Magnetic resonance imaging of total body fat

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THOMAS, E. Louise, Nadeem Saeed, J oseph V. H ajnal, Audrey Brynes, Anthony P. Goldstone, Gary Frost, and J immy D. Bell. Magnetic resonance imaging of total body fat. J. Appl. Physiol. 85(5): 1778–1785, 1998.—In this study we assessed different magnetic resonance imaging (MRI) scanning regimes and examined some of the assumptions commonly made for measuring body fat content by MRI. Whole body MRI was used to quantify and study different body fat depots in 67 women. The whole body MRI results showed that there was a significant variation in the percentage of total internal, as well as visceral, adipose tissue across a range of adiposity, which could not be predicted from total body fat and/or subcutaneous fat. Furthermore, variation in the amount of total, subcutaneous, and visceral adipose tissue was not related to standard anthropometric measurements such as skinfold measurements, body mass index, and waist-to-hip ratio. Finally, we show for the first time in the case of dual-energy X-ray absorptiometry, peripheral, body fat content with varying degrees of accuracy. However, until the advent of X-ray computed tomography and magnetic resonance imaging (MRI), it had not been possible to differentiate between subcutaneous and internal fat reserves. This is important as it is thought that internal fat, in particular visceral fat, may be a key factor in disease development.

Computed tomography has been used to measure total body fat content and to study different fat compartments, giving relatively accurate measurement of internal and subcutaneous fat depots (22). However, a fundamental drawback of the method is exposure to ionizing radiation, making whole body fat measurements, especially for serial studies, impractical.

MRI has been proposed as an alternative technique of determining internal and subcutaneous body fat. It has been validated in phantoms, animals, and human cadavers and has been shown to accurately measure adipose tissue in vivo, showing good agreement with values produced by dissection and chemical analysis (1, 8, 9, 17). Furthermore, it has been shown that MRI can be used serially to measure different body fat depots in human subjects (9, 18, 23). However, a review of published literature shows that a wide variety of methodologies has been used, with different MRI parameters and methods of data collection, ranging from extrapolation of single-slice acquisitions (2) or multiple-slice acquisitions over selected regions of the body (9, 23, 24, 26) to whole body fat measurements (18). Single or spatially limited multiple-slice acquisitions, as opposed to whole body data sets, have been used as a compromise between accuracy and cost, including scanning and/or analysis time. This, in part, has precluded the possibility of determining the relationship between different body fat depots.

In this study we have used whole body MRI data sets to determine the relationship between different body fat depots. We have also examined the effect of subsampling and the consequences of limited scanning on the measurements of body fat content. Finally, we used the MRI results to demonstrate individual variability of internal fat content in a cohort of 67 female volunteers over a wide range of total body fat content.

MATERIALS AND METHODS

Volunteer assessment. Written informed consent was obtained from all volunteers. Permission for this study was obtained from the Ethics Committee of the Royal Postgraduate Medical School, Hammersmith Hospital, London (Rec. 92/3995).

A cohort of 54 otherwise healthy female volunteers were studied. They were divided into four groups according to their body mass index (BMI): group A, BMI 19–24.9 kg/m² (n = 19); group B, BMI 25–29.9 kg/m² (n = 12); group C, BMI 30–39.9 kg/m² (n = 17); and group D: BMI >40 kg/m² (n = 6).

Whole body MRI was obtained from all volunteers. In addition, 13 (non-diabetic) female subjects with Prader-Willi syndrome (PWS; BMI range 23.61–51.63 kg/m²) were studied. These subjects are known to have an unusually high body fat content and an atypical body fat distribution. The PWS subjects were included to investigate a possible upper limit for body fat content. Body fat content was also measured by skinfold anthropometry in all 67 subjects and bioelectric impedance in 58 of these volunteers.

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THE ACCURATE DETERMINATION of total body fat, both internal and subcutaneous, has become an important issue as their differential contribution to diseases such as non-insulin-dependent diabetes mellitus and coronary heart disease becomes clearer. There are many techniques, including underwater weighing (11), anthropometry (7), body water dilution (20), impedance (21, 6), and dual-energy X-ray absorptiometry (16), that have long been used to give estimates of total, and, in the case of dual-energy X-ray absorptiometry, peripheral, body fat content with varying degrees of accuracy. However, until the advent of X-ray computed tomography and magnetic resonance imaging (MRI), it had not been possible to differentiate between subcutaneous and internal fat reserves. This is important as it is thought that internal fat, in particular visceral fat, may be a key factor in disease development.

