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Hylonome

Perspective Article

Functional assessment of the muscle-bone unit in the lower leg

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Abstract

Based on the mechanostat theory and the muscle-bone hypothesis, a methodological assessment of the musculoskeletal status in health and disease should relate maximum muscle force in relation to bone mass and geometry. While useful (*i.e.* three-dimensional) measures of tibial bone parameters can be obtained by peripheral quantitative computed tomography (pQCT), intrinsic plantarflexor muscle force cannot be directly measured under *in vivo* condition in humans. Instead, tissue size, torque and ground reaction force have been used as proxy markers of intrinsic muscle force. However, most of these proxy markers are not or insufficiently representative of maximum force. Based on our recent research, we describe a novel approach for the assessment of the lower leg muscle-bone unit in health and disease. It incorporates multiple one-legged hopping (m1LH) to assess maximum voluntary ground reaction force acting on the forefoot (F_{m1LH}) and bone mineral content at the 14%-site of tibia length ($vBMC_{14\%}$) as assessed by pQCT. Using the quantitative relationship between these two variables in conjunction with F_{m1LH} per body weight, we present a two-step quantitative diagnostic algorithm to discriminate between primary and secondary bone disorders in children and adults.

Keywords: Maximum Voluntary Muscle Force, Multiple One-Legged Hopping (m1LH), Bone Strength, peripheral Quantitative Computed Tomography (pQCT), Primary and Secondary Bone Disease

The muscle-bone unit

Frost[1] proposed a negative feedback system, *i.e.* the mechanostat, to explain how mechanical usage might influence bone mass and geometry, and postulated that structural adaptation is driven by the experienced bone strains[2,3]. However, the mechanostat theory makes no assumption about the nature of the mechanical forces causing bone strain. In line with Thompson's conception that bone mass is influenced by the developing musculature[4] and based on the data of Zanchetta *et al*.[5], Schiessl *et al*.[6] suggested that except for traumata it must be maximum muscle forces that cause the largest bone strains, primarily due to the poor lever arms most muscles work against. Consequently, muscle and bone form a functional unit, the so-called muscle-bone unit[7].

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Proxy markers of maximum intrinsic muscle force

Following the previous line of reasoning, a very strong relationship should exist between maximum muscle force and bone mass/geometry, and maximum muscle force should be a better predictor for bone mass/geometry than any other proxy marker for maximum force (*e.g.* muscle cross-sectional area, volume, mass, and torque). In this context, it should be realized that the term "muscle strength" is inappropriately nebulous and should be abandoned, because: 1) it has no basis in classical mechanics as outlined by Isaac Newton in 1687 in his three-volume Philosophiae Naturalis Principia Mathematica, and 2) it is not recognized by the Système International d'Unités (SI). We are not free to use terms, nomenclature, units, quantities and forms of expression other than those defined and described in the SI. As previously noted by others[8], "…To do so is simply not science and that non-science can become nonsense…".

Because of the theoretical connotation between (maximum) intrinsic muscle force and bone mass/geometry, any assessment of the musculoskeletal status in health and disease should put maximum muscle force in relation to bone mass/geometry to assess whether bone is properly adapted to muscle, and at which level this is eventually the case. Bone mass and geometry

of the human tibia and radius can accurately and reliably be estimated by peripheral quantitative computed tomography (pQCT), although only at distinct sites and not at the whole-organ level. Conversely, maximum intrinsic muscle force cannot be directly measured under *in vivo* condition in humans. Instead, two types of proxy markers of intrinsic muscle force have been used: a) tissue size (*i.e.* muscle volume[9], cross-sectional area[10] or lean mass[11]) and/or b) torque[12] or ground reaction force[13,14].

Is a bigger muscle a stronger muscle?

The use of tissue size as a surrogate of maximum intrinsic muscle force is limited, mainly because of four reasons. First, the force that a muscle exerts depends on the amount of motor unit activity, the latter changing with the number of motor units that are active (motor unit recruitment) and the rates at which motor neurons discharge action potentials (rate coding). Of these two force coding strategies, only the former is related to muscle fibre size. In fact, for any given motor recruitment at unchanged firing rates, force output will be higher if the number of parallel actin-myosin crossbridges (*i.e.* the number of parallel sarcomeres or fiber cross-sectional area) is increased. Since the upper limit of motor unit recruitment in larger muscles is ~85-95% of maximum voluntary isometric force/torque[15,16], increases in force above this level can only be achieved with adaptations in discharge rate and/or radial muscle fiber hypertrophy, but not by an increase in recruitment (*i.e.* an increase in the number of recruited motor units and thus activated muscle fibers)[15]. It follows that muscle force output can be increased without any concurrent change in muscle size.

Second, muscle force also depends on the type of motor unit activity, and as such on the mix of activated muscle fiber types. In fact, skeletal muscles consist of a continuous spectrum of myofibers, which differ in their myofibrillar proteins (myosin isoforms, in the first place), metabolic enzymes (predominance of glycolytic or mitochondrial activities), but also in any subcellular system, including transmembrane ionic fluxes and intracellular calcium signaling[17]. In particular, force-velocity experiments for human slow and fast fibers show that maximum isometric tension (*i.e.* force per cross-sectional area) is up to 2-fold higher in fast relative to slow fibers[18]. Similarly, optimum shortening velocity and maximum shortening power are markedly higher in type 2 relative to type 1 fibers. Interestingly, mechanical and kinetic properties of the actin-myosin interactions under stretch (active lengthening) are independent of the myosin heavy chain isoform[18]. Consequently, for any given muscle cross-sectional area operating during shortening or isometric contractions, force output at fixed recruitment and discharge rates will be higher, if the share occupied by type 2 fibers is higher. Hence, the same muscle cross-sectional area can lead to different maximum force output, owing to differences in fiber type distribution with respect to the number of fibers and total area per fiber type.

