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BONE MARROW AND ADIPOSE TISSUE (G DUQUE AND B LECKA-CZERNIK, SECTION EDITORS)

Bone Marrow Adipose Tissue Quantification by Imaging

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Abstract

Purpose of Review The significance and roles of marrow adipose tissue (MAT) are increasingly known, and it is no more considered a passive fat storage but a tissue with significant paracrine and endocrine activities that can cause lipotoxicity and inflammation.

Recent Findings Changes in the MAT volume and fatty acid composition appear to drive bone and hematopoietic marrow deterioration, and studying it may open new horizons to predict bone fragility and anemia development. MAT has the potential to negatively impact bone volume and strength through several mechanisms that are partially described by inflammaging and lipotoxicity terminology.

Summary Evidence indicates paramount importance of MAT in age-associated decline of bone and red marrow structure and function. Currently, MAT measurement is being tested and validated by several techniques. However, purpose-specific adaptation of existing imaging technologies and, more importantly, development of new modalities to quantitatively measure MAT are yet to be done.

Keywords Yellow marrow · Osteosarcopenia · Red marrow · Hematopoietic marrow · Lipotoxicity · CT · MRI

Introduction

Our understanding of the adipose tissue is expanding, and fat is no longer considered a simple energy reserve or a space-filler for atrophied tissues. Adipose tissue is an active endocrine and paracrine organ with universal distribution and typically high volumes in the body (usually 30–45% of whole-body mass in older adults). It displays a variety of biological roles and processes from general metabolism to inflammation and aging.

With aging, and development of osteopenia/osteoporosis, marrow adipose tissue (MAT) increasingly expands. In severe osteoporotic cases, MAT comprises over half of the tissue volume in the femoral neck, and bone volume fraction is only around 30%, with MAT replacing almost all trabecular bone (unpublished data).

Therefore, quantifying MAT can be a key factor in understanding bone biology and possibly a biomarker of early changes in the skeletal tissue and a diagnostic tool to predict bone fragility. This review aims to provide basic knowledge on the importance of fat in the decline of musculoskeletal system, common imaging techniques of quantifying MAT, their advantages, and limitations.

This article is part of the Topical Collection on *Bone Marrow and Adipose Tissue*

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Why Fat Storages, and Particularly MAT, Are Increasingly Important

Bone marrow normally contains some fat at birth; however, marrow fat infiltration increases with age and is highly and negatively associated with bone loss and hematopoietic marrow (HPM) atrophy [1••], and hence, it is considered an ectopic fat depot. Age-associated fat relocation from peripheral (subcutaneous) sources into ectopic depots (defined as triglyceride storages in tissues other than primary adipose tissue that normally contain only small amounts of fat) has been documented for decades. However, their impact on the target organs and particularly bone marrow (e.g., effects on osteoblast function) has recently gained the interest of health researchers [2, 3, 4•, 5•, 6, 7].

This increased volume of ectopic fat can potentially rise the local concentration of paracrine factors such as inflammation mediators, adipokines, and fatty acids [1••, 8–12]. However, in addition to the increased fat volume, the secretory profile of MAT changes significantly compared to the subcutaneous fat within the same subject, and equally importantly, compared to younger subjects [6, 9]. An increased volume and a more inflammatory and lipotoxic profile of ectopic fat in general and MAT, in particular, can have significant consequences for the target (host) tissues. High volumes and vast areas of contact between infiltrated fat versus muscle and bone (Fig. 1) make musculoskeletal tissues particularly vulnerable to the lipotoxic effects of age-associated infiltration of fat.

Importantly, not only MAT expansion is associated with bone and HPM atrophy in both humans [1••] and progeria mice [13••], but also blunting MAT's lipotoxic and inflammatory properties by lifelong provision ω3 fatty acids in mice can prevent age-related bone and HBM loss [13••]. High dietary inflammatory index [14] is associated with development of sarcopenia [15], osteoporosis, and risk of fractures [16, 17], and theoretical evidence of associations with osteosarcopenia, particularly in obese people, is available [18]. Considering the close associations of the index with risk factors for obesity and ectopic fat infiltration and lipotoxicity, it is likely that an inflammatory diet (high in carbohydrates, meat, and trans fatty acids and low in fibers and vegetables [19]) can predispose subjects to a variety of fat-related conditions including osteosarcopenia. In fact, mid-thigh fat mass, independent of muscle mass, is associated with increased risk of falls in older subjects [20].

