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GROWTH AND MATURATION IN FEMALE NON-ATHLETES AND ATHLETES

Thesis for the degree of Doctor of Sport Sciences in the branch of
Sports Training supervised by Prof. Dr. Manuel J. Coelho-e-Silva,
Prof. Dr. João Valente-dos-Santos and Prof. Dr. Lauren B. Sherar,
submitted to the Faculty of Sport Sciences and Physical Education
of the University of Coimbra.

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“*Good teams become great ones when the members trust each other enough to surrender the Me for We*.”

Phil Jackson

To my family, friends and professors.

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Abstract

The current thesis was organized into eight chapters. In Chapter I, historical authors, data related with growth and biological maturation, basic concepts and literature in pediatric sciences were introduced, in addition to the overall purpose of this thesis. Chapter II detailed the samples, materials and procedures and data analysis included in each study. Subsequent chapters included study 1 to 5 and aimed: (*i*) to evaluate the growth, somatic, sexual and skeletal maturity of female youth soccer players; (*ii*) to examine inter and intra-observer agreement using different approaches of Greulich-Pyle (GP) method: overall (wholeGP) or bone-by-bone approach inspection of all bones (30-boneGP) or solely the pre-mature bones (GP_{pmb}). In order to obtain an overall skeletal age (SA), this analysis was conducted considering the median or the mean of previous mentioned GP alternatives; (*iii*) to compare GP and Fels SAs in a sample of female soccer players and to test the agreement of maturity status classifications with both protocols; (*iv*) to describe the growth and skeletal maturity status, assessed by Fels protocol, and maturity-associated variation in body size and composition among female soccer players; (*v*) to investigate the contribution of body size descriptors, isolated and combined with skeletal maturity status and training, to explain the inter-individual variability in left ventricular mass (LVM). A general discussion, summarizing the findings of each study, combined with their practical applications, were presented in Chapter VIII. Overall, the mentioned chapters highlighted that: (*i*) literature using SA as an indicator of biological maturation is limited, while predicted age at peak height velocity (PHV) was the most used method among female soccer players. This non-invasive estimation was positively related to ages at prediction; (*ii*) although the procedures to estimate a global SA with GP method are not totally clear, differences between bone-by-bone approaches were negligible. In contrast, an overall examination of the x-ray tended to increase inter-observer discrepancies; (*iii*) SA differences between concurrent protocols had practical applications to classify players according to their maturity status; (*iv*) female soccer players tended to be compared with general population in stature and were systematically heavier from under-13 to under-17. Additionally, players were mainly advanced in biological maturation; (*v*) fat-free mass emerged as the best single size descriptor to normalize LVM, but, when available, skeletal maturation and training parameters should be obtained. The current thesis underpins the impact of body size, composition and biological maturation assessment to the long-term athlete development among females involved in organized sport. It has the potential to provide useful information for clinicians, coaches and researchers in the clarification of concepts related to growth and maturity indicators. Finally, growth and maturation are parameters that should be regularly assessed to examine inter-individual differences among female soccer participants.

Keywords: Growth; Biological Maturation; Skeletal Age; Greulich-Pyle; Fels; Somatic Maturation; Sexual Maturation; Female Athlete; Youth; Echocardiography; Scaling; Allometry

Resumo

A presente tese está organizada em oito capítulos. No primeiro capítulo foram abordados os autores históricos, os principais estudos longitudinais e transversais, os conceitos de crescimento e maturação. O segundo capítulo detalhou as amostras estudadas, assim como os materiais, procedimentos e técnicas de análise estatística. Os capítulos subsequentes incluem os estudos que constituem a presente tese. Os seus objetivos foram: (*i*) avaliar o crescimento e a maturação biológica (somática, sexual e esquelética) em jovens jogadoras de futebol; (*ii*) examinar a reprodutibilidade e a concordância entre observadores no método *Greulich-Pyle* recorrendo a diferentes abordagens: global ou para cada um dos ossos (considerando só os pré-maturos ou incluindo os 30 ossos). Para o cálculo da idade óssea foi testado o impacto da média ou da mediana; (*iii*) comparar as idades ósseas estimadas por dois métodos concorrentes (*Greulich-Pyle* e *Fels*) e avaliar a sua concordância na classificação das futebolistas femininas por grupo maturacional; (*iv*) descrever os parâmetros de crescimento e a maturação biológica, estimada pelo método de *Fels*, e a variação do tamanho e da composição corporal por categoria maturacional; (*v*) investigar a influência do tamanho corporal isoladamente e, combinado com a maturação esquelética e a experiência de treino, para explicar a variabilidade inter-individual na massa do ventrículo esquerdo. O capítulo final compreende uma discussão geral, na qual os principais aspetos e conclusões dos diversos estudos são sumariados, contextualizados e, com efeito, discutidas as suas implicações práticas. A partir da análise dos principais resultados, os estudos revelam que: (*i*) em futebolistas femininas, os estudos que reportam a idade óssea como um indicador de maturação biológica são escassos enquanto, o modelo preditivo do pico de velocidade de crescimento foi o método mais reportado; (*ii*) embora a estimativa da idade óssea pelo método *Greulich-Pyle* não seja totalmente concreta, as diferenças entre observadores foram negligíveis quando a análise radiográfica foi realizada osso a osso. No entanto, quando as avaliações foram efetuadas globalmente, as discrepâncias entre observadores aumentaram; (*iii*) as diferenças entre protocolos concorrentes tem aplicação práticas para classificar as jogadoras por categoria maturacional; (*iv*) as jogadoras de futebol feminino tendem a apresentar valores médios para a estatura comparados com a população geral e foram consideradas mais pesadas nas faixas etárias analisadas.

Adicionalmente, a maioria dos participantes foi considerada avançada na maturação biológica; (*v*) a massa isenta de gordura evidenciou-se como o melhor preditor para normalizar a massa do ventrículo esquerdo, mas quando possível, a idade óssea e os parâmetros de treino devem ser obtidos para a interpretação da mesma variável dependente. A presente tese demonstrou o impacto do tamanho, composição corporal e maturação biológica para o desenvolvimento a longo prazo das atletas femininas. A clarificação dos conceitos de crescimento e maturação é potencialmente útil tanto no âmbito clínico como no âmbito desportivo. Finalmente, estes parâmetros devem ser frequentemente avaliados para explicar as diferenças inter-individuais em jovens futebolistas femininas.

Palavras-chave: Crescimento; Maturação; Idade Óssea, Greulich-Pyle, Fels, Maturação Somática; Maturação Sexual; Atleta Feminina; Adolescência; Ecocardiografia; Scaling; Alometria

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List of Abbreviations

%95 CI	– 95% Confidence intervals
%FM	– Percentage of fat mass
%PMS	– Percentage of mature stature
30-boneGP	– Bone-by-bone inspections of 30-bones
A	– Agreement
a	– Constant
ANOVA	– Analysis of variance
ANCOVA	– Analysis of covariance
BMI	– Body mass index
BMIz	– Body mass index z-score
BSA	– Body surface area
CA	– Chronological age
D	– Disagreement
DEXA	– Dual-energy-x-ray absorptiometry
E	– Early
ε	– Error
FFM	– Fat-free mass
FIFA	– Fédération Internationale de Football Association
GP	– Greulich-Pyle
GP_{pmb}	– Bone-by-bone inspections of the pre-mature bones
ICC	– Intra-class correlation coefficient
ISWTd	– Interventricular septal wall thickness
k	– Allometric coefficient
L	– Late
Lin CCC	– Lin concordance correlation coefficient
LLTS	– Leuven Longitudinal Twin Study
LTAD	– Long-term athlete development
LVIDd	– Internal dimension of the left ventricle at end diastole
LVM	– Left ventricular mass

LVWT	– Left ventricular wall thickness
M	– Mean
max	– Maximum
Md	– Median
MHz	– Megahertz
min	– Minimum
MRI	– Magnetic resonance image
NHANES	– National Health and Examination Survey
OB1	– Observer 1
OB2	– Observer 2
PBMAS	– Pediatric Bone Mineral Accrual Study
PH	– Pubic hair
PHV	– Peak height velocity
PWTd	– Posterior wall thickness at end diastole
PWV	– Peak weight velocity
r	– Coefficient of correlation
R	– Model correlation coefficients
R^2	– Coefficient of determination
R^2 adjusted	– Adjusted coefficient of determination
RUS	– Radius, ulna and short bones
RWT	– Relative wall thickness
SA	– Skeletal age
SD	– Standard deviation
SEE	– Standard error of estimation
SGDS	– Saskatechewan Growth and Development Study
SWTd	– Septal wall thickness at end diastole
t	– Paired or independent t-tests
TM1	– Time-moment 1
TM2	– Time-moment 2
TOYA	– Training of Young Athletes
TW	– Tanner-Whitehouse
TW1	– Tanner-Whitehouse version 1
TW2	– Tanner-Whitehouse version 2

TW3 – Tanner-Whitehouse version 3

U.S. – United States

VIF – Variance inflation factor

VO_{2peak} – Peak oxygen uptake

wholeGP – Overall approach

x – Independent variable

ηp^2 – Eta squared

Chapter I

General introduction

1. General introduction

1.1. The study of human growth

The study of human growth emerges in the 19th century, and the term “auxological epidemiology” was cited by Tanner (1981) in order to define suboptimal conditions of health based on growth data. The first human growth curve was defined by Boas, Baldwin and Shuttleworth in 1930 and some important parameters were noted. For example, it was evident an interpersonal variation in times of maximal growth spurt. In other words, participants attained “age of maximal velocity” (Franz-Boas) or “peak height velocity (PHV)” (Tanner) in different timings. In parallel, Boas also noted that among adolescents, the adult stature did not vary. In this context, two different concepts were defined – growth and maturation. The International Association of Human Auxologists was created in 1977 and the Fifth International Congress of Auxology held in Exeter in 1988 reviewed the main literature about human growth. Currently, early longitudinal data from America (1930-1970) and Europe (1950-1980) are still reported in studies of children and adolescents and then represent a historical source of reference (Ulijaszek, Johnston, & Preece, 1997, Tanner, 1989).

Growth, maturation and development

The main North American longitudinal studies were led by educationists (e.g. Baldwin), psychologists (e.g. Bayley), doctors (e.g. Todd) and physical anthropologists (e.g. Krogman). The first one, conducted at Iowa by Baldwin and Meredith (1917-1970), was aimed to develop physical measurements. The Atlas of Skeletal Maturation was initially created by Thomas Wingate Todd (1937) and later it was the basis for the Greulich-Pyle (GP) atlas. At the University of California Institute of Child Welfare, in 1928, Nancy Bayley included a birth-to-maturity sample and examined growth parameters and intellectual development - Berkeley Growth Study (Jones & Bayley, 1941).

The most important longitudinal study was developed at the Fels Research Institute. Initially, Frank Falkner was the coordinator of the International Children Centre Studies in Paris and London and, in 1970, provided his experience as auxologist, pediatrician and leader to the institute. Additionally, three remarkable physical anthropologists integrated the Fels Research Institute: Earle Reynolds, Stanley Garn and Alex Roche (Tanner, 1989). The first European longitudinal study was the Harpenden Growth Study, conducted in an orphanage and led by James Mourilyan Tanner with Whitehouse as an anthropometrist (Tanner, Whitehouse, Marubini, & Resele, 1976). Before that, Tanner visited the U.S. and learned measurement procedures with Meredith. The Harpenden Growth Study uniformed the techniques, and all the studies coordinated by the International Children Center in Europe applied the same procedures. This aspect highlighted the historical connection between the European and the American studies. Whitehouse created the anthropometric apparatus for Harpenden and, in parallel, a new protocol for assessing skeletal maturation was implemented – Tanner-Whitehouse (TW) method (Tanner, Whitehouse, & Healy, 1962; Tanner et al., 1975). Based on the first studies of Reynolds and Wines (1948, 1951) at Fels Research Institute, the stages of secondary sex characteristics were developed in the Harpenden Growth Study. Additionally, differences between cross-sectional and longitudinal standards were constantly discussed between authors. In this context, two important authors interested in growth data analysis collaborated with Tanner – Michael Healy and Harvey Goldstein. The contribution of these authors was relevant in pediatric exercise sciences and particularly in studies that used growth and maturity indicators. The interest of growth and maturation are particularly pivotal during the adolescence. In this period, an acceleration in the rate of growth is noted and most of bodily systems attain the maturity. Obviously, growth and maturation should be interpreted in parallel with the term development or behavioural context (Tanner, 1989).

1.2. Growth and maturation

Growth is the main biological activity during the first two decades of life and implies an enhancement of the body as a whole or in specific segments. Consequently, children and adolescents become taller and heavier. Changes in body mass correspond to transformations in different bodily tissues – skeletal, muscle, fat and organs (Malina, Bouchard, & Bar-Or, 2004). Maturation is a process towards the adult or maturity state (status). The process occurs in all tissues, though it presents different timings and rates. Timing refers to the moment when a maturational event happens (e.g. age at PHV, age when 90% of the predict adult stature was attained), while tempo is the rate of specific maturational event (Malina et al., 2004; Malina, 2014). Maturity occurs in all organs, systems and tissues, but it is specific and variable according to the biological-system concerned. In other words, sexual maturity is defined as the fully functional reproductive capability, while skeletal maturity is related to the total ossified adult skeleton. Two other important biological systems are: the endocrine and nervous systems. Both are associated with sexual, somatic and skeletal maturation. Even though they are often expressed as synonyms, growth and maturation represent different concepts and their study is based on measurements and observations of specific outcomes (Malina et al., 2004; Malina, Rogol, Cumming, Coelho e Silva, & Figueiredo, 2015). Whole-body, partial lengths, circumferences, breadths and skinfolds are commonly used to describe growth status in youth samples. Stature and body mass were extensively reported as the whole-body size descriptors, and both present comparable age-specific paths from infancy to adulthood. For example, stature tends to increase during infancy and early childhood, stabilizes during middle childhood, rises during adolescence and slow increases until adulthood. A similar pattern was noted for body mass, but after adolescence it continues to increase until adult age (Malina et al., 2004). The growth curves or reference data provide indications about the size attained at a specific age. Based on national surveys, the curves describe age and sex-specific percentiles among youth. The growth charts developed in the U.S. samples combined national surveys from 1959 to 2000 (Kuczmarski et al., 2002). Two important aspects related to the development of growth charts should be noted: data from different ethnic populations was combined and body masses of children measured between 1976-198 (National Health and Nutrition Examination Survey (NHANES II)) and 1998-1994 (NHANES III) were not included in the formulation of the growth charts. The exclusion of this data was related to the increasing of obesity among American youth and, consequently, considered undesirable from a public health perspective to avoid an upward shift of the percentile curves for body mass. Overall, sex and age and sex-specific percentiles are effective to plot the growth status of youth samples (Kuczmarski et al., 2002). As previously mentioned, maturation is a process towards the final stage (maturity or adult) and it is often viewed in terms of status and timing. Maturity status refers to the state of maturation at the chronological age (CA) of observation, while the latter refers to the CA at which specific maturational landmarks occur. The terms are related, but express different concepts (Malina et al., 2015).

Sexual indicators

The two most common indicators to examine the biological maturity status are secondary sex characteristics and skeletal maturation. Based on earlier studies of Reynold and Wines (1948, 1951), written descriptions were produced for each stage of secondary sex characteristics (Tanner, 1962). Sexual indicators include breasts and menarche in females; genitalia, voice change and facial hair in males; pubic and axillary hair in both sexes. Five stages are used to describe breast and genital development and six stages define the development of pubic hair (PH) in males and females. The stage 1 for breast, genitalia or PH indicates the absence of each characteristic, and thereby represents the prepubertal state. The subsequent stages describe the development of the secondary sex characteristics, while the stage 5 corresponds to the adult or mature state. In the meantime, the limitations of theses stages should be noted. For example, a female may be classified as in stage 2 for breast development and stage 1 for PH, which suggests that stages are not equivalent. An additional problem is the definition of the beginning and the end of a particular stage. For example, a boy rated as stage 4 for genitals could be entering at the beginning of stage 4 or at the end of the same stage.

The CA at entry into a stage and the period of time (rate) that a child spends in each stage (timing) are highly variable (Sherar, Baxter-Jones, & Mirwald, 2004). Therefore, intervals between observations should be short (3-6 months). Finally, stages of genitals and PH were combined into a continuous score, e.g., 1.8 or 2.6, and reported using the mean and standard deviation (Quiterio, Carnero, Silva, Bright, & Sardinha, 2009). Stages are discrete categories, thus there are no equivalent intermediate stages.

Menarche is defined as the first menstrual period and the timing (age) at which menarche occurs is often reported as a sexual indicator of female adolescents. There are three methods to predict age at menarche: longitudinal, status quo and retrospective. Longitudinal observations include regular visits (3-6 months of interval) and two parameters are retained: if the menarche has or has not occur, and, if so, the timing of the occurrence. Status quo method includes a sample aged 9-17 years old and estimates the age at menarche for a sample based on a prohibit analysis. The method also calls for two predictors: if the girl is premenarcheal or postmenarcheal and the CA when the first menstrual cycle occurred. Essentially, the method estimates the median age at menarche and also provides percentiles for the sample. The retrospective method can only be applied to postmenarcheal females and they are asked to recall the age at menarche (Malina et al., 2004). Obviously, the method is based on the memory of recall and excludes the premenarcheal girls. Status quo or retrospective questionnaire are mainly used in samples of female adolescents (Baxter-Jones & Helms, 1996; Padez & Rocha, 2003) and the data is discussed later in the Chapter III.

Somatic indicators

Growth parameters are not indicators of maturation. However, using longitudinal data, important parameters can be estimated: the initial inflection of the growth curve and age at maximal growth spurt. Additionally, the percentage of mature stature (%PMS) attained at different ages could be used as a potential maturity indicator. Briefly, the current stature is expressed as percentage of adult stature. Among adolescents of the same CA, the one closer to adult stature is closer to the mature state than the other farther from adult stature (Malina et al., 2004).

Specific mathematical models fitted the repeated measurements of stature in European and American samples. Allowing for variation in different modelling methodologies, age at PHV and PHV were estimated for European and American longitudinal samples. For example, age at PHV for the 63 females from Fels Longitudinal Study (Malina, Choh, Czerwinski, & Chumlea, 2016) was 11.61±0.73 and 12.01±1.00 for 131 females examined in the Zurich Longitudinal Growth Study (Prader, Largo, & Holf, 1974). Substantial variance was noted: age at PHV range from 9.59 to 13.18 years in American girls (Malina et al., 2016). Based on Preece-Baines Model 1, age at PHV among females from Wroclaw Growth Study was 11.61±0.73 years old and the observed age at PHV ranged 9.03-14.82 years (Malina & Kozieł, 2014). The rate or centimeters gained during the maximal growth spurt (PHV) was, on average, consistent in different longitudinal studies. Using a 5th order polynomial function, American females (Byard & Roche, 1993) tend to gain, on average, 7.94 cm per year. Among Polish non-active, females gained 7.5 cm per year (Malina et al., 1997). The results were consistent across different samples, although data including young females following longitudinally is lacking. Other possible indicator of somatic maturation is the %PMS attained at a particular age (Malina et al., 2004). In the meantime, age at PHV and %PMS attained require longitudinal data. Of interest, non-invasive estimations of the previous mentioned indicators are systematically applied in studies of young male and female athletes (Westbrook, Taylor, Nguyen, Paterno, & Fordo, 2020; Baxter-Jones et al.,2020; Martinho et al., 2021). Based on the differential timings of sitting height and leg length, sex-specific equations were created to predict the time before or after PHV (in years). The equation was based in 115 girls from the Saskatchewan Pediatric Bone Mineral Accrual Study (PBMAS) followed between 1991 and 1997. Two other samples from the Saskatchewan Growth and Development Study (SGDS) and the Leuven Longitudinal Twin Study (LLTS) were used to cross-validate the original equation, but the final equation combined the three longitudinal studies. Measurements and the time between observations varied between studies: Canadian girls were followed semi-annually, while Belgian females were measured annually. The standard error of estimation (SEE) for females was 0.569 (Mirwald, Baxter-Jones, Bailey, & Beunen, 2002). Given the interest in non-invasive estimations, two recent equations emerged to the predicted age at PHV (Moore et al., 2015; Fransen et al., 2018). Using two Canadian studies and data from Harpenden Growth Study, a simple model, including solely age and sitting height, was calculated for girls. The SEE was 0.528 years (Moore et al., 2015).

Finally, and more recently, also including Canadian youth and Belgian soccer players, a maturity ratio (CA/age at PHV) with age, stature, body mass and leg was established (Fransen et al., 2018). The model was developed uniquely in males.

% PMS attained at a given CA requires current stature and is later on expressed as a percentage of the predicted adult value. The participant who is closer to adult stature is more upfront in maturity status compared to the one who is farther from mature height (Bayer & Bayley, 1959; Beunen et al., 1997; Roche, Tyleshevski, & Rogers, 1983). Initially, some equations including skeletal age (SA) were proposed. However, a sample of 210 white females from the Fels Longitudinal Study produced a non-invasive estimation of percentage of the predicted adult height based on CA, stature, body mass and midparent stature (Khamis & Roche, 1994). The error between actual and predicted mature stature from 4 to 17 years using the Khamis–Roche protocol was, on average, 1.52 cm in females (Khamis & Roche, 1994). The protocol has been used in studies of young males and females involved in competitive sport (Malina et al., 2010; Howard, Cumming, Atkinson, & Malina, 2016; Westbrook et al., 2020).

Skeletal indicators

The secondary sex characteristics are limited to pubertal years, while SA is applicable from infancy until adulthood. Therefore, skeletal maturity is probably the best indicator of biological maturity status. It is often assessed by an examination of a standard radiograph (progresses from initial ossification to the mature state) of the hand-wrist bones in relation to the sample upon which the method of assessment was created (Malina et al., 2004). The three commonly methods reported for estimating SA are: GP (Greulich & Pyle, 1959), TW (Tanner et al., 1962; Tanner et al., 1975; Tanner, Healy, Goldstein, & Cameron, 2001) and Fels (Roche, Chumlea, & Thissen, 1988). It should be noted that the methods are comparable in relation to principle (the bones are matched with specific criteria), but they differ in criteria, samples and procedures (Malina et al., 2015).

The GP protocol was developed in white American children of high socio-economic status (31 males and 29 girls) in Brush Foundation Study. The GP was based on early studies of Thomas Wingate Todd (1937) and it is often known as atlas method.

The assessment of SA calls for a detailed examination of hand and wrist bones comparing them with the photographs of Atlas. The instructions of the original book include a global analysis of x-rays in order to find the most approximate standard. Using the same book, a detailed examination of the 30 bones should be done separately. The estimate of SA refers to the CA of the child from the Brush Foundation Study whose plate coincides with the one under-examination (Greulich & Pyle, 1959). By inference, 30 bones were considered for the estimation of SA. Information about the SA estimation is lacking, particularly the exclusion of mature bones and which measure of the central tendency should be used (i.e., median or mean). This topic is discussed in Chapter IV.

The TW method was originally developed in ~3000 British children from a medium socio-economic level. It corresponds to a biological weight method and features of 20 bones should be rated according to the written criteria proposed by the original authors. Radius, ulna, carpals, metacarpals and phalanges of the first, third and fifth digits are assigned as discrete stages, and scores for each stage were derived. An overall maturity score, ranged from zero to 1000 (maturity), was obtained by summing the score of each bone. Carpal bones and radius, ulna and short bones systems explained separately 50% of the total maturity score. The total of 20 scores can be transformed in SA, which is labelled as TW1-20 bone SA (Tanner et al., 1962). The first revision of the method eliminated some stages of the long and round bones, and sex-specific stages were re-weighted. Additionally, an arithmetic mean of systems did not fit the information given by all bones. For example, finger bones provide more information than carpals, radius and ulna, so then separate scoring systems established for carpals (TW2 carpals), radius, ulna and short bones (TW2 RUS), in addition to TW2-20 bone, were created. Based on the conversion of these scores, specific SAs were obtained: TW2 carpals SA, TW2 RUS SA and TW2-20 bone SA (Tanner et al., 1975). The most recent version of Tanner-Whitehouse method (TW3) maintained the grades and respective scores for each bone. Additionally, samples of Great Britain, Belgium, Italy, Spain, Argentina, America and Japan were included for the conversion of maturity scores to SA. 20-bone SA was eliminated, but the tables for the conversion of the skeletal maturity scores to SA were updated for carpals (TW3 carpals SA) and RUS systems (TW3 RUS SA) (Tanner et al., 2001).

The Fels protocol was developed based on a longitudinal sample from the Fels Longitudinal Study (355 and 322 girls). The sample, analyzed between 1932 to 1937, was from middle-class families in south-central Ohio (U.S.). Briefly, age and sex-specific categorical grades for different maturity indicators of radius, ulna, carpals, metacarpals and phalanges of the first, third and fifth fingers were verbally described. The linear ratios of the epiphysis and metaphysis and the absence or presence of the adductor sesamoid and pisiform were also considered. Grades and linear measurements are introduced into a software that estimates SA and error associated with the assessment based on the maximum likelihood method (Roche at al., 1988).

1.3. Pediatric exercise sciences: growth and biological maturation

Inter-individual variation in growth and maturation is a common issue in pediatric exercise sciences, including studies involving male and female young athletes (Malina et al., 2015; Malina, Figueiredo, & Coelho-e-Silva, 2017). For example, among 135 elite male soccer players aged 10.7-16.5 years, whose stature and body mass were plotted on percentile 50th at 11-12 years and fluctuated the 50th and 75th percentiles at 13-14 and 15-16 years. In parallel, SA, assessed using the Fels method, approximated, on average, CA at 11-12 years, while SA was, on average, advanced CA in 13-14 and 15-years. Thus, the number of late maturing systematically decreased with CA, and the average and early maturing increased in the two year older groups (Malina et al., 2000).

Most recently, a compilation of 144 cross-sectional studies obtained from 1978-2015, involving male soccer players (Malina et al., 2017), examined secular changes in stature and body mass as well as in somatic indicators (age at PHV, estimates of stature attained at PHV and adult height). Secular trends in stature and body mass between 1978-1999 and 2020-2015 were noted. On the other hand, based on Preece-Baines model I, estimated age at PHV was 13.01 and 12.91 years for studies grouped in 1978-1999 and 2000-2015, respectively. A maturity-associated gradient was also noted in cross-sectional samples of British male tennis players (Myburgh, Cumming, Coelho E Silva, Cooke, & Malina, 2016) and

Portuguese and Polish table tennis players (Coelho-e-Silva et al., 2021). Both studies assessed SA using the Fels method. Among 47 elite Lawn tennis players, 17 of 27 under-12 players were classified as average in skeletal maturity status with equal numbers classified as delayed and advanced, while among under-14 and under-16 players, no player was classified as late maturing, and the numbers of players classified as average were comparable to the advanced in skeletal maturity status. Mean statures and body masses fluctuated between the 75th and 90th percentiles during adolescence (Myburgh et al., 2016). A cross-sectional sample of 68 Portuguese and 31 Polish table tennis players between 10.00–14.63 years of age, reported age-specific means for stature and body mass above the 50th percentile and, consistently, mean SA tended to exceed mean CA (Coelho-e-Silva et al., 2021). The selective nature of sport, among males, follows a maturity-related gradient which is not related to the indicator of maturation in use and the type of sport.

Consistent trends were noted when secondary sex characteristics (i.e., PH stages, testicular volume) were used as an indicator of maturity status in male soccer (Malina, Eisenmann, Cumming, Ribeiro, & Aroso, 2004) and basketball participants (Coelho e Silva et al., 2010a). Among 69 soccer players aged 13.2-15.1 years old, size parameters (i.e. stature and body mass) were plotted relative to U.S. data (Kuczmarski et al., 2002), in addition to sexual maturation that was assessed by secondary sex characteristics (i.e. PH stage). To be noted that approximately 48% were classified as PH4 or PH5. Consistent findings were reported among 80 male adolescent players aged 12-13 years old (Coelho e Silva et al., 2010a). 16 and 11 participants were classified as PH3 and PH4 at 12 years old, respectively, while 15 and 25 players were grouped as PH3 and PH4 at 13 years old, respectively (Coelho e Silva et al., 2010a).

One of the most popular longitudinal studies in youth sports was Training of Young Athletes (TOYA). Briefly, the participants (231 males) were followed annually during 3 consecutive years. The final sample of males included subjects from soccer, swimming, tennis and gymnastics. Growth parameters and testicular volume were plotted against British reference values. After controlling for CA and pubertal status, swimmers were, on average, +10.6 cm and +3.6 cm taller than gymnastics and soccer participants, respectively. Comparable results were reported for body mass: it was found that swimmers were heavier than gymnasts and tennis players from 14 to 16 years of age.

At 15 years old, swimmers and soccer players had, on average, greater testicular volume than gymnasts (Baxter-Jones, Helms, Mafulli, Baines-Preece, & Preece, 1995; Baxter-Jones & Helms, 1996). The authors of previous studies confirmed that swimming and soccer tend to select early and late maturing participants, respectively. These trends confirmed the early studies with youth players from Little League World Series (Hale, 1956; Krogman, 1959). Based on three criteria of PH, 112 participants (mean CA: 12.53 years) were classified as pre-pubertal, pubertal and post-pubertal. Approximately, 46% of the total sample was classified as post-pubertal (Hale, 1956). In parallel, SA, assessed according to Todd (1937) standards, exceeded CA in 39 of 55 male baseball players (Krogman, 1959). Overall, among males, sport seems to be selective. In other words, selection and retention in many sports tend to follow a maturity-related gradient, mainly during the maximal growth spurt. The proportion of early maturing or skeletally mature sport participants increased through the adolescence, while the frequency of late maturing participants tended to decrease with CA (Malina et al., 2015). Contrasting with the literature in male sport participants, studies of growth and maturity status including females are limited and mainly focused on individual sports (i.e. track and field, distance runners, swimming, tennis, dancers, figure skaters and mainly in gymnastics). Among 44 elite female tennis players from under-10 to under-16, stature and body mass were, on average, near to 75th centile in the four CA groups. SA, assessed by Fels method, was systematically in advance of CA, but 66% of the female sample was classified as on average (Mybrugh at al., 2016). In contrast, 16 female distance runners, followed annually during 4 or 5 years, were plotted on the 50th percentile relative to U.S. data (Eisenmann & Malina, 2002). Additionally, SA estimation using the Fels protocol was only obtained at the baseline of 13 participants. Mean SA (11.7 years) lags behind -0.52 years compared to mean CA (12.2 years). The skeletal maturity and body size were examined in cross-sectional samples of 26 female late adolescents: track and field late adolescents aged 15.2 to 17.9 years old, specifically. Consistent with the previous studies involving female sport participants, nine of 24 athletes had SA lower than CA and the success in youth sport was associated with normal or delayed biological maturation (Malina, Beunen, Wellens, & Claessens, 1986).

Furthermore, studies that reported growth and maturity status of female gymnasts were extensively examined and documented.

Mean stature of gymnasts that participated on TOYA study was systematically below the median of reference data (Erlandson, Sherar, Mirwald, Mafulli, & Baxter-Jones, 2008) and, apparently, they had lighter body masses (Baxter-Jones & Mafulli, 2002). Interestingly, swimmers (13.3 years) and tennis players (13.2 years) attained the menarche approximately 1.0 year earlier than gymnasts (14.3 years) (Baxter Jones et al., 1994). Similar ages at menarche were reported among European gymnasts. Among Polish (Zíemilska et al., 1981), Swiss elite (Tönz Stronski, & Gmeiner, 1990) and highly trained (Theintz, Howald, Weiss, & Sizonenko, 1993), Swedish (Lindholm, Hagenfeldt, & Ringertz, 1994), expressed in years, means and standard deviations ages at menarche were 15.1±0.9, 14.4±0.2, 14.5±1.2, 14.5±1.4, respectively. Overall, mean statures were below the 50th percentile and later maturation noted in female artistic gymnasts has often been attributed to the specific-selective nature of sports (Malina et al., 2013).

1.4. Growth and maturity-associated variation in left ventricular mass

The interpretation of cardiac outputs is central to understand the response of children and adolescents to exercise. To be noted that left ventricular mass (LVM) has been described as an important morphological characteristic of the heart (Malina et al., 2004). Changes in LVM output during adolescence are related with growth and heart adaptations promoted by the exercise (de Simone et al., 1998). Adaptations in response to the hemodynamic loading conditions induced by the participation in organized and regular sport include changes in the cavity size, wall thickness, which, in return, will predict the total mass of the left ventricle (Krysztofiak, Młyńczak, Folga, Braksator, & Małek, 2018). Intra- and inter-individual variability in cardiac variables in general and LVM in particular are associated with participation in sports, but observed changes depend in large part on the type of sport (Golbidi & Laher, 2012). Chronic volume loads are generally associated with an increase in end-diastolic diameters and by inference in LVM; these consequently result in eccentric hypertrophy (Demirelli et al., 2015; Krysztofiak et al., 2018). On the other hand, when posterior wall thickness increases and not the absolute size of cavity, the ventricle is defined by a concentric remodelling (Krysztofiak et al., 2018). Cross-sectional and longitudinal data have reported that LVM increases from childhood to adulthood (Malina et al., 2004; Dekkers, Treiber, Kapuku, Van Den Oord, Snieder, 2002; Sabo, Yen, Daniels, & Sun, 2014), which may suggest that cardiac dimensions are strongly related to body size. Adolescence is a critical period with significant changes in body proportions indicating that LVM should be normalized for the body dimensions (Malina et al., 2004). Although American Society of Echocardiography and the European Association of Cardiovascular Imaging stated that (Lang et al., 2015): "*The indexing of LV mass allows comparisons in subjects with different body sizes. However, whether to use height, weight, or BSA as the indexing term remains controversial. Studies suggest that indexing to height raised to allometric powers such as 1.7, 2.13, and 2.7 has advantages over indexing to BSA, especially when attempting to predict events in obese patients (p. 248)*", fat-free mass (FFM) was claimed as the most prominent size descriptor to normalize cardiac output (Kryztofiak et al., 2018; George, Birch, Pennel, & Myerson, 2009; Giraldeau et al., 2015; Koorenman et al., 2018). Non-linear interrelationships between growth, maturation and sport participation were examined among male adolescents but few studies using females involved in organized sport (Utomi et al., 2013).

1.5. Adjusting and interpreting physiological functions for variation in body size

Assumptions for ratio standards and allometric modelling

From childhood to adulthood, significant changes in body size and proportions are noted and, as expected, these transformations are followed by increases in absolute values of physiological outputs. As previously mentioned, LVM is routinely expressed by stature or body surface area (BSA), while peak oxygen uptake ($VO2_{peak}$) is adjusted for body mass. However, FFM has been claimed as the best denominator of physiological outputs. Note, this issue needs to be examined, particularly among females. This process of eliminating the effect of body size is denominated scaling. In other words, the aim of scaling is to produce a size-free output (Welsman & Armstrong, 2008). Consequently, the scaled variable should appropriately account for body size without retaining any residual correlation with the original size variable (Nevill, Ramsbottom, & Williams, 1992). The robust associations between size and performance also highlight a significant inter-individual variation in body size within CA groups. Additionally, a simple scatter-plot indicated that data points are more concentrated in the lighter children, becoming progressively more spread with the increase of body mass (fanning plot that represents non-constant error or heteroscedasticity). Traditional ratios are expressed as a division of physiological parameters by the size descriptor. Although the limitations of ratios standards are well described in the literature, they are often used to create a size-free expression. To be noted that statistical procedures indicate that correlation coefficient of physiological performance normalized for size descriptor, and respective body dimension, should not be significantly different from zero. If the correlation remains statistically significant, simple ratios standards fail to accommodate the impact of body size. Additionally, using this type of approach, the error is additive or constant. Unfortunately, graphical visualization of performance and body size suggests a heteroscedasticity, which means the error increases as performance and size increases. The error is multiplicative. In order to solve the limitations of traditional ratios, non-linear alternatives emerged as the best techniques to create a size-free expression (Welsman & Armstrong, 2008). Indeed, allometric scaling was initially adopted in biological sciences for describing and interpreting size-related changes in resting metabolic rate among mammals (Schmidt-Nielsen & Knut, 1984). This non-linear approach successfully accommodates the relationship between body size descriptors and physiological parameters, it overcomes the heteroscedasticity (more points are clustered of the regression line) and represents a better fit model than the traditional ratios (Nevill & Holder, 1994).

$$y = a \cdot x^k \cdot \varepsilon \qquad \textbf{(1.1)}$$

$$\ln(y) = \ln(a) + b \times \ln(k) + \ln(\varepsilon) \qquad \textbf{(1.2)}$$

The association between a physiological variable scale and respective size descriptor is subsequently tested using Pearson product-moment correlation coefficients. If the allometric model was successful in partitioning out the influence of body size parameters, the Pearson correlation coefficients correlation should approach zero. The normal distribution of residuals (predicted output – observer output) is statistically tested and graphically inspected to confirm homoscedasticity of error (Nevill et al., 1992). The variance of a dependent variable is not totally fitted by the predictor included in simple linear regression. Then, multiplicative allometric models are needed to fit the unknow variance of the physiological performance. These models included more than one predictor, and often consider indicators of biological maturation and training. Multiple linear regression is the statistical technique used to fit the unknow variance (Equations 1.3, 1.4, 1.5):

$$y = \text{size descriptor}_1{}^{k1} \times \text{size descriptor}_2{}^{k2} \times \varepsilon \qquad \textbf{(1.3)}$$

$$\ln(y) = k_1 \times \ln(\text{size descriptor}_1) + k_2 \times \ln(\text{size descriptor}_2) + \ln(\varepsilon) \qquad \textbf{(1.4)}$$

$$\ln(y) = k_1 \times \ln(\text{size descriptor}_1) + k_2 \times \ln(\text{size descriptor}_2) + k_3 \times \ln(\text{size descriptor}_3) + a + b_1 \cdot (\text{chronovarible}_1) + \ln \varepsilon \qquad \textbf{(1.5)}$$

Theoretical exponents

Static or phylogenetic allometric coefficients describe the dimensional relationship between physiological parameter and size descriptors among participants in the same stage of development (Beunen, Rogers, Woynarowska, & Malina, 1997). Standardized allomeric coefficients were proposed to normalize LVM (Lang et al., 2015) or VO_{2peak} (Kleiber & Rogers, 1961), but this topic received consideration in a recent systematic review (Lolli, Batterham, Weston, & Atkinson, 2017). Overall, the calculation of standardized coefficients seems to be related with large sample sizes. Given the sample-specificity of the *k* exponent, the application of a theoretical exponents value should not be adopted (Welsman & Armstrong, 2008). Afterwards, studies involving children and adolescents need to detail allometric modelling procedures.

Note, relevant authors of pediatric exercise sciences (Bar-Or and Thomas Roland) explored the advantages of power function ratios. They stated "*ratio scaling is not the ideal way to compare the maximal aerobic power of people who differ in body size" (13, p. 6) but noted that as, "the most convenient and traditionally accepted way a majority of studies still express maximal O2 uptake per kilogram body mass" (13, p. 7)*". In the present time, most of the studies normalized physiological parameters using simple ratios because they are simple to be applied (Welsman &Armstrong, 2019).

