Imaging of the Muscle-Bone Relationship

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Abstract: Muscle can be assessed by imaging techniques according to its size (as thickness, area, volume, or alternatively as a mass), architecture (fibre length and pennation angle), with values used as an anthropometric measure or a surrogate for force production. Similarly, bone's size (as area or volume) can be imaged using MRI or pQCT, although typically bone mineral mass is reported. Bone imaging measures of mineral density, size and geometry can also be combined to calculate bone's structural strength – measures being highly predictive of bone's failure load *ex vivo*. Imaging of muscle-bone relationships can hence be accomplished through a number of approaches by adoption and comparison of these different muscle and bone parameters, dependent on the research question under investigation. These approaches have revealed evidence of direct, mechanical muscle-bone interactions independent of allometric associations. They have led to important information on bone mechanoadaptation and the influence of muscular action on bone, in addition to influences of age, gender, exercise and disuse on muscle-bone relationships. Such analyses have also produced promising diagnostic tools for clinical use, such as identification of primary, disuse-induced and secondary osteoporosis and estimation of bone safety factors. Standardisation of muscle-bone imaging methods is required to permit more reliable comparisons between studies and differing imaging modes, and in particular to aid adoption of these methods into widespread clinical practice.

Keywords: Muscle, Bone, Imaging, BMD, DEXA, pQCT.

A – IMAGING THE PROBLEM

The dynamic muscle-bone relationships can be analyzed by imaging masses, structures, and their interactions [1]. Imaging *masses* requires analysis of anthropometric features related to non-directional magnitudes. Imaging *structures* involves analyses of direction-related properties. Imaging *interactions* requires the analysis of mathematical relationships between image-derived, directional and non-directional variables. There are three particularly attractive approaches to those purposes, namely, 1. imaging muscle mass and structure as force generators, and imaging bone mass/structure concerning their ability to 2. transform forces into stress, and 3. adapt to stress (force/area values).

1. Imaging muscles as force-generators

The power-generating step in muscle contractions is the cross-bridge interaction between myosin and actin filaments [2]. Force, and velocity, of a given contraction are therefore determined by the number of active cross-bridges. Within skeletal muscle fibres, actin and myosin filaments are parallel-aligned and neatly bundled into sarcomeres, thus endowing the many cross-bridges with a direction. Muscle force increases with the number of sarcomeres-in-parallel, whilst contraction velocity increases with the number of sarcomeres-in-series [3]. Therefore, as an anatomical approximation of a muscle's, or a muscle group's force generating capacity we can identify the cross-sectional area (CSA). The anatomical CSA (aCSA), *i.e.* the CSA perpendicular to the origin-insertion axis, is easily identifiable with conventional X-ray based or conventional T1 or T2 –weighted magnetic resonance (MR) images. The physiological CSA (pCSA) takes into account the pennation angle in those muscles (Figure 1), where the fibres' direction deviates from the anatomical axis of the muscle, which is the case for most of our muscles. Mathematically speaking, pennation packs more sarcomeres-in-parallel into a volume, albeit at the expense of reducing contraction speed. pCSA is a better predictor of a muscle's force generating capacity than aCSA [4]. To assess it requires measurement of the pennation angle, which can be done by ultrasound for parts of a muscle, or with MR diffusion tensor imaging [5].

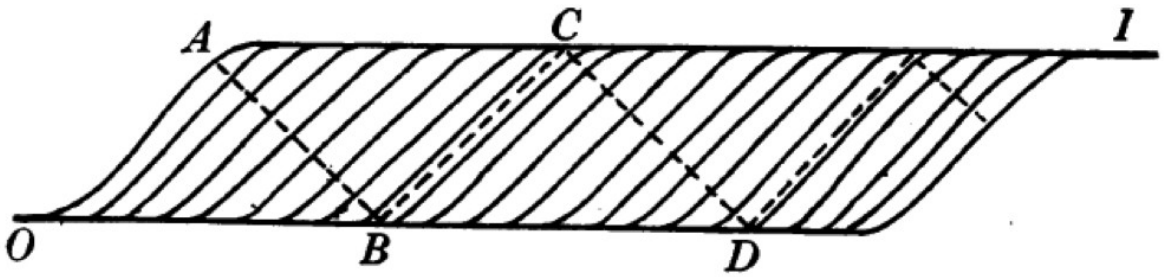


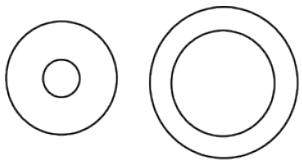
Fig. 1 Diagram of a unipennate muscle indicting fibre orientation (solid diagonal lines), line BC indicates appropriate plane for measurement of physiological cross-sectional area (pCSA). Anatomical cross-sectional area (aCSA) is that resulting from a section orthogonal to the upper aponeurosis line ACl. Reproduced with permission from [4].

2. Imaging bone ability to receive forces

At the tissue level, bone has properties of material stiffness (the degree of deformation under a given stress), material strength (ability to resist fracture), and toughness (ability to dissipate energy). The stiffness of cortical and trabecular bone as a material is strongly associated with bone mineral density [6, 7], which can be measured by cross-sectional densitometric techniques such as peripheral quantitative computed tomography (pQCT).