Third, skeletal muscle hypertrophy, *i.e.* the increase in cell volume/protein content (but not number) of muscle fibers, and atrophy result from a homeostatic shift favoring either muscle protein synthesis (MPS) or protein breakdown (MPB), respectively. If the net protein balance is positive (MPS>MPB), muscle protein (and thus mass) is accumulated. One of the most widely recognized mechanisms for controlling muscle mass involves mechanical tension[19]. Mechanical stimuli such as active and passive force can regulate the rate of MPS through changes in translational efficiency and/or translational capacity. Although mechanical stimuli have been shown to affect both of these processes, the primary effect of mechanical stimulation appears to occur at the level of translational efficiency[19]. Therefore, in the case of high mechanical stress, there obviously exists a linkage between muscle mass and bone mass/geometry in that increased mechanical usage concomitantly leads to mechanotransduction in muscle and bone (triggered by bone strain magnitude and/or rate), followed by structural adaptation (increase in muscle mass and bone mass/geometry), and one would expect that these adaptations occur proportionally.

However, mechanical stimuli are not the sole stimuli influencing skeletal muscle mass. In fact, translation is a highly complex process requiring the continuous molecular integration of multiple positive (*e.g.* exercise, amino acids, hormones) and negative stimuli (*e.g.* glucocorticoids), and, furthermore, transcriptional regulation of skeletal muscle can occur through modulation of *e.g.* neural signaling[20]. This is logical if one appreciates that besides locomotion, skeletal muscle serves other fundamental functions during the human life span. For example, muscle plays a central role in whole-body protein metabolism by serving as the principal reservoir for amino acids to maintain protein synthesis in vital tissues and organs in the absence of amino acid absorption from the gut[21] and by providing hepatic gluconeogenic precursors[22]. It follows that if the muscle fibers undergoing hypertrophy due to non-mechanical reasons are not (partly or completely) the same that are also activated during a maximum force task, then the accumulated muscle mass will not lead to an increase in maximum force.

There will also be a disconnect between muscle mass and bone mass/geometry, if the signals leading to muscle hypertrophy are not mechanical in nature. This is nicely exemplified by myostatin null mice, where myostatin deficiency causes an approximate doubling of muscle mass compared with normal mice. In adult myostatin-deficient mice, however, cortical area, bending moment of inertia, and polar moment of inertia of the femora are unaltered relative to normal mice[23]. Given that the body mass of myostatin null mice is identical to the wild-type control mice, their peak femoral strain should be similar to the control, if their activity level and type were the same[24]. This suggests that normal activity behavior (in the cage) against normal body weight is insufficient to activate the surplus of muscle mass in myostatin deficient mice, which in turn may explain why femoral size and shape is unaltered in the "mighty" mouse[24].

Fourth, as noted earlier[25,26], there probably exists a link between growth plate closure and joint size adaptability. If joint size is determined at the end of puberty and hyaline cartilage cannot be enhanced, then the peak joint forces must be controlled in

order not to exceed those that the joints had adapted to at the end of puberty[26]. It follows that in healthy, non-deconditioned adults, maximum force may strongly depend on joint area, and that increases in muscle mass will hardly ever lead to increases in maximum force.

The meaning of peak vs. maximum force – how strong is strong?

Besides tissue size, torque and ground reaction force have been obtained during various contraction modes [concentric (*i.e.* shortening), isometric, and eccentric (*i.e.* lengthening)], and used as proxy markers of intrinsic muscle force. However, one of the most fundamental facts, yet often neglected, is that for a given activation state, maximum muscle force is generated during lengthening contraction[27,28]. Thus, in order to be maximal, the force or torque recording must *a priori* have been obtained during lengthening muscle contraction at maximum muscle activation. Furthermore, force (and torque) output depend on contraction velocity, as shown by the Hill/Katz' force-velocity curve[27,28].

Torque and ground reaction force can be assessed by the use of an isokinetic dynamometer and by different jumping maneuvers on a force plate, respectively. With respect to isokinetic dynamometry, the variability in peak (*i.e.* eccentric) plantar flexion torque output is relatively high, as indicated by the high SD for this isokinetic contraction mode[29], mainly because of methodological difficulties (*e.g.* fixation of body parts, range of motion, variable hip and ankle joint angles). Furthermore, there are no consensus angular velocities at which torque should be assessed, and at peak eccentric angular velocity [about -5.23 rad/s (-300°/s)] subjects produce even lower torque values because complete mechanical recruitment cannot occur in the very short time interval (high velocity and very short distance). Consequently, eccentric plantar flexion torque is difficult to assess using isokinetic dynamometry and consequently is not practical.

It follows, that only submaximal muscle forces are obtained using isokinetic dynamometry. For example, Maganaris *et al.*[30] calculated that the force of soleus muscle during electrically stimulated (and therefore not voluntary) isometric contraction amounts to ~2400 N in young men with a body mass of ~75 kg, corresponding to a force of ~3.2 times body weight. Assuming that the force produced during eccentric (lengthening) contractions can exceed the isometric force up to ~50%[31], a peak force of ~4.8 times body weight would result. In contrast to dynamometry-derived peak force, maximum voluntary ground reaction force (F_{m1LH}) during multiple 1-legged hopping (m1LH) is in the range of 3-3.5 times body weight (approximately 2300-2600 N for a person with 75 kg body mass). Due to the unfavourable lever arm relationship between toes-rotational axis of the ankle joint and rotational axis of the ankle joint-achilles tendon (typically 3:1[32]) plantarflexor muscle force must be approximately 3 times F_{m1LH} to keep the heel from the ground. Thus, during m1LH plantarflexor muscle force amounts to approximately 9-10.5 times body weight (6800-7900 N for a person with 75 kg body mass, Figure 1). This value is 2.8-3.3 times higher than that obtained by Maganaris *et al.*[30] for isometric contractions, and would be still twice the calculated value for eccentric contractions.

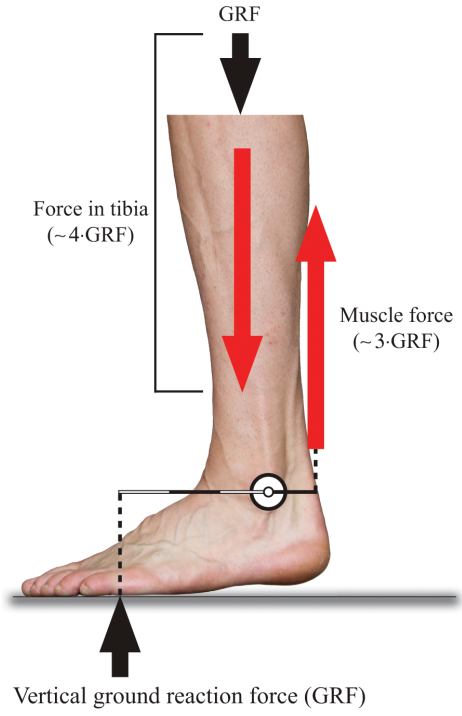

Figure 1. Approximation of the force in the tibia during multiple one-legged hopping (m1LH). Vertical ground reaction force (GRF) during m1LH corresponds to ~3.5 time body weight (~2600 N for a man weighing 75 kg). Due to the unfavorable lever arm relationship between toes-rotational axis of the ankle joint and rotational axis of the ankle joint-achilles tendon (typically 3:1) plantarflexor muscle force must be approximately 3 times GRF. This force (~7900 N for a man weighing 75 kg) also acts on the tibia. During m1LH, the peak force acting on the tibia corresponds to four times GRF (~14 times body weight, or ~10500 N for a man weighing 75 kg).