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MRI. Magnetic resonance (MR) images were acquired on a Picker 1.0T HPQ system by using a rapid T1-weighted
spin-echo sequence with repetition time = 36 ms, echo time = 14 ms, flip angle 120°, field of view = 60 cm, 256 × 256 matrix, phase-conjugate symmetry, and a slice thickness of 10 mm. Images in the arms and legs were collected with two averages, those in the torso with one average. Subjects lay in the magnet in a prone position with arms straight above the head. This position was chosen to minimize respiratory motion, a potential source of artifact, and to ensure that large subjects would fit in the bore of the magnet. All images were acquired as single slices at the isocenter to avoid image distortion, and the volunteers were moved through the magnet on a purpose-built platter (3).

Effect of subsampling. The efficiency of data-acquisition schemes for assessing body fat may potentially be increased by subsampling and then extrapolating to estimate the total. A convenient method of achieving this is to image transverse slices with gaps between them. To investigate the consequences of this strategy, we studied three volunteers and acquired contiguous 10-mm-thick slices covering the torso from the head to the pelvis. The image data were analyzed slice by slice to measure the fat content, and the effects on the estimate of total and compartmental fat of omitting slices were examined. In each case the range of fat volumes achieved was calculated for all permutations of slice subsets with a chosen fraction of slices omitted. This provided a measure of the uncertainty introduced by each level of subsampling.

Whole body MRI. Subjects were scanned from their fingertips to their toes by acquiring 10-mm-thick transverse images with 30-mm gaps between slices in the arms and legs and 10-mm gaps between slices in the trunk. An illustrative figure from part of a data set is shown in Fig. 1.

Image analysis. With the scanning parameters employed, fat appears as a high signal against a muted background of other tissues and noise. The images were segmented and analyzed by using a software program that employed knowledge-based image processing to label voxels as fat and nonfat components. The image-processing and/or -analysis procedure employed a contour-following algorithm (12) to isolate individual structures from binary images produced by thresholding. The threshold needed to identify the voxels associated with fat components was computed automatically from gray-intensity histogram analysis and background-noise computation as described below.

Eight small, circular regions of interest of 5-mm radius were automatically placed at the edges of the image frame for

![Fig. 1. Transverse magnetic resonance images showing distribution of internal and subcutaneous fat from a whole body magnetic resonance imaging (MRI) data set from a healthy female volunteer [age = 21 yr, body mass index (BMI) = 27.9 kg/m², waist-to-hip ratio = 0.81, subcutaneous fat = 29.6 liters, visceral fat = 2.25 liters. Images were acquired by using a rapid T1-weighted spin-echo sequence from the volunteer's fingertips to her toes by acquiring 10-mm-thick transverse images with 30-mm gaps between slices.](http://jap.physiology.org/10.220.33.5)
where $M$ is the mean intensity and $SD$ is the standard deviation of intensity in the regions of interests.

Voxels with intensities less than $N$ were regarded as contributing only noise and were excluded from further analysis. A gray-level histogram was computed for the whole image; this had a bimodal appearance, which distinguished fat voxel intensity from the background noise. An initial fat threshold value ($T_F$) selected from the histogram by monitoring the global dip in the histogram, was employed as a first estimate of the fat voxel threshold. This was compared with $N$ to make sure that it was not less than $N$. If $T_F \leq N$, then $T_F$ was set to a value just above $N$. The image was thresholded by using $T_F$, and the outer subcutaneous fat boundary was extracted by employing the contour-following algorithm. An updated threshold value ($T_C$) for the fat voxels was computed on the basis of the distribution of voxel intensity along this extracted contour

$$T_C = T_M - T_{SD}$$

where $T_M$ is the mean intensity along the contour and $T_{SD}$ is the standard deviation along the contour. This equation was employed provided $T_C > N$. However, if this condition was not satisfied, then $T_C$ was increased to $N + 0.1T_{SD}$. This was taken as the optimum fat threshold for a particular slice, and the image was thresholded to create a binary image that identified all adipose tissue compartments. The outer subcutaneous fat boundary corresponding to this updated threshold value was extracted by using the contour-following algorithm. This contour was coded by using the Freeman eight-way chain code to give dimensional measurements and compressed storage of boundary data (10).