1.6. Rationale, objective and outline of the thesis

The literature about growth and maturation among males is mainly focused in soccer players (Malina et al., 2000; Coelho-e-Silva et al., 2010b; Valente-dos-Santos et al., 2015; Ostojic et al., 2014; Malina et al., 2015; Teixeira et al., 2018; Hill, Scott, Malina, McGee, & Cumming, 2020; Parr et al., 2020). Obviously, other sports have received consideration, such as basketball (Coelho-e-Silva, Figueiredo, Carvalho, & Malina; 2008; Coelho-e-Silva et al., 2010a; Carvalho et al., 2011; Martinho et al., 2021), tennis (Myburgh et al., 2016), table tennis (Coelho e Silva et al., 2021), judo (Detanico, Kons, Fukuda, & Teixeira, 2020), and roller-hockey (Coelho-E-Silva et al., 2012; Valente-dos-Santos et al., 2013a; Valente-dos-Santos et al., 2013b).

The three common methods to estimate SA were adopted in male soccer players. For example, TW2 RUS was applied in players from Belgium (Duarte et al., 2018), TW3 RUS was used among 55 elite young Serbian players (Ostojic et al., 2014), SA of elite players from a regional academy in France was obtained with GP protocol (Carling, Le Gall, & Malina, 2012), and, finally, Fels method was often reported in Portuguese male soccer players (Malina et al., 2000; Figueiredo, Gonçalves, Coelho E Silva, & Malina, 2009; Coelho-e-Silva et al., 2010b). Moreover, secondary sex characteristics and two non-invasive estimations (i.e., predicted age at PHV and %PMS) were also used in studies about young soccer players (Malina et al., 2004b; Malina, Coelho E Silva, Figueiredo, Carling, & Beunen, 2012; Hill et al., 2020). Unfortunately, studies among female soccer players are lacking.

Simple ratios fail to create a size-free expression and, consequently, they provide spurious results. As previously mentioned, allometric scaling approach facilitates the construction of appropriately size-adjusted expressions. In the meantime, the best denominator to normalize LVM is still under discussion, particularly during adolescence, when significant changes in size and composition are evident. Among 73 roller hockey players aged 14.5 to 16.5 years old, the sitting height and FFM were the best size descriptors to explain inter-individual variability in LVM. When two size descriptors were included in the model, in addition to a chronovariable (i.e. CA or SA), biological maturation estimated by Fels method was not a significant predictor. Even the explained variance of LVM increased comparing to the simple models (Valente-dos-Santos et al., 2013a). Comparable results were noted among male non-sport participants aged 11.15-15.12 years old (Valente-dos-Santos et al., 2014). Meanwhile, allometric coefficients differed between the two studies. The slopes between LVM and body size descriptors were 2.64, 2.49, 0.76 and 0.22 for stature, sitting stature, FFM and fat mass (FM) among roller hockey players, while in school adolescents the respective coefficients were 2.33, 2.18, 0.80 and 0.17. These results confirm the necessity to calculate specific coefficients for each sample and it seems central not generalize the coefficients for males and females.

The adolescent growth spurt differentially impacts adult stature and mass in males and females. Maximal growth spurt occurs, on average, two years earlier in females than in males and it is less intense in girls. PHV seems to have a marked impact on sex differences in body mass and composition. Among males, a linear increase in metabolic active tissues (i.e., FFM) from childhood to adulthood was noted among boys during adolescence, while in contrast, the increase in FFM reaches a plateau in girls, in addition to a linear increase in FM (Malina et al., 2004). Nevertheless, TW method also considered that males and females attain skeletal maturity at different timings. The most recent version of TW protocol defined the adult status at 16.5 and 15.0 years old, in males and females, respectively (Tanner-Whitehouse et al., 2001).

With this in mind, generalizations of youth males involved in organized sport should not be applied for females. Moreover, research dealing with the female soccer players is limited and does not systematically consider variation associated with inter-individual differences in growth, biological maturation and training.

In summary, the aims of this thesis are to describe the growth and maturity status of female youth soccer players, to examine the agreement between concurrent protocols to estimate SA, and, finally, to create allomeric models to explain inter-individual variability in LVM. The thesis is presented considering the five manuscripts which address a specific component of the overall purpose of the study.

Chapter II describes a comprehensive characterization of the sample included in the five studies as well as the materials, methods and analysis.

In Chapter III, based on an organized search, a narrative review about growth and maturity status of female soccer players was initially prepared. Manuscripts that reported height, weight and/or an indicator of biological maturation were considered. Studies spanning the years 1992-2020 and, consequently, secular trends in height and weight were examined considering two different periods.

Given the lack of details provided by some authors that frequently used the GP method, Chapter IV examines the reproducibility and inter-observer agreement of the respective protocol. In Chapter III only one study reported SA obtained by GP protocol, which, considering the preceding, is central to access data quality.

The agreement between concurrent protocols is mainly to focus on male soccer players (Malina, Chamorro, Serratosa & Morate, 2007; Malina et al., 2018). Among soccer players, TW3 RUS tended to be systematically lower than Fels and TW2 RUS. In Chapter V, the comparison between Fels and GP methods was tested among female soccer players.

In the Chapter VI, growth and skeletal maturity status (obtained by Fels method) of female Portuguese soccer were examined. Furthermore, maturity-associated variation in body size was analyzed.

Finally, simple and multiplicative allometric models for scaling LVM are presented; and, in addition, the independent contribution of body size descriptors as well, combined with the effects of biological maturation (assessed by the Fels method), to explain variability of LVM among female soccer players, are explored in Chapter VII.

Chapter VII includes the general discussion in which the findings of the various are summarized.

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Chapter II

Methods

2. Methods

2.1. Study design and sampling

The five studies presented in the current thesis were partially supported by the Portuguese Foundation for Science and Technology (SFRH/BD/121441/2016). The data collection is linked with a signed protocol between the University of Coimbra and the Portuguese Institute of Sports and Youth (IPDJ/FCDEF.UC/2017–01). The research was conducted in accordance with ethical procedures of the Declaration of Helsinki for human studies by the World Medical Association (2013) as well as in accordance with ethical standards for sports medicine (Harris, MacSween, & Atkinson, 2019).

Female soccer players were recruited from clubs affiliated to the Portuguese Soccer Federation. Data was collected at the Porto Sports Medicine Center as part of medical examinations required for the registration in the Federation (Law 204/2006; act 11/2012). Participants or legal guardians were informed about the objectives, methodological procedures, risks and, afterwards, they provided informed consent. Participation was voluntary and participants could withdraw the study at any time. Table 2.1 summarizes the characteristics (design, sampling and variables) of each study.

Table 2.1. Basic characteristics of each study.

Study	Design	Characteristics	Age	Studied variables
1	Review	161 studies 293 observations	9.0-18.0 years	stature, body mass, biological maturation
2	Cross–sectional	100 female soccer players	12.0–16.7 years	skeletal age assessed by Greulich-Pyle protocol
3	Cross–sectional	441 female soccer players	10.1–16.7 years	skeletal age assessed by Greulich-Pyle and Fels protocols
4	Cross–sectional	441 female soccer players	10.1–16.7 years	stature, body mass, fat mass, fat-free mass, skeletal age assessed by Fels protocol
5	Cross–sectional	228 female soccer players	11.8–17.1 years	stature, body mass, fat mass, fat-free mass, training experience, left ventricular mass

2.2. Anthropometry

Anthropometric measurements were collected by a single experienced observer following standardized procedures (Lohman, Roche, & Martorell, 1988). Stature was measured to the nearest 0.1 cm using a stadiometer (model 98.603, Holtain Limited Crosswell, Crymych, UK) and body mass was measured to the nearest 0.1 kg using a digital scale (SECA, model 770, Hanover, MD, USA), respectively. The body mass index (BMI) was calculated and plotted as a BMI-for-age z-score (BMIz) for a reference population (Kuczmarski et al., 2002).

Skinfolds measured using a Lange caliper (Beta Technology Incorporated Cambridge, Maryland, USA) at two sites, triceps and medial calf, were used to estimate fat mass percentage (%FM) from a sex and age-specific equation (Slaugther et al., 1988). The equation was developed in participants aged 8-29 years old from Illinois and Arizona based on a multicompartmental model. Body density and body water were estimated by underwater weighing and deuterium oxide dilution, respectively. Additionally, bone mineral content was measured using a photon absorptiometry technique on the upper limbs (i.e. radius and ulna). Nine skinfolds (triceps, biceps, subscapula, midaxillary, suprailiac, anterior suprailiac, abdominal, mid-thigh and medial calf) were obtained from each participant and considered for the multiplicative models. Then, %FM was predicted following the equation 2.1 developed for females and, consequently, absolute FM and FFM were derived.

$$\%\ \text{FM} = 0.610 \times (\text{triceps skinfold} + \text{medial calf skinfold}) + 5.1 \qquad \textbf{(2.1)}$$

2.3. Biological maturation

Sexual maturation

Among females, studies that reported secondary sex characteristics and age at menarche were extracted for Study 1. Assessments of secondary sex characteristics according to the adapted protocol of Tanner (1962) were considered for the review. Age at menarche (an indicator of maturity timing) and frequencies of players by menarcheal status (yes/no) were examined for Study 1.

Somatic maturation

Age at peak height velocity (PHV) is an indicator of maturity timing. However, it requires longitudinal data. Based on 293 data-points extracted from literature, Preece-Baine model was applied to estimate growth parameters (Preece & Baines, 1978). The model is based on a multiplicative exponential logistic equation and respective derivative function enables to obtain the velocity of specific growth landmarks. The main variables obtained from this analysis are: age at PHV (years), maximal height during peak height velocity (cm), maximum velocity of growth (cm per year), percentage of mature stature (% PMS) at PHV, and standard error of estimate (SEE) associated with the model (Hauspie & Molinari, 2004). Longitudinal studies among females are lacking and, for this reason, non-invasive estimations of maturity timing or status have been applied in studies of female soccer players as shown in Study 1.

Firstly, the algorithm based on Canada and Belgium longitudinal data (Mirwald, Baxter-Jones, Bailey, & Beunen, 2002) was frequently adopted to predict the time before or after maximal growth spurt (expressed in years) and it is known as maturity-offset (equation 2.2 is specific for females):

$$\text{maturity offset (years)} = -9.376 + (0.0001882 \times (\text{leg length} \times \text{sitting height})) + 0.0022 \times (\text{age} \times \text{leg length}) + (0.005841 \times (\text{age} \times \text{sitting height})) - ((0.002658 \times (\text{age} \times \text{mass})) + (0.07693 \times (\text{mass-by-stature ratio} \times 100)) \qquad \mathbf{(2.2)}$$

The need to multiply body mass/stature by 100 was not reported in the original publication. Predicted age at PHV (years) was calculated as the difference between chronological age (CA) and offset. The SEE of predictive model was 0.569. Secondly, predicted adult or mature stature (%PMS) was calculated based on CA, current stature, body mass and midparent stature (Khamis & Roche, 1994). Age-specific beta coefficients were developed from the sample of Fels Longitudinal Study. The SEE for female participants was 1.67 cm. Then, current stature was expressed as a %PMS attained as an indicator of maturity timing (equations 2.3 and 2.4):

Adult stature = $\text{ß}_{0} + (\text{ß}_{1\text{stature}} \times \text{stature}) + (\text{ß}_{2\text{body mass}} \times \text{body mass}) + (\text{ß}_{3\text{midparent stature}} \times \text{midparent stature})$ **(2.3)**

% PMS = $(\text{current stature} \div \text{adult stature}) \times 100$ **(2.4)**

Both indicators also have been used to group children adolescents that participate in organized sport by maturity status. For example, maturity-offset (time before or after PHV) was used in 157 female soccer players (Emmonds, Morris, Murray, Robinson, Turner, & Jones, 2017) to classify as pre-PHV (offset <-1 years), circa-PHV ($\leq\pm1$ years) or post-PHV (offset $>+1$ years). Additionally, maturity-bands based on % PMS were used to organize players in maturational categories. Considering 138 adolescent female soccer players, pre-pubertal participants were classified as pre-pubertal < 87% adult stature, pubertal from 87 to 94% adult stature and post-pubertal as > 94% adult stature (Westbrook, Taylor, Nguyen, Paterno, & Ford, 2020).

Skeletal maturation

Studies reported that skeletal age (SA) or skeletal maturity status were considered for Study 1. Details regarding Greulich-Pyle (GP) and Fels protocols were documented in Studies 2, 3 and 4. Finally, to explain left ventricular mass (LVM), the predictive models included the skeletal maturity status, assessed by Fels method, as a dummy variable (Chapter VII).

SA was assessed by a posterior-anterior radiograph of the left hand and wrist. This technique exposes the participants to a minimal dose of radiation, which is equivalent to watching TV for 3 hours every day (Malina, Rogol, Cumming, Coelho e Silva, & Figueiredo, 2015).

GP requires the assessment of SA 30 bones of hand and wrist. SAs were assigned by comparing individually each bone of the radiograph with the standards included in the original Atlas (Greulich & Pyle, 1959). The details to calculate an overall SA for each participant are not totally clear. Hence, different estimations of SA were obtained following the instructions of relevant authors (Roche & Johnson, 1969; Malina, Bouchard, & Bar-Or, 2004). However, GP method is often applied clinically by comparing the radiograph as a whole to the illustrations presented on the Atlas. (Malina et al., 2015) An additional question is the lack of details when GP method is applied. In summary, GP SAs were calculated using the mean, median and when the radiograph was examined as a whole. The mean and median were also obtained excluding the mature bones.

The Fels protocol (Roche, Chumlea, & Thissen, 1988) was also applied to assess SA. It requires grades of bone-specific maturity indicators for the radius, ulna, carpals (i.e. capitate, hamate, triquetral, lunate, scaphoid, trapezium and trapezoid), metacarpals and phalanges of the first, third and fifth rays. The absence or presence of pisiform and adductor sesamoid is observed. Additionally, linear widths of epiphysis and metaphysis of long and short bones are measured. Then, grades are computed into a software (Felshw 1.0 Software, Lifespan Health Research Center, Departments of Community Health and Pediatrics, Boonshoft School of Medicine, Wright State University, Dayton, Ohio). The method weights the grades according to age and sex and consequently a SA and associated-standard error are provided. Note, the error associated with the assessment is an exclusive characteristic of this protocol and it systematically tends to increase with age (Roche et al., 1988). For example, among Portuguese male soccer players, the standard error ranged at 11–12 years: 0.28– 0.32; 13–14 years: 0.28–0.51; 15–17 years: 0.28–0.72 (Malina et al., 2000). The same trend was noted among male Portuguese and Polish table tennis players (Coelho-e-Silva et al., 2021). Standard errors tend to increase with age, particularly, as the adult status is near to be attained; this reflects the progressive reduction in the number of bone indicators available for SA assessment.

Considering the SAs of GP and Fels methods, players were grouped into maturity categories: late or delayed, average or on time, early or advanced (Malina, 2011). This classification was based on the difference between SA and CA. A band of ±1.0 year is commonly used in studies of young athletes (Malina, 2011). In this context, late maturing was defined as SA < CA by one year or more; on time or average maturing was defined as players with SA within ±1.0-year band of CA; early maturing was defined as SA above 1.0 year of CA. SA is not assigned if a player was classified as skeletally mature. The interval of 1.0 year approximates the standard deviation (SD) of SA within CA groups among adolescent females included in the Fels Longitudinal Study (Roche et al., 1988; Malina, 2011).

2.4. Echocardiography

Resting echocardiography was conducted using Vivid 3 ultrasound machine with a 1.5 to 3.6 MHz transducer (GE Vingmed Ultrasound, Horten, Norway). Based on two-dimensional images (recorded at 100mm/s), M-mode echocardiograms were derived for direct visualization. Measurements of the internal dimension of the left ventricle at end diastole (LVIDd), septal wall thickness at end diastole (SWTd), and posterior wall thickness at end diastole (PWTd) considered. Data quality for LVIDd was 0.17mm (95% LOA, 1.95–2.28 mm, %CV = 0.3, 95% LOA: 4.1–4.8%); for SWTd, 0.02mm (95% LOA 0.30–0.34 mm, %CV = 0.3, 95% LOA, 4.2–4.8%); and for PWTd was 0.06mm (95% LOA, 0.45–0.56 mm, %CV = 0.8, 95% LOA, 6.5– 8.1%).

2.5. Statistical analyses

Data analysis was executed according to the specific goals of each study (Table 2.2). Statistical procedures were done using IBM SPSS version 23.0 software (SPSS, Inc., IBM Company; NY, USA), GraphPad Software, San Diego California USA, www.graphpad.com), and Dell Statistica 13 version 13. (Dell, Inc. Data Analysis Software System, software.dell.com). Significance level was kept at 0.05.

Table 2.2. Statistical analysis used in each study.

Analyses	Study				
	1	2	3	4	5
Preece-Baines model 1	●				
Analysis of variance	●			●	
Analysis of co-variance	●				
Bland-Altman	●				●
Agreement / disagreement		●			
Cohen-d		●		●	●
Paired samples *t*-tests		●	●		
Intra-class correlation coefficient		●			
Correlation coefficient			●		●
Kappa coefficient			●		
Lin concordance correlation coefficient			●		
Independent t-test				●	
Effect size correlation	●			●	
Kolmogorov-Smirnov					●
Simple linear regression					●
Multiple linear regression					●
Homoscedasticity of residuals					●

2.6. References

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Chapter III

Study 1

Growth and Maturity Status of Female Soccer Players:
A Narrative Review

3. Growth and Maturity Status of Female Soccer Players: A Narrative Review

3.1. Abstract

Reported mean ages, heights and weights of female soccer players aged <19 years in 161 studies spanning the years 1992–2020 were extracted from the literature or calculated from data available to the authors; 35 studies spanning the years 1981–2020 also included an indicator of biological maturation. Heights and weights were plotted relative to U.S. reference data. Preece–Baines Model 1 was fitted to moving averages to estimate ages at peak velocity. Maturity indicators included skeletal age, pubertal status, age at menarche, percentage of predicted adult height and predicted maturity offset. Heights and weights showed negligible secular variation across the time interval. Heights were slightly above or approximated the reference medians through 14 years old and then varied between the medians and 75th percentiles through 18 years old. Weights were above the reference medians from 9 to 18 years old. Mean ages at menarche ranged from 12.7 to 13.0 years. The trend in heights and weights suggested the persistence and/or selection of taller and heavier players during adolescence, while estimated age at peak height velocity (PHV) and ages at menarche were within the range of mean ages in European and North American samples. Data for skeletal and sexual maturity status were limited; predicted maturity offset increased linearly with mean ages and heights at prediction.

Keywords: Height; Weight; Youth Athletes; Puberty; Maturity Offset; Predicted Adult Height

3.2. Introduction

An early review of the growth and maturity status of youth athletes (Malina, 1994) indicated only two studies of female soccer players. One considered skeletal age (SA) among adolescent female athletes in several sports, including soccer (Novonty, 1981), while the other described the somatotypes of 50 female athletes, including six soccer players, aged 10–15 years (Liu, Plowman, & Wells, 1989). This is somewhat surprising given the rapid increase in the number of girls participating in soccer in both Europe and North America since the 1970s (National Federation of State High School Associations, 2019; U.S. Youth Soccer, 2014; UEFA, 2017). In contrast, injuries in youth soccer tournaments have been of interest and studies have noted higher injury rates among females than in males (Nilsson & Roaas, 1978; Schmidt-Olsen, Bünemann, Lade, & Brassøe, 1985; Andreasen, Faunø, Lund, Lemche, & Knudsen, 1992; Kibler, 1993). These studies focused on injury statistics and types of injuries; growth and maturity were not considered.

An early review of the physiological and physical characteristics of adult female soccer players included six studies between 1986 and 1992 that reported heights and weights (Davies & Brewer, 1993). Subsequent reviews (Datson et al., 2014; Martínez-Lagunas, Niessen, & Hartmann, 2014) noted an increase in studies of body size and composition and the functional characteristics of adult players, but one (Martínez-Lagunas et al., 2014) included eight studies reporting age, height, weight and/or the functional characteristics of players aged ≤18 years, while the other (Datson et al., 2014) noted that “ …the anthropometric profile of elite youth players has yet to be fully examined” (p. 5). In contrast, a systematic review of the reliability and utility of change of direction speed tests among female adolescent soccer players did not consider potential variation in test performances associated with chronological age (CA) per se (mean CAs of the samples ranged from 9.3 to 17.3 years) and with growth and maturity status (Pardos-Mainer, Casajus, Julian, Bishop, & Gonzalo-Skok, 2020).

Although variation in growth and maturation is indicated as central to the long-term development (LTAD) of female youth players (The Football Association, 2020), it is essential to recognize the interactions among CA, growth and maturity status, and the functional development of youth players.

Given the increased popularity of soccer among girls and the selectivity of sport and competition for potentially talented players at relatively young ages, the purpose of this narrative review is to evaluate the growth and maturity status of female youth soccer players. Studies reporting heights and weights spanned 1992 through 2020, and, except for a study in 1981, studies reporting maturity indicators spanned 1995 through 2020.

3.3. Material and methods

Search methods

Studies of female soccer players aged <19 years were extracted from several databases (Pubmed, Web of Knowledge and Scopus) using the following terms:" youth female soccer", "youth soccer female", "young soccer female", "soccer female" and "female soccer". In addition to age, height and weight, the search also identified studies that included an indicator of biological maturation. Initial screening was done by title and abstract, and when needed the full manuscript was consulted. If the data combined players from multiple sports or combined males and females, the manuscript was excluded. All materials were reviewed by two individuals (D.V.M. and R.M.M.), but the final decision was made by the first author. The extracted studies were complemented with papers on female athletes accumulated over the years by the first author.

Age, height and weight

Mean CAs, heights and weights were extracted from 155 full-text articles spanning 1992 through November 2020. In addition, raw data for CAs, heights and weights were available to the first author from six studies: three theses dealing with youth players, a study of collegiate athletes that included a sample of soccer players aged 18 years and two surveys of adolescent participants in recent bio-banded soccer tournaments. Age-group-specific means were calculated for each of the six data sets. Thus, 161 studies were deemed eligible. The studies are indicated in chronological order in Supplementary Table S1. Several studies reported the same age, height and weight data in one or more reports. The earliest report was retained for this review; the others are also indicated the Supplementary Table S1.

The 161 studies included 293 observations (age-group means) for the age, height and weight of female youth players spanning 8.2 to 18.9 years old. The majority of studies and data points were, respectively, from Europe (75 and 140) and North America (61 and 106), followed by Latin America (10 and 20), Asia-India (9 and 18), the Pacific (3 and 6) and Africa (3 and 3). The U.S., England, Spain and Canada accounted for 58% of the studies and 60% of the data points. The median sample size was 20 with a range from 6 to 4556, but sample sizes ranged from 10 to 75 for 85% of the data points. The largest sample included youth players from 230 clubs participating in a prospective study of knee injuries (Hägglund & Walden, 2016).

Prior to considering the growth status of the female players, two preliminary analyses were done. The first considered potential secular variation in body size. For the analysis of secular change, studies were grouped into three time intervals, 1992 to 2009 (27 studies, 49 data points), 2010 to 2017 (63 studies, 114 data points) and 2018 to 2020 (71 studies, 130 data points). Within each interval, composite means of the mean CAs, heights and weights were also calculated for several CA groups (<11, 11–12, 13–14, 15–16 and 17–18 years) and compared within each CA group using analyses of variance (ANOVAs). Reported means were not adjusted for sample size to avoid the risk of age-adjusted means being influenced by few studies with large samples.

The mean heights and weights from each study in the three intervals were also plotted by CA relative to U.S. reference medians and the 25th and 75th percentiles for girls (Kuczmarski et al., 2002). The reference was developed from nationally representative samples of American girls of European, African-American and Hispanic-American ancestry surveyed between 1963 and 1994, but weights increased from the late 1970s to the early 1990s. As a result, the body weights of children and youth aged ≥6 years in the 1988–1994 national survey were not used in the development of the charts (Roche, 1999). This was done because the gain in weight was considered undesirable from a public health perspective and to avoid an upward shift of the percentile curves for weight and BMI. Changes in heights of American girls and boys have been relatively small across subsequent U.S. national surveys through 2014, but weights have increased over time (McDowell, Fryar, Ogden, & Flegal, 2008; Fryar, Gu, & Ogden, 2012; Fryar, Gu, Ogden, & Flegal, 2016).

Since sport is selective (Malina, 1994), a second preliminary analysis addressed potential size variation by competitive level. Players aged >11 years were classified as local or elite based on descriptions in the respective studies. Classifying players aged 8–10 years as elite is not warranted (Kirland & O´Sullivan, 2018). Age, height and weight of local and elite players were compared by two-year CA groups with analyses of covariance (ANCOVA), with year of study as the covariate. All data were then combined, and composite means of the mean CAs, heights and weights were calculated by single-year CA groups from 9 to 18 years; the single 8-yearold sample was included with the 9-year-olds. Mean heights and weights were plotted by CA relative to U.S. reference medians and the 25th and 75th percentiles for girls (Kuczmarski et al., 2002). Allowing for limited numbers at the younger ages moving averages, with a window of three measurements, of mean heights and weights were also calculated and fitted with Preece–Baines Model 1 (Preece & Baines, 1978; Hauspie & Roelarts, 2012) to estimate ages at peak height velocity (PHV) and peak weight velocity (PWV) using Dell Statistica 13 (2016). Although designed for longitudinal data, the Preece–Baines model has been applied to cross-sectional data (Zemel & Johnston, 1994).

Maturation

Skeletal age (SA) and secondary sex characteristics (breasts, pubic hair) are established indicators of maturity status—state of maturation at the time of observation. Both are often labeled as “invasive” as the former requires a small dose of radiation and the latter is often considered as intruding an adolescent’s privacy. Ages at menarche and at PHV are indicators of maturity timing—ages at which specific maturational event are attained. Both indicators of timing require longitudinal data that span adolescence (Malina, Bouchard, & Bar-Or, 2004). Unfortunately, longitudinal data for female soccer players that span adolescence are not available.

In this context, non-invasive predictions of maturity status and timing are increasingly used in studies of youth athletes (Malina, 2014). The use of current height expressed as a percentage of predicted adult height without SA was proposed as an indicator of maturity status in the 1980s (Roche, Tyleshevski, & Rogers, 1983) and sex-specific equations for the prediction of adult height based on the age, height and weight of the individual and the mid-parent height of her/his biological parents were developed (Khamis & Roche, 1994). The latter equations are often used in the context of bio-banding (Malina et al., 2019). As commonly applied, the heights and weights of players are measured, while the parental heights are reported. Sex-specific equations for the prediction of maturity offset, defined as time before PHV (Mirwald, Baxter-Jones, Bailey, & Beunen, 2002), are also increasingly used. The equations require CA, height, weight, sitting height and estimated leg length. Modified prediction equations using age and height in girls and age and sitting height or age and height in boys are also available (Moore et al., 2015). By definition, predicted maturity offset is an indicator of maturity timing; predicted age at PHV is estimated as the difference between CA at prediction and maturity offset.

The earliest study of maturity status (skeletal age) in female soccer players was published in 1981 (Novotny, 1981), while an indicator of maturity was reported in 36 studies (22%) spanning 1995 to 2020. The use of established maturity indicators, however, was limited (Supplementary Table S1). Only one recent study in 2020 considered SA, while pubertal status was evaluated in nine studies spanning 2010 to 2019. The ages at menarche, with one exception, were based on the retrospective (recall) method in seven studies of adolescents and young adults spanning 1995 to 2017. Predicted maturity offset was used in 16 studies spanning 2012 to 2020, while the percentage of predicted adult height attained at the time of observation was used in three studies: 2002, 2018 and 2020.

3.4. Results

Height and Weight

The mean heights of soccer players in studies spanning the years 1992–2009, 2010–2017 and 2018–2020 overlapped and spanned the 25th to 75th percentiles of the U.S. reference (Supplementary Figure S1), although most means were at or above the reference medians at 15–18 years. The trend was similar for body weight, but most means in the three intervals were at or above the reference medians beginning at about 12–13 years (Supplementary Figure S2). The ANOVAs indicated only one significant difference among the CAs, heights and weights of players in the three time intervals (Table 3.1). Players 13–14 years of age in 1992–2009 were significantly heavier than players in 2010–2017 ($p < 0.05$), but the effect size was small ($\eta p^2 = 0.07$). By inference, secular changes in heights and weights of female soccer players were negligible between 1992 and 2020. The ages, heights and weights of players aged 13–14 and 17–18 years classified as elite or local did not differ (Table 3.2), while local players aged 11–12 years were significantly taller and heavier than elite players ($p < 0.05$) and elite players aged 15–16 years were significantly taller than local players ($p < 0.05$). The effect sizes (ηp^2), 0.22 to 0.36, suggested a moderate effect. The mean heights of the total sample of soccer players by CA group and the estimated fit of the Preece–Baines model to the moving averages are illustrated in Figure 3.1. The mean heights were, on average, taller than the reference medians among players aged <11 years, approximated the medians through 14 years, and were above the medians through 18 years. The estimated age at PHV based on the Preece–Baines model was 11.55 years with a standard error of estimate (SEE) of 0.56 cm. The latter is an estimate of the goodness of fit of the model. The corresponding trends for body weight are shown in Figure 3.2. The weights of soccer players were consistently above the reference medians across the age range. The estimated age at PWV was 12.22 years with an SEE of 0.67 kg.

Table 3.1. Number of data points (N) †, means (M) and standard deviations (SD) based on the means for chronological age (CA), height and weight of female soccer players by two-year age groups in studies spanning three intervals, 1992 to 2009, 2010 to 2017 and 2018 to 2020, and results of ANOVAs and effect sizes (η_p^2).

	Intervals of Studies											
Age	**1992–2009 (a)**			**2010–2017 (b)**			**2018–2020 (c)**					**Significant**
Groups	**N**	**M**	**SD**	**N**	**M**	**SD**	**N**	**M**	**SD**	**F**	**η_p^2**	**Differences**
CA, years												
<11	5	10.1	0.3	9	9.9	0.8	6	10.4	0.3	1.62	0.16	
11–12	7	12.0	0.4	17	12.1	0.5	29	12.2	0.6	0.51	0.02	
13–14	11	14.2	0.6	38	14.1	0.5	47	13.9	0.7	0.97	0.02	
15–16	18	15.8	0.6	35	15.8	0.6	42	15.9	0.6	0.58	0.01	
17–18	8	17.8	0.4	15	17.9	0.6	6	17.8	0.7	0.04	<0.01	
HEIGHT, cm												
<11		142.8	3.6		140.1	3.4		143.2	4.5	1.51	0.15	
11–12		153.3	2.1		152.2	3.7		153.4	5.5	0.36	0.01	
13–14		162.0	3.2		160.5	3.4		160.5	2.5	1.30	0.03	
15–16		164.0	2.1		164.2	2.1		164.6	2.5	0.31	0.01	
17–18		164.9	2.5		166.4	2.8		165.1	3.6	0.87	0.06	
WEIGHT, kg												
<11		36.9	1.5		35.1	3.0		36.4	3.1	0.77	0.08	
11–12		46.1	3.8		44.2	4.8		45.3	5.5	0.39	0.01	
13–14		55.6	3.4		52.5	3.7		52.9	3.5	3.44 *	0.07	a > b
15–16		57.5	2.2		57.3	2.6		58.1	2.6	0.92	0.02	
17–18		60.5	2.5		60.5	2.9		59.6	3.2	0.24	0.02	

† data points are the means for mean ages, heights and weights reported in the available studies or calculated from data available for six studies; * $p < 0.05$.

Table 3.2. Number of data points (N), means (M) and standard deviations (SD) based on means for the age, height and weight of female soccer players classified as local or elite in four chronological age groups, results of age-group-specific ANCOVAs controlling for the year of study, and effect sizes.

Age Groups		**Local**			**Elite**			
	N	**M**	**SD**	**N**	**M**	**SD**	**F**	**η_p^2**
AGE, years								
11–12	32	12.2	0.5	21	12.0	0.6	2.31	0.04
13–14	56	14.1	0.6	40	14.0	0.6	0.96	0.03
15–16	40	15.8	0.6	55	15.8	0.6	0.01	<0.01
17–18	6	18.0	0.6	23	17.8	0.6	0.37	0.01
HEIGHT, cm								
11–12		154.1	3.5		151.2	5.6	6.17 *	0.11
13–14		160.6	3.0		160.9	2.9	3.22	0.03
15–16		163.7	2.0		164.7	2.4	4.33 *	0.05
17–18		164.8	3.4		166.0	2.8	2.40	0.08
WEIGHT, kg								
11–12		46.4	4.7		42.8	4.9	7.27 *	0.13
13–14		53.4	3.9		52.5	3.2	2.49	0.03
15–16		57.3	1.8		58.0	2.9	1.29	0.01
17–18		60.5	3.5		60.3	2.7	1.36	0.05

* $p < 0.05$.

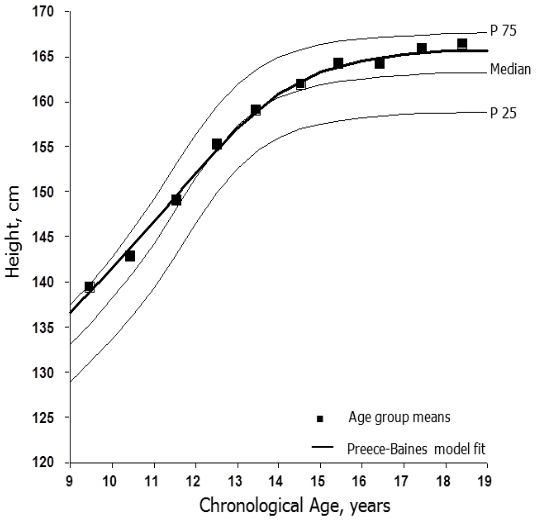

Figure 3.1. Age-group means for heights of female soccer players (■) and the fit of the Preece-Baines Model 1 (solid line) applied to the moving averages of mean heights plotted relative to reference medians and the 25th and 75th percentiles for U.S. girls.

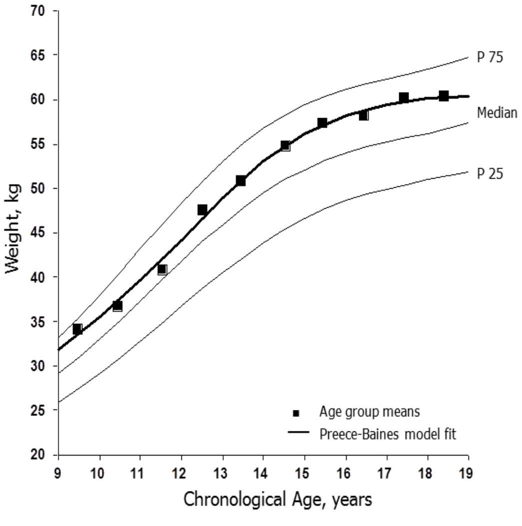

Figure 3.2. Age-group means for weights of female soccer players (■) and the fit of the Preece–Baines Model 1 (solid line) applied to the moving averages of mean heights plotted relative to reference medians and the 25th and 75th percentiles for U.S. girls.

Skeletal age (SA)

Two studies, separated by 40 years, considered the skeletal maturity of female soccer players. The earlier study (Novotny, 1981) involved 23 players observed at 13 and 16 years of age and used a local adaptation (Kapalin & Picko, 1964) of the Greulich–Pyle (GP) method (Greulich & Pyle, 1959) to estimate SA. CAs and SAs of the soccer players were, on average, similar at both ages (Novotny, 1981). The more recent study of a large sample of Portuguese players indicated Greulich–Pyle SAs that were, on average, in advance of CAs at 12 and 13 years, equivalent at 14 years and slightly delayed at 15 and 16 years; when assessed with the Fels method (Roche, Chumlea, & Thissen, 1988), the SAs were advanced, on average, relative to CAs from 12 to 16 years (Martinho et al., 2021).

Pubertal Status

Nine studies evaluated pubertal status. The status was assessed at a clinical examination in one study (Nicolao, Pedrinelli, Martino Zogalb, Orbetelli, & Lelte de Barros Neto, 2010) while self-assessments of stage of breast (Unnithan, Roche, Garrard, Holloway, & Marwood, 2015) or pubic hair (PH) (Sigward, Pollard, Havens, & Powers, 2012) development or of “Tanner stages” (Lozano-Berges et al., 2017; Lozano-Berges et al., 2019a; Lozano-Berges et al., 2019b; Plaza-Carmona et al., 2016; Ubago-Guisado, Gómez-Cabello, Sánchez-Sánchez, García-Unanue, & Gallardo, 2015) were used in seven studies. The latter did not differentiate between breasts and PH. The samples spanned variable CA ranges: 8–14, 9–13, 9–18, 11–14, 12–15 and 13–15 years. Players were generally classified as pre-pubertal, pubertal or post-pubertal (though procedures and labels varied) or the distributions of stages were simply described for the sample. The variation in pubertal status by CA or variation in CA within and between pubertal groups was not addressed. It is thus difficult to ascertain the sexual maturity status of the samples relative to a reference for the general population. One study used self- and parental-assessments of pubertal status to identify pre-pubertal (9.9 ± 0.3 years) and post-pubertal (14.6 ± 0.5 years) players (Lyle, Sigward, Tsai, Pollard, & Powers, 2011).

Age at Menarche

Seven studies (Siegel, 1995; Düppe, Gärdsell, Johnell, & Ornstein, 1996; Casper, Michaels, & Simon, 1997; Petterson, Nordström, Alfredson, Henriksson-Larsén, & Lorentzon, 2000; Södermann, Bergström, Lorentzon, & Alfredson, 2000; Prather et al., 2016; Brännström et al., 2017) reported ages at menarche among soccer players (Table 3.3). Only one study was focused on growth and maturity status per se (Siegel, 1995) and provided estimates of age at menarche with the status quo and retrospective methods (Malina et al., 2004). The status quo method was used with players aged 10–18 years; it required the CA of the player and whether or not she had attained menarche (yes/no). Based on probit analysis, the median age at menarche of the sample was 12.9 ± 0.9 years and was identical with the mean recalled age at menarche among adolescent and collegiate players aged 14–23 years in the study (12.9 ± 1.3 years) (Table 3.3). In one study (Prather et al., 2016), 48 of 75 players aged 10 through 14 years (64%) had not yet attained menarche, but the distribution of players by menarcheal status (yes/no) within single-year CA groups was not reported. A study of pubertal status in 35 Spanish players of 11 through 13 years of age (12.8 ± 0.8 years) noted that 20 players had already attained menarche but did not report the distribution by CA (Lozano-Berges et al., 2019).

The ages of players in studies using the retrospective method ranged from 13 to 30 years and sample sizes were small (*n* = 13–16) in three studies. The seven mean ages varied between 12.7 and 13.0 years with standard deviations of 0.7 to 1.3 years. The studies spanned 1995 to 2017 and thus indicated no evidence of secular change.

Table 3.3. Descriptive statistics for chronological age and age at menarche (mean (M) or median (Md) and standard deviation (SD)) in female soccer players; studies are listed by year of publication.