As an organ, a bone also has properties of structural stiffness and toughness, both of which together determine its strength (the maximal stress the bone can stand until fracture). There are three principle kinds of strains and stresses, namely compression, tension and shear. Bones as structures can experience many different kinds of deformations, of which loading in bending and torsion are common examples. Strength/stiffness in compression is related to bone mass in the cross-section orthogonal to the applied stress. Assuming invariant density, a larger bone cross-section results in greater bone mineral content (BMC) - measures which are more strongly correlated with axial bone strength than BMD measures alone [8, 9]. Measurement of bone diameters, or periosteal and endocortical perimeters describe changes in bone shape or cortical thickness which can evaluate some relative or combined effects on the corresponding, periosteal and endocortical

surfaces. However, the distribution of bone mass relative to the centre of mass also affects its stiffness and strength in bending and torsion. Cross-sectional moment of inertia (CSMI) of the cortical bone area in long bones indicates bone's stiffness in bending (axial CSMI) or torsion (polar CSMI) and is calculated as the sum of the voxel areas multiplied by the square of the distance (either in a single plane in the case of axial CSMI, or absolute distance in the case of polar CSMI) of each voxel from the centre of bone mass (Figure 2). Moment of resistance (alternatively known as section modulus) is calculated by dividing the moment of inertia by the outer radius and indicates bone's strength in bending or torsion. Therefore – for example – the same bone mass organized into a large diameter, thin-walled tube will be stiffer in torsion (and indeed in bending) than a small diameter, thick-walled tube.



CSA:	6.3 cm^2	6.3 cm^2
I_p:	6.28 cm^4	7.47 cm^4
R_p:	20.1 cm^3	20.9 cm^3
$Radius_{out}$:	1.5 cm	2.0 cm
$Radius_{in}$:	0.5 cm	1.4 cm

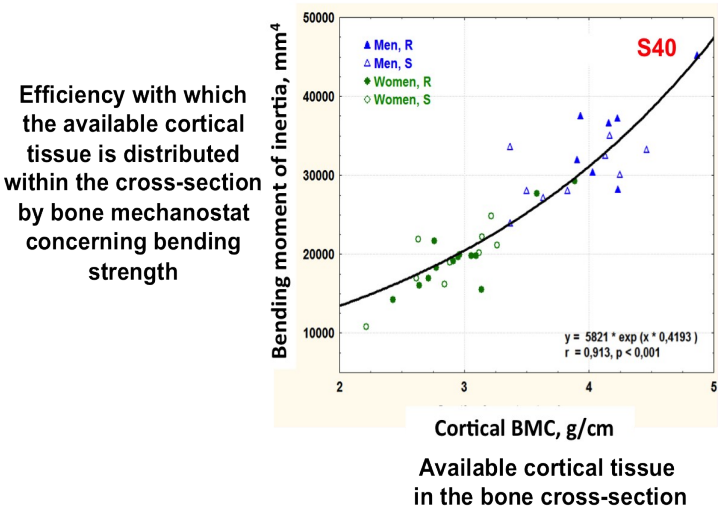
Fig. 2 Geometrical properties and strength in cylinders. CSA – Cross-sectional area. I_p – Polar moment of inertia. R_p – Polar moment of resistance. $Radius_{out}$ – outer cylinder radius. $Radius_{in}$ – inner cylinder radius

The relationships between CSMIs and bone strength tend to vanish when bone diameter is proportionally much larger than cortical thickness. In this situation, CSMIs can be very large, but the bone can fail in buckling. The tendency to do so is estimated by the *buckling ratio* = periosteal diameter / cortical thickness. Within these limitations, the CSMIs can be combined with tissue mineral density to create density-weighted moments of inertia (also known as Bone Strength Index BSI). BSI has been shown to be a stronger predictor of bone breaking strength than its components CSMI or cortical

BMD , explaining 89% of variance in strength [10]. Similarly, density-weighted section modulus (strength-strain index, or SSI) has been derived, and explained 98% of variance in fracture load in human tibia – this association being stronger than those between fracture load and other bone measures such as cortical area and section modulus without density weighting [11].

3. **Imaging bone ability to adapt to the mechanical environment**

- *Within-bone relationships*. Bone modeling and remodeling can be both modulated and spatially-oriented as a function of mechanical usage by bone *mechanostat*. This can result in re-orientation of cortical shells and trabecular networks as structural adaptations of bone tissue distribution to the induced stresses. These bone properties can be assessed by correlating different image-derived indicators of geometric properties (y) as a function of indicators of bone tissue mass (x_1), as “distribution/mass” (d/m) relationships, or stiffness (x_2), as “distribution/quality” (d/q) relationships (Figure 3).



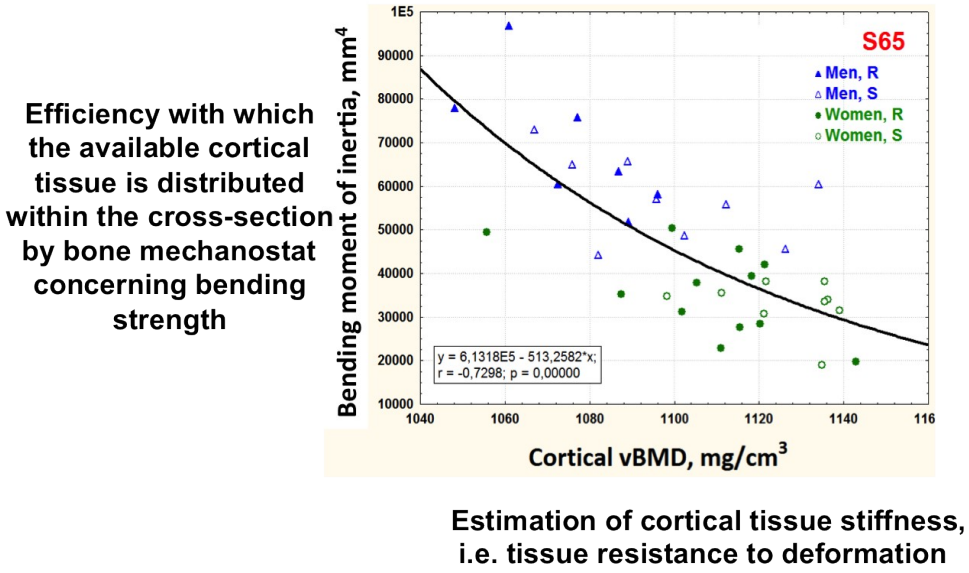


Fig. 3 Typical “distribution/mass” (d/m, left) and “distribution/quality” (d/q, right) curves obtained from scans taken at 40% (d/m) and at 65% (d/q) of the tibia height in male and female individuals who were untrained (S) or trained in long-distance running (R) [12]