Prior knowledge was employed to extract the inner subcutaneous fat boundary, the internal fat compartments, and the bone marrow from the binary image created above. The knowledge base held information on the approximate size and position of these structures relative to the outer subcutaneous fat. In addition, shape constraints were introduced to identify the bone marrow. For example, in the arms, the bone marrow, when viewed in the transverse plane, was described as being closely related to a circular-shaped structure.

Each slice was then manually reviewed by using an interactive routine within the software program, and unwanted voxels were deleted. This was particularly useful in deleting pixels associated with the liver and bowel content that appear as bright, high-intensity structures in the images. These signals arise from tissue components with similar intensities and range (minimum-maximum). The relationship among groups was tested with Bland and Altman’s method (4). Differences among groups was tested by using the Student’s unpaired $t$-test.

**RESULTS**

The healthy subject population scanned in this study was selected to reflect a wide range of body sizes, with BMI ranging from 19 to 57 kg/m$^2$. The subject population was divided into four groups according to their BMI. A summary of the results for each group of healthy volunteers and the PWS subjects is shown in Tables 1 and 2. The percent total body fat was similar for subcutaneous, internal, and total fat measurements. From this, and previous work, we have adopted operating conditions that use 10-mm slices with 30-mm gaps (3). This scanning method resulted in an accurate measurement of fat distribution in a scan time of $\sim 20$ min, with a total analysis time of $\sim 4–5$ h, depending on a subject’s height. Therefore, this approach was used throughout the study.

Measurement of body fat by MRI. The percent total body fat content of each subject for the full cohort of 67 volunteers is shown in Fig. 3. The percent total body fat content of each of the four groups of healthy volunteers and PWS subjects is shown in Table 1. The same data are presented twice in Table 1, reflecting the effects of the alternative assumptions that can be made to interpret the results from the MR images. These assumptions relate principally to the nature of the content of each voxel identified as “fat tissue” in the MR images. The two assumptions are that each voxel identified is interpreted as containing only fat tissue (model A), or, alternatively, the voxel volume is assumed to reflect the adipose tissue itself, which is composed of triglycerides, water, proteins, and minerals (model B). For model B, we have employed a mean triglyceride volume fraction of 80%, as calculated from published values (23, 26).
Table 1. Body composition results for the different BMI groups and subjects with PWS

<table>
<thead>
<tr>
<th>Group</th>
<th>No. of subjects</th>
<th>BMI group</th>
<th>PWS</th>
<th>P &lt; 0.01</th>
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<tr>
<td>A</td>
<td>n = 19</td>
<td>23.11–40.86</td>
<td>59.12–2.16</td>
<td>58.70–2.41</td>
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<tr>
<td>B</td>
<td>n = 11</td>
<td>29.44–59.68</td>
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<td>D</td>
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Thus, depending which model is used, the resulting absolute measurement of total body fat content is substantially altered, leading to a range of percent body fat content for our volunteer population of 23.11–68.09 (model A) and 18.49–54.47% (model B). For clarity, we present the results using both methods wherever necessary.

Table 2. Body fat content determined by MRI for different BMI groups and subjects with PWS

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Figure 4 shows percent total body fat for each individual, grouped according to their BMI. Although the mean value for each group is significantly different, there is substantial variation within each group. Furthermore, some individuals in the lower BMI ranges have similar percent body fat as do those in the higher BMI groups.
Total internal and subcutaneous fat. Overall, there was an increase in total internal fat with increasing subcutaneous fat. However, there were some marked individual variations. A significant number of lean subjects (group A) had similar or higher percent total internal fat than did some obese individuals (groups B and C). This appeared to be true even when the fat content of the individuals was compared in absolute terms (liters) (Table 2).

Visceral and nonvisceral internal fat. The total internal fat content of each subject was subdivided into visceral and nonvisceral internal body fat. Visceral fat content was obtained by quantifying fat signals in the slices from the femoral heads to the slice containing the top of the liver or the base of the lungs (T10). Subcutaneous fat in these slices was labeled as “abdominal” subcutaneous fat. All other internal fat was labeled as “nonvisceral” internal fat. Interestingly, although we found a significant correlation between percent visceral fat and the waist-to-hip ratio ($r = 0.66, P < 0.01$) for the total group and for groups B ($r = 0.66, P < 0.01$) and C ($r = 0.53, P < 0.02$), at the extremes of BMI (groups A and D) there was no significant correlation (group A: $r = -0.13, P = 0.29$; group D: $r = 0.24, P = 0.32$). The same relationships were found between total internal fat and the waist-to-hip ratio.