							Age at Menarche, years			
Year,				**Chronological Age, years**			**Recall**		**Status Quo**	
Reference	**Country**	**Method**	**N**	**Range**	**M**	**SD**	**M**	**SD**	**Md**	**SD**
1995 (Siegel)	US	Probit	82	10–18					12.9	0.9
		Recall	62	14–23	17.1	2.3	12.9	1.3		
1996 (Düppe et al.)	Sweden	Recall	96	13–28	18.3	4.0	12.8	1.3		
1997 (Casper et al.)	US	Recall	16		15.1	0.7	12.9	1.2		
2000 (Petterson et al.)	Sweden	Recall	15		17.6	0.8	12.7	0.7		
2000 (Södermann et al.)	Sweden	Recall	51	14–19	16.3	1.4	13.0	0.9		
2016 (Prather et al.)	US	Recall	145	15–30			13.0	1.0		
2017 (Brännström et al.)	Sweden[1]	Recall	13	13–16	15.3	0.7	13.0	0.8		

[1] The initial sample was 19 players; only 15 completed the questionnaires and, of the 15, ages at menarche were available for 13 players.

Percentage of Predicted Adult Height

Three studies used the percentage of predicted adult height attained at the time of observation as the indicator of maturity status. Maturity status so defined was used as a continuous variable among players aged 9–15 years and was unrelated to perceptions of adult autonomy support (Cumming, Battista, Standage, Ewing, & Malina, 2006). The method was used in two studies to classify players by pubertal status. Among 34 players (13.3 ± 1.5 years), nine were classified as pubertal (88–94%) and 25 as post-pubertal (>94%); the ages of players in each group were not indicated (Mullen et al. 2018). In the other study, players aged 9–17 years were classified as pre-pubertal (<87%, *n* = 17, 10.3 ± 0.6 years), pubertal (87–94%, n = 32, 11.9 ± 0.8 years) or post-pubertal (>94%, *n* = 90, 14.6 ± 1.6 years) (Westbrook, Taylor, Nguyen, Paterno, & Ford, 2020).

Predicted Maturity-Offset/ Age at PHV

Predicted maturity offset was used in 16 studies spanning the years 2012–2020. The original sex-specific equation (Mirwald et al., 2002) was used in all but one study (Supplementary Table S1). The ages of players at prediction ranged from 8 to 18 years, but the mean CAs largely spanned 11–15 years. Most studies reported predicted maturity offset, while four reported predicted age at PHV; one study reported both. Several studies compared the characteristics of players classified as pre-, at- or post-PHV (i.e., maturity status); the results for predicted offset or age at PHV were largely descriptive. The means for predicted maturity offset in soccer players increased linearly with mean CA (Figure 3.3A) and mean height (Figure 3.3B) at prediction. The mean predicted ages at PHV are shown in Figure 3.4, which also includes corresponding data for girls in the Fels Longitudinal Study for comparison (Malina, Choh, Czerwinski, & Chumlea, 2016). The predicted ages at PHV in soccer players overlapped those for the Fels sample, tended to increase with CA at prediction and were later than the observed age at PHV in the Fels sample beginning at about 10 years of age.

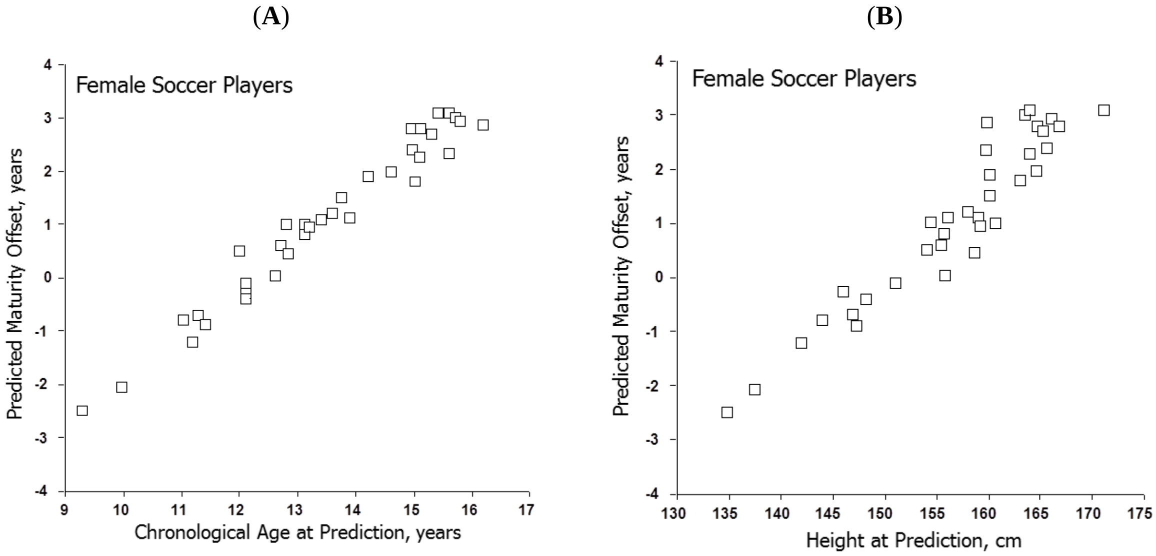

Figure 3.3. Means for predicted maturity offset (MO) in samples of female soccer players plotted relative to (**A**) mean chronological age and (**B**) mean height. References for the studies of soccer players are indicated in Supplementary Table S1: 2010–2017: [60–65]; 2018–2020: [58–68].

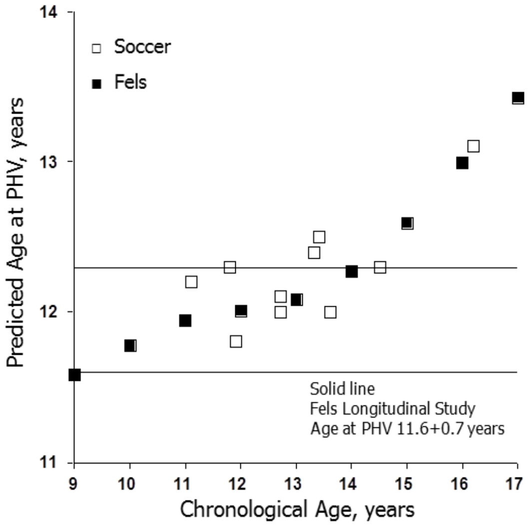

Figure 3.4. Mean predicted ages at peak height velocity (PHV) in girls aged 9 through 17 years from the Fels Longitudinal Study (Malina et al., 2016) and for female soccer players. The solid lines indicate the mean plus one standard deviation for observed age at PHV in the Fels Longitudinal Study. References for studies of soccer players are indicated in Supplementary Table S1: 2010–2017: [60]; 2018–2020: [59,64,66]; the means were calculated for select and non-select players combined in each CA group for reference [64].

3.5. Discussion

Height and Weight

The heights and weights of female soccer players showed no secular variation between 1992 and 2020 (Table 3.1). Although not directly comparable, male soccer players in 121 studies spanning the years 2000–2015 were taller and heavier than players in 23 studies spanning the years 1978–1999 (Malina, Figueiredo, & Coelho-e-Silva, 2017).

Female players aged <11 years were, on average, taller and heavier than the U.S. reference, although data were limited. The heights of the players approximated the reference medians through 14 years and were above the medians and approached the 75th percentiles in late adolescence (Figure 3.1). Body weights, in contrast, were above the reference medians throughout the age range (Figure 3.2). The trends suggested the persistence and perhaps selection of taller and heavier players during adolescence. The mean heights (158.0 to 169.3 cm) and weights (54.5 to 66.1 kg) of late adolescent players aged 17–18 years overlapped the mean heights (158.1 to 171.0 cm) and weights (55.2 to 65.7 kg) of adult soccer players aged 19–27 years surveyed between 1986 and 2020 (Supplementary Table S2).

The tallness in association with a heavier mass among late adolescent players probably reflected a more muscular physique and systematic training. Training programs aimed at the development of strength and power often enhance lean tissue mass (Malina, 2007; Malina & Geithner, 2011), while regular intensive training does not influence growth in height or the timing of the adolescent spurt (Malina & Coelho-e-Silva, 2017). Estimates of relative fatness among late adolescent soccer players were variable, likely reflecting variation among samples per se and in methodology—predictions from skinfolds, hydrostatic weighing, bioelectrical impedance and DEXA (Datson et al., 2014; Martínez-Lagunas et al., 2014; Malina & Geithner, 2011).

The estimated age at PHV based on modeling moving averages for the soccer players was 11.55 years.

Allowing for limitations associated with modeling cross-sectional data (Zemel & Johnston, 2016), the estimate was within the range of mean ages at PHV in longitudinal studies of European and North American girls (Malina et al., 2004; Beunen, Rogol, & Malina, 2006). The age at PHV occurs earlier, on average, among Japanese girls (Malina et al., 2004), although soccer players from Japan accounted for only 12 data points (5%) spanning 12–18 years of age. The estimated age at PHV for soccer players was somewhat earlier than the ages at PHV in longitudinal samples of youth of European ancestry participating in athletics, rowing and mixed sports, while ages at PHV for Japanese girls active in several sports, though not soccer, also tended to be earlier than the mean ages for athletes of European ancestry (Malina, Rogol, Cumming, Coelho-e-Silva, & Figueiredo, 2015). Consistent with observations in longitudinal samples, the estimated age at PWV of female soccer players (12.22 years) occurred, on average, after PHV, but was at the lower end of the range of mean ages noted in longitudinal samples of European and North American girls (Malina et al., 2004).

The Preece–Baines model also provided an estimate of height at PHV (149.6 cm) and young adult height (165.6 cm). The estimated height at PHV in soccer players was 90.3% of adult stature, which was similar to estimates in longitudinal studies of girls in Europe, 90.8% (Kozieł, 1998) and 91.5% (Molinari, Gasser, & Largo, 2013), and the U.S., 90.0% and 90.5% (Sanders et al., 2017). Overall, the estimated age at PHV and height at PHV as a percentage of adult height among female soccer players approximated the estimates for girls in the general population who were “on time” or average in these biological landmarks.

Skeletal maturation

Only two studies considered the skeletal maturity status of female soccer players. The earlier study observed no differences between the CA and SA of players at 13 and 16 years of age (Novotny, 1981), while the more recent study noted variation between the GP (Greulich & Pyle, 1959) and Fels (Roche et al., 1988) SAs and a tendency for soccer players to be somewhat advanced in SA relative to CA with the Fels method (Martinho et al., 2021). Comparative skeletal maturation data for female athletes in team sports are limited to the earlier study (Novotny, 1981), which noted no differences in the CA and SA of volleyball players at 14 and 17 years of age. SA data are more available for female athletes in several individual sports (Malina, 2011). Based on the difference between SA and CA, late-maturing girls were predominant during adolescence among artistic gymnasts. Although the majority of swimmers <14 years of age tended to be average in SA, equal numbers of swimmers aged 14–15 years were average or advanced in SA and the majority at 16–17 years of age were skeletally mature. Among track and field athletes, in contrast, maturity status varied by discipline (Malina, 2011), while the SAs of elite female tennis players were generally similar to the CAs among U12 players but advanced relative to the CAs among U14 and U16 players (Myburgh, Cumming, Coelho-e-Silva, Cooke, & Malina, 2016).

Pubertal status

Allowing for methodological inconsistencies, e.g., specifically combining stages of breast and PH or not specifying whether stage of breast or pubic hair was used, the studies of soccer players did not address sexual maturity status per se. Players were classified as pre-, peri- and/or post-pubertal or distributions of stages were simply described without considering variation in CA. Of relevance, Stage 1 of breast or pubic hair development is the pre-pubertal state, while Stage 2 of each characteristic marks the overt manifestation of puberty. Stages 2, 3 and 4 mark the pubertal state, while Stage 5 of breast or PH indicates the post-pubertal or mature state (Malina et al., 2004).

It is erroneous to classify players who have attained Stage 2 as pre-pubertal or players who have attained Stage 4 as post-pubertal (Nicolao et al., 2010; Lozano-Berges et al., 2019; Plaza-Carmona et al., 2016).

Age at menarche

The single estimate for youth soccer players based on the status quo method (12.9 years) was similar to the recalled ages at menarche (Table 3). The estimate was also consistent with estimates for youth athletes in several sports, including athletics, swimming and several team sports, while estimated ages at menarche for youth divers, figure skaters and artistic gymnasts were, on average, later (Malina et al., 2015).

The retrospective method requires the athlete to recall her CA at menarche. It relies on memory and may be influenced by recall bias and error, while some individuals recall only the whole year, presumably age at last birthday. Nevertheless, and allowing for small samples in three studies, mean recalled ages at menarche of soccer players spanned a narrow range (12.7 to 13.0 years) and indicated no secular effect. The means were also within the range of those for the general population (Whincup, Gilg, Odoki, Taylor, & Cook, 2001; Jull et al., 2006; Euling et al., 2008; Gohlke & Woelfle, 2009; Parent et al., 2013; Meng, Duan, Sun, & Jia, 2017) and were at the low end of recalled ages at menarche for athletes in team sports (Malina et al., 2004). In contrast, a study of adult soccer players from Kosovo reported a mean recalled age at menarche of 13.5 ± 1.3 years (Boshnjaku et al., 2016). Age at menarche was presumably obtained by questionnaire as one question asked, “Did you start exercising before menarche?” The mean for soccer players was identical to the mean ages at menarche among handball players (13.5 ± 1.0 years) and non-athletes (13.5 ± 1.0 years) (Boshnjaku et al., 2016).

Although the mean age at menarche in one sample of U.S. soccer players was 13.0 ± 1.0 years (Prather et al., 2016) and consistent with other estimates (Table 3.3), the authors noted: “This is the first study to describe a delay in the onset of menarche in a cohort of soccer athletes” (p 3). “Delay” implies that something associated with the sport influenced age at menarche. Although training is often indicated as a factor contributing to later ages at menarche in some athletes, the general consensus is that training per se does not affect menarche

(Loucks, Vaitukaitis, Cameron, Rogol, & Skrinar, 1992). Training, moreover, is only one aspect of the sport environment. Other factors include factors associated with selection, persistence and exclusion (cutting, dropout) and behaviors of coaches and trainers. The latter often place undue pressures upon young athletes in the context of diet and weight control, and extreme dietary restriction may delay menarche (Loucks et al., 1992). Age at menarche is also a heritable characteristic as evident in studies of twins (Malina et al., 2004), mother–daughter and sister–sister similarities in the general population and in athletes (Malina, Ryan, & Bonci, 1994) and ethnic variation (Chumlea et al., 2003; McDowell, Brody, & Hughes, 2007)

Percentage of Predicted Adult Height

The percentage of predicted adult height attained at the time of observation is increasingly used in studies of bio-banding among youth athletes, more so in males than in females (Malina et al., 2019). Data relating maturity status based on predicted adult height to SA are lacking for female soccer players, but the indicator had moderate concordance with SA in female tennis players (Myburgh, Cumming, & Malina, 2019).

The percentage of predicted adult height attained at the time of observation was used in two studies to classify players spanning a relatively broad CA range by pubertal status. Criteria for classifying youth players as pre-pubertal, pubertal or post-pubertal should be evaluated relative to observations from longitudinal studies. For example, among girls in the Zurich Longitudinal Study, estimated CA and percentage of adult height on entry into stage 2 of pubic hair (PH2) were 10.7 ± 1.1 years and 85.8 ± 2.9%, respectively (Gasser, Molinari, & Largo, 2013). In one study of soccer players (Westbrook et al., 2020), a percentage of predicted adult height of <87% defined the pre-pubertal state. Girls so classified were aged 10.3 ± 0.6 years with a percentage adult height of 84.0 ± 1.8%. Given the Zurich data, it is possible that some of the girls with a percentage of predicted adult height of <87% were in fact pubertal. Similarly, using a percentage of predicted adult height of >94% to indicate the post-pubertal state (Mullen et al., 2018; Westbrook et al., 2020) merits evaluation. Among girls in the Zurich study, estimated CA and percentage of adult height on entry into stage 4 of pubic hair (PH4), which is still the pubertal state, were 12.7 ± 1.2 years and 94.2 ± 2.1%, respectively, while estimated CA and percentage of adult height on entry into

PH5, the mature state, in Zurich girls were 13.7 ± 1.2 years and 97.0 ± 1.5%, respectively (Gasser et al., 2013).

Predicted Maturity Offset

Predicted maturity offset and/or predicted age at PHV (CA minus offset) was generally accepted as a maturity indicator or was used to classify players as pre-, at- or post-PHV without considering variation in CA per se. Predicted maturity offset increased linearly with CA (Figure 3.3A) and with height (Figure 3.3B) at prediction in the samples of soccer players. The trends in soccer players were consistent with CA-related trends in three longitudinal studies of girls (Malina et al., 2016; Kozieł & Malina, 2018; Malina, Kozieł, Kralik, Chrzanowska, & Suder, 2020), which also noted reduced variability in predicted maturity offset and in ages at PHV, and major limitations of the predictions among early and late-maturing girls defined by observed age at PHV.

The limitations of predicted maturity offset as a valid indicator of the time before or after PHV should be noted. The interval of PHV is central to the LTAD program for youth players in soccer (The Football Association, 2020) and other sports (Balyi & Hamilton, 2004; Bayli, Cardinal, Higgs, Norris, & Way, 2005). Awareness of the potential for misclassifications associated with predicted maturity offset and implications for player development require attention.

3.5. Summary

Relatively few of the studies considered in this narrative review (6%) were specifically focused on the growth and maturation of female youth soccer players. Of the studies reporting heights and weights, most (40%) were focused on changes in fitness and performance with CA and on training and match play, while many focused on the biomechanics and kinematics of the knee and hip, largely in the context of jumping (16%) and injuries and heading (16%). Other studies focused on bone health and body composition (10%), psychological variables (6%), nutritional status (4%) and general physiology (3%). Only one of seven studies reporting ages at menarche in soccer was focused on growth and maturation per se (Siegel, 1996), five were set in the context of bone health (Düppe et al., 1996; Casper et al.,1997; Pettersson et al., 2000) and one considered body perceptions (Prahter et al., 2016).

Allowing for the variety of studies, the heights and weights of players showed negligible secular variation between 1992 and 2020. Though limited to few observations, players aged <11 years were, on average, taller and heavier than the reference. Subsequently, the heights approximated the reference median through 14 years and approached the 75th percentile in late adolescence, while the weights were above the reference median from 12 through 18 years of age. The trends suggested the persistence of and/or selection for taller and heavier players during the adolescent years.

The estimated age at PHV based on the Preece–Baines model applied to the moving averages for height was 11.55 years and the mean ages at menarche ranged between 12.7 and 13.0 years. Both estimates of maturity timing among soccer players were within the range of mean ages at PHV and menarche in European and North American samples. Data for skeletal and sexual maturity status and for percentage of predicted adult height among female players are limited, while predicted maturity offset and age at PHV are increasingly used as a maturity indicator.

3.6. Implications

Individual differences in growth and maturation play a central role in the development of youth athletes, and the interval of PHV is central to the LTAD model proposed for female soccer players (The Football Association, p. 11): “Optimal aerobic training begins with the onset of Peak Height Velocity (PHV), more commonly known as the adolescent growth spurt. Strength training has two windows of accelerated adaptation in this phase. Window 1 is immediately after PHV and window 2 begins with the onset of menarche.” PHV specifically refers to the estimated maximal rate of growth in height during the adolescent spurt, while menarche occurs, on average, after PHV. Longitudinal data that span adolescence, 8 through 16–17 years of age in girls, are required to estimate PHV and age at PHV. Unfortunately, longitudinal data spanning the adolescent years are not available for female soccer players.

The adolescent growth spurt begins with an acceleration in the rate of growth in height during middle or late childhood (take-off); the rate of growth increases until it reaches a maximum (PHV), then decelerates and eventually ceases in late adolescence (Malina et al., 2004). Allowing for different methods of estimating parameters of the growth spurt, descriptive statistics and ranges of variability for ages at take-off of the spurt and in ages at PHV in three longitudinal studies of European girls are summarized in Table 3.4. The means are reasonably similar, but the ranges of variation are considerable. Corresponding statistics for the interval between age at take-off and age at PHV in the Polish longitudinal sample were 3.1 ± 0.8 years with a range of 0.5 to 5.4 years (calculated from data reported in Malina et al., 2020).

Table 3.4. Means (M), standard deviations (SD) and ranges for ages at take-off (initiation) of the growth spurt and at PHV in three longitudinal series of European girls.

		Age at Take-off, years			**Age at PHV, years**		
		M	**SD**	**Range**	**M**	**SD**	**Range**
Swiss (Largo et al., 1983)	110	9.6	1.1	6.6–12.9	12.2	1.0	9.3–15.0
British (Preece et al., 1978)	23	9.0	0.7	7.7–10.0	11.9	0.7	10.3–13.2
Polish (Malina et al., 2014)	198	8.9	1.1	6.3–12.0	11.9	1.0	9.0–14.8

Discussions of the need to monitor the adolescent spurt in the context of models for athlete development do not ordinarily address the range of individual differences in parameters of the growth spurt or the need to begin monitoring the growth of female athletes longitudinally at relatively young ages. The developmental model for female soccer (The Football Association, 2020) does not specify the method for identifying the onset of the growth spurt or for estimating age at PHV, while the LTAD model (Balyi et al., 2005; Largo & Prader, 1983) suggests monitoring growth in height over short intervals to estimate growth velocities and to monitor sudden change in velocity that may be indicative of the growth spurt. Estimated increments in growth over short intervals (3–4 months) must be interpreted with care. They should be adjusted for the interval between measurements and should consider measurement error at each observation (both inter- and intra-observer variability). Height measurements also vary with time of day (diurnal variation), tend to be reduced after intensive physical activity and also, vary with the season of the year (Malina et al., 2004).

Although predicted maturity offset (defined as the time before PHV) and estimated age at PHV (CA at prediction minus maturity offset) are increasingly applied in studies of youth athletes including female soccer players (Figures 3.3 and 3.4), intra-individual variation depending on CA at prediction is considerable. The predictions also have major limitations differentiating between early- and late-maturing youth defined by observed age at PHV (Boshnjaku et al., 2016; Malina et al., 2020; Bayli et al., 2004). Predicted ages at PHV are later than observed age at PHV among early-maturing girls and earlier than observed age at PHV in late-maturing girls. Practitioners using predicted maturity offset per se or variations of the method to identify when players enter and exit the interval of the adolescent growth spurt should be aware of these limitations and employ the methods with caution.

Predicted maturity offset also has major limitations when used to classify players by maturity status or to adjust fitness and performance scores to accommodate individual differences in maturation. If predicted offset is used to inform training design and prescription, it is essential that variation in CA and height at prediction and error associated with the prediction equations be considered. Perhaps additional or alternative methods might be considered as a complement, e.g., percentage of predicted adult stature attained at the time of observation. These and other issues related to the growth and maturity status and timing of youth athletes are discussed in more detail in several papers cited in this review (Malina, 2015; Malina et al., 2015; Malina et al., 2019).

3.6. References

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3.7. Supplementary material

Supplementary Table S1. Studies reporting ages, heights and weights of female youth soccer players by year within three intervals: 1992–2009, 2010–2017 and 2018–2020. Studies were initially sorted by those for which raw data were available to the first author. Then, all studies considered in the narrative review were sorted within each of the three time intervals by those which reported only age, height and weight and those which also included an indicator of maturity status: skeletal age, age at menarche, pubertal status, percentage of predicted adult height and predicted maturity offset or age at peak height velocity (PHV).

Studies for which the raw data for age, height and weight were available to the first author

1995–2020 (the references are also included in the three time intervals considered below)

1. Siegel SR (1995) Growth and maturity status of female soccer players from late childhood through early adulthood. Master of Science thesis, University of Texas, Austin, TX, USA. – also includes status quo and retrospective data for menarche which were re-analyzed
    a. Siegel SR, Katzmarzyk PT, Malina RM (1996) Somatotypes of female soccer players 10-24 years of age. In Studies in Human Biology, Bodzsár BE, Susanne C, (eds). Budapest, Hungary: Eötvös University Press, pp 277-85.
2. Kontos AP (2000) The effects of perceived risk, risk-taking behaviors and body size on injury in youth sport. Doctoral dissertation, Michigan State University, East Lansing, MI, USA.
    a. Kontos AP (2004) Perceived risk, risk taking, estimation of ability and injury among adolescent sport participants. J Pediatr Psychol. 29:447-55.

4. Cumming SP (2002) A bio-psychosocial investigation of self-determined motivation in recreational and travel youth soccer programs. Doctoral dissertation, Michigan State University, East Lansing, MI, USA.

    a. Cumming SP, Battista RA, Standage M, et al. (2006) Estimated maturity status and perceptions of adult autonomy support in youth soccer players. J Sports Sci. 24:1039-46. - percentage of predicted adult height attained at the time of observation

5. Stanforth PR, Crim BN, Stanforth D, et al. (2014) Body composition changes among female NCAA division I athletes across the competitive season and over a multiyear time frame. J Strength Cond Res. 28:300-7. – used only data for players 18 years of age.

6. US Soccer (2018), data for age, height and weight for participants in a 2018 tournament, with permission.

7. US Soccer (2020), data for age, height and weight for participants in a 2020 tournament, with permission.

1992–2009

Studies reporting only age, height and weight

1. McCulloch RG, Bailey DA, Whalen RL, et al. (1992) Bone density and bone mineral content of adolescent soccer athletes and competitive swimmers. Ped Exerc Sci. 4:319-30.

2. Kontos AP (2000) The effects of perceived risk, risk-taking behaviors and body size on injury in youth sport. Doctoral dissertation, Michigan State University, East Lansing, MI, USA.

a. Kontos AP (2004) Perceived risk, risk taking, estimation of ability and injury among adolescent sport participants. J Pediatr Psychol. 29:447-55.

3. Siegler J, Gaskill S, Ruby B (2003) Changes evaluated in soccer-specific power and endurance either with or without a 10-week in-season, intermittent, high-intensity training protocol. J Strength Cond Res. 17:379-87.

4. Cumming SP, Eisenmann JC, Smoll FL, et al. (2005) Body size perceptions of coaching behaviors of adolescent female athletes. Psychol Sport Exerc. 6:693-705.

5. Emery CA, Meeuwisse WH, Hartmann SE (2005) Evaluation of risk factors for injury in adolescent soccer: Implementation and validation of an injury surveillance system. Am J Sports Med. 33:1882-91.
6. Gómez López M, Moro MB (2005) Características fisiológicas de jugadoras españolas de fútbol femenino. KRONOS: Rendimiento en el Deporte 4(7):27-32.
7. Bellew JW, Gehrig L (2006) A comparison of bone mineral density in adolescent female swimmers, soccer players, and weight lifters. Pediat Phys Ther. 18:19-22.
8. Dickau L (2006) Examination of aerobic and anaerobic contributions to yo-yo intermittent recovery level 1 test performance in female adolescent soccer players. Master of Science thesis, University of Victoria, British Columbia, Canada.
9. Grandstrand SL, Pfeiffer RP, Sabick MB, et al. (2006) The effects of a commercially available warm-up program on landing mechanics in female youth soccer players. J Strength Cond Res. 20:331-5.
10. Pollard CD, Sigward SM, Ota S, et al. (2006) The influence of in-season injury prevention training on lower-extremity kinematics during landing in female soccer players. Clin J Sports Med. 16:223-7.
11. Landry SC, McKean KA, Hubley-Kozey CL, et al. (2007) Neuromuscular and lower limb biomechanical differences exist between male and female elite adolescent soccer players during an unanticipated side-cut maneuver. Am J Sports Med. 35:1888-1900.
12. Sporiš G, Čanaki M, Barišić V (2007) Morphological differences of elite Croatian female soccer players according to team position. Croatian Sports
13. Almagià Flores AA, Rodríguez F, Barraza Gómez FO, et al. (2008) Perfil antropométrico de jugadoras Chilenas de fútbol femenino. Internat J Morphol. 26:817-21.
14. Steffen K, Myklebust G, Andersen TE, et al. (2008) Self-reported injury history and lower limb function as risk factors for injuries in female youth soccer. Am J Sports Med. 36:700-8. Medicine Journal/Hrvatski Športskomedicinski Vjesnik. 22:91-6.

15. Sigward SM, Ota S, Powers CM (2008) Predictors of frontal plane knee excursion during a drop land in young female soccer players. J Orthop Sports Phys Ther. 38:661-7.

16. Vescovi JD, McGuigan MR (2008) Relationships between sprinting, agility, and jump ability in female athletes. J Sports Sci. 26:97-107.

17. Barber Foss KD, Ford KR, Myer GD, Hewett TE (2009) Generalized joint laxity associated with increased medial foot loading in female atheltes. J Athl Train. 44:356-62.

18. Barbero-Álvarez JC, Barbero Álvarez V, Gómez M, et al. (2009) Análisis cinemático del perfil de actividad en jugadoras infantiles de fútbol mediante tecnologia GPS. KRONOS: Rendimiento en el Deporte 8(14):35-42.

19. DiStefano LJ, Padua DA, DiStefano MJ, Marshall SW (2009) Influence of age, sex, technique, and exercise program on movement patterns after an anterior cruciate ligament injury prevention program in youth soccer players. Am J Sports Med. 37:495-505.

20. Ekegran CL, Miller WC, Celebrini RG, et al. (2009) Reliability and validity of observational risk screening in evaluating dynamic knee valgus. J Orthop Sports Phys Ther. 39:665-74.

21. Mujika I, Santisteban J, Impellizzeri FM, et al. (2009) Fitness determinants of success in men's and women's football. J Sports Sci. 27:107-14.

Studies including a maturity indicator in addition to age, height and weight: age at menarche

22. Siegel SR (1995) Growth and maturity status of female soccer players from late childhood through early adulthood. Master of Science thesis, University of Texas, Austin, TX, USA. –also includes status quo and retrospective data for menarche

    a. Siegel SR, Katzmarzyk PT, Malina RM (1996) Somatotypes of female soccer players 10-24 years of age. In Studies in Human Biology, Bodzsár BE, Susanne C, (eds). Budapest, Hungary: Eötvös University Press, pp 277-85.

23. Düppe H, Gärdsell P, Johnell O, et al. (1996) Bone mineral density in female junior, senior and former football players. Osteopor Internat. 6:437-41.

24. Casper RC, Michaels J, Simon K (1997) Body perception and emotional health in athletes: A study of female adolescents involved in aesthetic sports. In Simopoulos AP, Pavlou KN (eds) Nutrition and Fitness: Metabolic and Behavioral Aspects in Health and Disease. Basel, Karger, World Review of Nutrition and Dietetics 82:134-47.

25. Pettersson U, Nordström P, Alfredson H, et al. (2000) Effect of high impact activity on bone mass and size in adolescent females: A comparative study between two different types of sports. Calc Tissue Internat. 67:207-14.

26. Södermann K, Bergström E, Lorentzon R, et al. (2000) Bone mass and muscle strength in young female soccer players. Calc Tissue Internat. 67:297-303.

Studies including a maturity indicator in addition to age, height and weight: percentage of predicted adult height attained at the time of observation

27. Cumming SP (2002) A bio-psychosocial investigation of self-determined motivation in recreational and travel youth soccer programs. Doctoral dissertation, Michigan State University, East Lansing, MI, USA.
    a. Cumming SP, Battista RA, Standage M, et al. (2006) Estimated maturity status and perceptions of adult autonomy support in youth soccer players. J Sports Sci. 24:1039-46.

2010–2017

Studies reporting only age, height and weight

1. Bunc V (2010) Physiological and functional characteristics of adolescent athletes in several sports. In Coelho-e-Silva MJ, Figueiredo AJ, Elferink-Gemser M, Malina, RM (editors), Youth Sports: Growth, Maturation and Talent. Coimbra, Portugal, University of Coimbra Press, p. 115-25.

2. Castagna C, Impellizzeri FM, Manzi V, et al. (2010) The assessment of maximal aerobic power with the multistage fitness test in young women soccer players. J Strength Cond Res. 24:1488-94.

3. Ortiz A, Trudelle-Jackson E, McConnell K, et al. (2010) Effectiveness of a 6-week injury prevention program on kinematics and kinetic variables in adolescent female soccer players: A pilot study. Puerto Rico Health Sci. J 29:40-8.

4. Vescovi JD, Van Heest JL (2010) Effects of an anterior cruciate ligament injury prevention program on performance in adolescent female soccer players. Scand J Med Sci Sports. 20:394-402.

5. Ferry B, Duclos M, Burt L, et al. (2011) Bone geometry and strength adaptations to physical constraints inherent in different sports: Comparison between elite female soccer players and swimmers. J Bone Min Metab. 29:342-51.

   a. Ferry B, Lespessailles E, Rochcongar P, et al. (2013) Bone health during late adolescence: Effects of an 8-month training program on bone geometry in female athletes. J Bone Spine. 80:57-63. – same age, height, weight data as 2011 study

6. Gibson JC, Stuart-Hill L, Martin S, et al. (2011) Nutrition status of junior elite Canadian female soccer athletes. Internat J Sport Nutr Exerc Metab. 21:507-14.

7. Kontos AP, Dolese A, Elbin III RJ, et al. (2011) Relationship of soccer heading to computerized neurocognitive performance and symptoms among female and male youth soccer players. Brain Inj. 25:1234-41.

8. Parsons JL, Carswell J, Nwboa IM, et al. (2011) Athletic perceptions and physical performance effects of the FIFA 11+ program in 9-11 year-old female soccer players: A cluster randomized trial. Int J Sports Phys Ther. 14:740-52.

9. Rowland T, Unnithan V, Roche D, et al. (2011) Myocardial function and aerobic fitness in adolescent females. Eur J Appl Physiol. 111:1991-7.

10. Rubley MD, Haase AC, Holcomb WR, et al. (2011) The effect of plyometric training on power and kicking distance in female adolescent soccer players. J Strength Cond Res. 25:129-34.

11. Stieg JL, Faulkinbury KJ, Tran TT, et al. (2011) Acute effects of depth jump volume on vertical jump performance in collegiate women soccer players. Kinesiology 43:25-30.

12. Celebrini RG, Eng JJ, Miller WC, et al. (2012) The effect of a novel movement strategy in decreasing ACL risk factors in female adolescent soccer players. J Strength Cond Res. 26:3404-17.

13. Chrisman SP, O’Kane JW, Polissar NL, et al. (2012) Strength and jump biomechanics of elite and recreational female youth soccer players. J Athl Train. 47:609-15.

14. Dillern T, Ingebrigtsen J, Shalfawi SAI (2012) Aerobic capacity and anthropometric characteristics of elite-recruit female soccer players. Serb J Sports Sci 6:43-9.

15. Lindblom H, Waldén M, Hägglund M (2012) No effect on performance tests from a neuromuscular warm-up programme in youth female football: A randomized controlled trial. Knee Surg Sports Traumatol Arthrosc. 20:2116-23.

16. Portela Sarazola J (2012) Description and exemplary analysis of match running performance and a selected battery of tests in adolescent female soccer players. Master’s thesis, Institute of Movement and Training Science, University of Leipzig, Germany (as cited by Martinez-Lagunas et al. [J Sport Health Sci. 2014; 3:258-72]).

17. Walden M, Atroshi I, Magnusson H, et al. (2012) Prevention of acute knee injuries in adolescent female football players: Cluster randomized control trial. Br Med J. 344:e3042, doi: 10.1136/bmj.e3042.

18. Castagna C, Castellini E (2013) Vertical jump performance in Italian male and female national team soccer players. J Strength Cond Res. 27:1156-61.

19. Guagliano JM, Rosenkranz RR, Kolt GS (2013) Girls’ physical activity levels during organized sports in Australia. Med Sci Sports Exerc. 45:116-22.

20. Noyes FR, Barber-Westin SD, Tutalo Smith ST, et al. (2013) A training program to improve neuromuscular and performance indices in female high school soccer players. J Strength Cond Res. 27:240-51.

21. Adhikari A, Nugent J (2014) Anthropometric characteristic, body composition and somatotype of Canadian female soccer players. Am J Sports Sci. 2:14-8.

22. Celebrini RG, Eng JJ, Miller WC, et al. (2014) Effect of a novel movement strategy in decreasing ACL risk factors in female adolescent soccer players: A randomized controlled trial. Clin J Sports Med. 24: 134-41.

23. Gutierrez GM, Conte C, Lightbourne K (2014) The relationship between impact force, neck strength and neurocognitive performance in soccer heading in adolescent females. Pediat Exerc Sci. 26:33-40.

24. Haugen TA, Tønnessen E, Hem E, et al. (2014) VO2max characteristics of elite female soccer players 1989-2007. Internat J Sports Physiol Perf. 9:515-21.

25. Manson SA, Brughelli M, Harris NK (2014) Physiological characteristics of international female soccer players. J Strength Cond Res. 28:308-18.

26. Mathisen GE, Danielsen KH (2014) Effects of speed exercises on acceleration and agility performance in 13-year-old female soccer players. J Phys Educ Sport. 14:471-4.

27. Rumpf MC, Schneider AS, Schneider C, Mayer HM (2014) Training profiles and motivation of male and female youth soccer players. Int J Sports Sci Coach. 9:207-16.

28. Sogabe N, Okada M, Tokita Y, et al. (2014) Physical status and eating habits of junior/senior high school girls playing football. J Jpn Soc Shokuiku (nutrition education). 8:41-47. (in Japanese)

29. Stanforth PR, Crim BN, Stanforth D, et al. (2014) Body composition changes among female NCAA division I athletes across the competitive season and over a multiyear time frame. J Strength Cond Res. 28:300-7. – used only data for soccer players 18 years of age

30. Hirose N, Nakahori C (2015) Age differences in change-of-direction performance and its sub-elements in female football players. Internat J Sports Physiol Perf. 10:440-5.

31. Inoue Y, Otania Y, Naruse S, et al. (2015a) Relationships between lower-extremity muscle strength and the modified star excursion balance test in adolescent female players. Rigakuryoho Kagaku. 30:853-6 (in Japanese).

32. Inoue Y, Otani Y, Eusugi M, et al. (2015b) Characteristics of injuries with female junior high and high school soccer players in comparison with male players. Jpn J Phys Fit Sports Med. 64:345-50 (in Japanese).

33. Mathisen GE, Pettersen SA (2015) The effect of speed training on sprint and agility performance in 15-year-old female soccer players. LASE J Sport Sci. 6:61-70.

34. Nevado-Garrosa F, Suarez-Arrones L (2015) Comparación de las demandas fisicas de tareas de fútbol reducido y la competición en jugodoras de fútbol sub 13. Cultura, Ciencia y Deporte (Universidad Católica de Murcia) 10:235-43.

35. Ono E, Maegawa T, Kamei Y, et al. (2015) Investigating the method of evaluating the ability of power exertion at the lower limb for female collegiate athletes. Japan J Coaching Stud. 28:175-82.

36. Datson N (2016) An analysis of the physical demands of international female soccer match-play and the physical characteristics of elite players. Doctoral dissertation, Liverpool John Moores University, Liverpool, United Kingdom

37. Hägglund M, Waldén M (2016) Risk factors for acute knee injury in female youth football. Knee Surg Sports Traumatol Arthrosc. 24:737-46.

38. Lesinski M, Muehlbauer T, Granacher U (2016) Concurrent validity of the Gyko inertial sensor system for the assessment of vertical jump height in female sub-elite youth soccer players. BMC Sports Sci Med Rehab. 8:35, 1-9, doi: 10.1186/s13102-016-0061-x.

39. Oyón P, Franco L, Rubio FJ, Valero A (2016) Young women soccer players: Anthropmetric and physiological characteristics – evolution in a sports season. Archivos de Medicina del Deporte 33:24-8.

40. Póvoas SCA, Castagna C, da Costa Soares JM, et al. (2016) Reliability and construct validity of yo-yo tests in untrained and soccer-trained schoolgirls aged 9-16. Pediat Exerc Sci. 28:321-30.