- *Muscle-bone associations and interactions*. These can be assessed by imaging four different kinds of general (ubiquitous) muscle/bone associations, namely, 1. mass/mass (anthropometric), 2. structure/force (mechanical, translational), 3. structure/structure (mechanical, static) and 4. force/stress (mechanical, dynamic) relationships, as well as 5. some site-specific applications of the same, as follows.

B – OVERVIEW OF THE PARTICULAR MATTERS INVOLVED IN THE ANALYSIS OF MUSCLE-BONE ASSOCIATIONS AND INTERACTIONS

I – General approaches

1. Imaging muscle/bone mass/mass (anthropometric) relationships.

The biomechanical influences of muscles on bones as assessed by mass/mass relationships are blunted by natural morphogenetic associations [13-18], yet there is some evidence of a direct, mechanical interaction [19-23]. In fact, DXA studies of the whole-body and limbs (standard determinations of lumbar spine, femur and radius are unsuitable for this purpose) have shown that mineral (BMC, *y*) and lean (related to muscle, *x*) masses are linearly related in both sexes at any age with similar slopes [24-29] (Figure 4). However, the intercepts of those relationships differed in the order: children<men=post-MP women<pre-MP women [30, 31]. While similar slopes are compatible with the identity of bone *mechanostat* in the species [32], different intercepts would indicate the agonistic interference of sex hormones in the mechanical control of bone features [33].

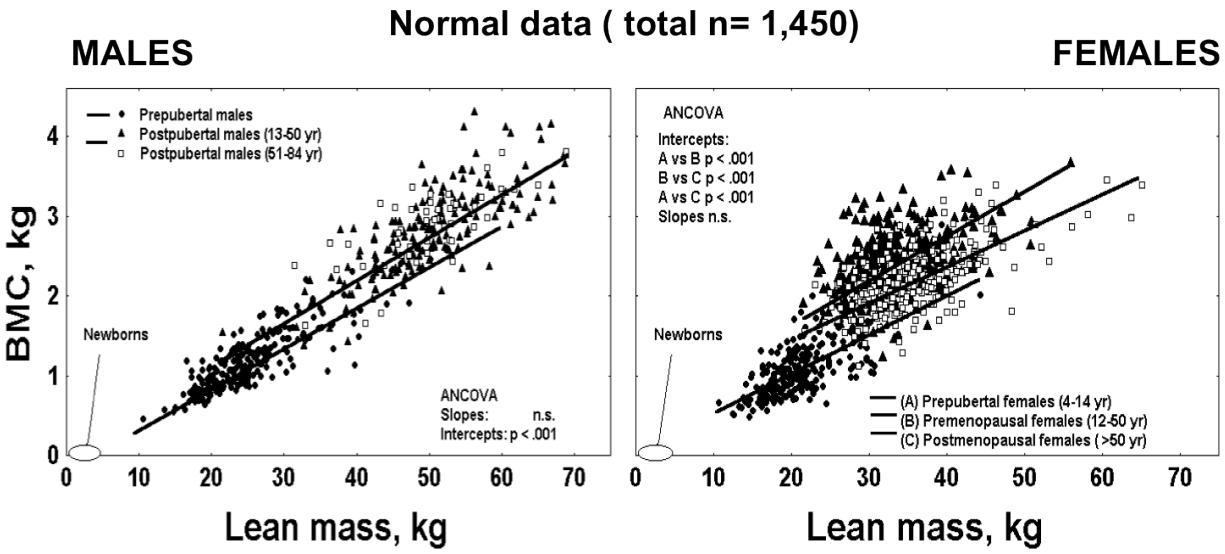


Fig. 4 Relationships between the DXA-assessed, whole-bone BMC (y) and lean mass(x) of a representative sample of healthy children, men, and pre- and post-menopausal women [30]

We have standardized those relationships for whole-body and limb measures in 3,000+ normal men and women and provided Z-scored charts for comparative diagnoses of *osteopenia* [34] which are specific to the type of device employed (Figure 5). Importantly, these relationships do not capture any structural variable related to actual muscle and bone *strength* [35]. However, the charts allow proposal of a predominantly “mechanical” or “metabolic” nature of the studied osteopenia (not osteoporosis) when compared to the values of BMC/lean mass ratio and lean mass data of the studied individual, respectively [36].