Moreover, it has been shown in ten healthy men (age range 31-43 years) that typical plantarflexion torque is about 1.5 to 2.0 $Nm/kg_{body\ mass}$, with higher values for an angular velocity of 0.52 rad/s (30°/s) as compared to 3.14 rad/s (180°/s)[33]. These values are considerably lower than those calculated for m1LH. For a healthy male subject weighing 75 kg, peak ground reaction force during m1LH corresponds to about 2600 N, or about 35 N/kg. Considering a lever arm in the ankle joint of about 0.12 m, the resulting plantarflexion torque during m1LH equals 4.2 Nm/kg.

Thus, a useful and reproducible (typical error expressed as coefficient of variation in percent corresponding to ~4.8%) approach to estimate maximum intrinsic muscle forces is to measure ground reaction forces during jumping[25,34]. However, the only two jumping maneuvers where peak voluntary ground reaction force (notably acting on the forefoot) occurs during landing (eccentric contractions) are multiple two-legged hopping (repeated maximal jumps on both forefeet with stiff extended knees

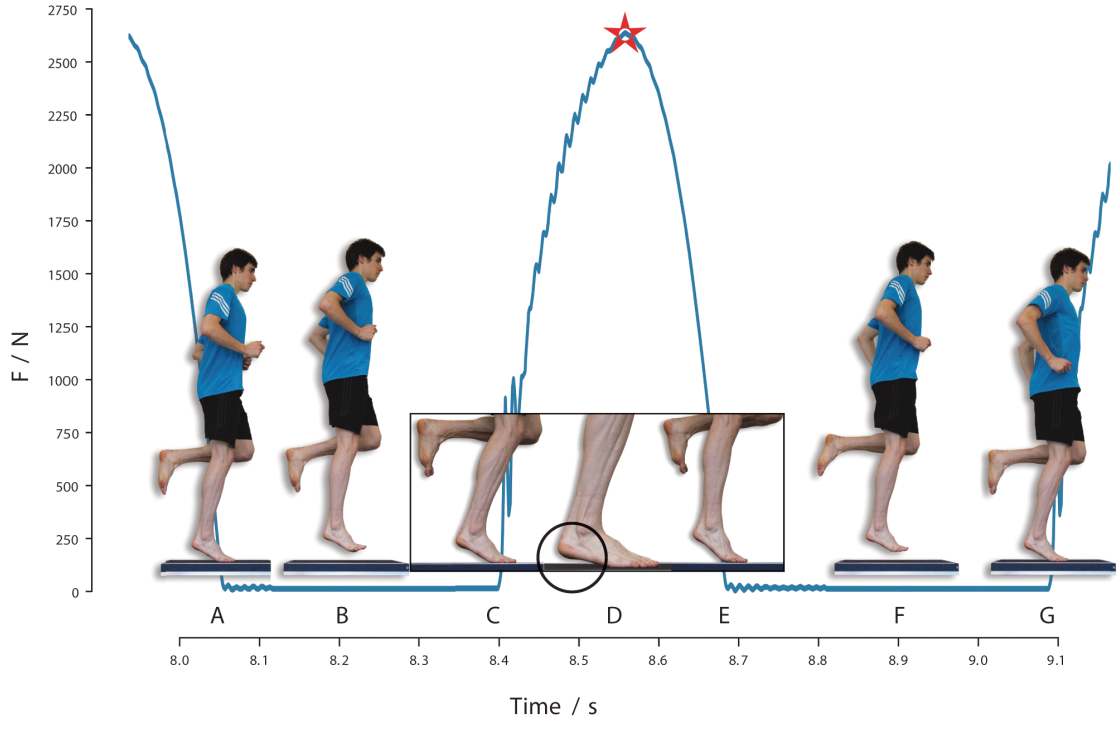

Figure 2. Maximum voluntary ground reaction force during multiple one-legged hopping (F_{m1LH}). The graph shows the phases of the movement corresponding to the indicated points on the force-time curve. A) Take-off; B) Highest point of the hop; C) Landing; D) Lowest point after landing: The magnified portion of the graph (see circle) highlights that the heel does not touch the ground (*i.e.* the force plate) during the landing phase, *i.e.* during lengthening ("eccentric") contraction; E) Take-off; F) Highest point of the hop; G) Landing. The asterisk on the force-time curve indicates F_{m1LH} for this test.

and without heel impact) and m1LH (Figure 2). Because mechanical output per leg is higher in one-legged jumps relative to two-legged jumps[34,35], and muscle activation during unilateral maximal tasks is 27-116% points higher relative to bilateral maximal tasks[36], m1LH should yield an even higher peak voluntary forefoot ground reaction force. In fact, during m1LH, peak voluntary ground reaction force corresponds to about 3-3.5 times body weight[25,34], while peak voluntary forefoot ground reaction force during multiple two-legged hopping is approximately 2.7 times body weight per leg in children[34]. Subsequently, peak voluntary ground reaction force during m1LH represents the largest peak force among all kinds of jumping maneuvers[34], and thus can be considered as maximum voluntary ground reaction force.