The devastating effects of fat infiltration into the musculoskeletal system are only a part of a systematic effect that ectopic fat has on all tissues. Ectopic fat-induced inflammation and lipotoxicity associated with high concentrations of inflammatory mediators and fatty acids (of which minimal amounts are secreted into the circulation [1••, 12, 21]) have been linked to malignancies in general [22, 23], leukemia [24], postmenopausal breast cancer [25], colorectal adenoma [26] risk, and a decline in lymphopoiesis (due to marrow and lymphoid organs adiposity) [27, 28]. Possibly, through similar mechanisms, the atrophy of red marrow with age-associated MAT

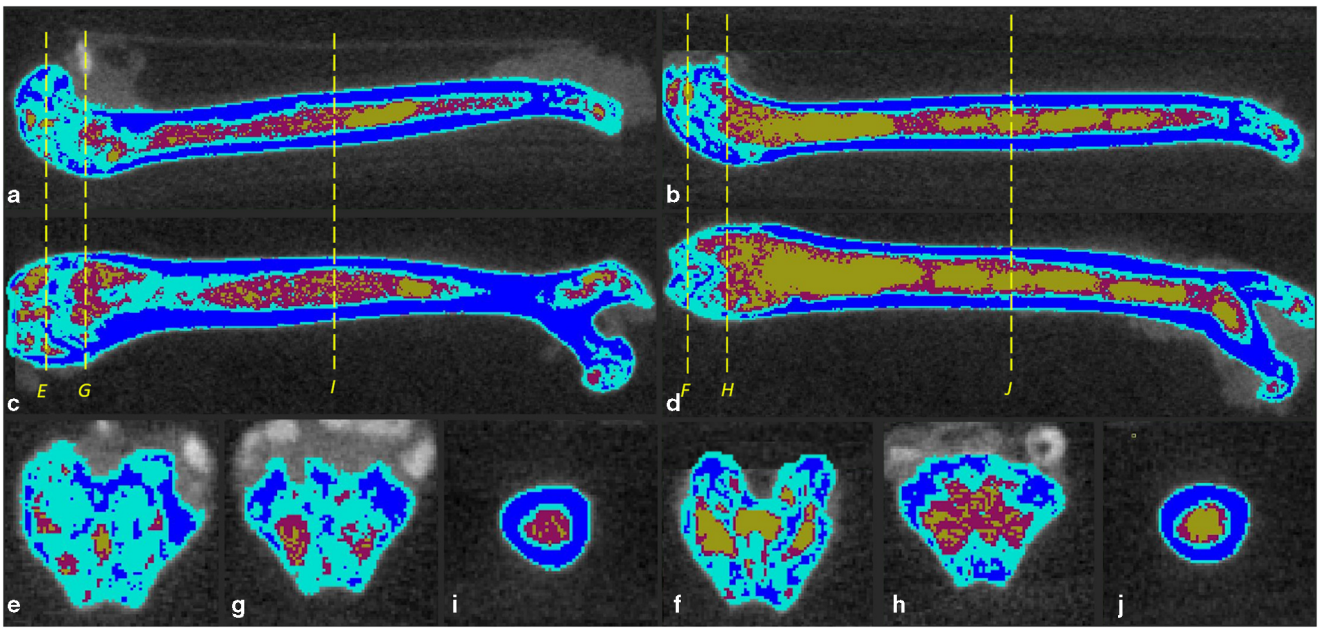

Fig. 1 A comparison between age-matched normal (left panels) vs osteoporotic (right panels) mice. Sagittal (**a**, **b**), coronal/frontal (**c**, **d**), and cross-sectional planes at epiphyseal (**e**, **f**), metaphyseal (**g**, **h**), and diaphyseal (**i**, **j**) regions all show higher marrow adipose tissue (MAT) and lower bone and hematopoietic marrow in osteoporotic group. Dark blue: dense bone, light blue: low-density bone, purple: hematopoietic marrow, and khaki: MAT

expansion in humans [1••] is potentially capable of inducing both immunodeficiency and anaplasia. In agreement with the above, possible molecular mechanisms by which ectopic fat infiltration causes inflammation and anaplasia have been suggested [29, 30].

Infiltration of fat into viscera, especially in women [10, 31, 32] and particularly into the pancreas that leads to beta cell lipotoxicity [8], has been associated with insulin resistance. However, older men appear to be an exception, and in fact, fat distribution seems to follow a different pattern in this age-sex group. Gynoid fat in older men appears to be protective against fractures [33], and unlike women [34–38], MAT volume does not follow the volume of visceral (VAT) and subcutaneous (SAT) adipose tissues, which is possibly due to significant fat relocation in older men [1••]. However, factors that affect fat relocation into the ectopic depots are not known yet, and their discovery may unravel the pathogenesis of many age-associated conditions including osteoporosis, sarcopenia, and insulin resistance.

Current Imaging-Based Methods to Quantify MAT

To diagnose, prognose, and advance our understanding of fat infiltration in the disease process, the development and validation of high-precision, sensitive, and specific imaging biomarkers are mandated. Like any other tissue, in situ imaging of MAT can be performed using two kinds of electromagnetic waves:

(a) High-energy shortwaves, i.e., X-rays that are absorbed at different rates in different tissues, depending on how electron-dense the tissues are with the resultant image reconstructed based on a virtual X-ray shade of the tissues. However, due to the high energy of the photons, this type of imaging has the potential for breaking DNA atomic bonds with the risk of mutations correlated with the energy and dose (exposure volume and time).
(b) Low-energy longer waves that are used in nuclear magnetic resonance (NMR) imaging (MRI) that detect contrasts in proton (basically water) density in different tissues. Unlike X-ray-based systems, it is not tissue absorption that provides the contrast between tissues, but the waves that the tissues emit after absorbing a particular wavelength. Magnetic resonance-based images provide best contrast between fat-based and water-based tissues. In the following, we discuss both X-ray and MRI techniques used in the quantification of tissues and MAT.