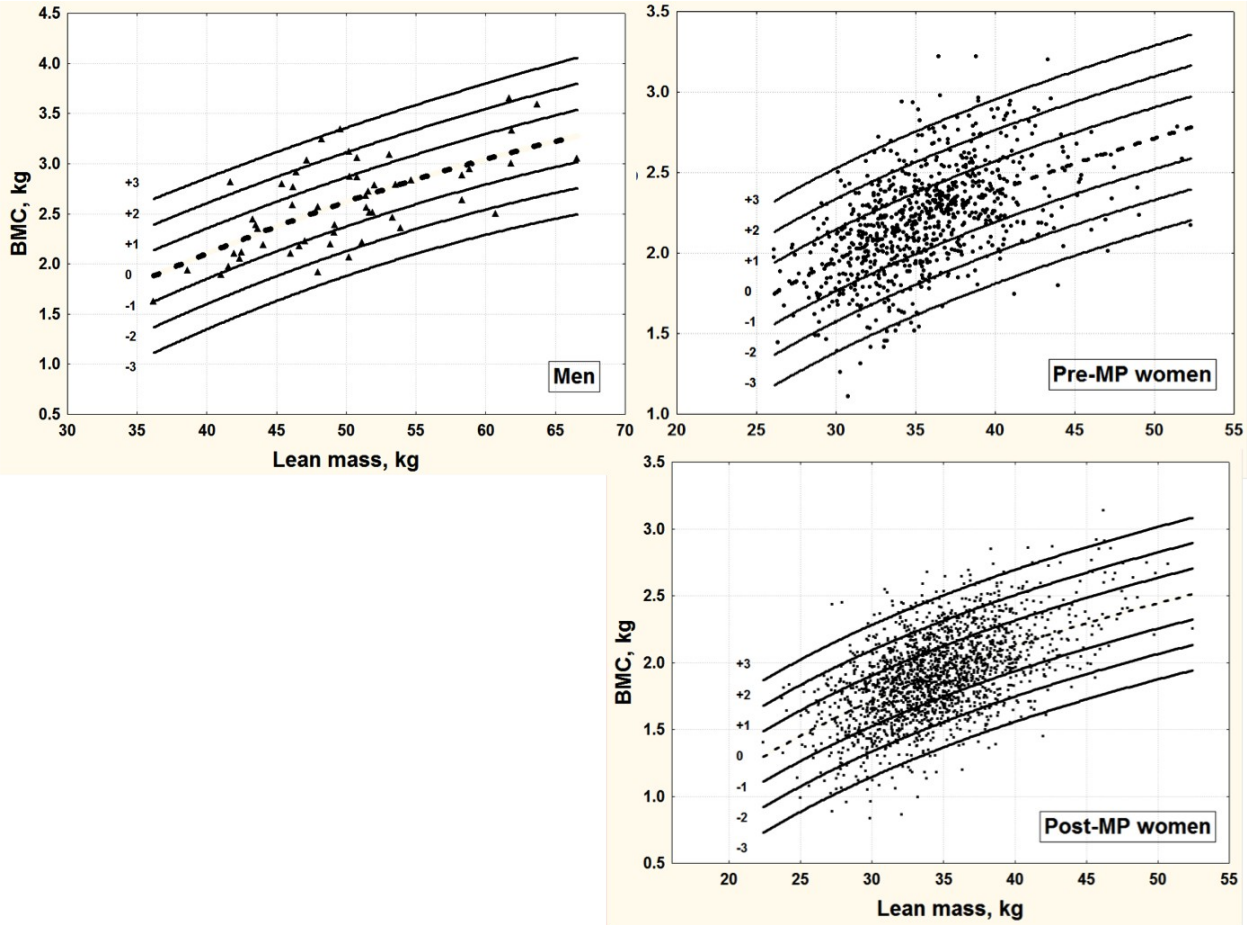


Fig. 5 Z-scored charts of the relationships between the DXA-assessed, whole-bone BMC (y) and lean mass (x) of a representative sample of healthy men and pre- and post-menopausal women [34]

This distinction may orient the therapeutic indications of physical or pharmacological treatments. Low bone/muscle proportionalities were observed in post-MP women with typically osteoporotic fractures [37]; in thyro-parathyroidectomized children [38]; in very lean, amenorrheic female ballet dancers, and in chronically haemodialized patients [unpublished].

2. Imaging muscle/muscle structure/structure (mechanical, translational) **relationships** (Combined cross-sectional (pQCT, etc) + dynamometric methodologies) -> **Muscle analysis**.

As discussed in A1 , and *ceteris parabus*, a larger muscle will produce greater force. Imaging of muscle size by MRI has revealed strong associations

between measures and maximal force. However, muscle size can be assessed as a three- (volume), two- (CSA) or one-dimensional (thickness) scalar measure. Due to time or equipment restrictions single site assessments of cross-sectional area or muscle thickness are commonly measured *in lieu* of muscle volume. The three values are unsurprisingly highly correlated [39] and are each also strongly related to maximal force [40-42]. Comparisons of the predictive ability of their different measures have produced varying results, with MRI-assessed elbow extensor and flexor volume explaining over 90% of variance in the isometric joint torques produced by their respective actions, whereas associations between torque and muscle CSA, and muscle thickness assessed by ultrasound in the same participants were weaker [42]. Conversely, other studies have shown similar relationships between maximal force and muscle CSA or volume [43].

Imaging muscle bone relationships makes the inherent assumption that all muscles have identical intrinsic strength, *i.e.* generate the same peak tension. This is, of course, not generally true. Old age and immobilization-induced atrophy, for example, entail reductions in the peak tension of isolated muscle fibres [44]. The latter effect seems to be caused by reduced concentration of contractile apparatus within the muscle cells. Moreover, muscle dystrophic disorders [45], multiple sclerosis [46] and possibly others. There is a long and undecided debate whether or not children have lower intrinsic strength than adults [47-49].