Test procedure for multiple one-legged hopping (m1LH)

Multiple one-legged hopping aims to assess maximum voluntary forefoot ground reaction force during landing (F_{m1LH}, Figure 2). Subjects start from an upright standing position with feet positioned hip-wide. To start the jumping maneuver, they lift one foot (usually the one of the dominant leg) off the force plate and start to jump repeatedly (approximately fifteen jumps, comparable to hopping during rope skipping) on the forefoot of the other (in this case nondominant) leg with a stiff knee. During the first few jumps, subjects are instructed to jump as fast as possible, whereas the subsequent ~ten jumps are performed as forcefully as possible. Importantly, subjects are advised never to touch the ground with their heels during the jumping maneuver. Any jumps with heel contact are excluded from the analysis. Heel contact is controlled visually during the jumping maneuver and/or detected automatically by the algorithm of the manufacturer's software. The m1LH is performed with freely moving arms. The best trial is the one in which the highest F_{m1LH} has been achieved. Maximum ground reaction force per body weight ($Frel_{m1LH}$) and F_{m1LH} are the main outcome variables for m1LH.

The test is practicable for both healthy males and females over a wide range of age and body size. In our laboratory, approximately one thousand males and females have been tested so far. All the recruited participants were able to perform m1LH as instructed. In fact, our oldest study participant was a 82-year-old woman weighing 57 kg. We only supported her occasionally by taking her at hand to keep the balance. Our youngest participants were 8 years old. They all performed m1LH unassistedly. None of our study participants ever got injured. However, it is uncertain how well patients and frail people will respond to m1LH, and future studies should aim at answering this question.

Maximum voluntary force during multiple one-legged hopping (F_{m1LH}) as one determinant of the muscle-bone unit

Based on the mechanostat and the muscle-bone hypotheses, we hypothesized that there should exist a very strong relationship between F_{m1LH} and tibial bone mass, and that F_{m1LH} should be a better predictor for bone mass than calf muscle cross-sectional area. To test these hypotheses, we recently investigated the relationship between F_{m1LH} and bone mineral content (*vBMC*) at the 4-, 14-, 38-, and 66%-site of tibia length in 323 healthy 8- to 82-year-old healthy males and females[25]. We found that the correlation was strongest between F_{m1LH} and $vBMC_{14\%}$ (R^2=0.840, P<0.001), and that the correlation between F_{m1LH} and *vBMC* at any site was stronger than the one between F_{m1LH} and muscle cross-sectional area (measured by pQCT at the 66%- site of tibia length). These findings are in concordance with the predictions of the mechanostat and muscle-bone hypotheses, as outlined in the introductory section of this article.

Furthermore, we found that in these 323 healthy individuals of different age, gender, and physical activity level, F_{m1LH} corresponded to 3-3.5 times body weight[25]. Hence, as mentioned earlier, m1LH, as compared to any other known jumping maneuver, elicits the highest peak ground reaction force acting on the forefoot[34], and, consequently, F_{m1LH} corresponds to maximum voluntary ground reaction force. The notion that F_{m1LH} indeed corresponds to maximum voluntary force is supported by unpublished data from our laboratory showing that F_{m1LH} cannot be further increased by loading. We analyzed peak forefoot ground reaction force per body weight during countermovement jumps (CMJ) and m1LH ($F\text{rel}_{CMJ}$ and $F\text{rel}_{m1LH}$, respectively) for three loading conditions corresponding to +5, +15 and +25% body weight in 14 well-trained men (age range: 20-26 years). Loads were applied by means of a weight vest. While $F\text{rel}_{CMJ}$ significantly increased during loaded (+15% and +25% of body weight) jumps as compared to the unloaded condition, $F\text{rel}_{m1LH}$ remained unchanged or even slightly decreased in the loaded conditions. These results lend further credence to the notions that: a) forefoot ground reaction force during m1LH corresponds to maximum voluntary ground reaction force, and thus reflects maximum voluntary muscle force, and b) in the healthy, non-deconditioned adult, maximum force cannot be increased (at least acutely), possibly due to joint size constraints. However, it remains to be determined whether the latter also holds true after long-term exercise interventions.

Having shown in the aforementioned large cross-sectional study that the correlation between F_{m1LH} and $vBMC_{14\%}$ generally is very strong, we were interested to see whether increases in F_{m1LH} and $vBMC_{14\%}$ during growth and exercise occur in proportion to each other. The reason for this is that the mechanostat theory predicts that the increasing muscle force during development provides the stimulus for the increase in bone mass/geometry. Thus, there should be a link between the magnitude of improvements in maximum force (due to jumping exercise and/or simple growth) and the improvements in bone mass/geometry. We thus performed a randomized, controlled 9-month school-based intervention study in children (age range 8 to 12 years)[37]. We were particularly interested: a) to compare the relationship between F_{m1LH} and $vBMC_{14\%}$ pre and post intervention, and b) to evaluate the relationship between the changes in F_{m1LH} (ΔF_{m1LH}) and the changes in $vBMC_{14\%}$ ($\Delta vBMC_{14\%}$). We hypothesized that the increase in F_{m1LH} and tibial structural changes (particularly at 14% of tibial length) from pre to post intervention would be higher for the jumping exercise-based intervention group as compared to the control group (no additional jumping exercise), and that ΔF_{m1LH} and $\Delta vBMC_{14\%}$ would be strongly correlated in both the intervention group and the control group.

Using this novel methodological approach with $vBMC_{14\%}$ as the dependent variable and F_{m1LH} as the predictor, we found that pre and post intervention, the correlation between F_{m1LH} and $vBMC_{14\%}$ was highly significant (P<0.001) both in the intervention group (R^2=0.840 and R^2=0.875 for pre and post, respectively) and the control group (R^2=0.631 and R^2=0.507 for pre and post, respectively). However, we observed no correlation (P>0.05) between ΔF_{m1LH} and $\Delta vBMC_{14\%}$ in either group. Moreover, although the intervention group tended to have greater gains for both F_{m1LH} and tibial bone strength/geometry (+2.1% and +1 to +3%, respectively) relative to the control group, the gains were not statistically different between groups (P>0.05).

It is curious that both variables, *i.e.* maximum force and bone mass, were tightly coupled at baseline, but did not increase in proportion to each other following growth/exercise. This situation resembles the one observed for the relationship between maximal oxygen consumption (as a measure of aerobic capacity) and time trial performance (as a measure of endurance capacity). Here, too, the variables are tightly linked at baseline (pre and post training period), but adaptations do not occur in proportion to each other[38]. These authors suggested that a common factor influences these capacities, but that the same factor does not appear to tightly couple the adaptive process that occurs during exercise[38]. The different coefficients of variations (CV) for F_{m1LH} and $vBMC_{14\%}$ might principally also explain why there was a lack of correlation between ΔF_{m1LH} and $\Delta vBMC_{14\%}$. However, as outlined in Anliker *et al.*[37], it seems unlikely that the lack of correlation between ΔF_{m1LH} and $\Delta vBMC_{14\%}$ was simply due to methodological issues related to the disparate magnitude of the related CVs. In fact, even when performing the regression analysis with only those individuals for whom the percent change in F_{m1LH} and $vBMC_{14\%}$ was higher than the least significant change (LSC=2 times the typical error expressed as a CV[39]), the correlation was still absent.