Computed Tomography Imaging

Computed tomography (CT) is a non-invasive imaging method that is based on the three-dimensional projection of X-rays. X-rays are electromagnetic waves, with wavelengths ranging from 0.01 to 10 nm. CT machines usually exploit a part of this range to calculate tissue attenuation coefficients, which after normalization to the attenuation of air and water, is called CT number and is expressed as Hounsfield units (HU) [39]. The degree of X-ray absorption depends on the electron densities (and atomic number [Z]) of a material and the inverse cube of the X-ray energy ($1/E^3$). X-ray absorption is highest in metals, e.g., calcium in bones, and negligible in the air. All other tissues, depending on their material density and metal concentrations (usually calcium [in bones and muscles] and iron [e.g., in blood and muscle]) take a value between the two extremes. Air has been arbitrarily given a HU value of −1000, and HU of water is zero. Adipose tissue, skeletal muscle, and bone (in ascending order) take Hounsfield unit ranges that permit their quantification in CT images (with some overlap between the tissues, especially in low-volume X-ray CT).

Conventional CT scans deliver a significant amount of radiation and scanning of live tissues is still considered risky due to the chances of mutagenesis. In the case of high-resolution CTs (micro- or nano-CTs) and synchrotron-based imaging that require much higher doses of X-ray, scanning of living beings and especially humans has not become a practice yet. However, advances in technology, e.g., automatic tube current modulation, have resulted in significantly smaller doses [40, 41]. With the development of high-resolution peripheral quantitative CT (HR-pQCT), current commercially available machines have an apparent maximum resolution of ~ 41–200 μm, and allow human distal limbs to be scanned with radiation doses under 5 μSv per scan.

Significant improvements in image analysis are still needed for the quantification of ectopic fat, particularly in muscle and bone. Our team has validated the use of a CT image analysis software to quantify MAT in small animal models (Fig. 1) [42•], while various techniques have been established to assess MAT in humans [1••, 43, 44]. We have also successfully measured SAT, VAT, MAT, and intramuscular adipose tissue (IMAT) in human subjects and validated the measurements visually (Fig. 2). Low resolution (and consequent partial voluming), beam hardening artefact, and high radiation doses remain as the main limitations of CT-based quantification of ectopic fat, particularly within the bones. Beam hardening leads to MAT not being "seen" by the X-rays hardened by surrounding radio-dense bone. The low resolution of current instruments (around 1 mm) leads to averaging of the Hounsfield values of thin trabeculae with smaller foci of fat or red marrow, particularly at the interfaces of these tissues. These limitations can lead to partial under- or over-estimation of bone and marrow constituents.

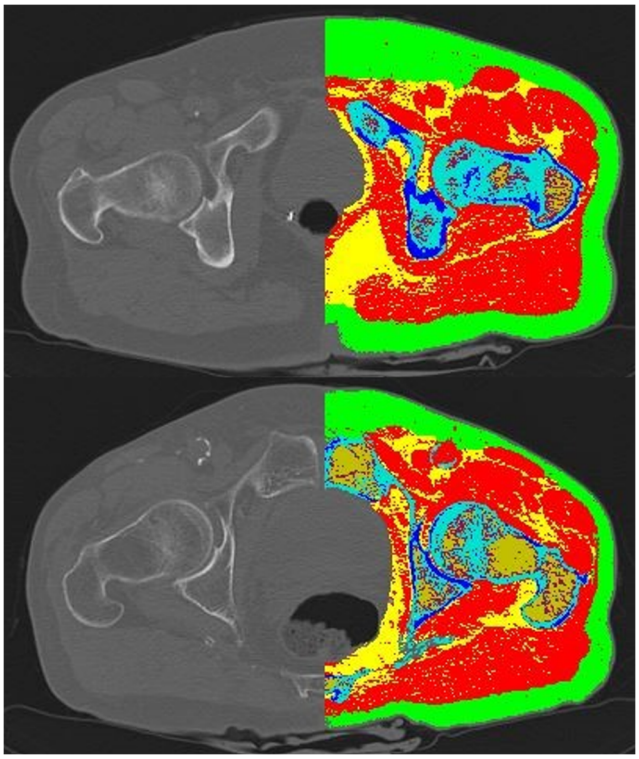

Fig. 2 Pelvic CT images segmented on the left side of the body in a normal (top) vs an osteoporotic subject (bottom). Color codes: green: subcutaneous fat, red: muscle, yellow: peri- and intramuscular fat, dark blue: dense bone, light blue: low-density bone, purple: hematopoietic marrow, and khaki: marrow adipose tissue (MAT). Note the significantly higher volumes of MAT and lower hematopoietic marrow in the osteoporotic subject. Comparing the segmented side to the non-segmented grey-scale image highlights the value of good image processing technique in quantifying features of tissues that are not easily detectable by naked eye

Dual-energy CT and micro-CT systems are being developed but are not used extensively. The technique has been validated using MR spectroscopy for quantification of MAT and bone volumes [45], or quantification of fat infiltration into liver [46]. Nevertheless, determining accurate thresholds to differentiate HPM from MAT and histologic validation of the technique, particularly on human specimens, is still to be carried out.