41. dos Santos Andrade M, Mascarin NC, Benedito-Silva AA, et al. (2016) Assessment of isokinetic peak torque reliability of the hip flexor, extensor, adductors and abductors muscles in female soccer players from 14 to 25 years. J Sports Med Phys Fit. 56:843-8.

42. Turner E (2016) Physical and match performance of female soccer players. Doctoral dissertation, University of Salford, Manchester, United Kingdom.

43. Chapelle L, Tassignon B, Aerenhouts D, et al. (2017) The hydration status of young female elite soccer players during an official tournament. J Sports Med Phys Fit. 57:1186-94.

44. Gradidge PJ-L, Constantinou D (2017) A comparative study on the cardiac morphology and vertical jump height of adolescent Black South African male and female amateur competitive footballers. Cardiovasc J Africa. 28: online publication. Doi: 10:5830/CVJA-2017-032.

45. Manore MM, Patton-Lopez MM, Meng Y, et al. (2017) Sport nutrition knowledge, behaviors and beliefs of high school soccer players. Nutrients 9:350, doi:10.3390/nu9040350.

47. Richardson A, Clarsen B, Verhagen EALM, Stubbe JH (2017) High prevalence of self-reported injuries and illnesses in talented female athletes. BMJ Open Sport Exerc Med. 3:e000199, doi:10.1136/bmjsem-2016-000199.

48. Taylor JB, Ford KR, Schmitz RJ, et al. (2017) Biomechanical differences of multidirectional jump landings among female basketball and soccer players. J Strength Cond Res. 31:3034-45.

49. Thompson JA, Tran AA, Gatewood CT, et al. (2017) Biomechanical effects of an injury prevention program in preadolescent female soccer athletes. Am J Sports Med. 45:294-301.

50. Watson A, Brickson S, Brooks A, et al. (2017) Subjective well-being and training load predict in-season injury and illness risk in female youth soccer players. Br J Sports Med. 51:194-99.

Studies including a maturity indicator in addition to age, height and weight: age at menarche

51. Brännström A, Yu J-G, Jonsson P, et al. (2017) Vitamin D in relation to bone health and muscle function in young female soccer players. Eur J Sport Sci. 17:249-56.

Studies reporting only age at menarche

52. Prather H, Hunt D, McKeon K, et al. (2016) Are elite female soccer athletes at risk for disordered eating attitudes, menstrual dysfunction, and stress fractures? Phys Med Rehab 8:208-13. – heights and weights of players were not reported

Studies including a maturity indicator in addition to age, height and weight: pubertal status

53. Anauate Nicolao AL, Pedrinelli A, Martino Zogalb PA, et al. (2010) Influência da maturação sexual no limiar de lactate em jogadoras de futebol. Revista Brasileira de Medicina do Esporte 16:335-38.

54. Lyle MA, Sigward SM, Tsai L-C, et al. (2011) Influence of maturation on instep kick biomechanics in female soccer athletes. Med Sci Sports Exerc. 43:1948-54.

55. Sigward SM, Pollard CD, Havens KL, Powers CM (2012) Influence of sex and maturation on knee mechanics during side-step cutting. Med Sci Sports Exerc. 44:1497-1503.

   a. Sigward SM, Pollard CD, Powers CM (2012) The influence of sex and maturation on landing biomechanics: implications for anterior cruciate ligament injury. Scand J Med Sci Sport 22:502-9. – same data set

56. Ubago-Guisado E, Gómez-Cabello A, Sánchez-Sánchez J, et al. (2015) Influence of different sports on bone mass in growing girls. J Sports Sci. 33:1710-8.

   a. Ubago-Guisado E, Mata E, Sánchez-Sánchez J, et al. (2017) Influence of different sports on fat mass and lean mass in growing girls. J Sport Health Sci. 6:213-8. – same data set b. Ubago-Guisado E, Garcia-Unanue J, López-Fernández J, et al. (2017) Association of different types of playing surfaces with bone mass in growing girls. J Sports Sci. 35:1484-92. – same data set

57. Unnithan V, Roche DM, Garrard M, et al. (2015) Oxygen uptake kinetics in trained adolescent females. Eur J Appl Physiol. 115:213-20.

58. Plaza-Carmona M, Vicente-Rodríguez G, Gómez-Cabello A, et al. (2016) Higher bone mass in prepubertal and peripubertal female footballers. Eur J Sport Sci. 16:877-83

   a. Plaza-Carmona M, Ubago Guisado E, Sánchez-Sánchez J, et al. (2013) Body composition and physical fitness in prepubertal girls swimmers and soccer players. J Sport Hlth Res. 5:251-8. – same data set

59. Lozano Berges G, Matute Llorente A, Gómez Bruton A, et al. (2017) Body fat percentage comparisons between four methods in young football players: Are they comparable? Nutrición Hospitalaria 34:1119-24.

Studies including a maturity indicator in addition to age, height and weight: predicted maturity offset or age at PHV

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Supplementary Table S2. Sources used in the compilation of heights and weights of adult female soccer players.

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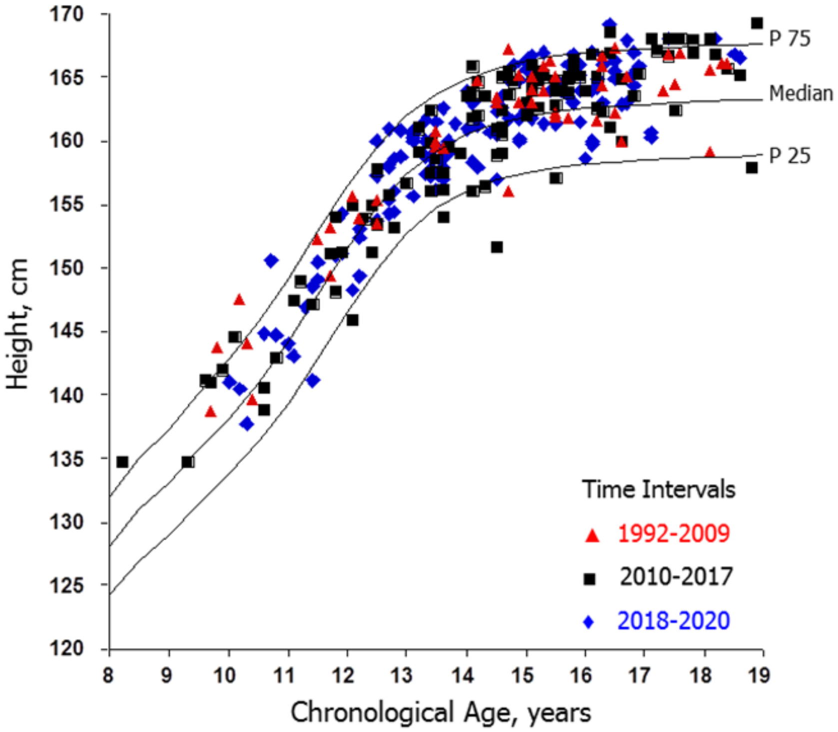

Figure S1. Mean heights reported in studies of female youth soccer players within three intervals, 1992–2009, 2010–2017 and 2018–2020, plotted by age relative to medians and 25th and 75th percentiles of reference data for U.S. girls (see text for details).

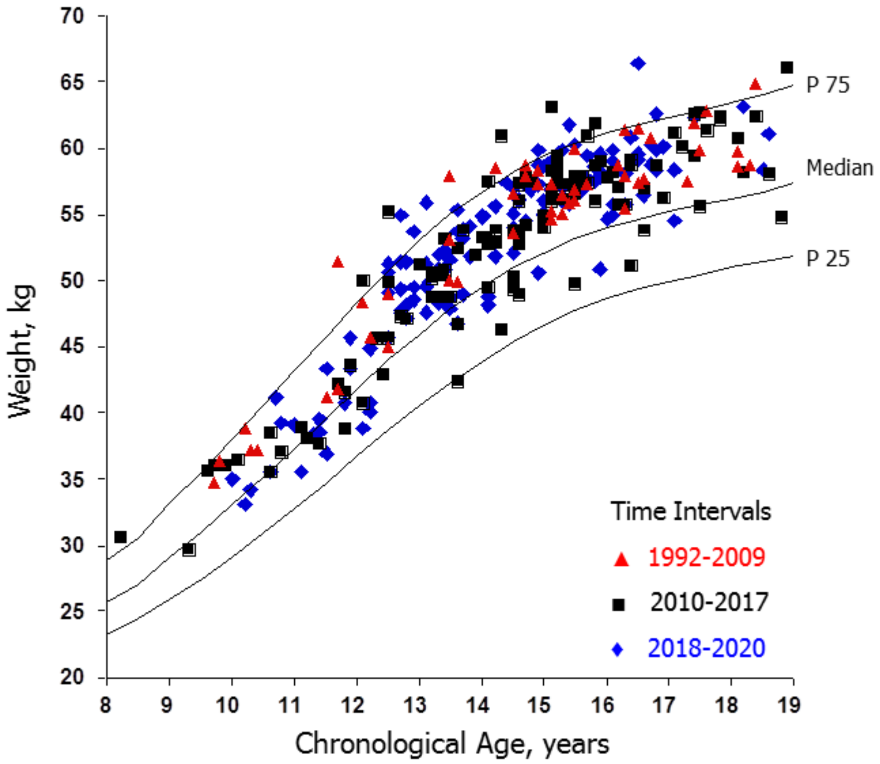

Figure S2. Mean weights reported in studies of female youth soccer players within three intervals, 1992–2009, 2010–2017 and 2018–2020, plotted by age relative to medians and 25th and 75th percentiles of reference data for U.S. girls (see text for details).

Chapter IV

Study 2

Reproducibility and inter-observer agreement of Greulich-Pyle protocol
to estimate skeletal age among female soccer players

4. Reproducibility and inter-observer agreement of Greulich-Pyle protocol to estimate skeletal age among female adolescent soccer players

4.1. Abstract

Background: Skeletal age (SA) is considered the best method of assessing biological maturation. The aim of this study was to determine intra-observer (reproducibility) and inter-observer agreement of SA values obtained via the Greulich-Pyle (GP) method. In addition, the variation in calculated SAs by alternative GP protocols were examined. Methods: The sample was composed of 100 Portuguese female soccer players aged 12.0–16.7 years. SAs were determined using the GP method by two observers (OB1: experience < 100 exams using GP; OB2: experience > 2000 exams using several methods). The radiographs were examined using alternative GP protocols: (wholeGP) the plate was matched to the atlas as an overall approach; (30-boneGP) bone-by-bone inspections of 30-bones; (GP_{pmb}) bone-by-bone inspections of the pre-mature bones only. For the 30-boneGP and GP_{pmb} approaches, SA was calculated via the mean (M) and the median (Md). Results: Reproducibility ranged 82–100% and 88–100% for OB1 and OB2, respectively. Inter-observer agreement (100 participants multiplied by 30 bones) was 92.1%. For specific bones, agreement rates less than 90% were found for scaphoid (81%), medial phalange V (83%), trapezium (84%) and metacarpal V (87%). Differences in wholeGP SAs obtained by the two observers were moderate (d-cohen was 0.79). Mean differences between observers when using bone-by bone SAs were trivial (30-boneGP: d-cohen less than 0.05; GP_{pmb}: d-cohen less than 0.10). The impact of using the mean or the median was negligible, particularly when analyses did not include bones scored as mature. Conclusion: The GP appeared to be a reasonably reproducible method to assess SA and inter-observer agreement was acceptable. There is evidence to support a recommendation of only scoring pre-mature bones during later adolescence. Further research is required to examine whether these findings are consistent in younger girls and in boys.

Keywords: Youth Sports, Female Athlete, Biological Maturation, Bone Age, Atlas Method

4.2. Introduction

The study of growth status, biological maturation, and physical performance is central to sports sciences, human biology, and pediatrics. Growth refers to changes in body size and has implications for proportionality, shape, and composition (Malina, Bouchard, & Bar-Or, 2004). Maturation is more difficult to define and refers to the progress toward the adult (mature) state. This occurs in all tissues and organs at different timing and rates, affecting functions and metabolism. Skeletal age (SA) refers to the degree of skeletal maturation and can be examined via a standardized radiograph (usually of the left hand and wrist). Although several indicators of biological maturation are available (e.g. secondary sex characteristics, age at peak height velocity (PHV), predicted percentage of mature stature(%PMS)), SA is frequently considered the gold standard indicator of biological maturity, partly because it can be applied from fetal life through childhood and the second decade of life (Malina et al., 2004).

Assessment of biological maturation is common in youth sports. It has been recognized that maturity status impacts performance (Coelho e Silva, Figueiredo, Carvalho, & Malina, 2008; Coelho e Silva et al., 2010a; Malina et al., 2005; Malina et al., 2019), injury (Malina et al., 2006; Le Gall, Carling, & Reilly, 2007; van der Sluis et al., 2014) and selection (Coelho-e-Silva et al., 2010b). Many studies have assessed the SA of adolescents participating in team sports (e.g. soccer, hockey) and found, in general, that players were early, maturing, taller, heavier and stronger (Figueiredo, Goncalves, Coelho-e-Silva, & Malina, 2009; Coelho-e-Silva et al., 2012). Malina et al. (2000) suggested that one reason for this observation is that, among male adolescent soccer players late maturers tended to be systematically excluded during years of maximal growth while those classified as early or average maturing are selected and/or promoted by coaches and club administrators and this became even more evident as the players got older and specialized in one sport. Although the same rationale could be applied to female adolescent athletes, the evidence is scarce.

The Greulich-Pyle (GP) method is often used to obtain a SA estimate and involves comparing each individual bone against pictorial standards (Greulich & Pyle, 1959). Across published literature, however, there are various versions of the GP protocol and often a lack of detail regarding the methods used to estimate SA. For example, one study (Fortes et al., 2014) stated: "*skeletal maturation was evaluated by the determination of bone age (BA) according to the GP method" (page 626) and did not detail the GP procedures, specifically whether the radiograph was examined as a whole (wholeGP) or via bone by bone*". A more recent study (Gouvea et al., 2016) described the biological maturity variation in body size, functional capacities, and sport-specific technical skills of 60 male Brazilian adolescent soccer players. The following was stated in the methodology: "*the left hand-wrist radiographs were obtained in a specialized laboratory and the GP method was adopted to estimate SA"* (page 465). The original authors prescribed that after identifying the standard which most closely resembles the film being assessed, one should proceed to make a more detailed comparison of the individual bones (Greulich & Pyle, 1959). In practice, GP SAs seemed not to be properly obtained based on the SA of the standard plate which the film of a young person most closely matches, thus excluding variation among bones. Research is required to ascertain the error associated with different methods of examination to inform future studies.

The present study aimed to examine the intra-observer reproducibility and inter-examiner agreement using a variation in GP protocols: a) overall (wholeGP) or bone-by-bone approach; b) inspection of all bones (30-boneGP) or solely the pre-mature bones (GP_{pmb}) c) calculating SA using the mean or the median values. It was hypothesized that agreement rates would be higher when observers follow a wholeGP and bone-by-bone approaches (30-boneGP; GP_{pmb}).

4.3. Methods

Ethics and procedures

This cross-sectional, descriptive study was approved by the Ethics Committee for

Sports Sciences in the University of Coimbra (Reference CE/FCDEF-UC/00122014). All data were collected in the Porto Sports Medicine Center as part of the medical exams for registration in the Portuguese Soccer Federation (Law 204/2006; act 11; 2012). An institutional agreement was signed between the University of Coimbra and the Portuguese Institute of Sports [IPDJ/FCDEF.UC/2017–01]. Parents or legal guardians were informed about the aims, testing protocols, risks and provided informed consent. Participants were also informed about the nature of the research and that they were allowed to withdraw from the study at any time.

A standardized radiograph of the left hand-wrist was obtained by an experienced technician. SA was assessed using the GP method, which is often called the atlas method (Greulich & Pyle, 1959). It is an inspectional protocol that was developed from a study of children from high socioeconomic families in Ohio (Cleveland, USA). The method involves the matching of a specific radiograph of an observed participant to the closest plate from the collection of illustrations (photographs) representing a sequence of biological (skeletal) maturation. The estimate of SA refers to the CA of the child from the Brush Foundation Study whose plate was classified as the closest to the one under-examination. Thus, if the radiograph of 13-year-old female soccer player matches the standard plate of the atlas obtained from a 11-year-old girl, the SA of the participant is 11 years. Each film was rated twice by an observer (OB1) who had completed a 45-h post-graduation course including the anatomy of hand and wrist, the biological basis of skeletal tissue, and the sequence of changes for each of the 30 bones assessed by the GP method. In parallel, measurements were also completed by a trained examiner (OB2) who had experience of conducting over 2000 assessments over the previous 3 years using the GP method in addition to other protocols (e.g. Tanner-Whitehouse; Fels). Examiners did not know the CA of the participants prior to applying the GP method.

Participants

The sample for this study were 100 Portuguese female soccer players aged 12.0–16.7 years. To be included in the study participants were required to have played competitive soccer for at least 2 years in a club affiliated to the Portuguese Soccer Federation. Exclusion criteria were: (i) ≥17 years of age; (ii) any traumatic bone injury in the hand and left wrist that causes radiopaque lines or areas; (iii) previous/current exposure to medicines (e.g. steroids, growth hormone) that affects growth acceleration.

Determination of SA

Each radiograph film was evaluated using three alternative protocols: inspection of the whole plate with the closest atlas photograph retained as the SA of the participant (wholeGP); 30 bones were individually examined and included in calculations of the SA (30-boneGP); 30 bones were individually examined and calculations to obtain the SA of the participant were based on premature bones only (GP_{pmb}). For 30-boneGP and GP_{pmb}, SA was calculated using the mean (M) or, in alternative, the median (Md). Consequently, the following scores were produced: 30-boneGP-M, 30-boneGP-Md, GPpmb-M, GPpmb-Md.

Analyses

Intra-observer reproducibility rates for each of the 30 bones were reported for each participant. Afterwards, inter-observer agreement was calculated for each bone and analyzed by maturity status (that is, agreement between two observers was examined separately for premature and mature bones). The error (OB1 minus OB2) was calculated for each individual bone. Mean differences of the SAs rated by OB1 and OB2 were calculated and magnitudes of the differences interpreted as follows (Hopkins, Marshall, Batterham, & Hanin, 2009): trivial ($r < 0.1$), small ($0.1 < r < 0.3$), moderate ($0.3 < r < 0.5$), large ($0.5 < r < 0.7$), very large ($0.7 < r < 0.9$), and nearly perfect ($r > 0.9$). Intra-class correlation coefficients were calculated to examine the variance between measurements for each observer. All analyses were performed using SPSS version 20 (SPSS, Inc. IBM Company; NY, USA) and Graphpad Prism (version 5 for Windows, GraphPad Software, San Diego California USA, www.graphpad.com).

4.4. Results

Tables 4.1 and 4.2 summarizes the intra-observer agreement rates, respectively for OB1 and OB2. The less experienced observer (OB1) agreement rates were < 90% (i.e. for radius: 82%; ulna: 86%; metacarpal V: 89%; proximal phalange II 89%; proximal phalange IV: 89%). Of these 66.8% were positive (time-moment 1 minus time-moment 2) and 87.5% of the total number of errors were −1 or+1 plates of the atlas (two consecutive stages – see Table 4.1). Identical analysis was performed by the more experienced observer (OB2) and results summarized in Table 4.2. When 30 bones were individually scored, intra-observer agreement rate was < 90% uniquely for proximal phalange I (88%) and 99% of the errors emerged from variation among two consecutive stages with a trend for lower SA values in the second examination (discrepancies −1 stage: 61.1%; discrepancies + 1 stage: 23.3%). In other words, increasing practice, particularly in OB1, tended to produce slightly lower SA scores.

Table 4.1. Intra-observer error (observer 1) on SAs estimates among female adolescent soccer players (n = 100).

			Agreement		disagreement						
	Time-moment 1 (TM1)	Time-moment 2 (TM2)	N	%	n	magnitude of discrepancies[a]					
						-3	-2	-1	+1	+2	+3
Radius	15.6 ± 1.2	15.5 ± 1.3	82	82%	18			5	12	1	
Ulna	16.1 ± 1.1	16.1 ± 1.1	86	86%	14			8	6		
Capitate	12.9 ± 0.3	12.9 ± 0.4	93	93%	7			4	3		
Hamate	12.8 ± 0.6	13.0 ± 0.2	92	92%	8	3	1	4			
Triquetral	12.8 ± 0.6	12.9 ± 0.5	90	90%	10	1	3	2	4		
Pisiform	-	-	100	100%							
Lunate	12.9 ± 0.4	12.9 ± 0.4	96	96%	4			1	2	1	
Scaphoid	12.9 ± 0.4	12.8 ± 0.5	91	91%	9			1	6	2	
Trapezium	12.8 ± 0.6	12.8 ± 0.6	90	90%	10	1	1		1	7	
Trapezoid	12.9 ± 0.4	12.9 ± 0.4	95	95%	5	1		1	2	1	
Adductor Sesamoid	-	-	100	100%							
Metacarpal I	14.9 ± 0.5	14.9 ± 0.5	94	94%	6			1	5		
Metacarpal II	14.7 ± 0.6	14.6 ± 0.6	91	91%	9			5	4		
Metacarpal III	14.7 ± 0.6	14.7 ± 0.6	92	92%	8			2	6		
Metacarpal IV	14.8 ± 0.5	14.7 ± 0.6	90	90%	10				10		
Metacarpal V	14.7 ± 0.5	14.7 ± 0.6	89	89%	11			3	6	2	
Proximal phalange I	14.4 ± 0.6	14.4 ± 0.6	91	91%	9		1	4	4		
Proximal phalange II	14.8 ± 0.5	14.7 ± 0.6	89	89%	11			1	10		
Proximal phalange III	14.8 ± 0.5	14.7 ± 0.6	91	91%	9				9		
Proximal phalange IV	14.8 ± 0.5	14.8 ± 0.6	89	89%	11		1	1	9		
Proximal phalange V	14.7 ± 0.6	14.8 ± 0.6	90	90%	10			8	2		
Medial phalange II	14.9 ± 0.4	14.8 ± 0.6	90	90%	10			1	8	1	
Medial phalange III	14.9 ± 0.5	14.8 ± 0.6	91	91%	9			1	7	1	
Medial phalange IV	14.9 ± 0.5	14.8 ± 0.5	91	91%	9			1	8		
Medial phalange V	14.8 ± 0.5	14.8 ± 0.6	92	92%	8			4	4		
Distal phalange I	14.3 ± 0.6	14.3 ± 0.6	97	97%	3			2	1		
Distal phalange II	14.3 ± 0.6	14.3 ± 0.6	95	95%	5			2	3		
Distal phalange III	14.3 ± 0.6	14.3 ± 0.6	96	96%	4			1	3		
Distal phalange IV	14.3 ± 0.6	14.3 ± 0.6	96	96%	4			1	3		
Distal phalange V	14.3 ± 0.5	14.3 ± 0.6	99	99%	1				1		
Total disagreements			2768		232	6	7	64	139	16	0
%			92.3%		7.7%	2.6%	3.1%	27.6%	59.9%	6.9%	0.0

[a] signs refer to TM1 minus TM2 and numbers corresponds to plates from the collection of photographs representing the sequence of skeletal maturation in atlas.

Table 4.2. Intra-observer error (observer 2) on SAs estimates among female adolescent soccer players (n = 100).

			agreement		disagreement						
	Time-moment 1 (TM1)	Time-moment 2 (TM2)	N	%	n	magnitude of discrepancies[a]					
						-3	-2	-1	+1	+2	+3
Radius	15.6 ± 1.2	15.5 ± 1.2	90	90%	10			1	9		
Ulna	16.0 ± 1.1	16.0 ± 1.1	94	94%	6				6		
Capitate	12.9 ± 0.3	13.0 ± 1.2	96	96%	4			4			
Hamate	12.9 ± 0.5	13.0 ± 0.4	96	96%	4			4			
Triquetral	13.0 ± 0.2	13.0 ± 0.1	96	96%	4			4			
Pisiform	-	-	100	100%							
Lunate	12.9 ± 0.4	13.0 ± 0.2	91	91%	9			9			
Scaphoid	12.7 ± 0.6	12.8 ± 0.5	93	93%	7			7			
Trapezium	12.8 ± 0.6	12.8 ± 0.5	96	96%	4		1	3			
Trapezoid	12.9 ± 0.3	13.0 ± 0.1	96	96%	4			4			
Adductor Sesamoid	-	-	100	100%							
Metacarpal I	14.9 ± 0.4	14.9 ± 0.4	92	92%	8			1	7		
Metacarpal II	14.7 ± 0.6	14.6 ± 0.4	91	91%	9			2	7		
Metacarpal III	14.6 ± 0.6	14.6 ± 0.6	95	95%	5			3	2		
Metacarpal IV	14.6 ± 0.6	14.6 ± 0.6	96	96%	4			4			
Metacarpal V	14.6 ± 0.7	14.6 ± 0.6	94	94%	6			3	3		
Proximal phalange I	14.4 ± 0.7	14.4 ± 0.6	88	88%	12			7	5		
Proximal phalange II	14.8 ± 0.6	14.8 ± 0.6	98	98%	2			1	1		
Proximal phalange III	14.7 ± 0.7	14.8 ± 0.6	98	98%	2			2			
Proximal phalange IV	14.7 ± 0.7	14.8 ± 0.6	96	96%	4			4			
Proximal phalange V	14.7 ± 0.6	14.8 ± 0.6	96	96%	4			4			
Medial phalange II	14.8 ± 0.6	14.8 ± 0.6	98	98%	2				2		
Medial phalange III	14.8 ± 0.6	14.7 ± 0.6	96	96%	4				4		
Medial phalange IV	14.8 ± 0.5	14.8 ± 0.5	94	94%	6			3	3		
Medial phalange V	14.8 ± 0.5	14.8 ± 0.5	98	98%	2			1	1		
Distal phalange I	14.3 ± 0.5	14.3 ± 0.6	94	94%	6			6			
Distal phalange II	14.3 ± 0.5	14.3 ± 0.5	100	100%							
Distal phalange III	14.3 ± 0.5	14.3 ± 0.5	93	93%	7			4	3		
Distal phalange IV	14.3 ± 0.5	14.3 ± 0.6	96	96%	4			4			
Distal phalange V	14.3 ± 0.5	14.3 ± 0.6	95	95%	5			3	2		
Total disagreements			2856		144		1	88	55		
%			95.2%		4.8%		1%	61.1%	23.3%		

[a] signs refer to TM1 minus TM2 and numbers corresponds to plates from the collection of photographs representing the sequence of skeletal maturation in atlas.

Inter-observer agreement rates are summarized in Table 4.3 for each bone and by maturity status (premature or mature). The two observers had 100% agreement when scoring mature bones. The number of participants who had pre-mature bones were greater for radius (n = 70), ulna (n = 52) and distal phalanges I-V (n = 65). The agreement rates among the two observers were lower for the pre-mature carpals (capitate, hamate, triquetral, lunate, scaphoid, trapezium, trapezoid). Excluding the pisiform and adductor sesamoid, disagreement between observers for the 28 bones was 7.9% (221 occasions) of the observations. Table 4.4 showed that 80.3% of disagreements were − 1 stage and + 1 stage, 17.1% were – 2 stages and + 2 stages and 2.5% were −3 stages and+3 stages. In general, when SAs were not identical, the less experienced observer (OB1) tended to score higher SAs (69.2%). For the total sample, mean SAs obtained from OB1 and OB2 attained identical value with mean differences classified as trivial, except for triquetral (d = 0.450; small), scaphoid (d = 0.390; small), metacarpal (d = 0.364; small) and medial phalange IV (d = 0.201; small).

Mean SAs calculated by the two observers are presented in Table 4.5. The mean SAs derived by the overall inspection (wholeGP) was 16.83 ± 1.30 years and 15.38 ± 1.22 years, respectively for OB1 and OB2 (d = 0.79; moderate mean differences). The bone-by-bone approaches attenuated differences between observers. When using the median, differences between observers resulted in no significant values and were considered as trivial (d = 0.10, for the pre-mature bones only; d = 0.05 for all 30 bones). Finally, independent from the observer, 30-boneGP using the median resulted in higher SAs compared to using the mean: + 0.36 years for OB1; + 0.38 years for OB2. When analyses used pre-mature bones only, mean and median values did not differ (Fig. 4.1 d and e). The correlation coefficient between observers ranged from 0.841 (95% CI: 0.690 to 0.922) to 1.00 with the narrowest overlapping variance occurring when the exams followed bone-by-bone approach and calculations included both pre-mature and mature bones using the mean (30-boneGP-M).

Table 4.3. Inter-observer agreement rates on the SAs according to skeletal classification as not mature or mature and for the total number of examinations among female adolescent soccer players (n = 100).

Bones	Age at mature state[1]	not mature[a]				mature[a]				total			
		n	D (n)	A		n	D (n)	A		n	D (n)	A	
				n	%			n	%			n	%
Radius	17	70	8	62	89%	30	0	30	100%	100	8	92	92%
Ulna	17	52	3	49	94%	48	0	48	100%	100	3	97	97%
Capitate	13	5	2	3	60%	95	2	93	99%	100	4	96	96%
Hamate	13	5	3	2	40%	95	6	89	94%	100	9	91	91%
Triquetral	13	4	4	0	0%	96	6	90	94%	100	10	90	90%
Pisiform	-									100	0	100	100%
Lunate	13	11	7	4	36%	89	0	89	100%	100	7	93	93%
Scaphoid	13	22	19	3	14%	78	0	78	100%	100	19	81	81%
Trapezium	13	17	15	2	10%	83	1	82	99%	100	16	84	84%
Trapezoid	13	5	4	1	20%	95	2	93	98%	100	6	94	94%
Adductor Sesamoid	-									100	0	100	100%
Metacarpal I	15	4	1	3	75%	96	0	96	100%	100	1	99	99%
Metacarpal II	15	26	4	22	85%	74	3	71	96%	100	7	93	93%
Metacarpal III	15	30	8	22	73%	70	0	70	100%	100	8	92	92%
Metacarpal IV	15	30	10	20	67%	70	0	70	100%	100	10	90	90%
Metacarpal V	15	32	13	19	59%	68	0	68	100%	100	13	87	87%
Proximal phalange I	15	53	4	49	92%	47	5	42	89%	100	9	91	91%
Proximal phalange II	15	14	4	10	71%	86	0	86	100%	100	4	96	96%
Proximal phalange III	15	16	9	7	44%	84	0	84	100%	100	9	91	91%
Proximal phalange IV	15	17	6	11	65%	83	0	83	100%	100	6	94	94%
Proximal phalange V	15	17	3	14	82%	83	7	76	92%	100	10	90	90%
Medial phalange II	15	16	8	8	50%	84	0	84	100%	100	8	92	92%
Medial phalange III	15	18	9	9	50%	82	0	82	100%	100	9	91	91%
Medial phalange IV	15	15	5	10	67%	85	1	84	99%	100	6	94	94%
Medial phalange V	15	15	11	4	73%	85	6	79	93%	100	17	83	83%
Distal phalange I	15	65	4	61	94%	35	0	35	100%	100	4	96	96%
Distal phalange II	15	65	4	61	94%	35	2	33	94%	100	6	94	94%
Distal phalange III	15	65	4	61	94%	35	1	34	97%	100	5	95	95%
Distal phalange IV	15	65	4	61	94%	35	1	34	97%	100	5	95	95%
Distal phalange V	15	65	2	63	97%	35	0	35	100%	100	2	98	98%
Total disagreement. n		819	178	641		1981	43	1938		3000	221	2779	
%			21.7%		78.3%		2.2%		97.8%		7.4%		92.6%

[a] skeletal maturity status by observer 2; D (disagreement between observer 1 and observer 2); A (agreement between observer 1 and observer 2); [1] Mature state defined as detailed in Radiograph Atlas of Skeletal Development of the Hand and Wrist.

Table 4.4. Descriptive (mean ± standard deviation) and comparison between observers for bone SAs among female adolescent soccer players (n = 100).

	observer 1 (OB1)	observer 2 (OB2)	Comparisons		disagreement						
					n	magnitude discrepancies[a]					
			d	(qualitative)		-3	-2	-1	+1	+2	+3
Radius	15.6 ± 1.2	15.6 ± 1.2	0.001	(trivial)	8			1	6	1	
Ulna	16.1 ± 1.1	16.0 ± 1.1	0.090	(trivial)	6				5	1	
Capitate	12.9 ± 0.3	12.9 ± 0.3	0.001	(trivial)	5			3	2		
Hamate	12.8 ± 0.6	12.9 ± 0.5	0.180	(trivial)	10	3	1	3	2		1
Triquetral	12.8 ± 0.6	13.0 ± 0.2	0.450	(small)	11		6	3	2		
Pisiform	-	-									
Lunate	12.9 ± 0.4	12.9 ± 0.4	0.001	(trivial)	7			1	5	1	
Scaphoid	12.9 ± 0.4	12.7 ± 0.6	0.390	(small)	20			2	14	4	
Trapezium	12.8 ± 0.6	12.8 ± 0.6	0.001	(trivial)	18	1	2	1	7	7	
Trapezoid	12.9 ± 0.4	12.9 ± 0.3	0.001	(trivial)	6	1	1	1	2	1	
Adductor Sesamoid	-	-									
Metacarpal I	14.9 ± 0.5	14.9 ± 0.4	0.001	(trivial)	2		1	1			
Metacarpal II	14.7 ± 0.6	14.7 ± 0.6	0.001	(trivial)	7		7				
Metacarpal III	14.7 ± 0.6	14.6 ± 0.6	0.168	(trivial)	8				8		
Metacarpal IV	14.8 ± 0.5	14.6 ± 0.6	0.364	(small)	13				12	1	
Metacarpal V	14.7 ± 0.5	14.6 ± 0.7	0.165	(trivial)	13				12	1	
Proximal phalange I	14.4 ± 0.6	14.4 ± 0.7	0.001	(trivial)	9			5	4		
Proximal phalange II	14.8 ± 0.5	14.8 ± 0.6	0.001	(trivial)	4				4		
Proximal phalange III	14.8 ± 0.5	14.7 ± 0.7	0.165	(trivial)	9				9		
Proximal phalange IV	14.8 ± 0.5	14.7 ± 0.7	0.165	(trivial)	9				9		
Proximal phalange V	14.7 ± 0.6	14.7 ± 0.6	0.001	(trivial)	11			7	4		
Medial phalange II	14.9 ± 0.4	14.8 ± 0.6	0.197	(trivial)	8				7	1	
Medial phalange III	14.9 ± 0.5	14.8 ± 0.6	0.182	(trivial)	10				9	1	
Medial phalange IV	14.9 ± 0.5	14.8 ± 0.5	0.201	(small)	6			1	4	1	
Medial phalange V	14.8 ± 0.5	14.8 ± 0.5	0.001	(trivial)	10			6	4		
Distal phalange I	14.3 ± 0.6	14.3 ± 0.5	0.001	(trivial)	5			3	2		
Distal phalange II	14.3 ± 0.6	14.3 ± 0.5	0.001	(trivial)	6		1	3	2		
Distal phalange III	14.3 ± 0.6	14.3 ± 0.5	0.001	(trivial)	5		1	2	2		
Distal phalange IV	14.3 ± 0.6	14.3 ± 0.5	0.001	(trivial)	5			3	2		
Distal phalange V	14.3 ± 0.5	14.3 ± 0.5	0.001	(trivial)	3			1	2		
Disagreement. n					234	5	20	47	141	20	1
%						2.1%	8.5%	20.1%	60.2%	8.6%	0.4%

[a]signs refer to OB1 minus OB2 and numbers corresponds to plates from the collection of photographs representing the sequence of skeletal maturation in atlas.

Table 4.5. Descriptive statistics (mean ± standard deviation) of skeletal age estimates obtained from two observers and mean differences among female adolescent soccer players (n = 100).

Criteria						mean difference		magnitude effect	
Reading	processing	calculations	n	Observer 1	Observer 2	value	(95% CI)	d	(qualitative)
Overall	GPwhole		100	16.83 ± 1.30	15.38 ± 1.22	1.45	(1.30 to 1.59)	0.79	(moderate)
Bone-by-bone	30-boneGP	M	100	14.20 ± 0.36	14.18 ± 0.37	0.01	(0.09 to 0.30)	0.05	(trivial)
		Md	100	14.56 ± 0.61	14.56 ± 0.61	0.00	(0.00 to 0.00)		
	GP_{pmb}	M	85#	14.24 ± 0.77	14.17 ± 0.74	0.07	(0.01 to 0.14)	0.10	(trivial)
		Md	85#	14.24 ± 0.76	14.16 ± 0.74	0.00	(0.00 to 0.00)		

GPwhole (30-bone assessment); 30-boneGP (all bones examined), GP_{pmb} (only pre mature bones examined), M (Mean), Md (Median), % 95 CI (95% confidence intervals), df (degrees of freedom); # 15 participants presented all bones as mature; The paired t-test cannot be computed because the standard error of the difference between means is 0.

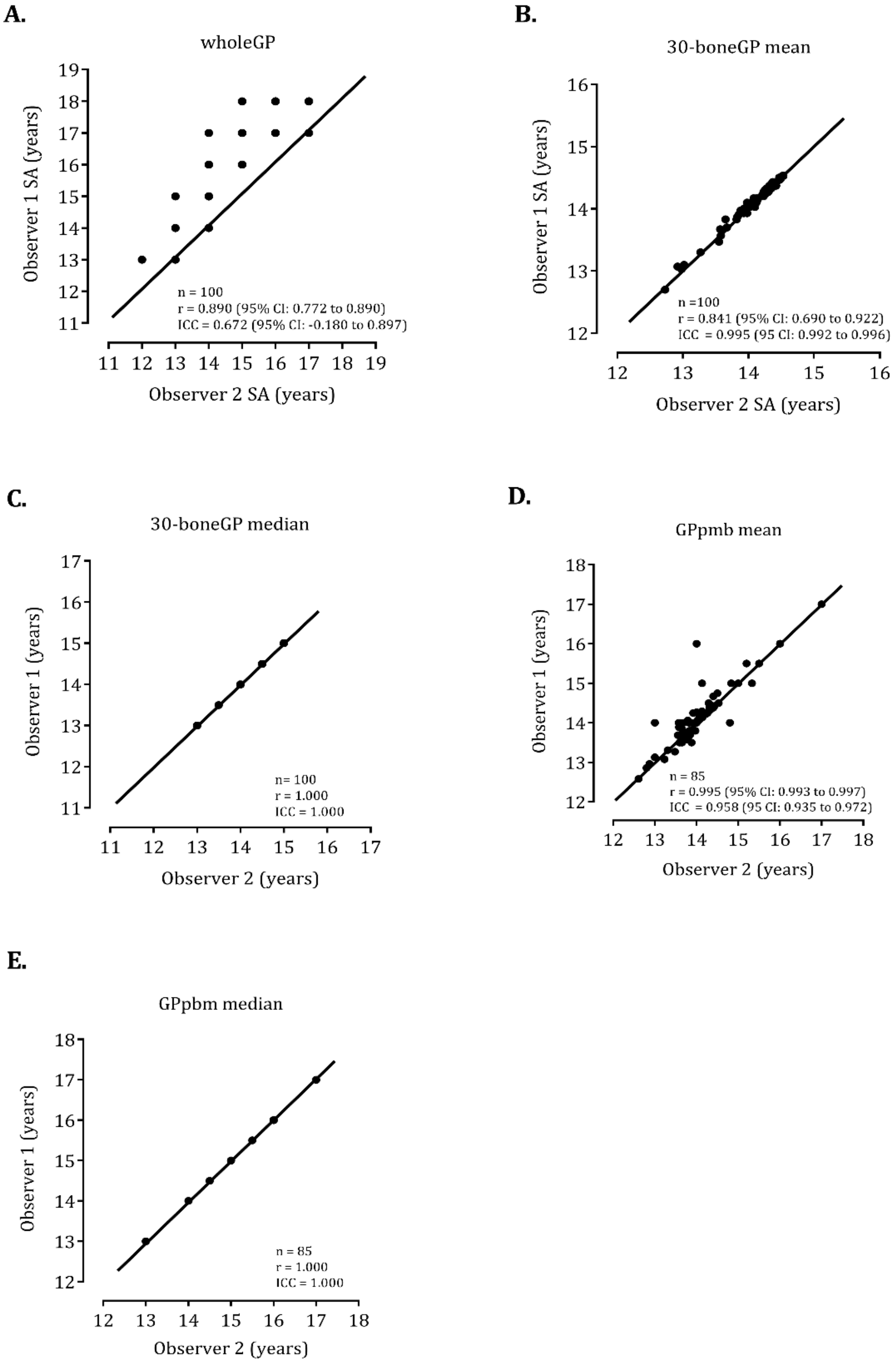

Figure 4.1. Scatterplot of the SA estimated by Observer 1 (y-axis) and Observer 2 (x-axis) in the whole inspection (a), a bone-by-bone approach using the mean (b) and, alternatively, the median (c) to calculate individual SA from all bone-specific SAs; and uniquely considering SAs of the premature bones (d using the mean; e using the median).