We further asked, whether the lack of correspondence between the magnitude of adaptation for maximum force and bone mass also was apparent in a system, in which the influence of modulators on the mechanostat would be absent. Hence, we analyzed F_{m1LH} and $vBMC_{14\%}$ for both the supporting and non-supporting lower leg of 66 12- to 18-year-old elite male soccer players. In line with our previous findings, we found a strong relationship between F_{m1LH} and $vBMC_{14\%}$ in both legs (preliminary data from

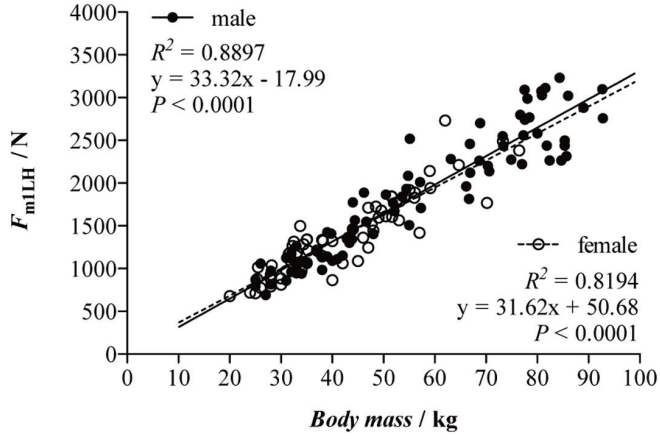

Figure 3. Correlation between maximum voluntary ground reaction force during multiple one-legged hopping (F_{m1LH}) and body mass in 81 males and 65 females (age range: 8 to 20 years).

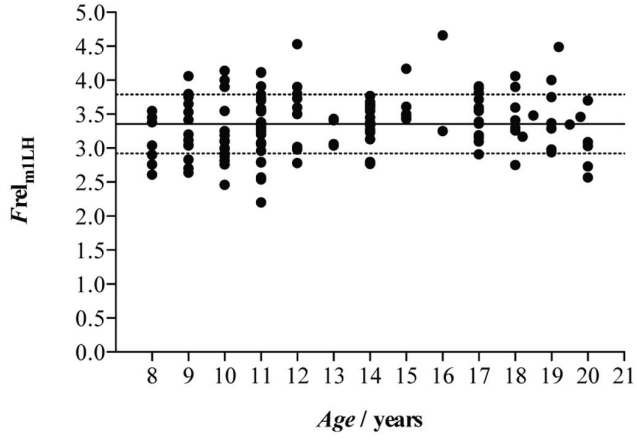

Figure 4. Maximum voluntary ground reaction force during multiple one-legged hopping normalized to body weight ($Frel_{m1LH}$) in 81 males and 65 females (age range: 8 to 20 years). The solid line shows the mean value and the dashed lines show ±1 SD.

our laboratory). However, we also found no relationship between ΔF_{m1LH} and $\Delta vBMC_{14\%}$ for side-to-side differences between the supporting and non-supporting leg in male adolescent soccer players. In this study, two-thirds of all elite male soccer players displayed differences in $vBMC_{14\%}$ greater than the LSC. On the contrary, only six players showed side-to-side differences in F_{m1LH} greater than the threshold for the detection of "real" differences when monitoring single individuals. Therefore, we cannot exclude that the disparate CVs for F_{m1LH} and $vBMC_{14\%}$ might explain the lack of correlation between ΔF_{m1LH} and $\Delta vBMC_{14\%}$ in this case. Clearly, further research and methodological advances are needed to answer the question whether ΔF_{m1LH} and $\Delta vBMC_{14\%}$ develop in proportion to each other.

Altogether, our data indicate that F_{m1LH} and $vBMC_{14\%}$ are tightly linked at all times, meaning that a common factor influences these parameters. However, in children and adolescents, growth and exercise does not increase these factors in proportion to each other, meaning that the adaptive processes are not tightly coupled or follow different time courses. The fact that the correlation between F_{m1LH} and $vBMC_{14\%}$ is very strong and robust over time (*i.e.* cross-sectionally and longitudinally stable), renders it ideal for the clinical assessment of the muscle-bone unit in health and disease. In particular, the system can be used to estimate, whether bone mass is properly adapted to maximum force, and to discriminate between primary and secondary bone disorders, as outlined below.

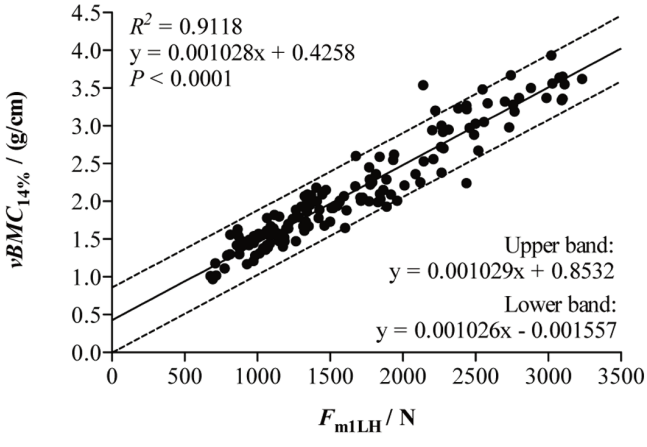

Figure 5. Bone mineral content at the 14%-site of tibia length ($vBMC_{14\%}$) in relation to maximum voluntary ground reaction force during multiple one-legged hopping (F_{m1LH}) in 81 males and 65 females (age range: 8 to 20 years). The dashed lines represent the 95% prediction bands.