Irrespective of the technique used, detection of fat infiltration into muscle and bone may be of predictive importance in diagnosing and determining the rate of progression of osteoporosis, sarcopenia, and osteosarcopenia. However, limitations of these methods include cost, sensitivity to patient motion, and lack of reference values and standards. In addition, their ability in quantifying marrow fat and particularly in contrast to HPM, and especially in humans, has not been validated yet.

Regarding labelling of MAT for imaging analyses, osmium is a heavy metal, and the radiopaque compound osmium tetroxide has high tissue penetration potential, which makes it ideal for staining lipid in bone [47] or close to bone [48]. One of the most important characteristics of osmium tetroxide is permeability, which produces uniformly stained block samples even at millimeter-scale thickness of samples [49]. Recently, osmium tetroxide (that has very high affinity to fatty tissue) has been shown to increase the contrast between MAT and MPM in micro-CT imaging and has been used in numerous MAT studies [50–59] as well as the study of other soft tissues and embryos [60–65]. This fixative/stain has been assessed as rapid, reproducible, and quantifiable [66–68]. Osmium-based micro-CT in comparison to other techniques such as micro-MRI is cheaper to perform scans with comparable contrast and spatial resolution [69]. It is claimed that this technique made it possible to distinguish between two MAT subpopulations known as constitutive MAT (cMAT) and regulated MAT (rMAT) [70, 71], but the nature, relevance, and significance of these subtypes remain to be proven. The highly toxic nature of the material is a major limitation for routine use for staining MAT and IMAT, and it can only be used for staining isolated specimens.

Magnetic Resonance Imaging

Magnetic resonance imaging (MRI) is based on the absorption and emission of energy in the radiofrequency (RF) range (42.58 MHz/T for proton) of the electromagnetic spectrum [72], which has insufficient energy to break chemical bonds or remove electrons. Therefore, MRI has been proven as a non-ionizing and safe imaging method [73]. MR-computed or reconstructed images are based on spatial variations in the phase and frequency of the RF energy being absorbed and emitted by the scanned object. A few biological elements, such as oxygen, fluorine, sodium, and phosphorus, have the potential to be used as elements for producing RF signals. However, since the human body is mainly comprised of fat and water (both having higher, but significantly different quantities of hydrogen atoms), MR images are typically acquired based on the proportion of hydrogen atoms within a tissue. Therefore, MR imaging is of particular interest in differentiating fat-based tissues from those with water as the main constituent.

MR imaging has become the preferred imaging method in evaluating diseases of the bone marrow or associated with bone marrow [74]. MRI-based bone marrow fat quantification techniques are non-invasive and facilitate imaging of large volumes of interest and different anatomical regions. Hence, it has been proposed as a reliable method to investigate the association between bone loss and bone marrow adiposity [75–77].

The appearance of the bone marrow images depends on the existence and proportions of trabecular bone, fat, and water within the scanned region of interest [78]. Each of these

elements of the bone marrow produces a unique radiofrequency signal, resulting in different signal amplitudes that are visualized as a grayscale level on the MR images. The level of brightness of each voxel represents the magnitude of the signal at that location.

Assessment of fat fraction in bone marrow can be performed using several techniques including routine sequences such as T1- and T2-weighted imaging, as well as dedicated sequences such as short T1 inversion recovery (STIR) and chemical shift imaging (e.g., the Dixon method) and magnetic resonance spectroscopy (MRS) [79•].

HPM is more cellular, containing the hematopoietic and blood cells and contains approximately 40% fat, whereas yellow marrow contains 80% fat [3], and hence the two tissues contain significantly different proton densities and arrangements. This results in different T1 times for HPM and yellow marrow which is exploited in T1-weighted imaging to generate contrast between the two tissue types [80]. In addition to qualitative assessment of MAT, limited quantitative assessment can also be performed using routine T1 imaging [81]. Compared to T2-weighted imaging, T1-weighted imaging provides improved visualization of bone marrow (Fig. 3). Nevertheless, with proper adjustment and acquisition, T2 images are also useful in assessing MAT (Fig. 4) although thresholding-based quantification of fat in such images remains a challenge, particularly at lower resolutions.

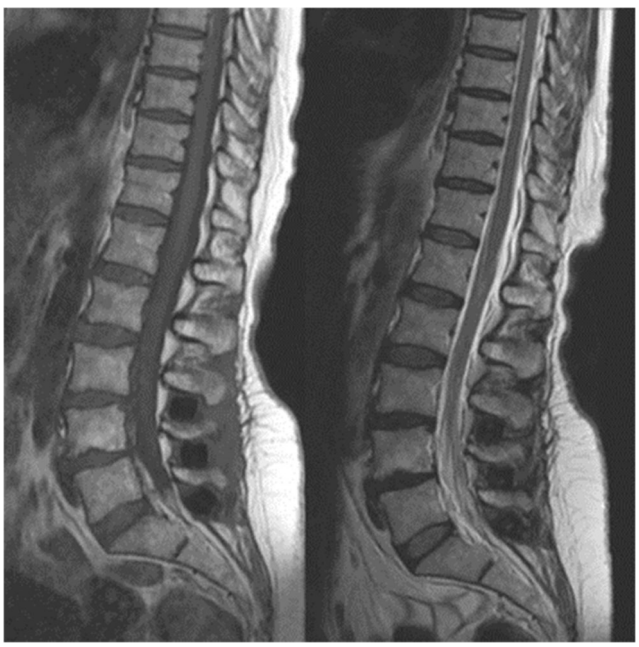

Fig. 3 T1- (left) and T2 (right)-weighted images of thoraco-lumbo-sacral vertebrae in the same subject. Better contrast of marrow fat (brighter voxels) is one of the desired properties of T1-weighted sequences that makes qualitative and quantitative assessment easier