4.5. Discussion

Although Greulich-Pyle method has been often used to estimate SA from hand-wrist radiographs, little attention has been given to the impact of adopting different methodological approaches. The current study examined the reproducibility and agreement between two observers who assessed SAs of 100 female adolescent soccer players using the GP protocol. Disagreement between observers mostly occurred on carpal bones. Intra-observer agreement rates were acceptable, although the reproducibility was slightly lower for the less experienced. When differences existed, lower SAs were more likely to be derived in the second time-measurement. Finally, comparison between observers noted that the more experienced observer tended to produce slightly lower SA scores.

A previous study (Roche & Davila, 1976) of North-American children aged 4–15 years noted a lack of intra-observer agreement among carpal bones. It is plausible that agreement rates were associated with age, particularly before round bones (i.e. carpals) reach the mature state (Tanner, Healy, Goldstein, & Cameron, 2001). Carpals are more complex to rate compared to long bones (Roche, 1989), whose examinations concentrate on the centers of ossification and fusion of the epiphyseo-diaphysial. The examination of the carpal bones includes the inspection of the shape and radiopaque lines or zones which may help explain the poorer inter-observer agreement rates in the present study. Although mean differences between examiners in bone-specific SAs tended to be trivial or small, the number of disagreements (> 10%), were particularly apparent in the scaphoid and trapezium, metacarpals IV-V, and proximal phalange V. In general, the less experienced observer overestimated the ratings when compared to the experienced observer. Thus, less experienced examiners may need to adopt a conservative decision (i.e. when unsure match the radiograph to the younger of the two standard plates in the atlas). The literature includes considerable discussion about the exclusion of the carpal bones when a hand-wrist radiograph is assessed to obtain an estimate of SA (Roche, 1989; Tanner et al., 2001; Malina, 2011).

The atlas method involves assigning a SA to each of the 30 bones of the hand-wrist (Greulich & Pyle, 1959). The literature is not consistent regarding the appropriate utilization of the protocol. Consequently, GP SA is often assessed on the basis of an the basis of an overall approach, that is, matching a film ignoring potential variation among bones (Malina, 2011; Malina, Rogol, Cumming, Coelho-e-Silva, & Figueiredo, 2015). The current study examined alternative approaches such as only including 30 bones or pre-mature bones. The exclusion of mature bones is common in the literature (Roche & Johnson, 1969; Roche, Chumlea, & Thissen, 1988). For example, Todd (1937) recommended retention of the most advanced bones when calculating SA. In the present study, within each observer, the SA using the mean did not show fluctuations when considering the mature bones or not. Among Australian females, differences between GP SA mean from all bones and GP SA mean excluding the carpals were 0.02 and 0.05 for 12 and 13-year-old groups, respectively (Roche & Johnson, 1969). In the present study, the inter-individual variance was substantially reduced when the calculations were based on the mean and included mature bones (standard deviation was 0.36–0.37 years, depending of the observer). The largest standard deviation was seen in the overall (wholeGP) approach (standard deviation: 1.22–1.30 years). Recently, the median has been recommended as an alternative to the mean (Malina et al., 2004) to obtain the final SA from bone-specific SAs: "*The SA of the standard plate is the assigned SA of the bone in question. The process is repeated for all bones that are visible in the hand-wrist x-ray and the child's SA is the median of the skeletal ages of each individually rated bone*" (pages 279–280).

There are a few limitations to note. The present study only included adolescent females, many of whom has mature carpals. Future studies need to consider younger samples and males. However, it should be noted that sport tends to focus on the middle and later adolescent years (Malina et al., 2006; Coelho-e-Silva et al., 2008; Coelho e Silva et al., 2010a; Figueiredo et al., 2009), particularly in team sports such as soccer where competitive and organized participation tends to start after 11 years. Future research should focus on whether the impact of observer and methodological approach differs via child maturity status (i.e. early, average or late maturing).

4.6. Conclusions

In summary, the GP method showed acceptable reproducibility and agreement between observers, suggesting that 45-h of training (rating 100 radiographs) is adequate. Where an observer is less experienced, he/she should be encouraged to select the younger age of two standards when the decision is not obvious. The BbB approach has a greater inter-observer agreement compared to the overall approach. Observers should organize their readings following a particular bone, instead of completing the scores by participant. Finally, the estimate of individual SA for each youth participant should use either the mean or the median of the pre-mature bones.

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Chapter V

Study 3

Assessment of skeletal age in youth female soccer players: Agreement between Greulich-Pyle and Fels protocols

5. Assessment of skeletal age in youth female soccer players: Agreement between Greulich-Pyle and Fels protocols

5.1. Abstract

Objective: The purpose of the study was to evaluate the agreement between the Fels and Greulich-Pyle methods for the assessment of skeletal age (SA) in female youth soccer players. Methods: The sample included 441 Portuguese players 10.08–16.73 years of age who regularly participated in organized and competitive soccer. Standardized radiographs of the left hand-wrist were obtained and analyzed by an experienced examiner. SA was estimated with the Fels and Greulich-Pyle (GP) methods. Differences between SA and chronological age (CA) were used to define skeletal maturity groups: late, average and early maturing. In addition to descriptive statistics, Cohen's kappa and Lin concordance correlation coefficients were used to evaluate agreement between methods. Results: Intraindividual differences in SA based on the two methods varied between 0.10 to 1.47 years among age groups with larger mean differences at older ages. Agreement of maturity classifications between methods was 74% at younger ages (under-13: kappa = 0.48; under-14: kappa = 0.39; Lin CCC = 0.68) and declined with increasing CA (under-17: 19% agreement; kappa = 0.001; Lin CCC = 0.11). About 19% of the total sample was skeletally mature with the Fels method and an SA was not assigned; in contrast, no players were skeletally mature with the GP method. Conclusions: GP SAs were systematically lower than Fels SAs among female soccer players. Intraindividual variability in SAs between methods was considerable. The findings highlight the impact of method on estimates of maturity status.

Keywords: Youth sports; Biological Maturation; Bone Age; Puberty; Female Athlete

5.2. Introduction

Physical growth and biological maturation are central to long-term athletic development (LTAD). The former refers to increases in body size, proportions, and composition, while the latter refers to progress towards the biologically mature state, which varies among body systems (Malina, Bouchard, & Bar-Or, 2004). The concept of maturation includes three dimensions, status — state of maturation at the time of observation, timing—ages at which specific maturational events occur, and tempo—rate at which maturation progresses. Skeletal age (SA), colloquially labeled "bone age", is a widely used indicator of maturity status (Malina, Rogol, Cumming, Coelho-e-Silva, & Figueiredo, 2015). It refers to the estimated level of maturity of the bones of the hand and wrist as evident in a standard radiograph relative to reference values for healthy children and youth. Three methods of SA assessment are commonly used, the Greulich and Pyle (1959) method, two versions of the Tanner-Whitehouse (TW) methods (Tanner et al., 1983; Tanner, Healy, Goldstein, & Cameron, 2001) and the FELS method (Roche, Chumlea, & Thissen, 1988). Other methods of SA estimation are available, but are generally limited to specific bones and often to specific clinical contexts (de Sanctis et al., 2014; Korde, Daigavane, & Shrivastav, 2015; Malina, 2017; Satoh, 2015). The Greulich-Pyle (GP) method is used most often in the clinical context.

SA is often used to verify CA in selected youth sport competitions (Malina, 2011). As an example, the stated CAs of 50 soccer players 15–16 years participating in a tournament in Bangkok were compared to their respective GP SAs; 39 players had an SA in excess of the age limit for the competition (Tritrakarn & Tansuphasiri, 1991). More recently, an estimate of SA based upon an magnetic resonance image (MRI) of the fusion of the distal epiphyses radius and ulna has been proposed for CA verification in international youth soccer competitions (Dvorak, George, Junge, & Hodler, 2007; Timme, Steinacker, & Schmeling, 2017). However, use of the MRI protocol for age verification with female soccer players in U16 and U17 competitions years has not been recommended (Tscholl, Junge, Dvorak, & Zubler, 2016).

There is a reasonably extensive literature on the SAs of youth athletes of both sexes in a variety of sports (Malina, 2011). Two earlier studies of youth baseball

(Krogman, 1959) and American football (Rochelle, Kelliher, & Thornton, 1961) used the atlas method of Todd (1937), which was the precursor of the GP method. Subsequent studies of youth athletes of both sexes and in a variety of sports have used the GP, TW, and FELS methods (Malina, 1994; Malina, Coelho-e-Silva, & Figueiredo, 2013). SA data are more available for male than female athletes in several sports with the exception of artistic gymnasts (Malina et al., 2013). Inter-individual variation in biological maturity status is a significant factor affecting the physical characteristics and performances of youth athletes and is also a factor in talent development and selection (Coelho-e-Silva et al., 2010). Maturity-associated variation in body size and composition, strength and performance of young athletes is increasingly a topic in interest in human biology and the sports sciences (Malina et al., 2004). Data for female soccer players are lacking, except for a sample of players from the former Czechoslovakia for whom SA was assessed with a local version of the GP method (Novotny, 1981). Central to many studies are comparisons of youth of the same CA classified as late (delayed), average (on time) or early (advanced) maturing (Coelho-e- Silva et al., 2010; Martinho et al., 2020). In this context, agreement rates of maturity status classifications based on different methods of assessment are of relevance. The relevance also extends beyond growth and performance. For example, after participating in a tournament biobanded for biological maturation, late maturing players from four professional soccer clubs considered the games to be less physically challenging but appreciated having more opportunities to use, develop and demonstrate their technical, physical, and psychological competencies (Cumming et al., 2017).

Concordance of SA assessments based on the GP, TW, and FELS methods was examined in an Italian sample of 171 males and 156 females 1–17 years of age (Aicardi et al., 2000; Vignolo et al., 1992). The results suggested a trend for higher FELS SAs compared to CAs during late adolescent years. FELS SAs were, on average, significantly higher than TW3 RUS SAs (Tanner et al., 2001) among Spanish soccer 12–15 years (Malina, Chamorro, Serratosa, & Morate, 2007). However, among 15-year-old players, 11 of 14 were classified as skeletally mature with the TW3 method compared to only two with the FELS method. By inference, although SAs based on the three methods are related, they are not equivalent.

In the context of the preceding and given the lack of data addressing skeletal maturation among youth female soccer players, the purpose of this study is to compare

GP and FELS SAs in a sample of female adolescent soccer players. A secondary purpose was to compare maturity status classifications of the players (late, average, and early) based on the two methods. It was hypothesized that estimates of SA were specific for each method, which in turn would impact in the distributions of players when classified by skeletal maturity status.

5.3. Materials and methods

Procedures and ethics requirements

The study was approved by the Coimbra University Ethics Committee (Reference: CE/FCDEF-UC/00122014). A signed institutional agreement was also obtained between the University of Coimbra and the Portuguese Institute of Sports and Youth (Ministry of Education). Players were recruited from clubs registered in the Portuguese Soccer Federation. All data were collected at the Porto Sports Medicine Center as part of medical examinations required for registration in the Federation (Law 204/2006; act 11/2012). Examination procedures followed the Declaration of Helsinki produced by the World Medical Association for research with humans. In addition, parents or legal guardians were informed about the aims, protocols and risks of the methodology, and provided informed consent. Assent was obtained from each participant, after information about the nature of the research was provided, and players were able to withdraw from the study at any time. During the visit to the medical unit, a posterior–anterior radiograph of the left hand and wrist with the fingers fully extended was taken by a qualified radiology technician.

Sample

The sample included 441 Portuguese female soccer players 10.08 to 16.73 years of age (14.38 ± 1.24 years). The difference of date of medical examination minus date of birth defined CA for each player. Participants were grouped by CA as follows: under-12 (<12.00), under-13 (12.00 to 12.99), under-14 (13.00 to 13.99), under-15 (14.00 to 14.99), under-16 (15.00 to 15.99), and Under-17 (16.00 to 16.99).

Fels skeletal age (Fels SA)

The posterior–anterior radiographs of the left hand and wrist of each player was evaluated by an experienced assessor (DVM) with both the FELS (Roche et al., 1988) and GP (Greulich & Pyle, 1959) methods to derive a SA with each method. The FELS method requires the evaluation of specific written descriptions of maturational grades for each indicator (the illustrations serve as a guide): radius, ulna, seven carpals, and the metacarpals and phalanges of first, third, and fifth rays. In addition, the method requires the measurement of epiphyseal and metaphyseal widths of the radius, ulna and metacarpals and phalanges of the first, third, and fifth rays. The absence/presence (ossification) of the pisiform and adductor sesamoid is also noted. The grades for each indicator and width measurements are entered into the Felshw 1.0 software (Felshw 1.0, Software Lifespan Health and Research Center, Departments of Community Health and Pediatrics, Booshoft School Medicine, Wright State University Dayton, Ohio) to obtain an estimate of SA and its associated standard error. SAs of a sample of 40 players were assessed on a second occasion within 7 days. The intraobserver technical error of measurement was 0.15 year and the reliability coefficient (Mueller & Martorell, 1988) was 0.99. The intraindividual mean difference and intraclass correlation coefficient were, respectively, 0.04 years (SD = 0.08 years) and 0.988 (95% CI: 0.977–0.994).

Greulich-Pyle skeletal age (GP SA)

The maturity status of each individual bone of the hand and wrist was assessed relative to the sex-specific plates in the Atlas. The SA of the Atlas plate that most closely matched the individual bone under evaluation was the assigned SA of the specific bone. The procedure was repeated for each bone in the radiograph (Roche et al., 1988; Roemmich et al., 1997). The assigned SA for the individual was the median of the SAs of all rated bone. When a specific bone was rated as mature, the CA of the player was accepted the SA of the bone under examination, and was used for the calculation of the overall SA based on the median of the 30 bone-specific SAs. All radiographs were assessed by a single observer (DVM) who was trained by individuals experienced with both the Fels and GP methods (RMM, MJCS).

Intraobserver reproducibility in GP SA assessments were reported elsewhere for 100 female adolescents aged 12.0–16.76 years (Faustino-da-Silva et al., 2020): 2856 agreements among 3000 SA assessments (95.2%).

Maturity status

CA was subtracted from the respective SA (i.e., SA minus CA) with the Fels and GP methods to classify each player as late (SA < CA by more than 1.0 year), on time or average (SA within ±1.0 year of CA), or early (SA > CA more than 1.0 year). The band ±1.0 year approximates the standard deviations for SAs in samples of youth, both nonathletes and athletes (Malina, 2017). If the bones of the hand and wrist had attained the mature state, the individual was simply noted as skeletally mature and SA was not assigned.

Statistical analyses

Descriptive statistics were calculated by competitive age groups for CA, FELS SA, GP SA. Differences between SA and CA with each method and between FELS and GP SA were also calculated. Intraindividual mean differences between FELS and GP SAs were compared by age group using paired t-tests. CAs of the 12 players in the under-12 group spanned 10.1 to 11.9 years, as such, the age group was retained for the descriptive statistics, but was excluded in cross-tabulations of players by skeletal maturity status based on the two estimates of SA. The SAs of individual players with GP and FELS methods were plotted relative to their respective CAs, and bivariate correlation coefficients between SA and CA were calculated. Lin concordance correlation coefficients (Lin CCC) (Lin, 1989) were used to determine the agreement of SAs with the FELS and GP protocols for the total sample and by CA groups. Consistent with previous studies (Malina et al., 2015; Malina et al., 2018), skeletally mature players were not included in analyses requiring SAs. Agreement between maturity status classifications based on the FELS and GP methods was estimated with Cohen kappa coefficients (Viera & Garrett, 2005) in the under-13 through under-17 age groups, and in the total sample. All analyses were performed using SPSS version 20.0 (SPSS Inc., IBM Company, NY) and Graphpad Prism (version 5.00 for Windows, GraphPad Software, San Diego, California, www.graphpad.com).

5.4. Results

Descriptive statistics by CA are summarized in Table 5.1, while corresponding statistics for the difference between SA and CA with each method and between the Fels and GP SAs are summarized in Table 5.2. For each CA group, mean values for Fels SAs were, on average, greater than corresponding means for GP SAs. Standard deviations for GP SAs were systematically lower than corresponding estimates for FELS SAs. Standard deviations for GP SAs declined from under-12 through under-17 players, while those for FELS SAs were reasonably stable (≥1.0 year) except among under-17 players. Frequencies of players classified as mature with the Fels method were as follows: 5 (under-14); 15 (under-15); 29 (under-16); and 34 (under-17). In contrast, no players had a GP SA of 18 years, although the plate for 18 years in the GP Atlas was indistinguishable from that of an adult (Table 5.1).

Table 5.1. Descriptive statistics (means [M], standard deviations [SD], ranges) for CA, Fels SA and GP SA by age group among adolescent female soccer players (n=441) by CA group.

	n	CA (years)		Fels SA (years)				Greulich-Pyle SA (years)		
				not mature			mature			
CA group (years)		M	SD	M	SD	Range	(n)	M	SD	Range
Under-12	12	10.79	0.64	11.31	1.40	(8.58 to 14.35)	0	10.67	1.25	(8.75-14.00)
Under-13	39	12.70	0.29	13.75	1.09	(12.12 to 16.81)	0	13.64	0.76	(12.00-15.00)
Under-14	110	13.56	0.29	14.77	1.32	(11.61 to 17.98)[a]	5	14.17	0.72	(12.00-15.00)
Under-15	151	14.44	0.27	15.38	1.25	(11.72 to 17.78)[b]	15	14.56	0.57	(13.00-15.00)
Under-16	75	15.54	0.25	16.38	1.00	(13.52 to 17.90)[c]	29	14.94	0.23	(14.00-15.00)
Under-17	54	16.33	0.20	17.06	0.71	(15.67 to 17.95)[d]	34	16.00	0.01	(16.00-16.50)

Descriptive statistics for CA (mean±sd) when the skeletally mature players were excluded for the Fels method: [a]CA (13.54±0.30); [b]CA (14.45±0.26); [c]CA (15.52±0.27); [d]CA (16.29±0.22).

Table 5.2. Descriptive statistics (means [M], standard deviations [SD], ranges) for the differences between Fels SA and CA, between GP SA and CA, and between Fels and GP SAs among female adolescent soccer players (n=441) by CA group.

CA group (years)	SA_{FELS} - CA difference (years)[a]				$SA_{Greulich-Pyle}$ - CA difference (years)				SA_{FELS} - $SA_{Greulich-Pyle}$ (years)[a]					
													paired t-test	
	n	M	SD	Range	n	M	SD	Range	n	M	SD	Range	t	p
Under-12	12	0.52	1.20	(-1.50 to 2.42)	12	-0.10	1.03	(-1.33 to 2.07)	12	0.63	0.76	(-0.24 to 2.46)	2.901	0.014
Under-13	39	1.04	1.11	(-0.82 to 3.85)	39	0.93	0.81	(-0.84 to 2.51)	39	0.10	0.69	(-1.71 to 1.81)	0.991	0.328
Under-14	105	1.23	1.24	(-1.78 to 4.04)	110	0.61	0.66	(-1.34 to 1.95)	105	0.65	0.91	(-1.48 to 3.92)	7.495	0.000
Under-15	136	0.93	1.22	(-2.57 to 3.70)	151	0.12	0.56	(-1.57 to 0.99)	136	0.84	0.86	(-1.28 to 2.78)	11.519	0.000
Under-16	46	0.86	1.00	(-2.38 to 2.61)	75	-0.59	0.29	(-1.90 to -0.07)	46	1.47	0.85	(-0.48 to 2.90)	11.607	0.000
Under-17	20	0.77	0.70	(-0.70 to 1.85)	54	-0.34	0.20	(-0.73 to -0.02)	20	1.06	0.72	(-0.33 to 1.95)	6.531	0.000

[a]Skeletally mature players are not included in the analysis.

SAs of individual players with each method are plotted relative to CA in Figure 5.1. Allowing for the small sample of under-12 players, the majority (seven of 12) have a FELS SA that was higher than CA (Figure 5.1A). The distribution of FELS SAs highlighted the trend for advanced SA relative to CA. The trend in the distribution of GP SAs relative to CA (Figure 5.1B) is affected by the scale. According to GP method, SAs approximated the whole year or the mid-year of the Atlas plates; as such the median of the 30-bone SAs for an individual player was not a continuous variable. In the FELS method, SA was expressed using decimal numbers as for CA.

Differences between FELS SA and CA were, on average, systematically larger than corresponding mean differences between GP SA and CA in each age group (Table 5.2). Standard deviations of the difference between FELS SA and CA were also larger than corresponding estimates for GP SA minus CA. The mean difference between Fels and GP SAs is, on average, lowest among under-13 players (0.10 year) and largest among under-16 players (1.47 years), and the mean differences increase linearly from under-13 to under-16 players. No players were classified as mature with the GP method, while 83 players (19%) were skeletally mature with the FELS method.

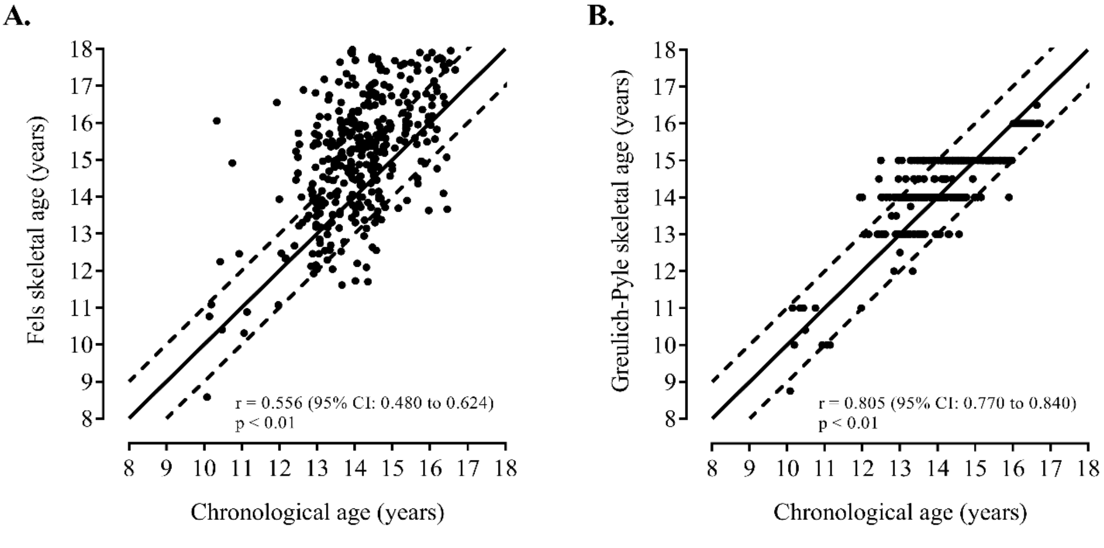

Figure 5.1. Individual skeletal ages (SAs—years) of female soccer players by (a) Fels (n = 358) and (b) Greulich-Pyle (n = 441) protocols plotted relative to respective chronological ages (CAs—years).

Cross-tabulations of skeletal maturity status based on FELS and GP SAs are summarized in Table 5.3. Agreement of maturity status classifications for the total sample of players was moderate (46%) and significant (K = 0.15, p = 0.000; Lin CCC = 0.67). Concordance of classifications based on the kappa statistic was highest among under-13 (74%; K = 0.48, p = 0.002) and under-14 (65%; K = 0.39, p = 0.000) players, but systematically declined and was not significant among under-15, under-16 and under-17 players. The trend was generally similar for Lin CCCs, that is, highest among under-13 and under-14 players, and declined with age. The five skeletally mature under-14 players with the FELS method were classified as early maturing with the GP method; in contrast, all skeletally mature under-15, under-16, and under-17 players with the Fels method were classified as average with the GP method.

Table 5.3. Concordance of maturity status classifications based upon GP and Fels SAs within CA groups and in the total sample of female soccer players (n = 441), kappa coefficients and concordance correlation coefficients (Lin CCCS) and respective 95% confidence intervals.

		Maturity status							
		Greulich-Pyle				Total	Agreement		Lin CCC[#] (95%CL)
	Fels	Late	On time	Early	Mature		%	Kappa (p)	
Under-13	Late	**0**	0	0	0	0	74	0.48*	0.68
(12.00-12.99)	On time	0	**16**	5	0	21		(0.002)	(0.24; 0.89)
	Early	0	5	**13**	0	18			
	Mature	0	0	0	**0**	0			
	Total	0	21	18	0	39			
Under-14	Late	**0**	4	0	0	4	65	0.39*	0.73
(13.00-13.99)	On time	1	**39**	1	0	41		(0.000)	(0.57; 0.83)
	Early	1	26	**33**	0	60			
	Mature	0	0	5	**0**	5			
	Total	2	69	39	0	110			
Under-15	Late	**4**	2	2	0	8	44	0.08*	0.53
(14.00-14.99)	On time	2	**61**	0	0	63		(0.004)	(0.43; 0.61)
	Early	2	61	**2**	0	65			
	Mature	0	15	0	**0**	15			
	Total	8	139	4	0	151			
Under-16	Late	**1**	1	0	0	2	25	-0.01	0.45
(15.00-15.99)	On time	4	**18**	1	0	23		(0.060)	(0.37; 0.52)
	Early	0	21	**0**	0	21			
	Mature	0	29	**0**	**0**	29			
	Total	5	69	1	0	75			
Under-17	Late	**0**	0	0	0	0	19	0.01	0.11
(16.00-16.99)	On time	0	**10**	0	0	10		(1.000)	(0.05; 0.17)
	Early	0	10	**0**	0	10			
	Mature	0	34	0	**0**	34			
	Total	0	54	0	0	54			
Total †	Late	**6**	7	2	0	15	46	0.15*	0.67
(10.08-16.73)	On time	9	**148**	7	0	164		(0.000)	(0.62; 0.71)
	Early	3	127	**49**	0	179			
	Mature	0	78	5	**0**	83			
	Total	18	360	63	0	441			

[a]SA value was not assigned for female adolescent soccer players classified as mature and, consequently they were not considered in Lin CCC analysis. [b]Includes 12 players aged <12.00 years; Lin CCC (Lin concordance correlation coefficients); 95% CL (95% confidence limits).
*($p < 0.01$).

5.5. Discussion

Both the GP and FELS methods were based on sample of American children and youth of European ancestry (American Whites) from upper and middle classes families in the state of Ohio, specifically Cleveland in the north of the state for the GP method and the central region of the state for the Fels method (Greulich & Pyle, 1959; Roche et al., 1988). It has also been suggested that the samples used to develop the respective methods may no longer be considered representative of the U.S. population (Satoh, 2015). However, estimated ages at peak height velocity (PHV) for girls and boys in the Brush Foundation Study (Sanders et al., 2017) and in the Fels Longitudinal Study (Guo, Siervogel, Roche, & Chumlea, 1992; Thissen, Bock, Wainer, & Roche, 1976) were generally consistent with contemporary and more recent longitudinal studies of US and European youth (Malina et al., 2004). Allowing for the small sample of under-12 players in the present study, GP SAs were, on average, in advance of CA among mid-adolescent players, but lagged behind CA among late adolescent players. The present study also indicated a trend for FELS SAs to be in advance of GP SAs in all CA groups. The trend in mean differences between Fels and GP SAs, that is, Fels SA > GP SA contrasted observations for the FELS Longitudinal Study sample among whom GP SAs were, on average, in advance of FELS SAs at 12 through 15 years (Chumlea, Roche, & Thissen, 1989). The differences likely reflect variation in the application of the GP method (see below).

The FELS method was developed on radiographs of participants in the Fels Longitudinal Study who were followed from the 1930s through the 1980s. The method requires specific grades (maturity indicators) for the radius and ulna, seven carpals (the pisiform is simply noted as not ossified [absent] or ossified [present]), and for the metacarpals and phalanges of the first, third and fifth digits. Each bone has a variable number of indicators and each indicator is assigned using specific grades. Redundant indicators were excluded in an effort to reduce the amount of information to be assessed. Linear measurements of the epiphyseal and diaphyseal widths of long bones are also required.

Meantime, the GP atlas includes plates (standards) representing boys and girls resident in the city of Cleveland who were followed from 1931 through 1942 (Greulich & Pyle, 1959). The protocol includes sex-specific plates for boys and girls from birth to maturity. Although the method should be applied by assigning a SA to each individual bone of the hand-wrist, it is commonly applied by matching the radiograph of the child “as a whole” to the reference plates and the assigned SA is the reference plate to which the radiograph most closely matches (Creo & Schwenk II, 2017; De Sanctis et al., 2014; Korde et al., 2015; Martin et al., 2011; Mughal, Hassan, & Ahmed, 2014). As such, variation among individual bones is overlooked. Based on the reproducibility of bone-specific comparisons in 100 female players 12.0–16.7 years of age, agreement rates were >90%, except for proximal phalanx I, 88%

(Faustino-da- Silva et al., 2020). Unfortunately, the GP (1959) Atlas failed to indicate whether the assigned SA should be based on the mean or the median of the SAs assigned to the 30 bones. By inference, the impact of using different criteria to determine GP SAs needs further study, for example, using all bones or only those bones that are not mature. In the present study, the GP method was applied by examining all bones, that is, the SA of the plate to which each bone in the radiograph most closely matched was the SA of the bone. The protocol was repeated for all bones of the hand-wrist. During late adolescent years, interindividual variation is largely concentrated on developmental stages of the radius, ulna, metacarpals and phalanges, and using the mean may be a critical decision.

None of the players was classified as skeletally mature with the GP protocol. On the other hand, 83 players were mature with the FELS method (Roche et al., 1988). Table 5.4 summarizes the number of participants in the present study classified as mature for carpals bone using the GP method. A large percentage of players in the current sample already attained the bone-specific mature state for the carpals with the GP method. However, instructions in the Atlas are not specific about how should be used the information from mature bones in the calculation of individual SAs. As such, the determination of SA would be affected if the calculation is limited to the premature bones (the illustration corresponding to GP mature state was not attained). If both premature and mature bones are supposed to be included in the calculations, the mean of the 30 bone-specific SAs would be affected, specifically when introducing the age at mature state in the Atlas for players who already attained mature state (See Table 5.4). As such, it was decided to use the median of all bone-specific SAs instead of the mean as the final GP SA.

Table 5.4. Age at mature state for each individual carpal bones according to Greulich-Pyle protocol and number of female soccer players classified as mature.

Carpal	Age of mature state in Atlas[a] (years)	Mature state attained	
		(n)	%
Capitate	13.0	412	93
Hamate	13.0	407	92
Triquetral	13.0	404	92
Pisiform[b]	9.5	441	100
Lunate	13.0	274	62
Scaphoid	13.0	317	72
Trapezium	13.0	351	80
Trapezoid	13.0	404	92
Adductor sesamoid[b]	11.0	441	100

[a] Maximum skeletal age to be assigned according to Greulich & Pyle protocol.
[b] For pisiform and adductor sesamoid assessment was absent or present and this was noted for all subjects and all participants in the current study.

Relatively few studies have specifically addressed the growth and maturity characteristics of female soccer players; the majority of studies of youth players are focused on fitness, training and match play, and on injuries (Malina, Martinho, Valente-dos-Santos, Coelho-e- Silva, & Kozieł, 2021). As noted earlier, comparative data on the skeletal maturation of female players are limited to a sample from the former Czechoslovakia (Novotny, 1981). Using a local modification of the GP method, mean SAs and CAs of 23 soccer players did not differ 13 years (CA: 13.5 ± 0.9; SA: 13.0 ± 1.0 years) and also at 16 years (CA: 16.8 ± 0.9, SA: 16.8 ± 0.7 years). In the present study, GP SA at 13 years was, on average, in advance of CA, while GP SA lagged behind CA at 16 years (Table 5.1). Corresponding results were generally similar for 12 Czech volleyball players at 14 years (CA: 14.2 ± 0.9; SA: 14.2 ± 1.2 years) and 17 years (CA: 17.0 ± 0.5; SA: 16.7 ± 1.0 years). The majority of data addressing SA in female athletes are limited to artistic gymnastics, swimming and track and field (Malina, 2011). In contrast, predicted estimates of maturity status are increasingly used in recent studies of female soccer players (Malina et al., 2021).

Bio-banding is a current version of maturity-matching in youth sports (Malina et al., 2019). It specifically refers to the process of grouping athletes for competitions on the basis of estimated biological maturity status rather than chronological age groups (Cumming, Lloyd, Oliver, Eisenmann, & Malina, 2017). Percentage of predicted adult stature attained at the time of observation is the most used indicator for bio-banding (Malina et al., 2019).

Sport is selective and exclusive, especially during adolescence (Coelho-e-Silva et al., 2010), and selectivity in many sports is related in part on maturity status. The topic has received more attention in boys than girls. According to bio-banding strategies, restricting maturity associated variation in size, by inference in strength and motor performance, contributes to greater competitive equity and, potentially, enhances the overall development of youth players. A recent application of the protocol evaluated the reactions of male players who participated in "bio-banded tournaments." For example, in a tournament limited to players aged 11–14 years whose heights at the time of observation were between 85% and 90% of their respective predicted adult heights (Cumming et al., 2017), those players classified as advanced in predicted maturity status described the games as a more challenging and imposing greater physical demand compared to age group competitions. In contrast, late maturing players described the tournament as providing a greater opportunity to demonstrate and develop their technical, physical and psychological attributes, to exert their influence on the game, and to adopt positions of leadership. The bio-banding protocol is still in an experimental stage and results of applications in female players have not been published to date.

Many national soccer federations generally follow similar developmental tracks for both male and female players. In Portugal (Federação Portuguesa de Futebol, 2020), co-ed participation is permitted in the under-13 (11–12 years), under-15 (13–14 years), and under-17 (15–16 years) competitive age groups, while sex-specific participation in competitive soccer in limited to under-19 players and seniors. As a result, female players aged 11–16 years often train and compete with males or may request re-classification as juniors. Both strategies, however, are potentially associated with reduced opportunities to play and perhaps increased risk for injury among female players.

Research is needed to support a long-term developmental model for girls participating in soccer, particularly decisions related to age-associated variation in recommended age at beginning competition, ball size, field and goal dimensions, structure of the matches (two halves or four periods as in basketball), transition from 7-player into 11-player soccer, specialization by playing position, international competition, and clarification of the definition of senior level. It is well documented that females, on average, attain the adult state (adult stature, skeletal, and sexual maturity) before males (Malina et al., 2004) and this should perhaps be considered by federations regarding ages for the transition to senior level. In the context of the latter, SAs with the FELS method are not assigned to players who attained skeletal maturity. The latter included five under-14 players and the percentage of skeletally mature players increased with CA: 9.9% (under-15), 38.7% (under-16), and 63.0% (under-17).

The current study was based on a large cross-sectional sample of adolescent female soccer players from all districts of Portugal. This is a major strength of the study as data addressing the skeletal maturity status of female participants in team sports are lacking. The data are, of course, limited to soccer so that generalizations to female athletes in other sports should be made with caution. The analysis is also limited to the GP and FELS methods. Future analyses should consider assessments of SA with the different versions of the TW method, 20-bone SA, Carpal SA and RUS SA (Tanner et al., 1983; Tanner et al., 2001; Tanner, Whitehouse, & Healy, 1962). As often noted, SA assessments are considered invasive, given the exposure to a low dose of radiation, and costly (Malina et al., 2015; Rogol, Cumming, & Malina, 2018). In this context, there is a need for further research to addressing the relationship between estimates of SA based on the GP, FELS, and TW methods of assessment and noninvasive estimates of maturity status in female athletes. Such data are limited to studies of Fels SA and percentage of predicted adult height attained at the time of observation similar to studies of male youth participants in American football (Malina, Dompier, Powell, Barron, & Moore, 2007) and soccer players (Malina, Coelho-e-Silva, Figueiredo, Carling, & Beunen, 2012).

In summary, GP SAs were systematically lower than FELS SAs in the present sample of female soccer players. Intraindividual differences in SA based on the two methods fluctuated from 0.10 to 1.47 years among age groups with largest mean differences at older ages. Given the relatively large number of players classified as skeletally mature with the FELS method, many CA eligible players for youth soccer competitions would likely be questioned by sport authorities if SAs are used for CA verification (Malina, 2011). The findings have implications beyond soccer and perhaps beyond youth sports (Malina et al., 2007). Nevertheless, generalization of the current findings to other sports and to the general populations warrants care.

5.6. References

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Chapter VI

Study 4

Body size, fatness and skeletal age in female youth soccer

6. Body size, fatness and skeletal age in female youth soccer

6.1. Abstract

Background: Growth and maturation are crucial aspects to adequately organize training and competitions in youth sports. Aim: This study was aimed to examine maturity associated variation in body size and adiposity among adolescent girls participating in competitive youth soccer by age group. Methods: The sample included 441 players aged 10.08-16.73 years. Stature and body mass were measured and body composition estimated. Fels method was used to determine skeletal age (it was not assigned for mature participants). Skeletally maturity was derived from differences between chronological and skeletal ages as follows: late, average or early maturing. Results: Mean stature approximated the 50th percentiles of the general population at all competitive age groups, while mean weight fluctuated between 50th and 75th percentiles. Age-and maturity-specific means for fat mass ranged 18.0%-28.2%. The number of players classified as skeletally mature increased with competitive age groups (under-13: 0%; under-15: 8%; under-17: 49%). Conclusion: In general, early maturing girls tended to be heavier than respective peers of the same competitive age group, particularly when compared to the late maturing group.