Algorithms to quantify the "fitness" of the muscle-bone unit

As shown by our results, there is a strong and robust relationship between F_{m1LH} and $vBMC_{14\%}$ at any time (*i.e.* in a 'static' view). Therefore, F_{m1LH} and $vBMC_{14\%}$ are well suited to quantify the lower leg muscle-bone unit. However, we only measured healthy, "asymptomatic" participants. In our view, pain and limited motor skills, which might be present in various clinical situations, most probably preclude a correct estimation of maximum force by m1LH. It is thus difficult to extrapolate the practical feasibility in healthy subjects to all kinds of clinical situations. Based on a previously published qualitative two-step diagnostic algorithm to evaluate musculoskeletal adaptation in the forearm of children and adolescents[40,41], we pooled and reanalyzed children and adolescents from our previously published data set[25], and constructed new algorithms to quantify the lower leg muscle-bone status. Importantly, instead of using muscle cross-sectional area of the forearm as a proxy marker of muscle force we used F_{m1LH}, and alternatively to radial $vBMC$, we used tibial $vBMC_{14\%}$.

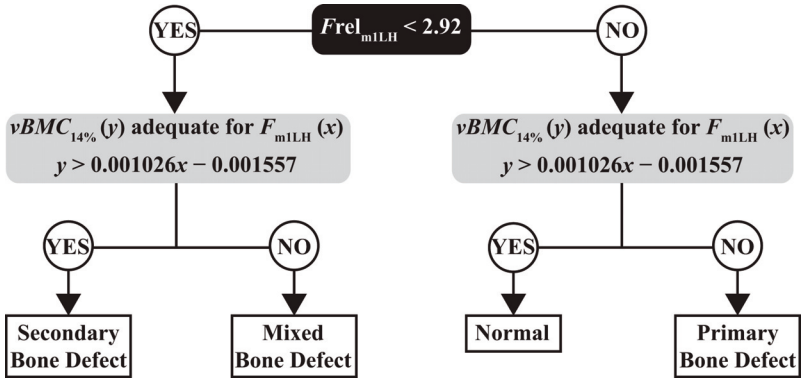

Figure 6. Prospective algorithm for children. $Frel_{m1LH}$, maximum voluntary ground reaction force per body weight; F_{m1LH}, maximum voluntary ground reaction force; $vBMC_{14\%}$, bone mineral content at the 14%-site of tibia length.

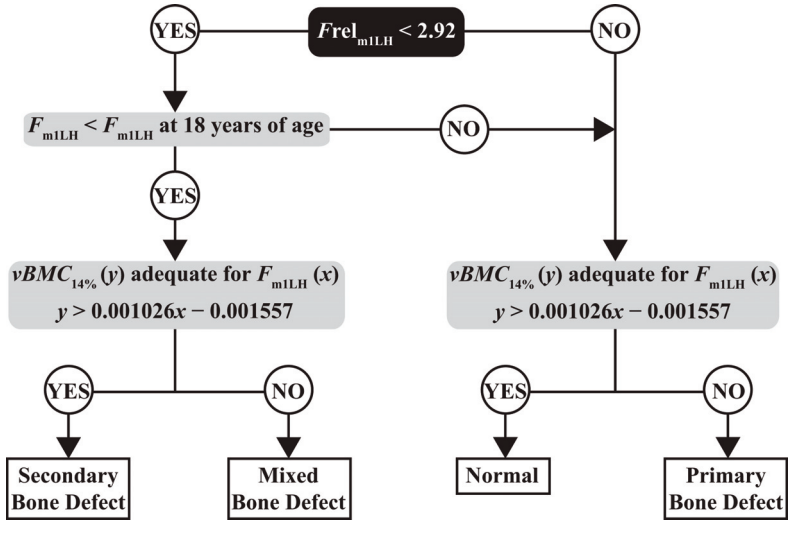

Figure 7. Prospective algorithm for adults. $Frel_{m1LH}$, maximum voluntary ground reaction force per body weight; F_{m1LH}, maximum voluntary ground reaction force; $vBMC_{14\%}$, bone mineral content at the 14%-site of tibia length.

Prospective algorithm for children

Since in 8- to 20-year-old children and adolescents, F_{m1LH} and body mass are strongly correlated (Figure 3), and the parameters of the regression lines are independent of gender, F_{m1LH} must be expressed relative to body weight in order to obtain useful reference data. We found that in children and adolescents F_{m1LH} normalized to body weight ($Frel_{m1LH}$) was constant, *i.e.* did not depend on age. Over all ages, $Frel_{m1LH}$ was 3.35±0.43 (mean±SD, n=146), (Figure 4). Thus, $Frel_{m1LH}$ can be considered to be normal if 2.92 (*i.e.* mean − 1 SD) <$Frel_{m1LH}$<3.78 (*i.e.* mean + 1 SD). One SD is in accordance with the World Health Organization (WHO) definition of osteopenia, for which bone mineral content or bone mineral density values as measured by dual-energy X-ray absorptiometry (DXA) are more than 1 but less than 2.5 SD below the mean for young adults[42], as expressed in terms of the Z-score. As stated by Kanis[42], the reason for this threshold (mean − 1 SD) is to identify people at risk at an early stage. The first step of the algorithm evaluates whether $Frel_{m1LH}$ is sufficiently high, *i.e.* whether it is higher than the lower limit. The second step evaluates whether $vBMC_{14\%}$ is adapted and, thus, adequate for F_{m1LH}. Based on our data, this is the case if for any given F_{m1LH}, $vBMC_{14\%}$ is higher than the lower band of the 95% prediction interval, as determined by linear regression (Figure 5).

As proposed by Schoenau *et al.*[40], the results can be combined into four diagnostic groups subsequent to two situations. In the first situation, $Frel_{m1LH}$ is normal, *i.e.* reaches values above 2.92. If under these circumstance $vBMC_{14\%}$ is adequate for F_{m1LH} (*i.e.* $vBMC_{14\%}$ is above the lower band of the 95% prediction band for any given F_{m1LH}), this corresponds to the "normal" state. If, however, $vBMC_{14\%}$ is lower than expected