STIR imaging is a fat suppression sequence whereby imaging parameters are chosen to null the signal generated by fat with preservation of the signal generated by water. This sequence provides more homogenous fat suppression than T2-weighted imaging and is sensitive to bone marrow focal abnormalities [82].

Magnetic Resonance Spectroscopy

The resonant frequency of protons is affected by the structure of the molecule. When a proton, as part of a molecule, is slightly shielded by surrounding atoms or components from the applied magnetic field, the resonance frequency will be different. In a magnetic field, water protons resonate slightly faster than protons in triglyceride (fat) molecules (Fig. 5). This difference in resonance frequencies between water and fat is known as "chemical shift" and increases with increasing field strength. The chemical shift between water and fat is 3.5 parts per million (ppm) which is equal to approximately 220 and 440 Hz in MRI scanners of 1.5 and 3.0 Tesla, respectively.

Magnetic resonance spectroscopy (MRS) is an established technique used to measure fat content in localized regions of the vertebral bodies and long bones [76, 77, 83–85]. However, the distribution of bone marrow fat content can be spatially heterogeneous and there is a growing interest in applying chemical shift-based water/fat imaging techniques for measuring bone marrow fat content with high spatial resolution [83].

Bone Marrow MRI-Based Quantification and Bone Structure Assessment

The quantity, and most importantly composition, of MAT may characterize the conditions of skeletal health [86]. Yeung et al reported 29% MAT content in young control women, and 56, 63, and 65% in normal, osteopenic, and osteoporotic fracture free older women, respectively [87]. Interestingly, the highest and lowest proportion of unsaturated lipids in marrow fat were found in the younger controls and older women with osteoporotic spine according to their areal bone mineral density (aBMD). The unsaturation level and marrow fat content were negatively correlated ($r = -0.53$, $p < 0.0001$). In addition, Patsch et al. reported an association between a history of fracture and lower unsaturation levels, even after adjustment for spine volumetric BMD (vBMD; −1.7%; 95% CI −2.8 to −0.5%) [88]. Taking the existing MR- and non-MR-based imaging knowledge into account, it has been established that MAT expansion is associated with bone and red marrow atrophy. However, MRS provides further insight that

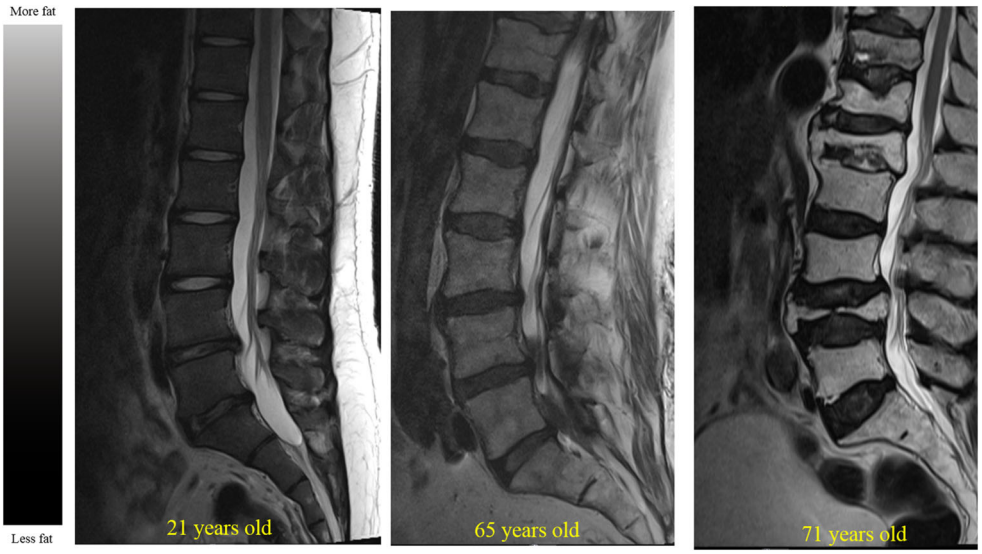

Fig. 4 Comparison of marrow adipose tissue (MAT) signals in a young (left), healthy older (middle) and an older subject with multiple vertebral compression fractures (right). Due to large voxel size, partial voluming, and inherent artefacts of conventional MRI images, threshold-based segmentation remains a challenge to quantify MAT infiltration, particularly on T2-weighted sequences