Boundaries such as age and clinical disorders must therefore be considered when using muscle CSA for clinical and scientific inferences. Attempts have been made to obtain detail of differences in size-adjusted force or 'muscle quality' by imaging methods. One such method is analysis of muscle X-ray attenuation (MXA) as obtained from computed tomography. MXA decreases with age [50] and is associated with specific tension of a muscle [51]. However, a lack of understanding of the physiological properties of muscle underlying MXA values is likely responsible for the lack of widespread adoption of the technique.

> **3. Imaging muscle/bone structure/structure** (mechanical, static) **relationships**
> (Cross-sectional methodologies only) -> *bone* mass, material quality, design and/or strength indicators vs *muscle* mass, cross-sectional properties, and/or force indicators ; muscle-bone strength indices) -> **Classification of osteoporoses**.

Beyond the meaningfulness of anthropometric muscle-bone mass/mass relationships (**B-I.1**), the biomechanical muscle-bone associations can be approached more specifically by comparing image analyses of bone features as described in **A-2,3** and muscle characteristics as referred to in **A-1**. To this purpose, it is generally preferable to select cross-sectional imaging data, as provided by QCT, pQCT, or similar techniques, rather than “areal” DXA data. Many of the variables measured cross-sectionally can be regarded as indicators of bone mass (total, trabecular or cortical BMC, total or cortical bone area, total or trabecular vBMD), bone material stiffnes (cortical vBMD), bone cross-sectional design (diameters, perimeters, cortical thickness, CSMIs, *buckling ratio*) or bone strength (BSIs, SSI). Bone trabecular structure can be assessed quasi-histomorphometrically by HR-pQCT. However, in this case the biomechanical interpretation of the data may be limited because the methodology does not capture the directional disposition of the trabecular network in relation to the direction of the forces which would break the bone. Muscle force can be easily (though indirectly) evaluated as the muscle cross-sectional area, which can be deprived of its fat content by filtration. Of course, dynamometrical data of the real muscle force are more suitable for this kind of analysis.

Image-derived muscle strength indicators usually correlate positively with all the above bone mass or strength indicators or with the CSMIs. Typically, in normal individuals, bone mass (tibial + fibula BMC in pQCT scans) correlates linearly with the maximal muscle CSA of the calf, comprising the origin (Figure 6a). Figure 6b shows the relationship between the A-P bending CSMI of the tibia and calf muscle CSA in the same cohort.

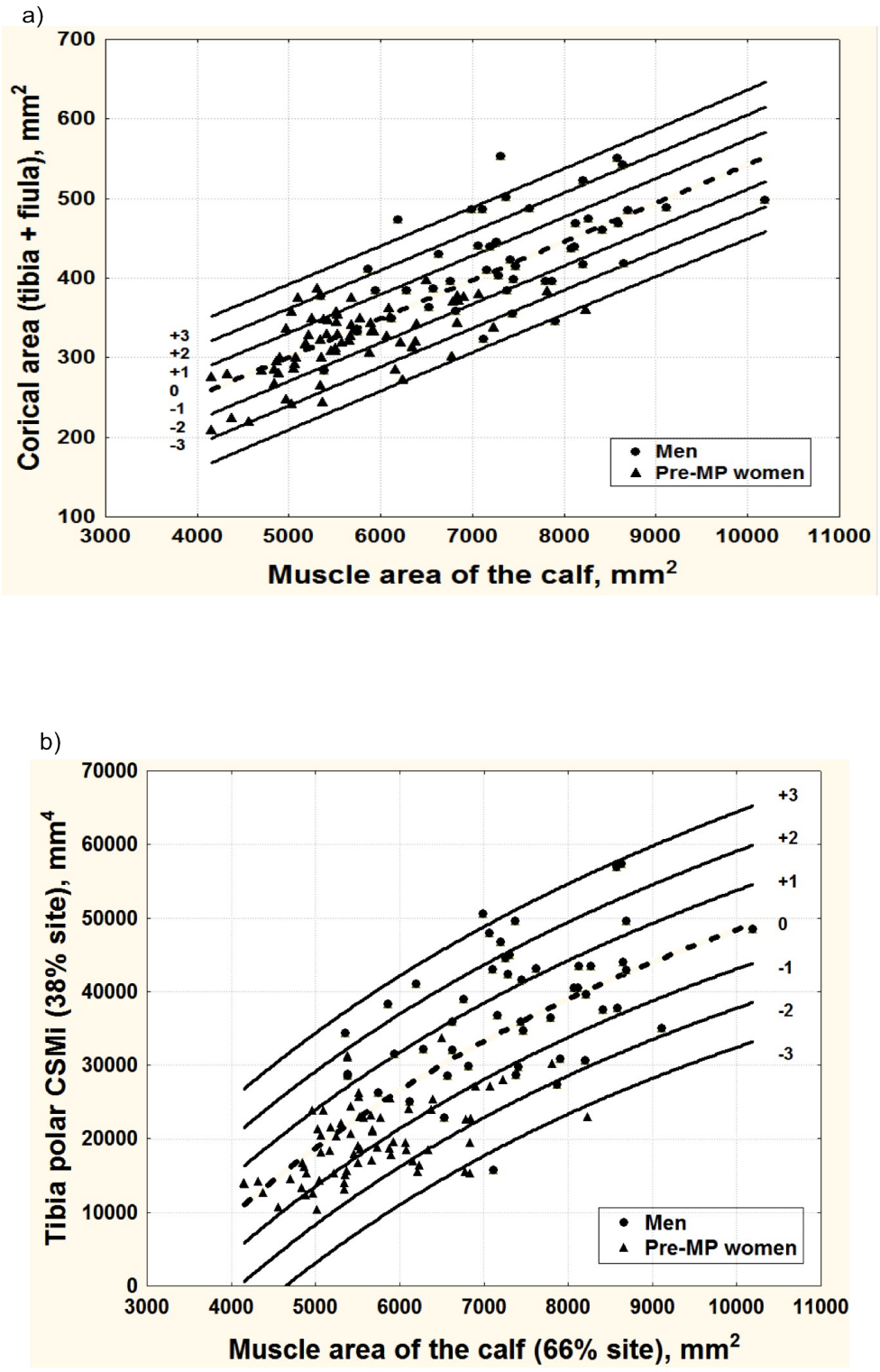


Fig. 6 a) Relationships between the maximal cross-sectional muscle area of the calf (x) and a) the cortical bone area of tibia+fibula (y) , b) polar CSMI of

