limb is scanned whereas the remaining body goes in the second scan. In the version B—meant for simultaneously wide and tall individuals—the head and upper left limb are scanned first, whereas the rest of the body is measured in the second scan. For both versions, subjects will need to be repositioned (offset in the horizontal or also in the vertical direction). An advantage of this summation strategy in comparison with the solutions of Nana et al. (2012) is that the region of the shoulder is less sensitive to human error in delimiting a segment for arm trunk than the spine. Moreover, the head does not need to be halved in any of the proposed versions,
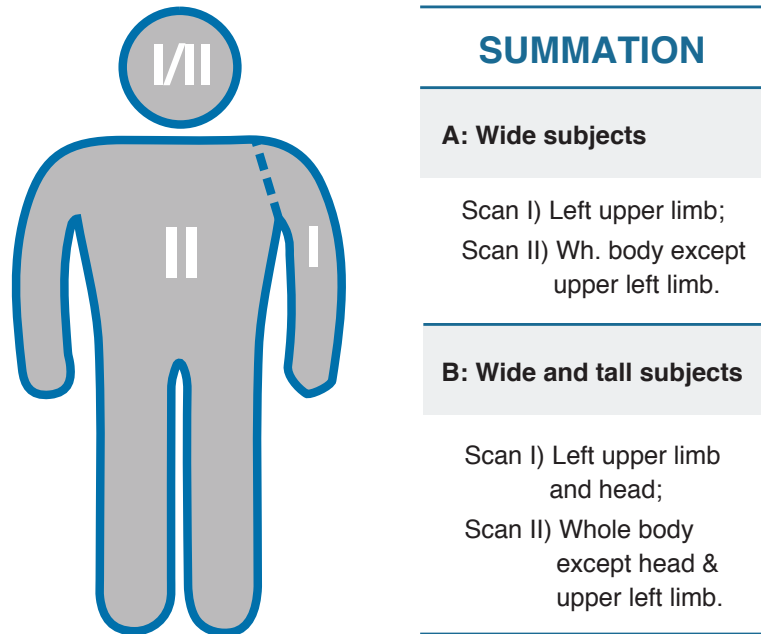
Figure 6.4: Alternative scanning protocol with palms facing the hip.

thus reducing summation errors even more. If the software provided support for such strategies, then the first scan could include only the region(s) of interest that will actually be needed and the overall evaluation time of two partial scans would be less than that of two whole-body scans. The summation process could be performed *automatically* by content-aware, feature-based matching and merging algorithms, thus also minimizing the scan analysis time requirements (Lowe, 2004; Szelinski, 2010).

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