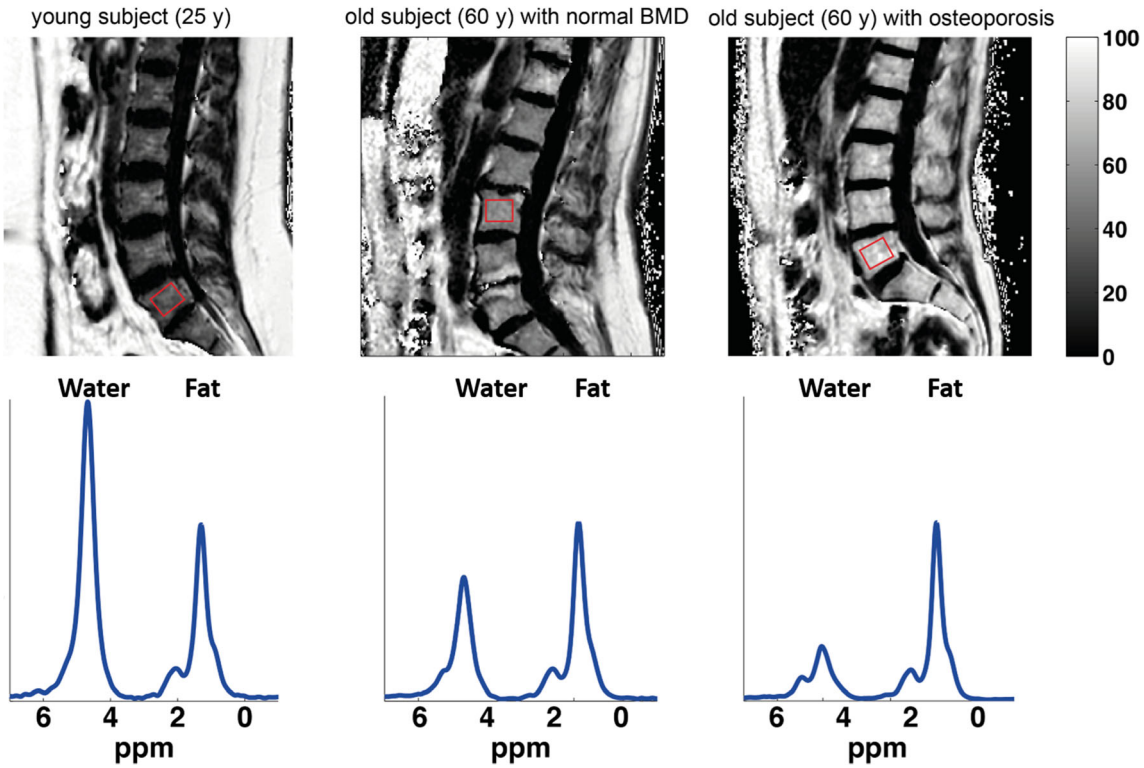

Fig. 5 *Upper panel:* Three sagittal MRI images of spines with increasing marrow fat from left to right in young, and older subjects (without and with osteoporosis), respectively. The red cubes are the regions of interest where MR spectroscopy has been carried out. *Lower panel:* Spectra indicating the proportional amounts of water- and fat-based tissues. The increasing marrow adipose tissue (MAT) with aging and bone volume decline is clearly evident. [Image modified from: Cordes et al. MR-Based Assessment of Bone Marrow Fat in Osteoporosis, Diabetes, and Obesity, Front. Endocrinol., 27 June 2016. https://doi.org/10.3389/fendo.2016.00074 (as per publisher's terms and conditions: https://www.frontiersin.org/legal/terms-and-conditions)]

in osteoporotic vs non-osteoporotic subjects other than MAT volume, its saturation also increases [87].

Future of MAT Imaging

MR Spectroscopy

It has been reported that inhibition of sclerostin by anti-sclerostin antibody (Scl-Ab) increases bone formation, and decreases bone resorption, leading to improved bone structure, bone mass, and bone strength while maintaining bone quality in multiple animal models of osteoporosis [89]. In addition, using MR spectroscopy, it has been shown that early Scl-Ab treatment reverts marrow fat expansion in ovariectomized rabbits, which could represent a further beneficial effect to those observed on bone mass and microstructural properties [89].

Fluorescent tags (e.g., quantum dots) and switchable molecular colors make in vivo imaging possible at the cellular level [90]. However, due to the hard-to-penetrate nature of calcified tissues for near infra-red lights (650–900 nm), alternative systems are yet to be developed to examine deep tissues. One such possibility is the development of radiopaque quantum dots with an affinity for MAT or HPM which can be systemically administered to facilitate MAT and HPM quantification and assessment.

Bone Porosity and Pore Fat Content Quantification

There is a growing interest in cortical porosity and bone marrow adiposity measurements to identify people at increased risk of fragility fracture. This has been performed using high-resolution peripheral computed quantitative tomography (HR-pQCT) [91–94]. Although a promising imaging method, the inherent limitation of X-rays to distinguish precisely fat from water means that HR-pQCT is unable to quantify pore content. Combining MRS with HR-pQCT may enable the quantification of pores size, distribution, and most importantly, pore content more accurately. By identifying pores with more water, from pores with a higher proportion of fat, this may provide a means to test the efficacy of anabolic therapies on osteoporotic patients and determine the effect of pore type on the progression of the pore sizes (trabecularization) and mechanical properties of cortical bone.