the tibia cross-sectional cortical bone area (y), both scanned by pQCT at 66% of the tibia height in a representative sample of healthy men and pre-menopausal women. The Z-scores of the corresponding distributions are indicated as a reference.

In agreement with the mechanostat theory, these correlations can evaluate the efficiency of the servo-controlled adaptation of bone design as a function of mechanical usage, and also distinguish between “mechanical” and “systemic” osteopenias as previously described. These relationships tend to differ greatly between men and women, chiefly because of 1. the larger values of all “extensive” indicators usually observed in male individuals, as a result of androgen-influences on both muscles and bones, with their obvious mechanical consequences, and 2. the estrogen-induced accumulation of bone mass (mostly trabecular tissue) per unit of muscle mass observed in pre-menopausal females, which tend to disappear after menopause. On the other hand, correlations of muscle indicators with the bone stiffness indicator, cortical vBMD, or other geometric indicators as the endocortical perimeter, cortical thickness, or the *buckling ratio* (i.e., variables which are not supposed to be directly regulated by bone mechanostat) are rather weak.

4. Imaging muscle/bone force/stress (mechanical, dynamic) relationships.

(Combined dynamometric + cross-sectional methodologies) -> safety factors estimation -> **True diagnosis of bone fragility**.

Stress (σ) is defined as a force divided by the transversal area on which it actuates. The maximal effective compression force, *usual* F_{max} induced by a contraction of the regional muscles on a long bone which can be assumed to resist mostly uniaxial compression can be measured dynamometrically. In normal conditions, *usual* σ_{max} is about 30 N / mm^2. It was estimated from experimental measurements that, also in normal conditions, the maximal stress a bone can resist in compression prior to fracture is *Fx* σ_{max} = 180 N /mm^2. Thus, and according to the Utah Paradigm of Skeletal Physiology [52], this would give a “safety factor” SF = *Fx* σ_{max} / *usual* σ_{max} = about 6 to normal bones working in compression. This permits calculation of the *theoretically necessary* cortical bone CSA_t required to support *Fx* σ_{max} in practical terms as CSA_t = 6 * *usual* F_{max} (kg) / 18 kg/mm^2, expressed in mm^2. As long as the *real* bone CSA of the individual studied (CSA_r) can be directly

determined by pQCT or similar techniques in the same area units, the percent relationship, 100 * CSA_r/CSA_t should estimate in what proportion the bone satisfies the SF predicted by the paradigm at the studied region and concerning the applied mode of deformation. This procedure is schematized in Figure 7 for a situation in which the distal tibia was selected for study as a bone region known to resist mostly uniaxial stress during customary mechanical usage [53, 54]. Whilst such approaches are only applicable in this manner to cortical bone (which varies little in density in adulthood outside of pathological conditions and old age), there also exists a relationship between trabecular bone compressive strength and its apparent density [55]. More precisely, the relationship is well described by power functions with exponents that are somewhat depending on the anatomical site. However, assuming an exponent of 2 seems to be a reasonable general approach [56]. Hence this idea could likely be adapted for epiphyseal bone. This or similar approaches (after the necessary validation and standardization) could evaluate the *degree of fragility* for a given bone, in biophysically reliable (stress) units. Such evaluation, performed in an osteopenic individual, can estimate to what extent his/her osteopenia has impacted bone strength in the studied region, with the corresponding specificity concerning site and mode of deformation. Thus, this should be a more reliable method to diagnose *osteoporosis* than the mechanically-irrelevant, -2.5 DMO T-score limit established for standard DXA determinations.