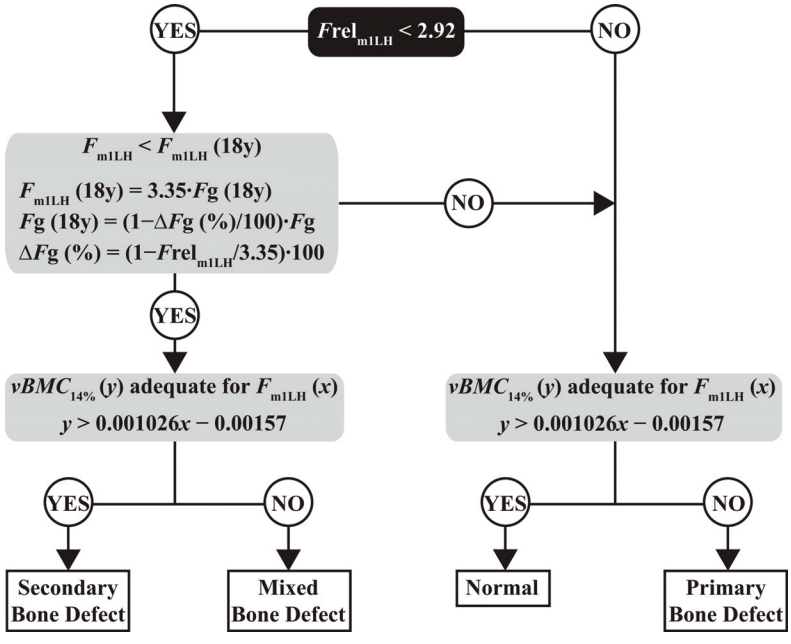

Figure 8. Retrospective algorithm for adults. $Frel_{m1LH}$, maximum voluntary ground reaction force per body weight; F_{m1LH}, maximum voluntary ground reaction force; F_G, force of gravity (body mass multiplied by the gravitational constant); ΔF_G (%), body weight gain/loss in percent; $vBMC_{14\%}$, bone mineral content at the 14%-site of tibia length.

for F_{m1LH}, the status is classified as a "primary bone defect". In the second situation, $Frel_{m1LH}$ is abnormally low, *i.e.* <2.92. If at the same time $vBMC_{14\%}$ is adapted to F_{m1LH}, a "secondary bone defect" exists. If, however, $vBMC_{14\%}$ is not sufficiently high for the given F_{m1LH}, a "mixed bone defect" (primary and secondary) is present (Figure 6).

Prospective/retrospective algorithms for adults

In theory, it is assumed that joint size adapts to maximum voluntary muscle force until the end of puberty, and, after growth plate closure, joint size cannot be further increased. As a consequence, the upper limit of F_{m1LH} is set by the given joint size after puberty, suggesting that F_{m1LH} has to be limited in order to prevent the system from damage[25,26]. It follows that in adults, a decrease in $Frel_{m1LH}$ can be due to an increase in body weight (*e.g.* through the gain of fat and/or muscle mass) and/or a decrease in F_{m1LH} (*e.g.* through deconditioning). For example, if an overweight person (*i.e.* a person who gained body weight in excess of that at 18 years of age) has a low $Frel_{m1LH}$ (but normal $vBMC_{14\%}$ relative to F_{m1LH}), the result could be misinterpreted as secondary bone defect, and subsequently, wrong recommendations could be issued. In fact, in such a case (contrary to the case of a true secondary bone defect), training to increase muscle force would not be expected to restore $Frel_{m1LH}$.

Therefore, the second step in the algorithm for adults is to determine whether F_{m1LH} decreased compared to F_{m1LH} at 18 years of age [F_{m1LH} (18y)]. In the ideal case of prospective assessments, F_{m1LH} and $vBMC_{14\%}$ are routinely measured at given time intervals during the entire life span. In this case, the individual value of F_{m1LH} (18y) is known, and it can be determined whether F_{m1LH} is reduced as compared to F_{m1LH} (18y) (Figure 7). The algorithm is then followed accordingly (Figure 7). If the effective value for F_{m1LH} (18y) is not known (essentially because it was not measured at the time), we suggest to estimate it as follows. First, the percent gain in body weight from the age of 18 years (ΔF_G) is calculated by using the measured $Frel_{m1LH}$ and the mean reference value of $Frel_{m1LH}$ (3.35, Figure 8). For all calculations, we assume that at the age of 18 years, the person was "normal" with respect to the parameters of the muscle-bone unit. Based on the calculated ΔF_G, body weight at 18 years of age [F_G (18y)] can be estimated (Figure 8). Finally, F_{m1LH} (18y) can be estimated by multiplying the mean reference value of $Frel_{m1LH}$ by F_G (18y) (Figure 8).

Conclusions

Multiple one-legged hopping in conjunction with pQCT represents a new functional system to assess the musculoskeletal status in children and adults. F_{m1LH} occurs during the landing phase of m1LH, where plantarflexor muscles are contracting eccentrically, and thus corresponds to maximum voluntary ground reaction force. Consequently, it reasonably estimates maximum voluntary plantarflexor muscle force. Moreover, multiple one-legged hopping is practicable and safe for both healthy males and females over a wide range of age and body size. In view of the motor demand and the high mechanical stress acting on the lower leg, future studies should evaluate the performance and suitability of m1LH in patients and frail people. Since the correlation between F_{m1LH} and

$vBMC_{14\%}$ is very strong and robust over time (*i.e.* cross-sectionally and longitudinally stable), these two parameters are well suited to be incorporated into the proposed two-step algorithm to quantitatively estimate whether bone mass is properly adapted to maximum force.