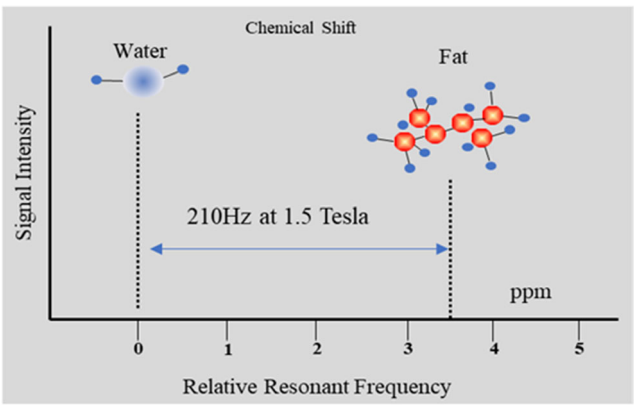

Fig. 6 A comparison of signal intensity and relative resonant frequency of protons in water and fat

Chemical Shift Imaging

Chemical shift encoding-based water-fat imaging has the advantage of overcoming spatial resolution and imaging time requirements for MRS. However, this type of quantification requires complex post-processing reconstructions and is prone to errors. Such a technique can be used to assess MAT composition [95, 96] (Fig. 6).

Limitations

Most X-ray-based imaging systems suffer high radiation doses, low resolution, and beam hardening artefacts and accessibility. Sophisticated image acquisition procedures, relatively higher cost, a variety of artefacts, and limited availability of MRI and MRS have made this modality not an accessible option for big cohort studies or as a diagnostic tool for marrow adiposity. Like most imaging modalities, another limitation is the resolution of the current clinical MR-based imaging systems that do not allow follow-up studies of the geography of fat expansion and bone and red marrow atrophy. The limitations of the conventional imaging systems have been summarized in Table 1.

Although MRI- and CT-based techniques have been shown to agree in quantifying tissues, it appears each is more appropriate in quantifying a particular type of tissue. For example, CT-based images are more apt for qualitative and quantitative assessment of hard tissue, but lack specificity at the assessment of soft tissues, particularly when enclosed within skeleton (e.g., red and yellow marrow). On the other hand, MRI images do not perform well in differentiating bone from fat or water-based tissue—depending on the sequence used (Fig. 7).

Detailing the image analyses and segmentation techniques for quantifying bone and marrow components is beyond the scope of this review, and we refer the readers to more specific reviews on the matter [97].

The Importance of MAT Quantification Standardization

Despite validation of MAT measurements in CT-based imaging systems of rodent bones [42•], applying these imaging techniques to quantify MAT in humans requires histological validation. Ectopic fat infiltration as a predisposing factor and possibly a predictor of osteoporosis [1••, 98], sarcopenia [98], diabetes [8], malignancies,

Table 1 The most commonly used imaging modalities and the advantages, limitations, and clinical potentials of each to quantify marrow adipose tissue (MAT)

Technique	Advantages	Limitations	Clinical potentials
CT-based techniques			
Conventional CT	Relatively widely available	High radiation dose, low resolution, averaging of voxels, beam hardening artefacts	With improved resolution and decreased radiation, MAT quantification is a likely potential Potential role for opportunistic screening for osteoporosis [96]
pQCT	Low radiation, relatively higher resolution	Expensive and limited availability; can only be used on distal extremities and not the trunk (e.g., spine). Beam hardening and averaging of the smaller bits of tissues (e.g., bone with fat) is still a limitation. Longer acquisition times make it sensitive to patient movement	High potentials to quantify MAT in higher resolutions, if the accessibility and price can be improved
HR-pQCT	Low radiation, relatively higher resolution	Very expensive and very limited availability; can only be used on distal extremities and not the trunk (e.g., spine). Beam hardening is an issue, but less probability of averaging of tissues due to decent voxel sizes in newer models (41 μm resolution). Longer acquisition times make it sensitive to patient movement	High potentials to quantify MAT in higher resolutions, if the accessibility and price can be improved
Micro-CT	High resolution	Very high radiation and long acquisition times have limited the use of μCT in vivo and especially in human subjects (currently only possible in lab animals or tissue samples). High price and lower availability have limited the access for researchers globally	Currently no clinical use is possible due to very high radiation dose and sensitivity to movement
Synchrotron nano-CT	Gold standard X-ray-based imaging. Best available phase-contrast images	Extremely high radiation doses, extremely limited access, extremely expensive to run experiments	Efforts to acquire phase-contrast images on living animals and humans are underway in the Australian synchrotron
Micro-CT + osmium tetroxide	Provides best contracts between MAT vs bone and red marrow	Extremely toxic. Also, suffers limitations of μCT	Alternative non-toxic chemicals are to be developed to avoid toxicity of the users. This may possibly extend the use of contrast materials to lab animals and possibly humans
MR-based techniques			
Routine care sequences (T1, T2, and STIR)	Part of routine clinical care	Limited availability Qualitative assessment	Can be assessed from routine care studies Potential for quantitative assessment
MRS	Most widely used method for MAT quantification	Limited availability Expensive	Emerging technique for MAT composition
Chemical shift encoding--based imaging	Emerging technique for MAT quantification	Limited availability Expensive Requires complex post-processing reconstructions	Emerging technique for MAT composition