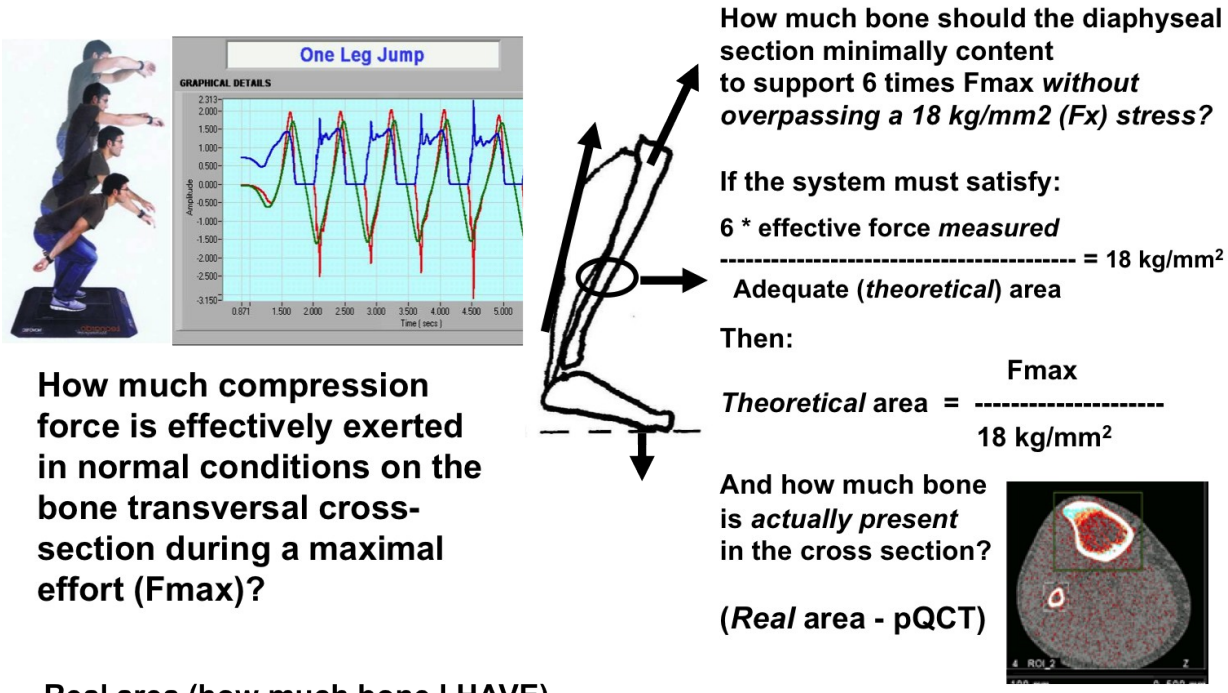


Fig. 7 Calculation of bone safety factor (SF)

II – Site-specific approaches

Imaging of the muscle-bone relationship has been used to provide information on both bone physiology in basic science, and pathophysiology within research and clinical settings. Muscle-bone strength indices (MBSIs) have been established previously [57]. Compressive MBSI is based on the relationship between muscle CSA (as a surrogate for maximal force) and bone CSA as a surrogate for bone mass – given the lack of significant variation in density of health cortical bone – which indicates bone's compressive strength. Similarly, bending MBSIs were calculated as the relationship between muscle bending moment (the product of muscle CSA and tibia length) and axial moment of resistance indicating bone strength in bending. Compressive MBSIs in the tibia vary throughout the limb length [57], suggesting that the influence of body mass on bone geometry is not pronounced. However, when considering bones adapted to the same compressive strength by controlling for bone mass, longer bones (*i.e.* those with a longer lever arm for bending moments) were stronger in antero-

posterior bending than short bones. Similar results have subsequently been found in the upper limbs, with the addition of much stronger relationships between wider bones (*i.e.* where the moment arm for torsional moments is greater) and strength in torsion when compressive strength was controlled for [58]. This suggests that antero-posterior moments may be the dominating influence on lower limb bones, whereas in the upper limbs torsional moments are most important.

The influence of factors such as exercise participation or disuse conditions, age, gender and pubertal stage on muscle-bone relationships have been considered. Examining the relationship between muscle and bone in exercise or disuse intervention studies is problematic. Firstly, rates of adaptation in muscle strength, size and bone strength are dischordant, particularly in exercise - significant changes in muscle size can be seen within 3 weeks of resistance training [59], whereas mechanoadaptation of bone has time-constants around 1 to 2 years [60]. Existing interventional exercise studies have reported only very meagre increases in bone strength [61], hence correlations between muscle and bone increases in these studies would likely be significantly weakened due to the compounding of large measurement errors relative to the absolute change in values. This could also explain the weak muscle-bone size side difference relationships found in studies of youth tennis players [62] and footballers .

In other studies of youth and elite tennis players [58, 63] whilst strong correlations between upper limb muscle and bone CSA were found (R^2 = 0.73-0.86) different muscle-bone relationships were found in the racquet and non-racquet arm, with bone:muscle ratio being greater in the racquet arm. The authors suggested that this may be a result of the influence of individual muscles on bone, or that the high-impact nature of tennis strokes requires the muscles to act in a very different way to habitual usage. Similarly, a study of derived muscle-bone indices in female controls and elite volleyball players found lower muscle-bone relationships in the athletes [57]. Conversely, in disuse studies the time course of detecting early bone and muscle loss is similar [64, 65], but muscle size changes are more pronounced [66]. No long-term (>1 year) controlled disuse intervention studies have been performed – however, muscle-bone relationships in spinal cord injury patients and controls were similar [67].

Another area of investigation is the effects of age and gender on muscle-bone relationships. Muscle-bone size indices have been shown to vary between genders and pubertal status in adolescents in both pQCT [68, 69]

and DXA studies [70], with pubertal effects likely a result (at least in part) of the dischordant timing of height velocity and lean mass and bone mass velocity during the pubertal growth spurt [71]. Gender effects have been proposed to be a result of additional bone mass accrued in women during childbearing years (potentially as a reservoir for calcium during foetal growth). Supporting this, effects of menopause on muscle-bone relationships have been found with women of child-bearing age having greater bone-muscle mass ratios [72]. Similarly, analysis of a DXA-based large cohort study found that whilst premenopausal women had a greater bone-muscle size ratio than postmenopausal women and men, the curves of the logarithmic equations for the relationships ran parallel to each other for all groups [73]. This is supportive of a common mechanical muscular influence on bone, with additional bone mass accrued during the childbearing years independent of muscular influence. Further to this, it has been suggested that extra bone mass in women is not stored in accordance with the mechanical 'need' of bone regions – this is supported by contrasting findings with regard to gender differences in pQCT-derived muscle-bone size indices at weight-bearing and non weight-bearing sites [57, 63] . However, when individuals within the same age and gender groups are considered, relationships between muscle and bone structure appear to be very consistent. For example, muscle-bone size ratios are similar within normal weight and overweight children [74], and even in the extreme disuse case of spinal cord injury patients [67]. Alongside the extreme exercise case of elite athletes, only in long-term anorexia nervosa patients are muscle-bone relationships found to differ from sedentary controls, with ratios decreasing with disease progression [75].