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References

1. Frost HM. The mechanostat: a proposed pathogenic mechanism of osteoporoses and the bone mass effects of mechanical and nonmechanical agents. Bone Miner 1987;2:73-85.
2. Frost HM. Skeletal structural adaptations to mechanical usage (SATMU): 2. Redefining Wolff's law: the remodeling problem. Anat Rec 1990;226:414-22.
3. Frost HM. Skeletal structural adaptations to mechanical usage (SATMU): 1. Redefining Wolff's law: the bone modeling problem. Anat Rec 1990;226:403-13.
4. Thompson D. On Growth and Form (abridged edition). Cambridge: Cambridge University Press; 1961 (original edition published in 1917).
5. Zanchetta JR, Plotkin H, Alvarez-Figueira, ML. Bone mass in children: Normative values for the 2–20-year-old population. Bone 1995;16(Suppl.):393-99.
6. Schiessl H, Frost HM, Jee WS. Estrogen and bone-muscle strength and mass relationships. Bone 1998;22:1-6.
7. Schonau E, Werhahn E, Schiedermaier U, Mokow E, Schiessl H, Scheidhauer K, *et al*. Influence of muscle strength on bone strength during childhood and adolescence. Horm Res 1996;45(Suppl.1):63-6.
8. Winter EM. "Critical power": time to abandon. Med Sci Sports Exerc 2011;43:552; author reply 3.
9. Ducher G, Courteix D, Meme S, Magni C, Viala JF, Benhamou CL. Bone geometry in response to long-term tennis playing and its relationship with muscle volume: a quantitative magnetic resonance imaging study in tennis players. Bone 2005;37:457-66.
10. Heinonen A, Sievanen H, Kannus P, Oja P, Pasanen M, Vuori I. High-impact exercise and bones of growing girls: a 9-month controlled trial. Osteoporos Int 2000; 11:1010-7.
11. Calbet JA, Diaz Herrera P, Rodriguez LP. High bone mineral density in male elite professional volleyball players. Osteoporos Int 1999;10:468-74.
12. Pettersson U, Nordstrom P, Lorentzon R. A comparison of bone mineral density and muscle strength in young male adults with different exercise level. Calcif Tissue Int 1999;64:490-8.
13. Binkley TL, Specker BL. Muscle-bone relationships in the lower leg of healthy pre-pubertal females and males. J Musculoskelet Neuronal Interact 2008;8:239-43.
14. Rantalainen T, Heinonen A, Komi PV, Linnamo V. Neuromuscular performance and bone structural characteristics in young healthy men and women. Eur J Appl Physiol 2008;102:215-22.
15. Duchateau J, Semmler JG, Enoka RM. Training adaptations in the behavior of human motor units. J Appl Physiol 2006;101:1766-75.
16. De Luca CJ, Hostage EC. Relationship between firing rate and recruitment threshold of motoneurons in voluntary isometric contractions. J Neurophysiol 2010;104:1034-46.
17. Schiaffino S, Reggiani C. Fiber types in mammalian skeletal muscles. Physiol Rev 2011;91:1447-531.
18. Linari M, Bottinelli R, Pellegrino MA, Reconditi M, Reggiani C, Lombardi V. The mechanism of the force response to stretch in human skinned muscle fibres with different myosin isoforms. J Physiol 2004;554:335-52.
19. Hornberger TA. Mechanotransduction and the regulation of mTORC1 signaling in skeletal muscle. Int J Biochem Cell Biol 2011;43:1267-76.
20. Berdeux R, Stewart R. cAMP signaling in skeletal muscle adaptation: hypertrophy, metabolism and regression. Am J Physiol Endocrinol Metab 2012; [Epub ahead of print].
21. Biolo G, Zhang XJ, Wolfe RR. Role of membrane transport in interorgan amino acid flow between muscle and small intestine. Metabolism 1995;44:719-24.
22. Felig P. The glucose-alanine cycle. Metabolism 1973; 22:179-207.
23. Hamrick MW, McPherron AC, Lovejoy CO, Hudson J. Femoral morphology and cross-sectional geometry of adult myostatin-deficient mice. Bone 2000;27:343-9.
24. Turner CH. Muscle-bone interactions, revisited. Bone 2000;27:339-40.
25. Anliker E, Rawer R, Boutellier U, Toigo M. Maximum ground reaction force in relation to tibial bone mass in children and adults. Med Sci Sports Exerc 2011;43:2102-9.
26. Rittweger J. Ten years muscle-bone hypothesis: what have we learned so far? - almost a festschrift. J Musculoskelet Neuronal Interact 2008;8:174-8.
27. Hill AV. The maximum work and mechanical efficiency of human muscles, and their most economical speed. J Physiol 1922;56:19-41.
28. Katz B. The relation between force and speed in muscular contraction. J Physiol 1939;96:45-64.
29. Olmo J, Jato S, Benito J, Martin I, Dvir Z. Identification of feigned ankle plantar and dorsiflexors weakness in normal subjects. J Electromyogr Kinesiol 2009;19:774-81.
30. Maganaris CN, Baltzopoulos V, Ball D, Sargeant AJ. *In vivo* specific tension of human skeletal muscle. J Appl Physiol 2001;90:865-72.
31. Gulch RW. Force-velocity relations in human skeletal muscle. Int J Sports Med 1994;15(Suppl.1):S2-10.
32. Runge M, Schiessl H, Rittweger J. Klinische Diagnostik des Regelkreises Muskel-Knochen am Unterschenkel. Osteologie 2002;11:25-37.
33. Moller M, Lind K, Styf J, Karlsson J. The reliability of isokinetic testing of the ankle joint and a heel-raise test

for endurance. Knee Surg Sports Traumatol Arthrosc 2005;13:60-71.
34. Veilleux LN, Rauch F. Reproducibility of jumping mechanography in healthy children and adults. J Musculoskelet Neuronal Interact 2010;10:256-66.
35. Bobbert MF, de Graaf WW, Jonk JN, Casius LJ. Explanation of the bilateral deficit in human vertical squat jumping. J Appl Physiol 2006;100:493-9.
36. Vandervoort AA, Sale DG, Moroz J. Comparison of motor unit activation during unilateral and bilateral leg extension. J Appl Physiol 1984;56:46-51.
37. Anliker E, Dick C, Rawer R, Toigo M. Effects of jumping exercise on maximum ground reaction force and bone in 8- to 12-year-old boys and girls: a 9-month randomized controlled trial. J Musculoskelet Neuronal Interact 2012; in press.
38. Vollaard NB, Constantin-Teodosiu D, Fredriksson K, Rooyackers O, Jansson E, Greenhaff PL, *et al*. Systematic analysis of adaptations in aerobic capacity and submaximal energy metabolism provides a unique insight into determinants of human aerobic performance. J Appl Physiol 2009;106:1479-86.
39. Hopkins WG. Measures of reliability in sports medicine and science. Sports Med 2000;30:1-15.
40. Schoenau E, Neu CM, Beck B, Manz F, Rauch F. Bone mineral content per muscle cross-sectional area as an index of the functional muscle-bone unit. J Bone Miner Res 2002;17:1095-101.
41. Schoenau E, Fricke O. Mechanical influences on bone development in children. Eur J Endocrin 2008;159(Suppl.1): S27-31.
42. Kanis JA, Melton LJ 3rd, Christiansen C, Johnston CC, Khaltaev N. The diagnosis of osteoporosis. J Bone Miner Res 1994;9:1137-41.