anemia, and immunodeficiencies [1••, 22–30] has the potential to be used as a marker for those conditions. This mandates development and standardization of current techniques and the development of more sensitive and specific methods that make ectopic fat quantification possible at a lower cost, with minimal or no radiation and with no toxicity. MAT is of particular interest here due to being encased within bone that decreases the accuracy of current X-ray-based imaging modalities, and causes a variety of artefacts in MRI. The resolution of most commonly used X-ray or MRI images is not suitable to differentiate the delicate bone trabeculae and adjacent MAT and hematopoietic marrow. A potential use of accurate noninvasive MAT measurement is assessment of the response to osteoporosis treatments, as according to our experiments, MAT decline in response to risedronate therapy is far greater than the negligible bone volume increase [99••].

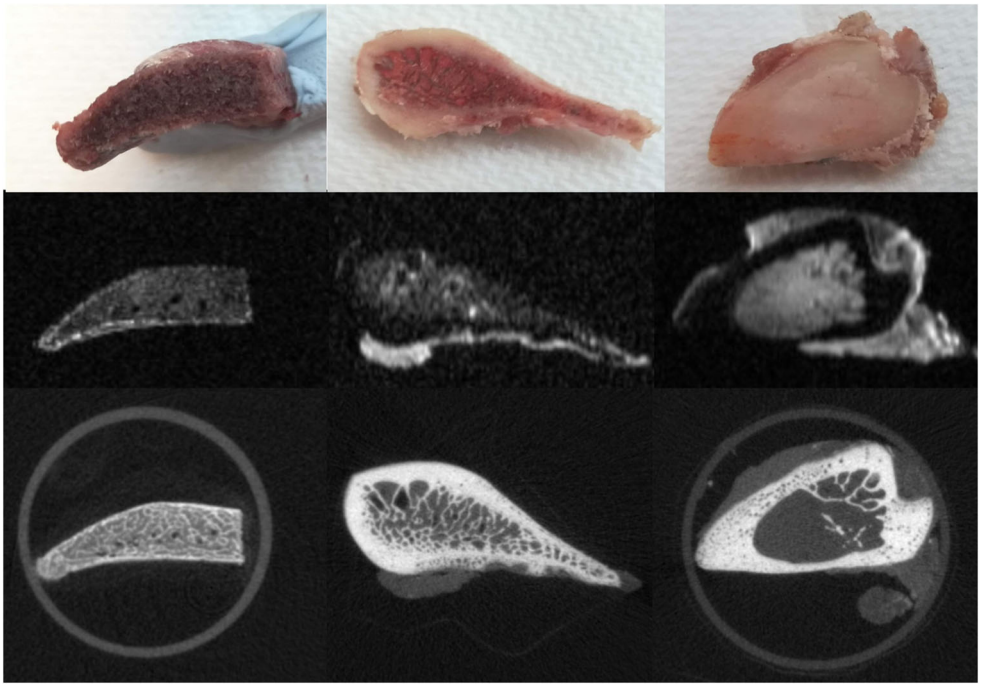

Fig. 7 Three samples (top row) with predominantly hematopoietic/red marrow (HPM; left), half/half HPM and marrow adipose tissue (MAT; middle) and predominantly MAT (right) scanned by micro-MRI (STIR; 190 μm resolution middle row) and HR-pQCT (81 μm resolution bottom row). As can be noted, MRI shows good contrast between MAT and HPM, but does not differentiate bone from either tissue. CT on the other hand shows excellent contrast between mineralized and soft tissues. Although the grey values of HPM and MAT overlap, they are still differentiable through good image analysis techniques

Conclusion

MAT is a major player in the pathophysiology of osteosarcopenia and anemia, and improved imaging techniques may play an important role in diagnosis, outcome prediction, and response to current and future treatments. Although current imaging modalities and analysis techniques permit the quantification of MAT, HPM, and bone, live tissue imaging is in its infancy and requires substantial improvements to non-invasively quantify all tissues, including MAT. A major limitation of the existing methods is their lack of capacity to quantify intra-myocytic fat, which can currently only be quantified by histological assessment and with oil-red-O staining. Future efforts should work towards the development of high-resolution and no or low-radiation in vivo imaging modalities with the capacity to differentiate tissue constituents at the molecular level (nano-scale). In particular, the development of non-toxic fat markers (non-toxic equivalents of osmium tetroxide) may prove useful in quantification of ectopic fat and MAT in particular. Development of better imaging techniques and modalities will require close collaboration between biomedical professionals, engineers, and physicists to develop such modalities.

Compliance with Ethical Standards

Conflict of Interest Ebrahim Bani Hassan, Ali Ghasem-Zadeh, Mahdi Imani, Numan Kutaiba, David K Wright, Tara Sepehrizadeh, and Gustavo Duque declare no conflict of interest.

Human and Animal Rights and Informed Consent This article does not contain any studies with human or animal subjects performed by any of the authors.

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Papers of particular interest, published recently, have been highlighted as:
- • Of importance
- •• Of major importance

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