Keywords: Stature; Body mass; Youth Sports; Female Athlete; Puberty

6.2. Introduction

In most European countries youth regularly participate in sport (Telama & Yang, 2000). The topic is relevant in terms of public health because sport is the most visible form of health enhancing physical activity among children and adolescents. The Portuguese Active Healthy Kids Report Card concluded that children and adolescents did not reach sufficient physical activity levels and compared to recommendations, Portuguese youth spent large amounts of time sedentary (Mota, Coelho e Silva, Raimundo, & Sardinha, 2016). The contribution of organized sport participation to daily energy expenditure was examined in youth from the United States (Katzmarzky & Malina, 1988). Authors used a 3-day diary and concluded that girls and boys expended, respectively, 20% and 16% of total daily energy expenditure in organized sports. Depending on age group, in Portugal sport was estimated to make up 11-13% of total daily energy expenditure (derived from accelerometry combined with a 3-day diary) which corresponded to 35-42% of the moderate-to vigorous physical activity (Machado-Rodrigues et al., 2012).

Among 12,568 Portuguese youth aged 10-18 years, soccer was shown to be the most popular sport among boys, while swimming and soccer were the most popular among girls (Seabra, Mendonça, Thomis, Malina, & Maia, 2007). The number of registered female players in FIFA (Fédération Internationale de Football Association) member countries approximated 1,356,000 and about 70% of female players were <18 years of age (UEFA, 2017). In Portugal, youth soccer players are grouped as infantiles or under-13 (aged 11-12 years), initiates or under-15 (aged 13-14 years), juveniles or under-17 (aged 15-16 years), juniors or under-19 (aged 17-18 years) and senior (19+ years) (Federação Portuguesa de Futebol, 2020). The infantiles, initiates and juveniles combine boys and girls for competition. Sex-specific games and competitions occur only at older ages (i.e. for juniors and seniors). Many female players aged 11-16 years often train and compete with males or, alternatively, request authorization to "move-up", that is, to play at junior level even though their age correspond to initiates or juveniles. Both strategies are potentially associated with reduction in opportunities for females to play and perhaps increased risk for injury (Ostenberg, Roos, Ekdahl, & Roos, 1998).

A predictor of physical activity and sports participation is biological maturity status (Sherar & Cumming, 2020). From a biological perspective, adolescence includes two major events: sexual maturation often assessed by pubic hair (PH) and/or gonadal development; and the adolescent growth spurt (Malina, Bouchard, & Bar-Or, 2004). Youth enter this crucial period at varying ages (differential timing) and proceed through it as variable velocities (differential tempo). Although both timing and tempo show substantial inter-individual variability, in general, girls mature earlier than boys (Malina et al., 2004). Longitudinal analysis of male youth soccer players from the Ghent Youth Soccer Project shows well-defined adolescent spurts in fitness test performance (Philippaerts et al., 2006). Studies addressing the growth and maturity characteristics of female soccer players, however, are relatively limited. Several studies (Düppe, Gärdsell, Johnell, & Ornstein, 1996; Södermann, Bergström, Lorentzon, & Alfredson, 2000; Pettersson, Nordström, Alfredson, Henriksson-Lársen, & Lorentzon, 2000) examined the interrelationship among height, weight, bone mass and muscle strength, but few examined maturity status of female players (Plaza-Carmona et al., 2016; Lozano-Berges et al., 2019). One fairly old study (published in Czech), assessed skeletal age (SA) of adolescent female athletes from several sports including 23 soccer players (Novotny, 1981). The present study aimed to assess growth status and skeletal maturity among female youth soccer players spanning under-13 through under-17 years. Within each competitive age group (under-13, under-15, under-17), body size and composition were compared among players of contrasting skeletal maturity status.

6.3. Methods

Procedures

The current research proposal was conducted in accordance with established ethical procedures (Harriss, MacSween, & Atkison, 2019). The project was approved by the Coimbra University Ethics Committee (CE/FCDEF-UC/00122014) and included a formal agreement between the University of Coimbra and the Portuguese Institute of

Portuguese Institute of Sports (IPDJ/FCDEF.UC/2017-01). All participants visited the Porto unit of Sports Medicine for a medical examination as part of the registration with the Portuguese Soccer Federation (Law 204/2006; act 11/2012). In addition to the medical examination, a battery of anthropometric dimensions was measured, and a radiograph of the left hand-wrist was obtained for the determination of SA. Parents or legal guardians provided written informed consent. Individual assent was obtained after players were informed about objectives and that participation was voluntary and they could withdraw from the study at any time.

Participants

The sample included 441 female soccer players 10.08-16.73 years of age who were registered with the Portuguese Soccer Federation. Chronological age (CA) was defined as the difference between date of the clinical examination and the date of birth (recorded as decimal age in years). Players were grouped as infantiles (under-13), initiates (under-15) or juveniles (under-17).

Skeletal age

Posteroanterior radiographs of the left hand and wrist of each player were taken by a trained and qualified technician. The distal forearm, palm and fingers were placed flat on the radiograph plate. SA was estimated using the Fels method (Roche, Chumlea, & Thissen, 1988), which considers specific indicators for each of the 22 bones of the hand-wrist: the radius, ulna, the seven carpals (capitate, hamate, triquetral, lunate, scaphoid, trapezium, trapezoid), pisiform, adductor sesamoid, metacarpals I, III and V, proximal phalanges I, III and V, medial phalanges III and V, and distal phalanges I, III and V. Maturity indicators for each bone include the presence or absence of the ossification center in the radiograph, changes in shape of the carpals and epiphyses of the long bones, ratios of epiphyseal and diaphyseal widths, and capping and fusion of the long bones. Assigned grades and width measurements were subsequently entered into the software (Felshw 1.0, Wright State University, Dayton Ohio, USA) to derive an SA and its standard error for each individual. The readings were performed by a single trained observer. A sample of 40 players was assessed on a second occasion within 7 days. Intra-observer technical error of measurement was 0.15 years and the reliability coefficient (Mueller & Martorell, 1988) was 0.99. The intra-individual mean difference and intraclass correlation coefficient were, 0.04 years (standard deviation=0.08 years) and 0.988 (95%CI: 0.977 to 0.994), respectively.

Anthropometry

A single qualified and experienced technician completed all assessments. Stature was measured using a stadiometer (Harpenden stadiometer, model 98.603, Holtain Ltd, Crosswell, UK) to the nearest 0.1cm. Body mass was measured using a digital scale (SECA balance, model 770, Hanover, MD, USA) to the nearest 0.1 kg. Skinfolds at the triceps and medial calf sites were measured with a Lange caliper (Beta Technology, Ann Arbor, MI, USA) to the nearest 0.5 mm. The protocols followed the guidelines of the international standardized reference (Lohman, Roche, & Martorell, 1988). A subsample of players (n=13) were measured on two occasions to estimate intraobserver technical errors of measurement (Malina & Hamill, 1973): stature (0.37 cm), sitting height (0.71 cm), body mass (0.56 kg) and skinfolds (0.47-0.72 mm); the errors were well within the range of intra- and inter-observer errors in several surveys in the United States and a variety of field studies, including studies of young athletes (Malina, 1995). Relative fat mass was estimated using the Equation 1 (Slaughter et al., 1998). Absolute fat mass (FM) and fat-free mass (FFM) were derived.

$$\%\ \text{FM} = 0.610 \times (\text{triceps skinfold} + \text{medial calf skinfold}) + 5.1 \qquad \textbf{(2.1)}$$

Analyses

Descriptive statistics (means, standard deviations) were calculated by age groups (under-13 through under-17) for CA, SA, SA minus CA, stature, body mass and estimated FM and FFM. The stature and body mass of individual players were plotted relative to U.S. reference data (Kuczmarski et al., 2002). The maturity status of each player was estimated from SA minus CA: players with a SA younger than CA by more than 1.0 year were classified as late maturing; players with a SA in advance of CA by more than 1.0 were classified as early maturing; finally, players with a SA within -1.0

year to +1.0 year of CA were classified as average maturing or on time. SA was not assigned for skeletally mature players. Descriptive statistics for early, average and late maturing players were calculated within competitive age groups, and analysis of variance also used to compare maturity groups. In addition, effect size correlations (ES-r) were determined and interpreted as follows (Hopkins, Marshall, Batterham, & Hanin, 2009): trivial (<0.100), small (0.100 to 0.299), moderate (0.300 to 0.499), large (0.500 to 0.699), very large (0.700 to 0.899), nearly perfect (0.900 to 0.999). When only two groups are under examination, t-test for independent samples was used and effect size estimated using Cohen d-values interpreted as follows (Hopkins et al., 2009): trivial (<0.200), small (0.200 to 0.599), moderate (0.600 to 1.199), large (1.200 to 1.999) and very large (2.000 to 3.999). Statistical analyses were done with SPSS version 20.0 (SPSS Inc, IBM Company, N.Y., USA) and Graphpad Prism (version 5.00 for Windows, GraphPad Software, San Diego California USA, www.graphpad.com).

6.4. Results

Stature and body mass of individual players were plotted by CA relative to U.S. references (Kuczmarski et al., 2002) for females in Figure 6.1 panels A and B, respectively. At younger ages, under-13 participants were between 10th and >90th percentiles for stature and body mass. Subsequently, about equal numbers of players under-15 and under-17 years had statures <10th and >90th percentiles. Proportionally more players under-13 and under-17 had body mass >90th percentiles compared to the proportion of players with body mass below 10th percentiles. Mean statures and body masses were plotted by CA group relative to the reference percentiles in Figure 6.2 (panels A and B). Mean statures of the female soccer players approximated the CA-specific reference medians whereas mean body masses varied between the 50th and 75th percentiles.

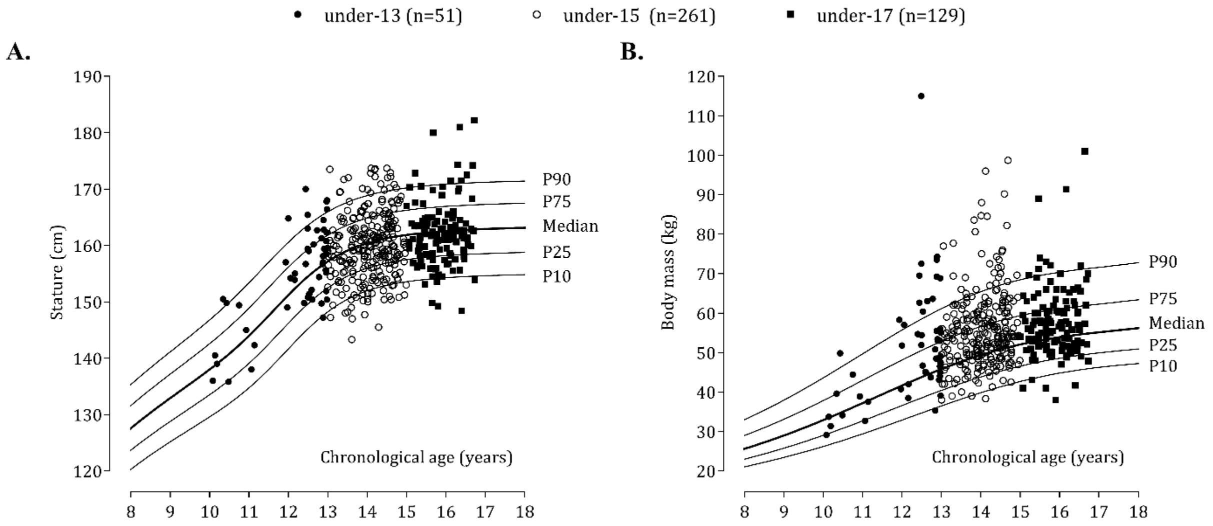

Figure 6.1. Statures (panel A) and body masses (panel B) of individual female soccer players plotted relative to U.S. percentiles (n=441).

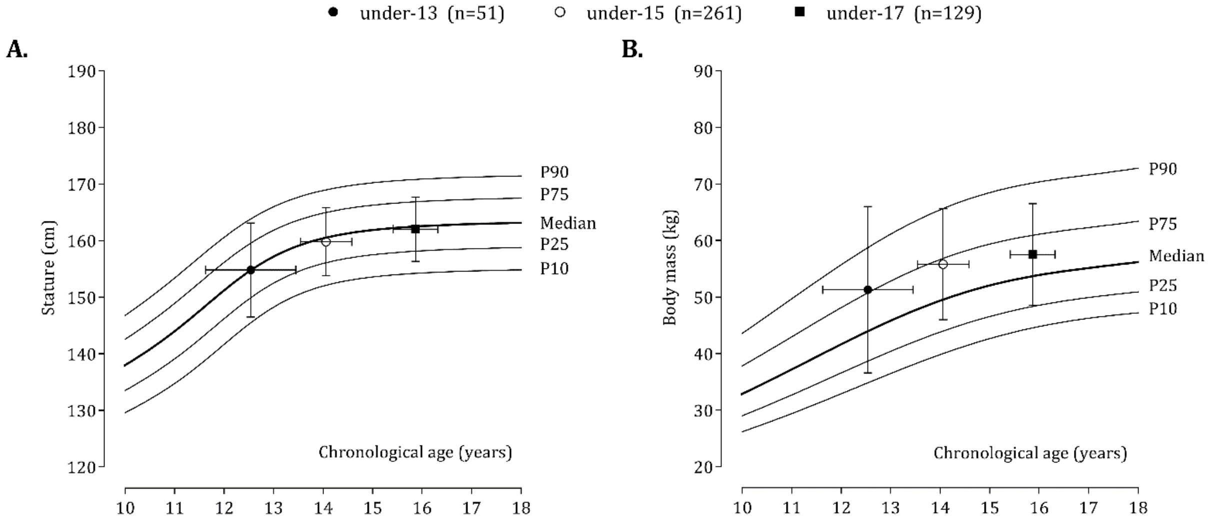

Figure 6.2. Mean stature (panel A) and mean body mass (panel B) of the competitive age-groups (under-13, under-15 and under-17) plotted relative to U.S. percentiles (n=441).

Descriptive statistics by competitive groups were summarized in Table 6.1. The number of skeletally mature players increased with CA, especially among under-17 players (n=63; corresponding to 49%). As expected, under-17 players were older, advanced in SA (F=99.382, p<0.01; ES-r=0.60), taller (F=24.036, p<0.01; ES-r=0.31) and heavier (F=6.720, p<0.01; ES-r=0.17) compared with younger groups. Adiposity-related variables, in contrast, did not differ among competitive age groups with mean values for % fat mass ranging 25.3%-26.5%. Late maturing players were minimally represented in the three competitive age groups; the majority were average or advanced in skeletal maturity status. Comparisons of players by maturity status within the competitive age groups were summarized in Table 6.2. Among under-13 participants, early maturing players were moderately heavier (t=2.783, p <0.01, d=0.79) and fatter (% fat mass: t=-4.586, p<0.05, d=0.72; fat mass in kg: t=-5.482, p<0.05, d=0.77) than players classified as average. At older ages (under-15 and under-17), mean differences in anthropometric variables of players contrasting in maturity status were small or trivial.

Table 6.1. Descriptive statistics and distributions of players by skeletal maturity status within competitive age groups, analyses of variance for age-associated variation in skeletal age, stature, body mass and estimated body composition among adolescent female soccer players.

Variable	Units	Under-13 (n=51)		Under-15 (n=261)		Under-17 (n=129)		ANOVA (analysis of variance)			
		n	M ± Sd	n	M ± Sd	n	M ± Sd	F	p	ES-r	(qualitative)
Chronological age	Years	51	12.54±0.91	261	14.06±0.52	129	15.87±0.45				
Skeletal age†	Years	51	13.17±1.56	241	15.12±1.32	66	16.59±0.97	99.382	<0.01	0.60	(moderate)
SA-CA†	Years	51	0.92±1.15	241	1.06±1.24	66	0.84±0.92	1.064	0.346	0.24	(small)
Skeletal maturity §											
late	F	1		12		2					
average	F	27		104		33					
early	F	23		125		31					
mature	F	0		20		63					
Stature	Cm	51	154.8±8.3	261	159.8±6.0	129	162.0±5.7	24.036	<0.01	0.31	(moderate)
Body mass	Kg	51	51.3±14.7	261	55.8±9.8	129	57.5±9.0	6.720	<0.01	0.17	(small)
Fat mass	%	51	25.7±6.7	261	26.0±6.5	129	25.3±6.6	0.569	0.57	0.05	(trivial)
	Kg	51	13.9±8.3	261	14.9±6.2	129	14.9±6.3	0.572	0.57	0.05	(trivial)
Fat-free mass	Kg	51	37.4±7.5	261	40.9±5.3	129	42.6±4.7	16.454	<0.01	0.26	(small)

M (mean); Sd (standard deviation); ES-r (effect size-correlation); SA (skeletal age); CA (chronological age)
† (Skeletally mature players are not included); § (Chi-square test: $\chi 2 = 112.30$, $p < 0.01$).

Table 6.2. Descriptive statistics (mean ± standard deviation) and multiple comparisons among players by skeletally maturity groups within competitive age groups.

			skeletal maturity status				comparison of mean differences					
			Maturing				test			magnitude effect		
Age group	Variable	units	late	Average	early	Mature	statistics	value	p	effect size	value	(qualitative)
			(n=1)	(n=27)	(n=23)	(n=0)						
Under-13	Chronological age	years	#	12.29±0.89	12.30±0.85							
(n=51)	Skeletal age	years	#	12.53±1.17	14.13±1.12		t	-4.704	<0.01	d	1.58	(large)
	SA-CA	years	#	0.24±0.76	1.82±0.79		t	-7.158	<0.01	d	2.08	(very large)
	Stature	cm	#	154.0±9.5	156.6±5.7		t	-1.167	0.25	d	0.40	(small)
	Body mass	kg	#	46.7±10.5	57.1±16.3		t	-2.783	<0.01	d	0.79	(moderate)
	Fat mass	%		23.7±5.5	28.2±7.3		t	-4.586	<0.05	d	0.72	(very large)
		kg		11.3±4.9	17.2±10.3		t	-5.842	<0.05	d	0.77	(very large)
	Fat-free mass	kg		35.4±7.0	40.4±7.0		t	-2.555	<0.05	d	0.73	(very large)
			(n=12)	(n=104)	(n=125)	(n=20)						
Under-15	Chronological age	years	14.10±0.47	14.10±0.56	14.01±0.50	14.22±0.34						
(n=261)	Skeletal age¶	years	12.58±0.69	14.33±0.84	16.01±0.87		†$F_{(2,238)}$	165.107	<0.01	ES-r	0.76	(very large)
	SA-CA¶	years	-1.52±0.68	0.22±0.56	1.99±0.74		†$F_{(2,238)}$	293.449	<0.01	ES-r	0.84	(very large)
	Stature	cm	160.2±7.5	160.4±6.0	159.5±5.9	158.2±5.6	‡$F_{(3,257)}$	0.866	0.45	ES-r	0.10	(small)
	Body mass	kg	48.9±6.4	55.5±10.6	56.3±9.1	58.1±9.4	‡$F_{(3,257)}$	2.623	0.05	ES-r	0.17	(small)
	Fat mass	%	22.3±3.5	26.0±6.7	26.1±6.4	27.6±7.1	‡$F_{(3,257)}$	1.745	0.16	ES-r	0.14	(small)
		kg	11.0±2.8	14.9±6.9	15.1±5.9	16.4±6.1	‡$F_{(3,257)}$	2.031	0.11	ES-r	0.15	(small)
	Fat-free mass	kg	38.0±4.4	40.5±5.6	41.3±5.1	41.7±5.6	‡$F_{(3,257)}$	1.771	0.15	ES-r	0.14	(small)
			(n=2)	(n=33)	(n=31)	(n=63)						
Under-17	Chronological age	years	15.52±0.52	15.75±0.43	15.75±0.43	16.00±0.45						
(n=129)	Skeletal age¶	years	13.60±0.12	16.06±0.58	17.34±0.46		§t	-9.751	<0.01	d	2.48	(very large)
	SA-CA¶	years	-1.92±0.64	0.29±0.41	1.59±0.45		§t	-12,012	<0.01	d	3.07	(very large)
	Stature	cm	157.8±4.7	161.7±6.0	162.1±4.5	162.3±6.1	♦$F_{(2,124)}$	0.095	0.91	ES-r	0.04	(trivial)
	Body mass	kg	44.3±8.9	54.4±5.8	57.3±6.6	59.7±10.1	♦$F_{(2,124)}$	4.122	<0.05	ES-r	0.25	(small)
	Fat mass	%	18.0±5.9	23.6±6.0	25.3±5.9	26.4±6.9	♦$F_{(2,124)}$	1.995	0.14	ES-r	0.18	(small)
		kg	8.3±4.2	12.9±3.9	14.8±4.9	16.2±7.5	♦$F_{(2,124)}$	3.239	<0.05	ES-r	0.22	(small)
	Fat-free mass	kg	36.1±4.7	41.4±4.9	42.5±3.4	43.4±5.0	♦$F_{(2,124)}$	1.980	0.14	ES-r	0.18	(small)

¶ (SA was not assigned for players classified as mature) ; † (multiple comparisons between late, average and early maturing under-15 female soccer players); ‡ (multiple comparisons between late, average, early maturing and mature under-15 female soccer players); § (since late maturing group was composed of two players, comparisons were only tested between average and early maturing under-17 female soccer players using t-test); ♦ (since late maturing group is uniquely composed of two players, comparisons were tested between average, early maturing and mature under-17 female soccer players using analysis of variance).

6.5. Discussion

The aim of this manuscript was to examine skeletal maturity variation in body size and estimates of body composition among Portuguese adolescent female soccer players Mean statures tended to approximate the reference medians for the general population, while mean weights varied between the medians and 75 percentiles. The mean statures of under-17 Portuguese players were slightly shorter than reported in a recent review of heights and weights of female soccer players from several countries spanning 1992 through mid-2020 (Malina, Martinho, Valente-dos-Santos, Coelho-E-Silva, & Kozieł, 2021). A report based on a representative sample of 22,948 Portuguese youth (Sardinha et al., 2011) stated that 21.6% of girls were classified as overweight or obese. In the present study mean values for stature and body mass of soccer players approximated the general Portuguese population (Santos et al., 2014), Azores Islands (Coelho-E-Silva et al., 2013) and Madeira Islands (Freitas et al., 2012) as presented in Table 6.3. In other words, female soccer did not seem to be particularly ‘selective’ in terms of height and weight. The current results suggested that Portuguese female soccer players tended to have elevated weight-for-height. The average %FM of the present cohort was 25% which is towards the upper end of the range (13.6%-29.1%) reported in a review of older adolescent and young adult female soccer players (Martínez-Lagunas, Niessen, & Hartmann, 2014). However, direct comparisons are affected by varied measures and protocols for estimating fat mass and interpretation needs caution.

Table 6.3. Mean values for stature and body mass in the current study among school girls in Portugal (Santos et al. 2014), Azores (Coelho-e-Silva et al. 2013) and Madeira (Freitas et al. 2013).

Body size	group	current study				Santos et al. (2014)*			Coelho-e-Silva et al. (2013)			Freitas et al. (2012)		
		CA (years)	n	mean	standard deviation	n	mean	d	n	mean	d	n	mean	d
Stature	under-13	12.54	51	154.8	8.3	1189	151	0.46	121	154.8	0.00	88	154.0	0.10
	under-15	14.06	261	159.8	6.0	1186	159	0.13	120	159.2	0.10	89	160.5	0.12
	under-17	15.87	129	162.0	5.7	1414	160	0.35	113	161.6	0.07			
Body mass	under-13	12.54	51	51.3	14.7	1189	46.4	0.33	121	50.7	0.04	88	46.3	0.34
	under-15	14.06	261	55.8	9.8	1186	54.1	0.17	120	56.3	0.05	89	55.2	0.06
	under-17	15.87	129	57.5	9.0	1414	56.2	0.14	113	59.1	0.18			

d (the size of the mean differences between the current study and studies of the general population was expressed divided by the standard deviation); Values were interpreted as follows (Hopkins et al., 2009): d<0.20 (trivial), 0.20≤d≤0.59 (small), 0.60≤d≤1.19 (moderate), 1.20≤d≤1.99 (large), 2.00≤d≤3.99 (very large), ≥4.00 (nearly perfect). *(The study reported means without decimal places for stature); CA (chronological age).

SAs of the female soccer players based on the Fels method were, on average, in advance of CAs across the competitive groups. Comparative data on the biological maturity status of female youth soccer players is limited. Based on an adaptation of the Greulich-Pyle, an early study in the former Czechoslovakia (Novotny, 1981) noted that CAs and SAs of 23 players did not differ at 13 years (CA: 13.5±0.9; SA: 13.5±1.0 years) and 16 years (CA 16.8±0.9, SA 16.8±0.7 years). Data are a bit more extensive for the sexual maturity status of female players. Stages of puberty were generally used to classify relatively small samples of players (n=20 to 40) as pre-, peri- or post-pubertal independently of CA (Plaza-Carmona et al., 2016; Lozano-Berges et al., 2019; Lyle, Sigward, Tsai, Pollard, & Powers, 2011) or the distributions of stages were simply described (Nicolao, Pedrinelli, Zogaib, Orbetelli, & Neto, 2010; Lozano Berges et al., 2017). With one exception, self-assessment was used, and stages of breast and pubic hair (PH) development were commonly combined. However, it should be noted that stages of breast and PH development are not equivalent (Malina et al., 2004; Sherar, Baxter-Jones, & Mirwald, 2004).

Several studies have reported ages at menarche, an indicator of maturity timing, in late adolescent and young adult soccer players (Malina et al., 2021). Menarcheal status, i.e., menarche has or has not occurred, was obtained during the medical examination for 297 players in the present study. Unfortunately, the number of participants aged 10 and 11 years was limited (n=8, all pre-menarcheal) so a probit analysis was not feasible. In contrast, the overwhelming majority of older players were post-menarcheal: 12 years (77%), 13 years (79%), 14 years (86%), 15 years (98%) and 16 years (97%). Though limited, the distribution suggested a trend towards somewhat earlier sexual maturation in the players which was generally consistent with the retrospective average age at menarche (12.4±1.3 years) of a sample of Portuguese university students aged 18-23 years surveyed between 1995 and 2001 (Padez, 2003) and a probit average age of 12.5±1.3 years based on a sample of Portuguese girls aged 9-19 years in 1990-1991 (Padez & Rocha, 2003).

As noted before, youth soccer participation in Portugal are organized by two-year CA groups, and only two competitive CA groups are isolated to females only juniors (under-19) and seniors (19+ years). As a result, co-participation with boys is the solution for many young girls to train and compete at under-13, under- 15 and under-17 levels. Many girls request to play up as a Junior. In this context, a comparison of the growth and maturity status of females in the present study and Portuguese male soccer players (Figueiredo, Goncalves, Coelho E Silva, & Malina, 2009; Malina, Coelho e Silva, Figueiredo, Carling, & Beunen, 2012) at different competitive CA levels is potentially of interest and summarized in Table 6.4. In all studies, SA was assessed with the Fels method. Though limited to few studies, the results suggested the following. Among under-13 players, females aged 12.5 years were, on average, slightly taller (+3.7 cm) and substantially heavier (+8.2kg) than males aged 12.3 years, and were advanced in SA relative to CA. There were proportionally more early maturing players among females and proportionally more late maturing players among males at these ages. Meantime, among under-15 players, both females and males were, on average, advanced in skeletal maturity status, but males emerged as +4.3 cm taller than females with late maturing players being underrepresented in both sexes at this particular competitive age groups. Twenty female initiates (under-15) were skeletally mature while no male players attained skeletal maturity. Finally, among under-17 players, proportionally more females (49%) than males (16%) were skeletally mature. Sex differences were discussed for both non-mature and mature juveniles (under-17 players) with mean values of the females in the current study, on average, shorter (non-mature: -11.7 cm; mature: -9.3 cm) and lighter (non-mature: -8.7 kg; mature: -10.3 kg) compared to Portuguese male soccer players of the same CA group. The preceding should be viewed in the context of sex differences in the timing of the adolescent spurt. Allowing girls than in boys. The mean statures of under-17 female players overlapped an for the cross-sectional nature of the Portuguese youth soccer studies, data confirms take-off and age at peak height velocity occurring about two years earlier in estimate derived from 25 samples of players 17-18 years of age, 166.1±2.5 cm, while the mean weight of non-mature players (55.4 kg) was lower and that of skeletally mature under-17 players (59.7 kg) was similar to the estimated mean weight for the 17-18 year-old players: 60.6±2.5 kg (Malina et al.,2021).

Table 6.4. Distributions of players by skeletal maturity status within competitive age groups and descriptive statistics (means ± standard deviations) for chronological age, skeletal age, height and weight among Portuguese female (this study) and male youth soccer players (Figueiredo et al., 2009; Malina et al. 2012). Statistics for under-15 and under-17 are presented for non- mature and skeletally mature players.

	Females									Males								
Group	N	CA	SA	Skeletal maturity status				Stature (cm)	Weight (kg)	n	CA	SA	Skeletal maturity status				Stature (cm)	Weight (kg)
				L	A	E	M						L	A	E	M		
Under-13	51	12.5±0.9	13.2±1.6	1	27	23	0	154.8±8.3	51.3±14.7	87	11.8±0.5	12.0±1.4	17	45	25	0	144.6±6.7	38.1±6.2
										63	12.3±0.5	12.5±1.3	13	37	13	0	151.1±7.5	43.1±7.0
Under-15	241†	14.1±0.5	15.1±1.3	12	104	125	20	160.0±6.1	55.6±9.8	93	14.1±0.5	14.7±1.1	4	55	34	0	164.4±9.1	54.5±9.4
	20¶	14.2±0.3	§					158.2±5.6	58.1±9.4	29	13.7±0.7	14.3±1.2	2	16	11	0	162.8±7.5	52.5±8.7
Under-17	66†	15.8±0.4	16.6±1.0	2	33	31	63	161.8±5.3	55.4±6.6	36	15.8±0.4	16.7±1.0	1	14	21	7	173.5±5.7	64.1±5.3
	63¶	16.0±0.5	§					162.3±6.1	59.7±10.1	7¶	16.1±0.2	§					171.6±6.8	70.0±8.7

CA (chronological age); SA (skeletal age); L (late); A (average); E (early); M (mature); † (non-mature); ¶ (mature); § (SA was not assigned for players classified as mature).

Means for estimated ages at peak height and peak weight velocities were summarized elsewhere (Malina et al., 2004) for nine longitudinal studies in the general population. Age at peak weight velocity (PHV) (ranged 12.1-12.9 years and always occurred 0.3 to 0.9 years later than age at PHV in the respective studies. Physical performance (sprinting, agility, jumping) of female soccer players aged 12-21 years has been reported to improve, on average, until approximately 15-16 years (Vescovi, Rupf, Brown, & Marques, 2011). Of potential relevance, menarche occurs about one year after PHV (Malina et al., 2004). Unfortunately, longitudinal data (including data on average age at PHV) among Portuguese female soccer players are not available. Age at PHV for small samples of girls participating in athletics, rowing and mixed sports were, on average, in the range of ages at PHV in the general population (Malina, Rogol, Cumming, Coelho e Silva, Figueiredo, 2015) an observation mirrored in male athletes (soccer, ice hockey, basketball, rowing, cycling, athletics). In contrast, estimated ages at PHV for artistic gymnasts of both sexes tended to occur at older CA. However, many of the studies did not include young adolescents (i.e. <11 y) and were of relatively short duration so that ages at PHV could not be estimated in some individuals, specifically those who were early and late maturing (Philippaerts et al., 2006; Malina et al., 2015). In the present study, there were fewer late maturing players across all CA groups with the majority of under-15 and under-17 players assessed as advanced in skeletal maturity or already skeletally mature.

Maturity status related dropout needs to be carefully studied and claims for longitudinal data of female soccer players. Many female adolescent soccer players in the present study experienced “playing up” to avoid co-participation in training session and competitions with males. Future research should explore anonymous interviews to understand whether adolescent females playing soccer in teams combining boys and girls perceived the experience as positive. The impact of playing and competing with older females as an alternative to training and competing with males of the same age also needs attention. For example, a recent study (Cumming et al., 2018) investigated experiences of male Premier League Academy soccer players aged 11-14 playing in a bio-banded tournament. Participants described the experience as positive and recommended the Premier League to integrate biobanding into the existing games. Early maturing players found that they had to adapt their style of play placing a greater emphasis on technique and tactics. In parallel, late maturing players appreciated having more opportunities to develop technical, physical, and psychological abilities.

The current study is not without limitations. Although the sample is relatively large, numbers are somewhat scarce at the younger ages. The sample is also cross-sectional which does not support generalizations, especially regarding the adolescent growth spurt. %FM was derived from two skinfolds which is not the best indirect measurement. Given advances in technology, future studies should consider alternative protocols to provide estimates of body composition, i.e., bioelectrical impedance, air displacement plethysmography and dual x-ray absorptiometry.

The increasing number of girls participating in soccer requires research addressing the growth and maturity characteristics of female youth soccer. This is clearly lacking compared to corresponding information already available for male youth soccer. The present study evidenced that Portuguese female soccer players have heights and weights similar to reference data and slightly higher percentage body fat compared to adolescent athletes. Throughout the age groups the players were more likely to be early maturing, and this became particularly evident at older competitive years. Body size, composition, biological maturation, in parallel to training, diet and habitual physical activity, have implications in female youth soccer, particularly to understand dropout, sport selection, injury risk and developmental changes.

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Chapter VII

Study 5

Scaling left ventricular mass in adolescent female soccer players

7. Scaling left ventricular mass in female adolescent soccer players

7.1. Abstract

Aim: The aim of the study was to examine the contribution of chronological age (CA), skeletal maturation, training experience and concurrent body size descriptors, to inter-individual variance in left ventricular mass (LVM) among female adolescent soccer players. Methods: The sample included 228 female soccer players 11.8-17.1 years. Training experience defined as years of participation in competitive soccer (range 2-9 years), was obtained by interview. Stature, body mass and skinfolds (triceps, medial calf) were measured. Fat mass was estimated; Fat-free mass was derived. LVM was assessed by echocardiography. Skeletal maturity status was as the difference of skeletal age (SA, Fels method) minus CA. Results: Fat-free mass was the most prominent single predictor of LVM (R^2=36.6%). It was associated with an allometric coefficient close to linearity (k=0.924, 95%CI: 0.737 to 1.112). A significant multiplicative allometric model including body mass, fat-free mass, CA, training experience and skeletal maturity status was also obtained (R=0.684; R^2=46.2%). Conclusion: Stature has limitations as a valid size descriptor of LVM. Body mass, fat-free mass, training experience, CA, body mass and skeletal maturity status were relevant factors contributing to inter-individual variability in LVM.

Keywords: Youth Sport; Female Athletes; Growth; Cardiac Function; Skeletal Age; Scaling

7.2. Introduction

Growth refers to changes in body size, and adolescence is the interval of major changes in height, mass, proportions and composition (Malina, Bouchard, & Bar-Or, 2004). The adolescent changes also influence the growth of specific organs, which in turn affects function. Cross-sectional (Janz, Dawson, & Mahoney, 2000) and longitudinal (Dekkers, Treiber, Kapuku, Van Der Oord, & Snieder, 2002; Sabo, Yen, Daniels, & Sun, 2014) studies have reported that left ventricular mass (LVM) increases during childhood through adolescence. Because age-associated variation in cardiac dimensions is due, in part, to growth related increments in body size, LVM is routinely expressed relative to stature, mass or body surface area (BSA) (George, Sharma, Batterham, Whyte, & McKenna, 2001; Dewey, Rosenthal, Murphy, Froelicher, & Ashely, 2008; Pressler et al., 2012). Systematic training for specific sports during childhood and/or adolescence may influence left ventricular wall thickness (LVWT) and/or increments in the left ventricular cavity. These training adaptations may lead to challenges in diagnosing conditions such as dilated cardiomyopathy or hypertrophic cardiomyopathy (Dewey et al., 2008).

The aforementioned structural and functional adaptive changes to the left ventricle have been labelled “athlete’s heart” (Fagard, 1996) with LVM being the most common indicator of these cardiac adaptations (Scharhag, Schneider, Urhausen, Rochette, & Kinderman, 2002; Haykowsky, Samuel, Nelson, & La Gerche, 2018). Chronic volume loads generally result in an increase in end-diastolic diameters and by inference in LVM; these consequently contribute to eccentric hypertrophy (Demirelli et al., 2015). Intra- and inter-individual variability in cardiac variables in general and LVM in particular are associated with participation in sport but observed changes vary with type of sport (Golbidi & Laher, 2012). It is also suggested that cardiac dimensions are associated with metabolically active tissues, mainly fat-free mass (FFM) (George, Birch, Pennell, & Myerson, 2009; Giraldeau et al., 2015; Kooreman et al., 2019). Among 73 male roller hockey players 14-16 years of age, for example, estimated FFM was the best single predictor of inter-individual variance in LVM (Valente-dos-Santos et al., 2013); however, the results also suggested that biological maturity status should also be considered alongside stature (the traditional size descriptor) to index LVM.

On the other hand, there is evidence that fat mass (FM) is also an independent and positive predictor of LVM in children and adolescents not engaged in youth sport (Chinali et al., 2006; Dai, Harrist, Rosenthal, & Labarthe, 2009).

Historically, ratio standards have been frequently used to interpret physiological and morphological dimensions among individuals, including athletes, who vary in body size and composition. Stature (cm) and BSA (cm^2) are, respectively, linear and bi-dimensional, while body mass and FFM are tri-dimensional variables. Allometric models have been suggested as an effective option for partitioning the effects of body size in order to derive a “size free” (dimensionless) expression of physiological parameters, e.g., maximum oxygen uptake in liters (Nevill, 1992; Nevill & Holder, 1994) or LVM expressed in grams (Dewey et al., 2008; Valente-dos-Santos et al., 2013; de Simone et al., 1992). Since variation in body mass and composition is associated with growth, maturity status and also systematic training (Malina et al., 2004; Malina, 2011; Malina, Figueiredo, & Coelho-e-Silva, 2017) proportional allometric models have been recommended among adult males and females (Nevill & Holder, 1994). Studies of youth athletes, particularly male hockey players, have addressed the independent and combined effects of variables such as chronological age (CA), maturity status (skeletal age, SA), and training experience with one or more body size descriptors (usually stature, body mass, FFM) on peak oxygen uptake (Valente-dos-Santos et al., 2013) and LVM (Valente-dos-Santos, 2013). However, studies of LVM relative to body size have focused on adults and male adolescent athletes. Data are still lacking for female adolescent athletes.

The adolescent growth spurt differentially impacts attained stature and mass in youth of both sexes. Peak height velocity (PHV) occurs, on average, 2 years earlier in girls than in boys and tends to be less intense in girls (Malina et al., 2004). Growth during the adolescent spurt has a marked impact on sex differences in body mass and composition. This is perhaps most marked in the linear increase in FFM among boys during adolescence, while the corresponding adolescent increase in FFM tends to reach a plateau in girls in association with a linear increase in adipose tissue. Therefore, it may be hypothesized that allometric models based on samples of male adolescent athletes may not be generalized to explain intra- and inter-individual variation in LVM among adolescent females. In this context, the objective of the present study was to examine the contribution of CA, skeletal maturity status, training experience and body size descriptors to inter-individual variability of LVM among adolescent female adolescent soccer players using an allometric modelling approach.