Within the clinical setting, muscle-bone ratios obtained from imaging have been proposed as a method to distinguish between primary and secondary bone disorders (Figure 8). Primary bone disorders or 'systemic osteopenias' can be attributed to dysfunction in bone metabolism/adaptation – hence, whilst muscle mass is normal bone mass is lower than expected. Conversely, secondary bone defects or 'disuse osteopenias' are where the bone's adaptive processes appear to function correctly but the low bone mass is *secondary* to so-called sarcopenia or dynopenia . Finally, mixed bone defects occur when muscle mass is low, and bone-muscle ratio is lower than expected – indicating dysfunction in bone adaptation in addition to a lower muscle stimulus to the bone. Indeed, a similar schema was employed in analysis of pQCT-derived muscle-bone ratios. The schema could discriminate between healthy children and primary bone defects in frequent

fracture patients and kidney transplant patients, and secondary bone defects in chronic renal failure patients [69]. DXA-derived indices of lean body mass and bone mass also showed children with osteogenesis imperfecta to have a primary bone defect, and frequent fracture patients and those with spinal muscular atrophy to have a secondary bone defect [76].

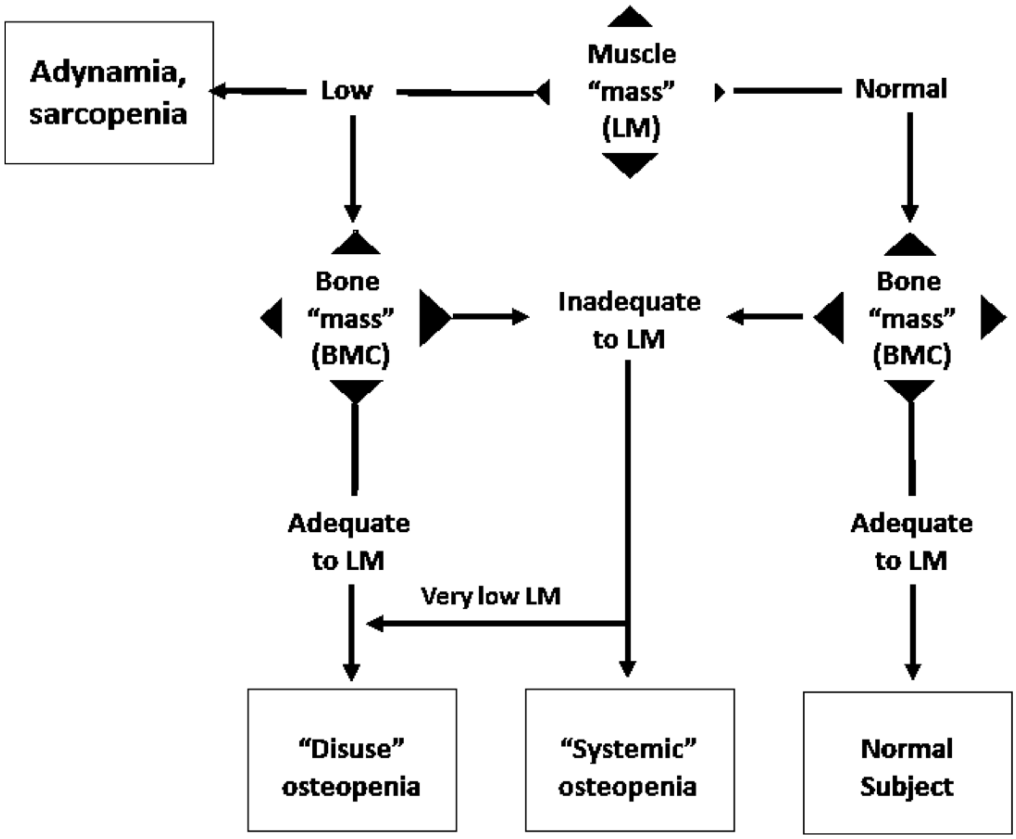


Fig. 8 Didactic representation of the three different etiologies proposed for all bone-weakening diseases, as referred to in the text.

This categorization of bone disorders has implications for patient treatment – *e.g.* in secondary disorders, exercise may function as a treatment route, whereas primary disorders may necessitate a nutritional or pharmacological intervention. Similarly study of muscle-bone relationships showed postmenopausal women with low muscle mass and high muscle-bone ratio having a very high risk of osteoporotic fracture [37]. A recent paper also

proposed MRI-derived muscle-bone volume ratios in the thigh as an index of sarcopenia [77]. However, in order for these assessment methods to transfer fully into a clinical setting (*i.e.* to be established as a typical tool in the clinician's arsenal), some standard methodology is required. Also, in the same way that osteopenia and osteoporosis have standardized definitions, a range of normative muscle-bone ratios can be established from large current datasets such in adults [78] and children [69, 76].

Therefore, it appears that even 'crude' measurements of muscle and bone size can be utilised in classification of bone disorders, and that more complex MBSIs have provided useful information pertaining primarily to bone mechanoadaptation and the influence of muscular action on bone. However, current imaging-based methods are likely limited in their ability to describe muscular influence – new methods such as 3D ultrasound of muscle may help future studies better approximate the muscle-bone relationship, both in research and clinical application.

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