7.3. Methods

Procedures

The research was approved by the Ethics Committee of the University of Coimbra and a signed institutional agreement with the Portuguese Institute of Sports. Participant voluntarily visited the Center for Sports Medicine as part of the required medical examination for registration in the Portuguese Soccer Federation (Law 204/2006; act 11/2012). Parents or legal guardians and the players provided written consent; the players were informed that their participation was voluntary and that they could withdraw from the study at any time. During the visit to the medical Center, a radiograph of the left hand-wrist was taken for the purpose of SA estimation, echocardiography was conducted, and a series of anthropometric dimensions were measured. Each of the protocols was conducted by qualified Center personnel in the respective domains.

Participants

The sample included 228 female soccer players 11.8– 17.1 years (14.6 ± 1.1 years). All players were registered in competitive clubs affiliated with the Portuguese Soccer Federation. Inclusion criteria were engagement in formal training and competition for at least one complete year, Caucasian ethnicity, no symptoms of underlying cardiovascular disease, and no family history of cardiovascular related mortality. Training experience was expressed as years of participation in competitive soccer at the club level, including registration with the Portuguese Soccer Federation. Individual information was obtained by interview on the day of observation and confirmed in consultation with institutional records of the Federation.

Chronological age (CA) and maturity status

CA was calculated as the difference between date of the clinical examination and date of birth. SA was estimated with the Fels method (Roche, Chumlea, & Thissen, 1988), which includes maturity indicators for each of the 22 bones of hand-wrist and ratios of epiphyseal-diaphyseal widths. Grades and measurements for each indicator were entered into the Felshw 1.0 software (Felshw 1.0, Software Lifespan Health and Research Center, Departments of Community Health and Pediatrics, Booshoft School Medicine, Wright State University Dayton Ohio, USA) to derive an estimate of SA and the associated standard error. The same trained observer assessed all radiographs. The maturity status of each individual was subsequently classified (Malina, 2011) as late (SA younger than CA by more than 1.0 year), average or on time (the difference between SA and CA was within the band of − 1.0 years to + 1.0 years), early (SA older than CA by more than 1.0 years), or mature (SA is not assigned).

Anthropometry

Body dimensions were measured following standardized procedures (Lohman, Roche, & Martorell, 1988). Stature was measured to the nearest 0.1 cm using a stadiometer (model 98.603, Holtain Limited Crosswell, Crymych, UK) and body mass was measured to the nearest 0.1 kg using a digital scale (SECA, model 770, Hanover, MD, USA). Skinfold thickness was measured to the nearest 0.5mm at two sites, triceps and medial calf using a Lange caliper (Beta Technology Incorporated Cambridge, Maryland, USA). Body fat (fat mass, FM) as a percentage of body mass (%FM) was estimated from the two skinfolds using equation 7.1 recommended for female adolescents of White/European ancestry (Slaughter et al., 1988). Absolute FM and FFM were derived.

$$\%\,FM = 0.610 \times (\text{triceps skinfold} + \text{medial calf skinfold}) + 5.1 \qquad \textbf{(7.1)}$$

Echocardiography

Resting echocardiographs were taken with a Vivid 3 ultrasound machine with a 1.5 to 3.6 MHz transducer (GE Vingmed Ultrasound, Horten, Norway). Two-dimensional images (recorded at 100mm/s) were used to derive M-mode echocardiograms for direct visualization. Measurements of the internal dimension of the left ventricle at end diastole (LVIDd), septal wall thickness at end diastole (SWTd), and posterior wall thickness at end diastole (PWTd) were made following the procedures of the American Society of Echocardiography. Intra-observer technical errors of measurement and variability based on echocardiograms of 20 randomly selected adolescents measured twice within a one-week interval were previously reported (Castanheira et al., 2017). Technical errors and 95% confidence levels were: LVIDd, 0.17 mm (95% LOA, 1.95–2.28 mm, %CV = 0.3, 95% LOA: 4.1–4.8%); SWTd, 0.02 mm (95% LOA, 0.30–0.34 mm, %CV = 0.3, 95% LOA, 4.2–4.8%); and PWTd, 0.06 mm (95% LOA, 0.45–0.56 mm, %CV = 0.8, 95% LOA, 6.5–8.1%). LVM was estimated using equation 7.2 (Devereux et al., 1986) and relative wall thickness (RWT) was calculated using equation 7.3 (Lang et al., 2005):

$$\mathrm{LVM} = 0.8 \times \{(1.04\ [\mathrm{LVIDd} + \mathrm{PWT} + \mathrm{SWTd})^3 - (\mathrm{LVIDd})^3]\} + 0.6 \qquad \textbf{(7.2)}$$

$$\mathrm{RWT} = (2 \times \mathrm{PWTd}) / \mathrm{LVIDd} \qquad \textbf{(7.3)}$$

Analyses

Descriptive statistics were calculated and normality of distributions checked. Pearson correlations were used to estimate relationships among CA, SA and training experience in years, on one hand, and body size descriptors (stature, body mass and FFM) and echocardiographic parameters, on the other hand. Pearson correlations were also used to examine associations between the body size descriptors and parameters of LVM (simple and derived variables). Magnitude of the correlation coefficients was interpreted as follows (Hopkins, Marshall, Batterham, & Hanin, 2009): trivial ($r < 0.10$), small ($0.10 \leq r < 0.30$), moderate ($0.30 \leq r < 0.50$), large ($0.50 \leq r < 0.70$), very large ($0.70 \leq r < 0.90$) and nearly perfect ($r \geq 0.90$). Simple allometric models following procedures proposed by Nevill et al. (1992) and Nevill and Holder (1994) were subsequently applied to the total sample:

$y = a \times x^{k} \times \varepsilon$ **(7.4)**

$\ln y = \ln a + k \times \ln x + \ln \varepsilon$ **(7.5)**

Equation 7.5 corresponds to the natural logarithmic transformation of equation 7.4. It permitted the determination of the constant and power function for each size descriptor. In both equations, y corresponded to LVM, while a and k were, respectively, the constant and scaling exponents. Simple allometric models were validated by the inspection of the correlations between scaled LVM and the respective independent variables (size descriptors). The influence of size descriptors was removed when the coefficients of correlation approached zero. Finally, multiplicative allometric models were derived by combining size descriptors (stature, body mass, FFM), CA, years of training and skeletal maturity status (coded as dummy variables; the 65 skeletally mature participants were not considered in the simple and multiplicative allometric models). Backward stepwise multiple regression with $p < 0.10$ as the criteria for removal was used to develop a parsimonious model. This procedure reduces collinearity among independent variables. Diagnostic statistics to evaluate the proportion of variability in an independent variable that was not explained by the other independent variables (tolerance) were used to examine multicollinearity for the final models. The variance inflation factor (VIF) was also calculated. Variables were excluded if tolerance was ≥0.1 and VIF was > 10 (to an R^2 of 0.90). For each allometric model, the coefficient of determination (R^2) was calculated to estimate the explained variance.

$\ln \text{LVM} = k_1 \times \ln(\text{stature in cm}) + k_2 \times \ln(\text{body mass in kg}) + k_3 \times \ln(\text{FFM in kg}) + a + b_1 \times (\text{CA in years}) + b_2 \times (\text{training experience in years}) + b_3$ (maturity status: late vs average; late vs early, with late maturing being zero) $+ \ln \varepsilon$ **(7.6)**

Differences between skeletal maturity groups in size descriptors (stature, body mass, FFM) and in absolute and scaled values of LVM were graphically compared. The magnitude of mean differences between maturity groups was interpreted using Cohen's d value as follows (Hopkins et al., 2009): < 0.20 (trivial), 0.20–0.59 (small), 0.60–1.19 (moderate), 1.20–1.99 (large), 2.00–3.99 (very large), > 4.00 nearly perfect.

Statistical analyses were done with SPSS version 20.0 (SPSS Inc., IBM Company, N.Y., USA) and Graphpad Prism (version 5.00 for Windows, GraphPad Software, San Diego California USA, www.graphpad.com). Alpha level was set at 0.05.

7.4. Results

Descriptive statistics for training experience, CA, SA, stature, body mass, BSA, body composition and echocardiographic parameters are summarized in Table 7.1. The distribution of players by maturity status (SA minus CA) was also indicated. CA was significantly correlated with stature ($r = 0.19$, $p < 0.05$), BSA ($r = 0.21$, $p < 0.01$), body mass ($r = 0.18$, $p < 0.05$), FFM ($r = 0.21$, $p < 0.01$) and LVM ($r = 0.13$, $p < 0.05$), but the correlations were low. Mean SA was advanced, on average, by approximately 0.65 year, relative to mean CA. SA was moderately correlated with FFM ($r = 0.39$, $p < 0.01$), BSA ($r = 0.41$, $p < 0.01$) and body mass ($r = 0.41$, $p < 0.01$). Correlations between training experience and several variables were lower but significant: negative with %FM ($r = -0.15$, $p < 0.05$); positive for cardiac variables: ISWTd ($r = 0.28$, $p < 0.01$), PWTd ($r = 0.25$, $p < 0.01$), LVM ($r = 0.22$, $p < 0.01$) and the LVM index ($r = 0.29$, $p < 0.01$). Means for LVIDd, ISWTd and PWTd were 43.5 mm, 7.6mm and 7.5 mm, respectively, in the skeletally mature players.

Correlations between size descriptors and dependent variables varied from moderate to large (Figure 7.1 a-c). Accordingly, simple allometric models between logarithmic transformations were calculated using stature, body mass and FFM as size descriptors to obtain dimensionless models aimed to explain inter-individual variability of LVM (Table 7.2). The allometric coefficients explained 16 to 37% of variance in LVM showing a linear relationship between LVM and FFM ($k = 0.924$, 95%CI: 0.737 to 1.112). Power function exponents for stature ($k = 1.930$, 95%CI: 1.240 to 2.620) and body mass ($k = 0.688$, 95%CI: 0.536 to 0.840) were, respectively, above and below, the unit corresponding to linearity. Finally, correlations between scaled variables and LVM were negligible for all size descriptors, suggesting that the simple allometric models were effective to evaluate LVM independent of body size (Figure 7.1 d-f).

Table 7.1. Descriptive statistics and correlations between chronovariables, size and echocardiograph parameters (n=228).

Variable	unit	descriptive statistics					normality (Kolmogorov-Smirnov)		correlations					
		range (min; max)	Mean		standard deviation	f			CA		SA		training experience	
			value	(95% CI)			value	*p*	r	p	r	p	r	p
Training experience	years	(2; 9)	5.3	(5.0 to 5.5)	2.1		0.149	<0.01						
Chronological age	years	(11.84; 17.05)	14.63	(14.49 to 14.77)	1.11		0.076	<0.01						
Skeletal age*	years	(11.46; 17.92)	15.28	(15.04 to 15.53)	1.54		0.067	0.07						
Skeletal maturity:														
delayed	f					25								
average	f					51								
advanced	f					87								
mature	f					65								
Stature	cm	(136.0; 182.2)	161.3	(160.4 to 162.2)	6.8		0.056	0.08	0.192	<0.05	0.241	<0.01		
Body surface area	m^2	(1.07; 2.05)	1.59	(1.57 to 1.61)	0.2		0.057	0.07	0.209	<0.01	0.413	<0.01		
Body mass	kg	(29.5; 101.0)	56.7	(55.2 to 58.1)	11.0		0.090	<0.01	0.176	<0.01	0.405	<0.01		
Fat mass	%	(7.5; 51.6)	25.4	(24.4 to 26.4)	7.7		0.075	<0.01			0.230	<0.01	-0.148	<0.05
	kg	(3.3; 51.4)	15.0	(14.0 to 16.0)	7.5		0.165	<0.01			0.292	<0.01		
Fat-free mass	kg	(26.2; 62.5)	41.7	(40.9 to 42.5)	5.8		0.051	0.20	0.213	<0.01	0.391	<0.01		
LVIDd	mm	(28.9; 56.1)	45.2	(44.7 to 45.7)	3.6		0.077	<0.01			0.242	<0.01		
ISWTd	mm	(5.4; 10.2)	7.7	(7.6 to 7.8)	0.9		0.064	0.03			0.203	<0.01	0.288	<0.01
PWTd	mm	(5.0; 9.6)	7.5	(7.4 to 7.6)	0.8		0.097	<0.01					0.249	<0.01
LVM	g	(50; 185)	107	(104 to 110)	22		0.049	0.20	0.131	<0.05	0.274	<0.01	0.222	<0.01
LVM index	$g.m^{-2}$	(33; 107)	67	(66 to 69)	11		0.023	0.20					0.286	<0.01

CA (chronological age); SA (skeletal age); f (frequency); LVIDd (left ventricular internal dimension at end of the diastole); ISWTd (interventricular septal wall thickness at end of the diastole); PWTd (posterior wall thickness at end of the diastole); LVM (left ventricular mass); LV index (left ventricular mass index = LVM / body surface area); f (absolute frequency); 95% CI (95 % confidence intervals).

*n=163; players classified as skeletally mature (n=65) were not considered in the analysis.

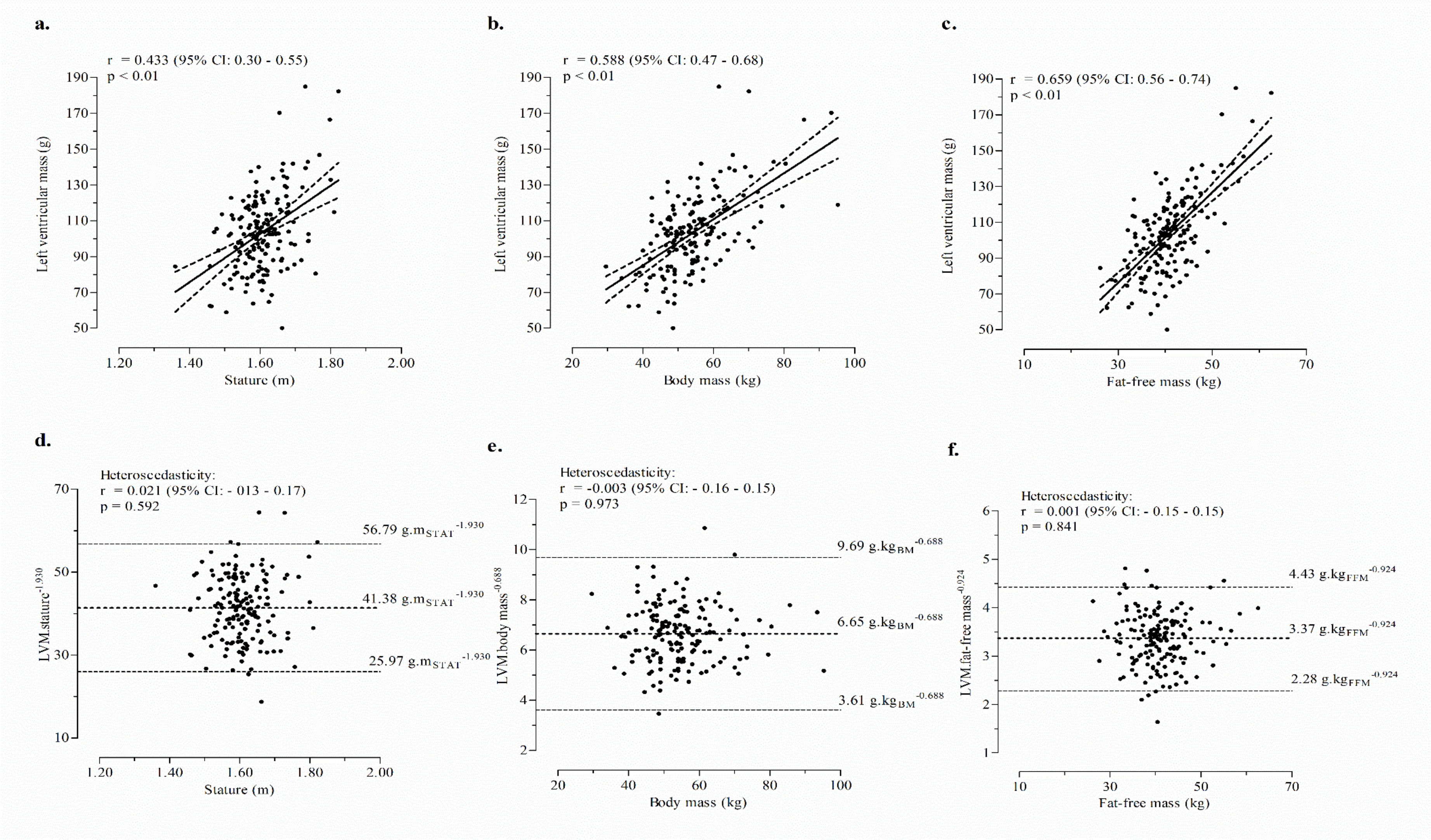

Figure 7.1. Relationship of LVM to stature (panel a), body mass (panel b) and fat-free mass (panel c), and correlations between power functions and respective size descriptors (panels d, e and f).

Table 7.2. Bivariate correlations and simple allometric models between LVM and size descriptors (n=163).

X_i: size descriptors	correlations between LVM and size X_i			simple allomeric models [ln (LVM) = ln (a) + k_i × ln (X_i) + log (ε)]						correlation (X_i, LVM/X_i^k)
	r	95% CI	(qualitative)	a	k_i		model summary			
					value	(95% CI)	R	R^2	*p*	
Stature	0.433	(0.299 to 0.550)	(moderate)	-5.185	1.930	(1.240 to 2.620)	0.399	0.159	<0.01	0.021
Body mass	0.588	(0.477 to 0.680)	(large)	1.878	0.688	(0.536 to 0.840)	0.576	0.332	<0.01	-0.003
Fat-free mass	0.659	(0.562 to 0.738)	(large)	1.199	0.924	(0.737 to 1.112)	0.608	0.366	<0.01	0.001

LVM (left ventricular mass); r (correlation coefficient); 95%CI (95% confidence intervals); k_i (scaling coefficient); ε (error); a (constant); R^2 (explained variance).

The panels of Figure 7.2 illustrate maturity-associated variation in stature, body mass, FFM and LVM. LVM showed the same maturity gradient (i.e. late < average < early) as noted for size descriptors. Comparisons of late and average maturing groups indicated consistently moderate Cohen's d values ($0.85 < d < 1.04$). Corresponding comparisons between late and early maturing indicated magnitude differences ranging from moderate ($d = 0.82$ for stature; $d = 1.12$ for body mass) to large ($d = 1.30$ for FFM). Finally, differences between average and early maturing players tended to be trivial (stature: $d <0.20$) or small (body mass: $d = 0.46$; FFM: $d = 0.36$). Taking into account interrelationships among size descriptors and skeletal maturity status, it was decided to examine their multiplicative effects on heart size. Table 7.3 summarizes the results of multiplicative allometric modelling combining size descriptors, CA and training experience with maturity status as a dummy variable. The explained variance for LVM increased to 46%; the resulting equation was as follows:

$\ln(\text{LVM}) = 1.070 + 0.412 \times \ln(\text{body mass}) + 0.621 \times \ln(\text{FFM}) - 0.028 \times (\text{CA}) + 0.022 \times (\text{training experience}) + 0$ (if maturity status = late), -0.137 (if maturity status = average) and -0.116 (if maturity status = early) **(7.7)**

When using scaled LVM values, the maturity-associated gradient was attenuated, and adolescent female soccer players classified as delayed (late maturing group) showed similar values compared to other maturity groups (Figure 7.3).

Table 7.3. Multiplicative allometric modelling* of LVM combining size, CA, skeletal maturation and training (n=163).

Predictors	Constant	coefficients	*P*	collinearity		model summary *			
				tolerance	VIF	R	R^2 adjusted	F	p
						0.694	0.462	24.146	<0.01
	1.070		<0.01						
ln (body mass)		0.412	<0.01	0.307	3.259				
ln (fat-free mass)		0.621	<0.01	0.288	3.470				
Chronological age		-0.028	0.05	0.845	1.184				
Training experience		0.022	<0.01	0.777	1.287				
Skeletal maturity status									
Late vs average		-0.137	<0.01	0.397	2.520				
Late vs early		-0.116	<0.01	0.339	2.952				

VIF (variance inflation factor); R^2 (explained variance)

* $\ln(\text{LVM}) = k_1 \times \ln(\text{stature}) + k_2 \times \ln(\text{body mass}) + k_3 \times \ln(\text{fat-free mass}) + a + b_1 \times (\text{CA}) + b_2 \times (\text{training years}) + b_3 \times$ (maturity status: late vs average; late vs early maturing, with late maturing being zero) $+ \ln \varepsilon$

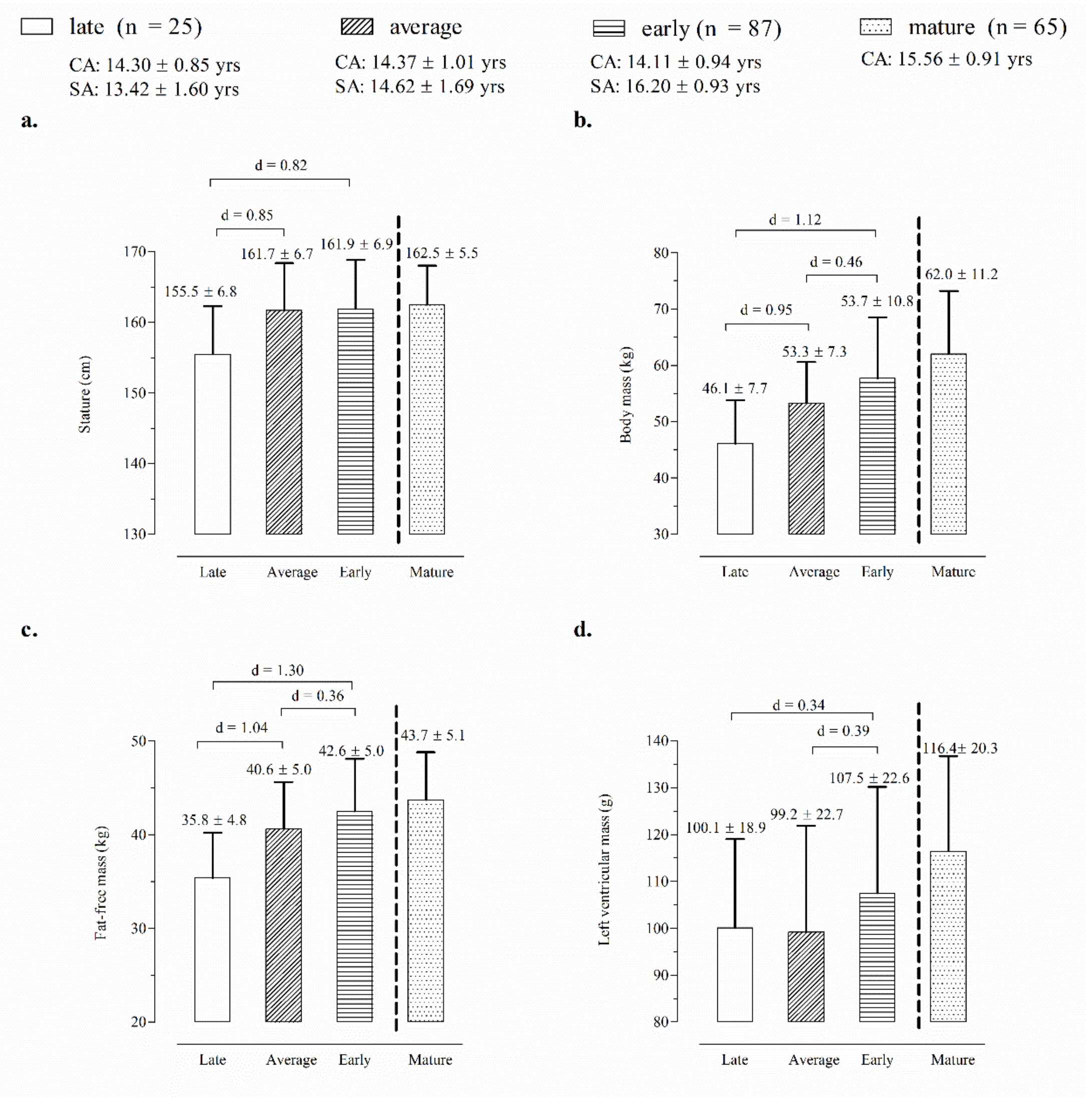

Figure 7.2. Mean values for stature (a), body mass (b), fat-free mass (c) and left ventricular mass (d) for the total sample of players by skeletal maturity status.

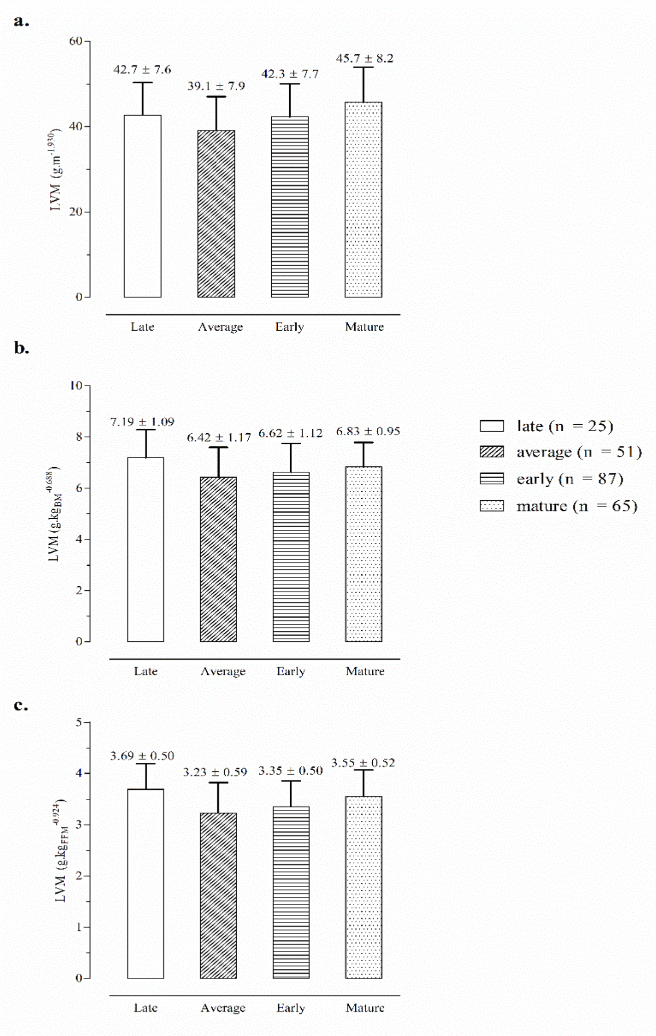

Figure 7.3. Means and standard deviations by skeletal maturity groups for scaled LVM expressed per unit of stature (a), body mass (b) and fat-free mass (c)

7.5. Discussion

The contributions of CA, skeletal maturity status, training experience in competitive soccer and indicators of body size to inter-individual variability in LVM was considered among Portuguese adolescent female soccer players 11.8–17.1 years of age. The predicted variable (i.e., LVM) was interpreted as tri-dimensional and, not surprisingly, the contribution of the stature (uni-dimensional size descriptor) to the explained variance in LVM was relatively low (about 16%). Stature did not consistently enter the final multiplicative allometric model. On the other hand, FFM (tri-dimensional indicator) was the best single predictor of LVM, explaining 37% of the variance. Its scaling coefficient, k = 0.924 (95%CI: 0.737 to 1.112), suggested a linear relationship with LVM (geometric similarity). Body mass, another tri-dimensional indicator, had a scaling coefficient, k = 0.688 (95%CI: 0.536 to 0.840), that departed from linearity and suggested an elastic relationship between body mass and LVM. The final multiplicative allometric model suggested that body mass, FFM and training experience in soccer were directly associated with LVM, and after controlling for the preceding, average and early maturing players had a proportionally smaller LVM compared to late maturing peers (reference group in the analysis).

The adolescent female soccer players had a mean stature at the 50th percentile of the US reference data for girls of the same age (Kuczmarski et al., 2002) but a mean body mass between the 50th and 75th percentiles of the reference. The tendency for greater mass-for-stature may reflect their advanced skeletal maturity status, consistent with cross-sectional observations for Portuguese adolescent male soccer players (Malina et al., 2000; Coelho-e-Silva et al., 2010; Malina, Coelho E Silva, Figueiredo. Carling, & Beunen, 2012; Valente-dos-Santos et al., 2015). The body mass index (BMI) of each individual participant was also plotted relative to US age-specific-scores (Kuczmarski et al., 2002) and the majority of female soccer players (n = 194) ranged between − 1.0 and + 1.0, while 26 players had BMIs that exceeded + 1.0. In addition, 103 of the soccer players were characterized by an excessive amount of fatness predicted from two skinfolds (> 25%), and the data showed a maturity related gradient in %FM: early>average > late. It is thus possible that excess body mass-for-stature may reflect increased FM. Nevertheless, future studies should consider alternative assessments of body composition such dual-energy x-ray absorptiometry (DEXA) or air displacement plethysmography, may provide more accurate estimates of FM and FFM.

Absolute values for LVM, LVIDd and PWTd in the current study were comparable to those reported for 32 American female soccer players 13–18 years (Watson et al., 2018). Interpretation and comparison of cardiac indicators across samples are influenced by body size and composition, but detailed information on the body dimensions of the American sample was not reported. Additionally, a slight increase in LV cavity and lower PWT were noted in the present sample of soccer players compared to female swimmers of the same age and similar average body masses (Csajagi, Szauder, Major, & Pavlik, 2015). It is possible that the results suggest eccentric remodelling independent of physiological adaptations to the haemodynamic loading associated with soccer participation (George et al., 2009; Lang et al., 2005; Naylor, George, O’Driscoll, & Green, 2008; Petek & Wasfy, 2018).

Theoretical allometric coefficients of k = 2.13 and k = 2.65 for stature have been adopted to normalize the effects of body size in LVM (Dewey et al., 2008; Pressler et al., 2012; Gireldeau et al., 2015; Lang et al., 2005; Pela et al., 2016). The simple allometric models in the current study, however, noted a lower exponent for stature (k = 1.930). The differences may reflect sampling variation, methodological constraints, statistical procedures, sex and/or age-associated variation. Moreover, stature only explained ≈16% of the variance in LVM and was not included in the proportional allometric model. Overall, the findings suggested that stature alone should not be considered to normalize or predict LVM. Similar results were also noted in 464 highly trained junior male and female athletes 14–18 years participating in cycling, soccer (males only), rowing, swimming and tennis, with small numbers in other sport (George et al., 2001).

It is possible that other size descriptors may be needed to normalize LVM. More recently, FFM based on DEXA was noted as the best size descriptor to compare LVM in 75 young adult females in static or dynamic sport activities (Kooreman et al., 2018). Among dependent variables in the current study, FFM was the best explanatory predictor of LVM, confirming the influence of metabolically active tissues on cardiac output. Results of the present study also suggested a linear relationship between the logarithmic transformations of LVM and FFM. The utility of simple ratios to estimate cardiac output of LVM per unit of FFM was noted in studies of trained (George et al., 2009) and untrained adults (Batterham, George, & Mullineaux, 1997) consistent with the theoretical range of geometric similarity, i.e., LVM is represented as a cubic expression and as such requires a 3-dimenisonal variable for normalization. On the other hand, the interpretation of LVM considering only FFM is limited by a lack of comparative studies in youth female sport participants.

Multiplicative allometric models are physiologically plausible and accommodate heteroscedasticity in the distribution of a variable, and thus provide a better statistical fit than simple models (Welsman & Armstrong, 2008). Not surprisingly, FFM combined with years of training in soccer and biological maturity status provided a better understanding of LVM than simple allometric models. The results were consistent with previous cross-sectional studies of adolescent sport (Valente-dos-Santos et al., 2013) and non-sport participants (Valente-dos-Santos et al., 2014) in showing that the interrelationships between growth and maturation are determinants of LVM. However, the contribution of SA per se was not a significant predictor of LVM in Portuguese male roller hockey players 14.5–16.5 years of age (Valente-dos-Santos et al., 2013). In contrast, results of the multiple backward regression analysis among adolescent female soccer players indicated that maturity status defined by SA minus CA was a significant determinant of LVM. SA provided perhaps the most accurate estimate of maturity status, i.e., the state of maturation of the hand-wrist bones at the time of observation (Malina et al., 2004). By inference, SA should be expressed relative to CA for inclusion in multiplicative allometric models.

A gradient of maturity associated differences in size and LVM were noted in the soccer players (early > on time > late maturing girls). Early maturing players tended to be heavier and relatively fatter and presented a larger LVM compared to average and late maturing players (Figure 7.2). This was consistent with observations for 6029 Flemish girls 6–16 years of age which showed a positive relationship between fatness and SA based on the Tanner– Whitehouse 2 method (Beunen et al., 1994). The trends thus suggested that absolute values of LVM were significantly influenced by early maturation which in turn was related to body composition, specifically pubertal gains in FM. Although FM does not have a strong relationship with LVM, sports participation was associated with changes in FM and FFM (Malina & Geithner, 2011) which may be a potential explanation for the inclusion of years of training in soccer and FFM in the final allometric model. Nevertheless, the multiplicative allometric model indicated that differences in LVM among maturity groups were reversed when body mass, FFM, CA and training experience were appropriately controlled. After controlling for body size descriptors (i.e., scaled LVM output), there were no substantial differences amongst female adolescent soccer players contrasting in maturity status (Cohen's d values were less than 0.20 as showed in Figure 7.3 a-c). A study of peak oxygen uptake among 54 adolescent females (10.7–13.5 years) to evaluate allometric models for concurrent size descriptors (stature, body mass and FFM) noted that scaled performance did not differ according to categories of self-assessed pubic hair development (Werneck et al., 2019). Among 59 male adolescent basketball players, those aged 14 years and classified in stage 3 for clinically assessed pubic hair development (mid-puberty) performed, on average, better on the 20-m shuttle run test than peers of the same age classified in stage 5 (post-pubertal) (Coelho E Silva, Figueiredo, Carvalho, & Malina, 2008). Overall, the available studies show that early maturing adolescents tend to be taller, heavier and stronger, but may not demonstrate superior performance in aerobic fitness tests.

Generally comparable results showing an influence of predicted maturity status based on predicted maturity offset, i.e., time before or after PHV on absolute values of peak force were noted in a cross-sectional study of 157 female soccer players combined across four competitive age groups, U10 through U16 (Emmonds et al., 2017). However, conclusions based on predicted maturity offset as an indicator of maturity status across this broad age range should be interpreted with caution given limitations of the equation used to predict maturity offset in girls. More specifically, predicted offset and in turn predicted age at PHV are affected by CA at prediction and has major limitations in early and late maturing girls defined by observed ages at PHV in two validation studies based on longitudinal samples (Malina & Koziel, 2014; Malina, Choh, Czerwinski, & Chumlea, 2016).

The distinction between physiologic increases in LVWT in athletes (i.e., athlete's heart) and hypertrophic cardiomyopathy accounts for about one-third of all exercise-related sudden cardiac deaths in trained athletes aged < 35 years old (Maron, Roberts, McAllister, Rosing, & Epstein, 1980; Maron, Epstein, & Roberts, 1986; Burke, Farb, Virmani, Goodin, & Smialek, 1991) and intense competitive sport is not recommended (Maron, Isner, & McKenna, 1994). To define physiologic limits of left ventricular hypertrophy in elite adolescent athletes, echocardiography was performed among 720 elite adolescent athletes (75% male) aged 14–18 years participating in ball, racket, and endurance sports, and in 250 healthy sedentary controls of similar age, sex, and body surface area (Sharma et al., 2002). Only a small proportion of athletes exhibited a LVWT exceeding upper limits and authors concluded that compared with controls, adolescent athletes had greater absolute LVWT. However, it should be noted that many sports tend to recruit/select and promote young athletes that have larger body sizes (Coelho E Silva et al., 2008; Coelho E Silva et al., 2010; Malina et al., 2017) and interpretation of both wall thickness and cavity diameter should be done according to principles of geometric similarity of heart size to body size (Nevill et al., 1992). LVM is a tri-dimensional variable and, consequently, it is not expected to have a linear relationship with stature in cm. Linearity is, however, expected between LVM and FFM which is the metabolically active component of body mass. Athletes exposed to systematic training tend to be characterized by a larger FFM (Malina et al., 2004; Malina & Geithner, 2011). Identification of athletes exceeding physiological limits is thus recommended. In addition, skeletal maturity status is an additional source of inter-individual variation in LVM, but does not correspond to any abnormality when LVM is scaled properly.

Although the present study considers a previously under studied population (adolescent female soccer players) and includes of a valid and established indicator of maturity status, specifically SA, several limitations of the present study should be noted. The sex-specific equation for predicting %FM from two-skinfold thicknesses has a standard error of estimate of 3.8% (Slaughter et al., 1988). FM was estimated as predicted %FM × body mass, and FMM was derived by subtraction (body mass - FM = FFM). Based on the two skinfolds used in the present study, %FM was 18.6 ± 7.2% in a combined sample 126 youth soccer players (mean age: 13.3 years, 86 boys, 40 girls) and was lower than estimated %FM based on DEXA, 21.9 ± 5.8% (Lozano-Berges et al., 2019). Unfortunately, the prediction equations are different for boys and girls so that comparisons with the combined sample should be interpreted with caution. The equation for boys (Slaughter et al., 1988) was also used in adolescent roller hockey players (Valente-dos-Santos et al., 2013a); estimates of FM and FFM derived from predicted %FM were significant contributors to inter-individual variability in LVM using allometry (FM: r = 0.56, 31% explained variance; FFM: r 0.51, 26% explained variance).

Future research is needed to examine intra- and interindividual variability in LVM associated with specific aspects of sport training and participation and internal and external markers of training load such as minutes and sessions, and ratings of perceived exertion. Moreover, characteristics of training process are generally specific for initiates, juveniles or juniors (competitive age groups by the Portuguese Soccer Federation). Although the sample size in the present study (n = 163) was larger than in previous studies, the cross-sectional design does not support a cause-effect relationship between size descriptors and cardiac remodeling. Finally, although echocardiography is still the most widely used method for assessing LVM, cardiac magnetic resonance imaging is considered the gold standard for determining LVM.

7.6. Conclusions

Results of this cross-sectional study of adolescent female soccer players indicated that inter-individual variance in LVM is, in part, explained by skeletal maturity status which affects body size and composition. Specifically, a larger body size tended to be associated with early maturing participants. Skeletal maturation, training experience, body size and composition should be considered in the interpretation of an athlete’s heart. The study also highlighted the utility of multiplicative allometric models for understanding LVM among adolescent girls participating in competitive soccer. Interpretation of echocardiography data from adolescent athletes apparently exceeding the physiologic limits of left ventricular size may require the assessment of body composition and SA.

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Chapter VIII

General discussion and conclusions

8. General discussion and conclusions

An extensive discussion of each study was considered in the respective chapters. This section summarized the main topics and proposed suggestions for future research and practical applications.

8.1. Growth and maturation in female soccer

A recent review on female soccer extracted few studies (n=8) that report stature and body mass among adolescent participants (Martinez-Lagunas, Niessen, & Hartmann, 2014). Additionally, the most used protocols to examine repeated sprint ability and associated-data quality were summarized based on 16 studies that followed the eligible criteria (Pardos-Mainer, Casajús, Julían, Bishop, & Gonzalo-Shok, 2020). Growth and maturity associated-variation in performance were ignored. These point is not consistent with the literature among males (Malina, Figueiredo & Coelho-e-Silva, 2017; Malina et al., 2019). Thereby, based on data compilation from 1981 to 2020, growth and maturity as characteristics of female players were described in **Chapter 3**.

Non-invasive indicators have been widely used in studies of female soccer participants. The time before or after peak height velocity (PHV), usually labelled as maturity-offset, and predicted age at PHV were systemically adopted. In the meantime, based on mean values, the algorithm was highly dependent on chronological (CA) and body size at prediction, confirming the limitations reported in longitudinal samples of English (Parr et al., 2020) and Dutch (Teunissen et al., 2020) soccer players, and American (Malina, Choh, Czerwinski, & Chumlea, 2016) and Polish non-sport participants (Malina & Kozieł, 2014a; Malina & Kozieł, 2014b; Kozieł & Malina, 2018; Malina, Kozieł SM, Králik, Chzanowska, & Suder, 2020). Additionally, among 198 Polish girls from Wroław Growth Study predicted age at PHV was substantially different from observed age at PHV when the participants were categorized as early, on time and late maturing (Malina & Kozieł, 2014a).

Given the interest of classifying players by maturity categories (Bayli, Cardinal, Higgs, Norris, & Way, 2005), predicted age at PHV categorized participants according to the predicted time of PHV or as pre-, at- or post-PHV. Two important points should be taking into account. Firstly, studies among female soccer players (Emmonds et al., 2017; Emmonds et al., 2020) compare the groups without considering variation in CA and body size. Secondly, a maturity-related gradient was previous reported that youth sports (Malina, 2011; Malina, Rogol, Cumming, Coelho e Silva, & Figueiredo, 2015) with early maturating players tend to persist during adolescence. The use of equation could be problematic in late and early maturing participants involved in organized and competitive sport.

Percentage of predicted adult (mature) stature (%PMS) attained at a given CA emerges from the current stature and a prediction of the adult stature. It was applied as a continuous variable (Cumming, Battista, Standage, & Ewing, & Malina, 2006) or to group players by pubertal status (Mullen et al., 2018; Westbrook, Taylor, Nguyen, Paterno, & Ford, 2020). Curiously, the same group of authors published two studies (Mullen et al., 2018; Westbrook et al., 2020) that grouped players by bands and used 87-94% of PMS to classify players as pubertal; <87% and >94% of PMS were the criteria to define pre-and post- pubertal players, respectively. In the meantime, the authors did not explain the criteria to define these bands. Previous studies in English elite male soccer players used bands that theoretically covered the maximal growth spurt: <85% of PMS; %PMS ranged from ≥85.0 to <90.0; ≥90.0% of PMS (Malina et al., 2019; Thomas, Oliver, Kelly, & Knapman, 2017; Malina, 2021). This approach was based on medians and percentiles (10th and 90th) at take-off and PHV of Zurich Longitudinal Study (Molinari, Gasser, & Largo, 2013). The 10th, 50th and 90th percentiles of %PMS at the beginning of maximal growth spurt were 78.3%, 81.5% and 84.5% respectively, while corresponding data for PHV was 89.8%, 91.5% and 92.9%.

Overall, it was suggested that <85% of PMS corresponds to the initial acceleration of stature, 85-90% of PMS is the interval between take-off and PHV, and 90-93% the interval of growth spurt (Malina, 2021). Percentiles of %PMS among females of the Zurich Longitudinal Study (Molinari et al., 2013) were obviously different compared to males (take-off: 10th percentile – 80%; 50th percentiles – 83.1%; 90th percentile – 87.3%; PHV: 10th percentile – 89.7%, 50th percentile: 91.5%; 90th percentiles: 93.8%). The bands proposed on two studies of female soccer players (Mullen et al., 2018; Westbrook et al., 2020) were within the range presented in Zurich Longitudinal data. To note that the bands are not fixed and should be modified.

Ages at menarche were retained from 6 studies. Only one study estimated age at menarche, based on a prospective approach (Siegel, 1995). The status quo method required two indicators: CA and a categorical variable (i.e. if the menarche occurred or not). It is applied to a sample aged 9-17 years and, consequently, median age at menarche is calculated. In the remaining studies (Düppe, Gärdsell, Johnell, & Ornstein, 1996; Casper, Michaels, & Simon, 1997; Petterson, Nordström, Alfredson, Henriksson-Larsén, & Lorentzon, 2000; Södermann, Bergström, Lorentzon, & Alfredson, 2000), players were asked to recall age at menarche. Although this retrospective approach is influenced by memory, ages at menarche were relatively consistent between studies (ranged 12.7-13.0 years). Unfortunately, the sample of female soccer players included in **Chapters V and VI** was limited to adolescent participants. The frequencies of players aged 10-11 years was not considerable (n=8), therefore, a probity or logistic regression could not be performed.

Secondary sex characteristics were also used to evaluate pubertal stages. The limitations of stages initially described by Reynolds and Wines (1948), and posteriorly adopted by Tanner (1962), were described in **Chapter I**. However, authors adopted the stages to classify players as pre-pubertal, pubertal and post-pubertal without considering age associated-variation (Lozano-Berges et al., 2016). Furthermore, stages were combined and analyzed as equivalent. For example, 35 female soccer players (age: 12.76 ± 0.59 years) were compared within pubertal stages in bone parameters (Plaza-Carmona et al., 2019). Adolescents were divided into two maturity groups: peripubertal (stages II and III) and post-pubertal (stage IV and V). Nevertheless, stages 2, 3 and 4 indicate the development of secondary sex characteristics and stage 5 corresponds to the mature state.

Comparisons of body composition assessment by dual-x-ray energy (DEXA) between type of sport (i.e. soccer, swimming, basketball, handball) and non-sport participants split the analysis of covariance by maturity groups (pre-pubertal and pubertal). These groups combined the assessment of breast development and pubic hair stages (Ubago-Guisado, Gómez-Cabello, Sánchez-Sánchez, García-Unanue, & Gallardo, 2015). Stages of secondary sex characteristics are not equivalent and are limited to adolescence (Malina, Bouchard, & Bar-Or, 2004).

On the other hand, skeletal age (SA) represents the best indicator to maturation once it is applicable from early childhood to young adulthood (Malina et al., 2004). Only one early study dated to 1981 used a local adaption of Greulich-Pyle (GP) protocol to assess SA of female soccer players. The method proposed by Kapalin was based on Czech children and it was analogous to GP technique (Novotny, 1981). Based on these findings, the **Chapter IV** explored the details of the GP protocol. This issue was central because the original Atlas did not clarify the procedures to obtain an overall SA for each participant (Greulich & Pyle, 1959). This topic was explored in 89 Australian females, but the sample was limited to children and adolescents aged 2-13 years (Roche & Johnson, 1969). Briefly, the mentioned study compared different approaches to obtain GP SA, suggesting the exclusion of carpal bones. The **Study 2** examined the reproducibility and inter-observer agreement bone-by-bone and, also, considering different methodological options. The findings were comparable to those reported among Australian females (Roche & Johnson, 1969). Differences contrasting 30 bones assessed using an individual approach, and solely pre-mature bones were negligible. By inference, the mature bones, including the carpals, can be excluded from SA assessment. Another additional question is the mathematical procedure to estimate a global SA for each participant. Relevant authors proposed the mean (Roche, Chumlea, & Thissen, 1988) or the median (Malina et al., 2004). GP SAs were, on average, comparable using the median or the mean. Finally, the protocol is constantly applied by clinics comparing the radiograph as a whole, with the photographs included in the Atlas (Malina et al., 2015). In parallel, studies in male soccer did not summarize the details associated with GP SA assessment (Le Gall, Carling, & Reilly, 2007; Gouvea et al., 2016). The results of **Chapter IV** found a substantially inter-individual variation between observers when the SA was obtained, by comparing the x-ray as a whole with the pictorial standards. Some important points need to be addressed, regarding the GP method.

Firstly, the method only considered 29 American girls followed by three months of interval during infancy, semi-annually from one to five years and annually until adulthood. Secondly, differences between GP atlas standards at 16, 17 and 18 years of age for females are slight, which do not enable to define mature or adult state. Thirdly, SA variability (expressed by standard deviation) is less than 1.0 year during adolescence, contrasting with concurrent protocols.

The protocols to estimate SA are comparable in principle but differed in criteria, procedures and samples. The aim of **Chapter V** was to test SA differences using concurrent methods (GP and Fels) among female soccer players. GP SAs were less than Fels SA, and differences between CA groups tended to increase with CA. The current study used the median of 30 bones to estimate GP SA (**Chapter IV**), but the differences in standards of 16, 17 and 18 years are concentrated in two bones (i.e. radius and ulna). This may explain less variability noted with GP protocol in late adolescence. SA represents, by definition, the CA of maturity level attained in the original sample. In light of the preceding, SA should not be interpreted as a continuous variable, but compared with CA. Consequently, maturity categories were defined. In **Chapter V**, players within CA groups were compared according to maturity status classifications with each method. As noted in **Study 1**, this issue was central for equalizing competitions, individualizing training programs and for long-term soccer development. The agreement between protocols seems to be reasonable at younger ages (under-13: 74%; under-14: 65%) but tended to decline with CA. Given the lower GP SA compared with Fels SA, the change of classifications after 14 years-old is stable. At 14 years-old, 61 players were classified as early maturing by Fels and only two had the respective classification with the GP method. The trends were similar at 15 and 16 years-old. For the total sample, 83 players were classified as skeletally maturity with Fels, while none was categorized as an adult with GP. These results highlighted the limitations of GP protocol to define adult status. More recently, SA was suggested for age verification (Timme, Steinacker, & Schemling, 2017). The topic was overviewed in youth male soccer with interesting comments that should be addressed. Firstly, the authors argued that an excessive amount of ionising radiation was used to obtain the hand and wrist film. In the meantime, with sophisticated technology, the radiation is minimal, equivalent to 3 hours of television (Radiological Society of North America, 2009). Secondly, it was questioned the applicability of the Fels protocol in samples of youth soccer. Although the sample from

Fels was assessed between 1932-1977, the age at PHV, predicted by triple logistic model in 74 boys, was 13.70±1.03 years (Malina et al., 2016), which is similar to age at PHV calculated from male soccer players (13.6 ±0.8 years) using the SITAR model (Malina et al., under review). Similar trends were noted in females. Using 63 girls from Fels sample, observed age at PHV 11.61±0.73 years (Malina et al., 2016). Unfortunately, longitudinal data spanning adolescence is not available for female soccer players. However, based on Preece-Baines model 1 applied to the points retained in **Study 1**, age at PHV was 11.55. Overall, contemporary changes between samples were not evident, and this seems a reasonable motive to use Fels protocol in youth samples involved in competitive sport. Considering the previous findings, the **Chapters VI and VII** assessed SA using Fels protocol.

The **Chapter VI** described the growth and maturity status, assessed by Fels method, of Portuguese female soccer players. Participants tended to approximate the 50th percentile for stature and were systematically plotted above the 50th percentile for body mass. Early maturing players were the most representative maturity category in three competitive groups. Interestingly, among juniors, 63 players (~49%) were classified as skeletally mature. It is documented that females, on average, attain the adult state, expressed as somatic, sexual and skeletal maturity, before males (Malina et al., 2004; Tanner, Healy, Goldstein, & Cameron, 2001), and this should perhaps be considered to organize competitive groups.

The study in **Chapter VII** investigated the influence of body size and estimated body composition, biological maturation, CA and training experience on left ventricular mass (LVM) in adolescent female youth soccer players using an allometric approach. The study confirmed that stature only fitted 16% of variance for LVM. Afterwards, it was the unique predictor that not entered in the multiplicative model. These findings indicated that stature did not partition adequately LVM and confirm previous studies of male (Valente-dos-Santos et al., 2013) and female sport participants (Kooreman et al., 2019). Additionally, fat-free mass (FFM) was identified as the best size descriptor to predict LVM. In studies of youth sport participants (Kooreman et al., 2018), adult athletes (George, Birch, Pennel, & Myerson, 2009) and adult non-athletes (Battherham, George, & Mullineaux, 1997), metabolically active tissue close to the unity (k=1.0) was also claimed to normalize cardiac output.

To be noted that, most recently, sex-specific data for LVM indexed to lean soft tissue-1 were produced, among 791 children and adolescents engaged in sport (Krysztofiak, Młyńczak, Folga, Braksator, & Małek, 2018). Additional independent variables were included in a multiplicative regression model to determine the impact on LVM. As explained in **Chapters V and VI**, SA should be compared with CA in order to classify players by skeletal maturity status (i.e. late, average and early) and, considering the previous, SA was included in the predictive model as a dummy variable. The combination of skeletal maturity status, metabolically active tissues and training experience worked as a significant predictor of LVM and increased the explained variance compared to simple allometric models. Subsequent comparisons suggested a maturity-related gradient in body size and cardiac output, with early maturing being taller, heavier and having greater LVM than average and late maturing peers. Similar trends were noted in adolescent male soccer players in Yo-Yo intermittent endurance test, velocity and jump protocols (Malina, Eisenmann, Cumming, Ribeiro, & Aroso, 2004). Players classified as pre-pubertal (i.e. PH1) tended to attain lower results in functional protocols compared to pubertal (i.e. PH2, PH3, PH4) and mature participants (i.e. PH5). In Chapter **VII**, the negative coefficients of maturity categories indicated that, after considering FFM and training, LVM in average and early maturing players showed smaller LVM compared to late maturing participants. Among 73 roller hockey players (Valente-dos-Santos et al., 2013), SA expressed as a continuous predictor was not a significant predictor of LVM. Nevertheless, SA per se is not an indicator of maturity status (see **Chapter III and V**) The consideration of body size parameters and biological maturation as potential exploratory variables of LVM were negligible in studies of adolescent female soccer players (Watson et al., 2018; Csajagi, Szauder, Major, & Pavlik, 2015).

8.2. Directions for future research

Limitations of each study were separately considered in the respective chapter. However, this section summarizes general concerns of the thesis and presents important topics for future research.

In **Chapter IV**, some of the details about GP method were clarified, but the sample used was limited to adolescence years. Data quality in GP need to be examined during childhood. Although the issue of maturity status in long-term athletic development (LTAD) programs in females was detailed in **Chapters III, V, and VI** the impact of GP approaches to classify players by maturity categories claims for future research.

Results of **Chapter V** highlight the influence of concurrent protocols on estimates of skeletal maturity categories. Tanner-Whitehouse method (TW) is often used to assess SA (Satoh, 2015), including in male soccer players (Malina et al., 2019). Thus, it is suggested that agreement between the TW method with Fels or the GP method in female soccer requires further examination. As reported in **Chapter III**, non-invasive estimations of maturity status obtained by offset protocol or %PMS were applicable among female soccer players. Interrelationship between skeletal maturity methods and predictive equations is lacking in females. In contrast, comparable data are available in Portuguese male soccer players (Malina, Coelho-e-Silva, Figueiredo, Carling, & Beunen, 2012). **Chapter VI and Chapter VII** estimated body mass compartments (i.e. fat mass, FFM) based on a sex-specific equation (Slaughter et al., 1988). Recently, a predictive equation has been proposed to estimate metabolically and non-invasive tissues in youth soccer players (Lozano-Berges et al., 2019). However, the mentioned study combined male and female soccer participants, which did not take into account sex-variation in FM and FFM during adolescence. Overall, modern technologies including air-displacement plethysmography or DEXA should be adopted in female soccer. Predictive equations of body composition assessment need to consider uniquely female samples. The findings of studies included in this thesis were limited to a cross-sectional design. Within competitive and organized sport, more longitudinal data combining growth, maturation, training or competition parameters are obviously necessary, particularly among females.

among females. Generalizations of conclusions showed in these five studies to other female samples in youth sports claims by longitudinal data and control group (i.e. non-sport participants).

8.3. Conclusions and implications

The current thesis provides the description of growth and maturity status of female soccer players, the impact of concurrent protocols in SA assessment and the interrelationship between body size, maturity status and training in physiological output (i.e. LVM). Unfortunately, studies reporting growth and maturity characteristics of female soccer participants are lacking, so data were compared with literature of non-sport participants or female involved in other competitive sports (i.e. swimming, tennis, gymnastics). The main conclusions and practical applications extracted from the current thesis were:

i. Long-term athlete development proposed age at PHV as an optimal window for coaches and athletes to organize the training of functional capacities. Age at PHV requires longitudinal data. Coaches should measure height every 3-4 months to obtain the growth velocity. Significant changes in velocity may be an indicative of growth spurt.

ii. Although maturity offset protocol has been widely adopted in youth soccer, the dependence of CA and body size suggested it utilization in samples with a narrow age-range.

iii. % PMS expressed as categorical variable seems to be a valid alternative to classify player according to age at PHV. This strategy was used in English players to equalize competitions (i.e. bio-banding tournaments).

iv. SA estimation is dependent of the method used and this issue had impact on classifying players by maturity categories. Fels protocol provides a standard-error associated with SA assessment which is a unique characteristic of the method.

v. The elevate percentage of FM in all CA groups suggested the necessity to include nutritional intervention in clubs.

vi. Adult status was attained in 81 participants with CA lower than 18.0 years. The organization of competitive groups should consider this point and readjust the senior level for female soccer participants.

vii. Given the association among body size, estimated body composition, and maturity status, LVM dimension is in part a function of maturity-associated variation in body size

viii. Allometric modelling technique created a “size-free expression”, providing a more plausible interpretation of cardiac output.

8.4. References

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Appendix A

Curriculum Vitae

Curriculum Vitae

Diogo Vicente Martinho

Finalizou a Licenciatura em Ciências do Desporto (2013) e o Mestrado em Treino Desportivo para Crianças e Jovens (2015) na Faculdade de Ciências do Desporto e Educação Física Universidade de Coimbra. No mesmo ano, finalizou uma pós-graduação em Treino Personalizado pela Universidade Lusófona de Humanidades e Tecnologias. No ano de 2016, obteve o estatuto de bolseiro de doutoramento pela Fundação para a Ciência e a Tecnologia (SFRH/BD/1241441/2016) apresentando como orientadores o Prof. Doutor Manuel João Coelho e Silva, o Prof. Doutor João Alberto Valente dos Santos e a Profª. Doutora Lauren Esliger Sherar. Em Maio e Julho de 2018 e 2019, realizou a primeira e segunda missão outgoing, respetivamente, à Universidade de Loughborough - School of Sport, Exercise, and Health Science. É membro do Centro de Investigação do Desporto e Atividade Física.

Identificação

Identificação pessoal

Nome completo
Diogo Vicente Martinho

Género
Masculino

Data de nascimento
1992/01/01

Nomes de citação

Martinho, D. V.
Martinho, D.

Identificadores de autor

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Pavilhão 3, 3040-248 Coimbra, Coimbra, Portugal (Profissional)

Domínios de atuação

Ciências Médicas e da Saúde - Ciências da Saúde
Ciências Médicas e da Saúde - Ciências da Saúde - Nutrição e Dietética

Idiomas

Idioma	Conversação	Leitura	Escrita	Compreensão	Peer-review
Português	Utilizador proficiente (C2)	Utilizador proficiente (C2)	Utilizador proficiente (C2)	Utilizador proficiente (C2)	Utilizador proficiente (C2)
Inglês	Utilizador proficiente (C2)	Utilizador proficiente (C2)	Utilizador proficiente (C2)	Utilizador proficiente (C2)	Utilizador proficiente (C2)

Formação

	Grau	Classificação
2018/08/17 - 2021/08/31 Em curso	Ciências do Desporto e Educação Física (Doutoramento) Especialização em Treino Desportivo Universidade de Coimbra Faculdade de Ciências do Desporto e Educação Física, Portugal Loughborough University School of Sport Exercise and Health Sciences, Reino Unido	
2021 Em curso	Dietética e Nutrição (Licenciatura) Escola Superior de Tecnologia da Saúde de Coimbra, Portugal	
2018 Concluído	Curso de estatística avançada (Outros) Universidade de Coimbra Faculdade de Ciências do Desporto e Educação Física, Portugal	
2017 - 2017 Concluído	Curso de análise de dados com SPSS (Curso médio) Universidade de Coimbra, Portugal	
2014 - 2015 Concluído	Treino Personalizado (Pós-Graduação) Universidade Lusófona de Humanidades e Tecnologias, Portugal	16

2013 - 2014 Concluído	Mestrado em Treino Desportivo para Crianças e Jovens (Mestrado) Universidade de Coimbra, Portugal *"Crescimento, maturação e especialização desportiva em Basquetebolistas masculinos dos 12 os 15 anos: variação por posição em protocolos maximais de curta duração realizados no laboratório e no campo de basqutebol."* *(TESE/DISSERTAÇÃO)*	18
2010 - 2014 Concluído	Ciências do Desporto (Licenciatura) Universidade de Coimbra, Portugal	15

Percurso profissional

Outros

2017/01 - 2017/01	Preparador Físico Juventude de Viana, Portugal

Projetos

Bolsa

	Designação	Financiadores
2017/09/01 - Atual	GROWTH AND MATURATION IN FEMALE ATHLETES and NON-ATHLETES: (1) concurrent assessment of skeletal age using Greulich-Pyle, Tanner-Whitehouse, FELS protocols and (2) interrelationship among skeletal and somatic indicators of biological maturation” SFRH/BD/121441/2016 Fundação para a Ciência e a Tecnologia, Portugal	Fundação para a Ciência e a Tecnologia, Portugal

Produções

Publicações

Artigo em revista	1	Diogo V. Martinho; Manuel J. Coelho-e-Silva; João Valente-dos-Santos; Cláudia Minderico; Tomás G. Oliveira; Inês Rodrigues; Jorge Conde; Lauren B. Sherar; Robert M. Malina. "Assessment of skeletal age in youth female soccer players : Agreement between Greulich-Pyle and Fels protocols". *American Journal of Human Biology* (2021): https://doi.org/10.1002/ajhb.23591. 10.1002/ajhb.23591

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4 Robert M. Malina; Diogo V. Martinho; João Valente-dos-Santos; Manuel J. Coelho-e-Silva; Slawomir M. Koziel. "Growth and Maturity Status of Female Soccer Players: A Narrative Review". *International Journal of Environmental Research and Public Health* (2021): https://doi.org/10.3390/ijerph18041448.

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8 Joana Oliveira-Rosado; João P. Duarte; Paulo Sousa-e-Silva; Daniela C. Costa; Diogo V. Martinho; Hugo Sarmento; João Valente-dos-Santos; et al. "Physiological profile of adult male long-distance trail runners: variations according to competitive level (national or regional)". *Einstein (São Paulo)* (2020): https://doi.org/10.31744/einstein_journal/2020AO5263.

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9 Rosado, Joana; Duarte, João P.; Sousa-e-Silva, Paulo; Costa, Daniela C.; Martinho, Diogo V.; Valente-dos-Santos, João; Rama, Luís M.; et al. "Body composition among long distance runners". *Revista da Associação Médica Brasileira* 66 2 (2020): 180-186. http://dx.doi.org/10.1590/1806-9282.66.2.180.

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10 Martinho, D. V.. "Abstracts From the 31st Pediatric Exercise Physiology Meeting: Children-Exercise-Physical Activity & Sport Performance (September 2019, Umeå, Sweden)". *Pediatric Exercise Science* 32 S1 (2020): S1-S12. http://dx.doi.org/10.1123/pes.2020-0002.

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13 Tavares, Óscar M.; Duarte, João P.; Werneck, André O.; Costa, Daniela C.; Sousa-e-Silva, Paulo; Martinho, Diogo; Luz, Leonardo G. O.; et al. "Body composition, strength static and isokinetic, and bone health: comparative study between active adults and amateur soccer players". *Einstein (São Paulo)* 17 3 (2019): http://dx.doi.org/10.31744/einstein_journal/2019ao4419.

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15 Tavares, Óscar; Duarte, João P; Costa, Daniela C.; Sousa-e-Silva, Paulo; Martinho, Diogo; Lus, Leonardo G. O.; Duarte-Mendes, Pedro; et al. "Agreement between dual x-ray absorptiometers using pencil beam and fan beam: indicators of bone health and whole-body plus appendicular tissue composition in adult athletes". *Revista da Associação Médica Brasileira* 64 4 (2018): 330-338. http://dx.doi.org/10.1590/1806-9282.64.04.330.

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16 Coelho-e-Silva, Manuel J.; Rebelo-Gonçalves, Ricardo; Martinho, Diogo; Ahmed, Alexis; Luz, Leonardo G. O.; Duarte, João P.; Severino, Vítor; et al. "Reproducibility of estimated optimal peak output using a force-velocity test on a cycle ergometer". *PLOS ONE* 13 2 (2018): e0193234. http://dx.doi.org/10.1371/journal.pone.0193234.

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Capítulo de livro

1 Manuel J. Coelho-e-Silva; Diogo V. Martinho; João Valente-dos-Santos. "Conceito de Desporto". In *e-Sports: O Desporto em Mudança?*, 1-17. Lisboa, Portugal: Visão e Contextos, 2020.

Publicado

2 Manuel J. Coelho-e-Silva; Diogo V. Martinho. "Dimorfismo sexual, androginia e desporto: factores conspícuos". In *Desporto, Género e Sexualidade*. Lisboa, Portugal: Visão e Contextos, 2018.

Publicado

Livro

1 Daniela C. Costa; Óscar M. Tavares; Joaquim M. Castanheira; Rui Soles-Gonçalves; Fátima Rosado; João Vasco-Miranda; Paulo Sousa-e-Silva; et al. *Composição corporal, futebol e saúde óssea*. Coimbra, Portugal: Universidade de Coimbra. 2018.

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Poster em conferência

1 Beatriz Ferreira; Bruna Pinheiro; Diogo V. Martinho; João Lemos. "DISCRIMINATION OF GLUTEN VS GLUTEN-FREE PRODUCTS BY NUTRICIONAL COMPOSITION AND COST". Trabalho apresentado em *Poster Week - Escola Superior da Tecnologia de Saude de Coimbra*, 2020.

2 Beatriz Ferreira; Diana Costa; Diogo Martinho; João Lemos. "MAIN APPLICATIONS AND NEGATIVE EFFECTS OF SENNA AS LAXATIVE". Trabalho apresentado em *Poster Week - Escola Superior da Tecnologia de Saude de Coimbra*, 2020.

3 Beatriz Ferreira; Diana Costa; Diogo V. Martinho; João Lemos. "ALENTEJO HEALTH PROBLEMS: A POLITICAL INTERVENTION". Trabalho apresentado em *Poster Week - Escola Superior da Tecnologia de Saude de Coimbra*, 2020.

4 Diogo V. Martinho; Joao Valente-dos-Santos; Manuel J. Coelho-e-Silva; João Gonçalves-Santos; Arturo O. Gutierrez; Leonardo G. O. Luz; Dalmo Machado; et al. "Allometric modelling of left ventricular mass in female adolescent players". Trabalho apresentado em *31st Pediatric Work Physiology Children and Exercise*, 2019.

5 Diogo V. Martinho; André O. Werneck; Jorge Conde; Manuel J. Coelho-e-Silva; Artur Pereira; Daniela C. Costa; João P. Duarte; et al. "Allometric scaling of aerobic fitness outputs in school-aged pubertal girls". Trabalho apresentado em *31st Pediatric Work Physiology Children & Exercise*, 2019.

6 Joana O. Rosado; Jose A. Duarte; Paulo Sousa-e-Silva; Daniela Costa; João Duarte; Joao Valente-dos-Santos; Hugo Sarmento; et al. "Body size and composition of ultra-trail runners: differences between recreational and competitive groups". Trabalho apresentado em *World Confederation for Physical Therapy Congress*, 2019.

7 Joana O. Rosado; Jose A. Duarte; Paulo Sousa-e-Silva; Daniela C. Costa; Diogo V. Martinho; Joao Duarte; Joao Valente-dos-Santos; et al. "PHYSIOLOGICAL PROFILE OF ULTRA-TRAIL RUNNERS: COMPARISON BETWEEN RECREATIONAL AND COMPETITIVE ATHLETES". Trabalho apresentado em *World Confederation for Physical Therapy Congress*, 2019.

8 Beatriz Ferreira; Diogo V. Martinho; João Lemos. "The influence of nutrigenomics: contribution to the athletic performance". Trabalho apresentado em *Poster Week - Escola Superior da Tecnologia de Saude de Coimbra*, 2019.

9 Sofia Antunes; Diogo V. Martinho; Diana Vieira; Beatriz Ferreira; João Lemos. "DIFFERENCES AMONG ANTIDEPRESSANTS AND FOOD-DRUG INTERACTIONS". Trabalho apresentado em *Poster Week - Escola Superior da Tecnologia de Saude de Coimbra*, 2019.

10 Catarina S. Santos; Mario J. Costa; Raul F. Bartolomeu; Tiago M. Barbosa; Joao P. Duarte; Diogo V. Martinho; Luis M. Rama. "Assessment of upper -limbs' symmetry in water fitness exercises". Trabalho apresentado em *XII SIMPOSIO INTERNACIONAL DE FUERZA Y PROYECTO IRONFEMME*, 2019.

11 Catarina Grumete; Diogo V. Martinho; Filipa Santos; Joana Fernandes; Joana Abreu; Joana Mesquita; Pamela Vazquez. "Bariatric Surgery and Satiety Mechanisms". Trabalho apresentado em *Poster Week - Escola Superior da Tecnologia de Saude de Coimbra*, 2018.

12 Lea da Cruz; Joao Lemos; Diogo V. Martinho. "THE EFFECTS OF BEEF AND WHEY PROTEIN ON BODY COMPOSITION". Trabalho apresentado em *Poster Week - Escola Superior da Tecnologia de Saude de Coimbra*, 2018.

13 Joaquim M. Castanheira; Joao Valente-dos-Santos; Daniela C. Costa; Diogo Martinho; Jorge Fernandes; Joao Duarte; Nuno Sousa; et al. "Remodelagem Cardíaca em Atletas Adolescents". Trabalho apresentado em *Jornadas de Cardiologia do Centro*, 2017.

Resumo em conferência

1 Valente-dos-Santos, J.; Martinho, D. V. ; Lourenço-Farinha, P.; Duarte, J.P.; Coelho-e-Silva, M. ou Coelho-e-Silva, M.J. ou Coelho-Silva, M ou Silva, M.; Oliveira, T.; Rodrigues, I.; et al. "ASSESSMENT OF BIOLOGICAL MATURATION IN YOUTH FEMALE SOCCER PLAYERS: AGREEMENT BETWEEN GREULICH-PYLE AND FELS PROTOCOLS". Trabalho apresentado em *25th Annual Congress of the EUROPEAN COLLEGE OF SPORT SCIENCE*, 2020.

Publicado

2 Diogo V. Martinho; Joao Valente-dos-Santos; Yuri V. Faustino-da-Silva; Jorge Conde; Tomás G. Oliveira; Enio R. V. Ronque; Ricardo R. Agostinete; et al. "Comparison of Greulich-Pyle skeletal ages in youth female soccer players". Trabalho apresentado em *XV SEMINÁRIO DE DESENVOLVIMENTO MOTOR DA CRIANÇA, Coimbra*, 2020.

Publicado

3 Martinho, D. V.. "Developmental analysis of antagonist-agonist strength relationship in knee joint muscle groups: isokinetic dynamometry assessment at 60°/s angular velocity in soccer players". Trabalho apresentado em *XV SEMINÁRIO DE DESENVOLVIMENTO MOTOR DA CRIANÇA*, 2020.

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4 Beatriz Ferreira; Bruna Pinheiro; Diogo V. Martinho; João Lemos. "DISCRIMINATION OF GLUTEN VS GLUTEN-FREE PRODUCTS BY NUTRICIONAL COMPOSITION AND COST". Trabalho apresentado em *Poster Week - Escola Superior da Tecnologia de Saude de Coimbra*, 2020.

5 Beatriz Ferreira; Diana Costa; Diogo V. Martinho; João Lemos. "MAIN APPLICATIONS AND NEGATIVE EFFECTS OF SENNA AS LAXATIVE". Trabalho apresentado em *Poster Week - Escola Superior da Tecnologia de Saude de Coimbra*, 2020.

6 Beatriz Ferreira; Diana Costa; Diogo Martinho; João Lemos. "ALENTEJO HEALTH PROBLEMS: A POLITICAL INTERVENTION". Trabalho apresentado em *Poster Week - Escola Superior da Tecnologia de Saude de Coimbra*, 2020.

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7 Catarina Grumete; Diogo V. Martinho; Filipa Santos; Joana Fernandes; Joana Abreu; Joana Mesquita; Pamela Vazquez. "BARIATRIC SURGERY AND SATIETY MECHANISMS". Trabalho apresentado em *Poster Week - Escola Superior da Tecnologia de Saude de Coimbra*, *Coimbra*, 2019.

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8 Sofia Antunes; Diogo V. Martinho; Diana Vieira; Beatriz Ferreira; João Lemos. "DIFFERENCES AMONG ANTIDEPRESSANTS AND FOOD-DRUG INTERACTIONS". Trabalho apresentado em *Poster Week - Escola Superior da Tecnologia de Saude de Coimbra*, *Coimbra*, 2019.

9 Sofia Antunes; Diogo V. Martinho; Diana Vieira; Beatriz Ferreira. "THE INFLUENCE OF NUTRIGENOMICS: CONTRIBUTION FOR THE ATHLETIC PERFORMANCE". Trabalho apresentado em *Poster Week - Escola Superior da Tecnologia de Saude de Coimbra*, 2019.

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10 Joao Valente-dos-Santos; Ricardo Rebelo-Gonçalves; Diogo V. Martinho; Joao P. Duarte; Daniela C. Costa; Paulo Sousa-e-Siva; Vitor Severino; et al. "Reproducibility of Force-Velocity Test Outputs Using 10-s Sprints Against Different Braking Forces". Trabalho apresentado em *American College of Sports Medicine*, *Minesotta*, 2018.

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11 Joao P. Duarte; Joao Valente-dos-Santos; Daniela C. COSTA; Sousa-e-Silva, P.; Diogo V. Martinho; Andre Seabra; Rui-Soles Gonçalves; Manuel J. Coelho-e-Silva. "Reproducibility Of Isokinetic Strength Assessment Of Knee Extensors And Flexors Adopting Concentric And Eccentric Contractions". Trabalho apresentado em *2018 Annual Meeting, World Congress on Exercise is Medicine*, *Minneapolis, Minnesota*, 2018.

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12 Luís Rama; Martinho, Diogo Vicente; Coelho e Silva, M.; Duarte, João; Costa, Daniela; Silva, Paulo; Teixeira, AM.. "Prevalence of the degradation cartilage marker, oligomeric matrix protein (COMP), in males cross training and ultra trail runners". Trabalho apresentado em *23rd Annual Congress of the European College of Sport Science*, *Dublin*, 2018.

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13 Lea Da Cruz; Joao Lemos; Diogo V. Martinho. "THE EFFECTS OF BEEF AND WHEY PROTEIN ON BODY COMPOSITION". Trabalho apresentado em *Poster Week - Escola Superior da Tecnologia de Saude de Coimbra*, 2018.

14. Arturo O. Gutierrez; Joao Valente-dos-Santos; Joao P. Duarte; Diogo V. Martinho; Paulo Sousa-e-Silva; Daniela C. Costa; Leonardo G. O. Luz; Joaquim M. Castanheira; Manuel J. Coelho-e-Silva. "ASSESSMENT OF SKELETAL MATURATION AMONG ADOLESCENT FEMALE SOCCER PLAYERS: agreement between FELS and TW3 protocols". Trabalho apresentado em *Pediatric Work Physiology*, *Katerin*, 2017.

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15. Joaquim M. Castanheira; Joao Valente-dos-Santos; Daniela C. Costa; Diogo V. Martinho; Jorge Fernandes; Joao Duarte; Vasco Vaz; et al. "INDICADORES DE REMODELAGEM CARDÍACA EM ATLETAS ADOLESCENTES DE ELITE". Trabalho apresentado em *XVI Congresso de Ciências do Desporto e Educação Física dos Países da Língua Portuguesa*, *Coimbra*, 2017.

16. Diogo V. Martinho; Maria J. Almeida; Manuel J. Coelho-e-Silva; Afonso A. Machado. "Orientação para a meta e ansiedade traço competitiva: Análise fatorial e as diferenças por tipo de esporte". Trabalho apresentado em *III Congresso Nacional de Psicologia de Motricidade Humana, Esporte, Recreação e Dança*, *Rio Claro*, 2014.

    Publicado

17. Ricardo Belli; Diogo V. Martinho; Gustavo H. Leso; Eduardo Barros; Vasco Vaz; Gonçalo Dias; António J. Figueiredo; Afonso A. Machado. "Orientação motivacional e ansiedade em jovens jogadores de futebol de diferentes nacionalidades". Trabalho apresentado em *III Congresso Nacional de Psicologia, Esporte, Recreação e Dança*, *Rio Claro*, 2014.

    Publicado

18. Ivo Rego; Rafael Baptista; Diogo V. Martinho; João M. Baptista-Almeida; João Pereira; João P. Duarte; Vitor Severino; et al. "Effects of size and maturation on cycling and running protocols before and after allometric scaling in youth basketball". Trabalho apresentado em *PWP*, *Coimbra*, 2013.

    Publicado

Atividades

Apresentação oral de trabalho

	Título da apresentação	Nome do evento Anfitrião (Local do evento)
2020/12/03	Assessment of biological maturation in youth female soccer players: (1) Reproducibility and inter-observer agreement of Greulich-Pyle protocol to estimate skeletal age among female adolescent soccer players; (2) Agreement between Greulich-Pyle and Fels protocols; (3) Scaling left ventircular mass in adolescent female soccer players	XXIVa International Scientific Conference Physical Activity of People at Different Age International Association of Sport Kinetics (Polónia)
2020/11/14	Comparison of Greulich-Pyle skeletal ages in youth female soccer players	XV SEMINÁRIO DE DESENVOLVIMENTO MOTOR DA CRIANÇA

Organização de evento

	Nome do evento Tipo de evento (Tipo de participação)	Instituição / Organização
2019 - 2019	XIX Forum Internacional do Desporto Simpósio (Outra)	

Participação em evento

	Descrição da atividade Tipo de evento	Nome do evento Instituição / Organização
2019/09/10 - 2019/09/14	31st Pediatric Work Physiology Children & Exercise Congresso	31st Pediatric Work Physiology Children & Exercise
2017 - 2017	Antropometria no Desporto Encontro	V Encontro Nacional de Estudantes de Dietética e Nutrição Escola Superior de Tecnologia da Saúde de Coimbra, Portugal
2017 - 2017	Bone Health and Sport Participation Encontro	Bone Health and Sport Participation Faculdade de Ciências do Desporto e Educação Física, Portugal

Arbitragem científica em revista

	Nome da revista (ISSN)	Editora
2021/01/04 - Atual	Scientific Reports	
2019/01/15 - 2021/01/01	BMC Pediatrics (1471-2431)	Springer (Biomed Central Ltd.)
2020/07/17 - 2020/12/20	Motriz, Journal of Physical Education	
2020/01/07 - 2020/04/23	Annals of Human Biology	

Curso / Disciplina lecionado

	Disciplina	Curso (Tipo)	Instituição / Organização
2018 - Atual	Estudos Práticos III - Basquetebol Auxologia e Cineantropometria Desporto Infanto Juvenil		
2021 - 2021	Fisiologia do exercício Bioenergética e metabolismo	(Outros)	Instituto Politécnico de Coimbra Escola

Membro de associação

	Nome da associação	Tipo de participação
2017/03 - Atual	Centro de Investigação do Desporto e Atividade Física	