Levels of visceral fat for the volunteers grouped according to their BMI are shown in Fig. 5. Although the mean value of visceral fat was significantly different among the four groups, levels of nonvisceral internal fat were relatively constant. For most volunteers the volume of nonvisceral internal fat was similar to or higher than that of their visceral fat, both as a percentage and in absolute terms (Tables 1 and 2). Again, as with total internal fat, a large range of visceral fat volumes was observed within each BMI group, with lean volunteers presenting visceral fat levels similar to or higher than those in overweight and obese subjects.

Assessment of standard techniques in comparison with MRI. Despite considerable individual variation, the standard techniques compared relatively well to MRI imaging. When all four groups of normal volunteers are taken together ($n = 54$), a significant correlation was found between MRI and impedance ($r = 0.93, P < 0.01$) and, to a lesser extent, between MRI and anthropometric ($r = 0.88, P < 0.01$) measurements of percent total body fat content. However, the correlation among variables in each group were quite different. There was a strong correlation between MRI and impedance in the overweight (group B: $r = 0.84, P < 0.01$) and obese volunteers (group D: $r = 0.90, P <
0.02) but a much weaker correlation in the lean volunteers (group A: r = 0.54, P < 0.02). Conversely, the correlation between MRI and anthropometry became weaker with increasing BMI (group A: r = 0.79, P < 0.01; group B: r = 0.59, P < 0.02; group C: r = 0.28, P < 0.14; group D: r = 0.29, P < 0.28).

The comparisons among the methods are also presented by using the method of Bland and Altman (4) by plotting the difference between the two measurements of a given subject against the mean. Figure 6 shows that there are significant differences among the methods. By using model B for the MRI data, agreement is slightly better between the pairs of techniques, although there is still considerable variance. It is notable that there are similar levels of individual discrepancy between each of the pairs of measurement techniques compared. Also, anthropometry appears to introduce a size-related effect compared with either MRI or impedance.

The MRI results for percent body fat content appear to level out for several of the PWS subjects, despite a very wide range of body weights (90–140 kg) (see Fig. 3). This phenomenon was independent of the MRI assumption used for estimating body fat content. By using model B, the MRI results gave an upper limit for maximum percent triglyceride content of ~54%, whereas model A leads to a maximum percent body fat content of ~68%, close to the theoretical maximum suggested by Webster et al. (27). Furthermore, despite the fact that subjects with PWS had the highest percent body fat, their visceral fat content was comparatively low. Indeed, there were no significant differences in the ratio of visceral to subcutaneous fat between the subjects with PWS (0.067 ± 0.01) and volunteers in group A (0.067 ± 0.01) or B (0.069 ± 0.01). However, the ratio of visceral to subcutaneous fat was significantly higher in volunteers in groups C (0.105 ± 0.01) and D (0.099 ± 0.01) than in those subjects with PWS.

DISCUSSION

This study demonstrates that the whole body MRI method gives an unbiased measurement of fat content, internal and subcutaneous, for a large range of body shapes and sizes. It also shows that there is significant variation in the percentage of total internal, as well as of visceral and nonvisceral fat, across the BMI range, which cannot be easily predicted from total body fat and/or subcutaneous fat.

The most accurate MRI method for measuring total body fat is no doubt an examination using contiguous slices covering the entirety of a volunteer’s body. However, this will demand considerable scanning and analysis time. At the other extreme is the use of a single slice at a predetermined level to extrapolate total fat content (2), thereby reducing scanning and analysis time at some cost in accuracy. We have shown that there is an increase in measurement uncertainty as slices are removed from a contiguous data set. This uncertainty highlights the possible inaccuracies in using single-slice data sets and is consistent with previous studies (19). For example, Ross et al. (19) found that, although a single-slice analysis produced significant correlation with total body fat, it only accounted for 81% of the variance in the group as a whole. It may be valid to use single-slice data to provide a rough estimate of internal fat, but potentially important information and subtle differences will be compromised. This is particularly important in studies where the relationship between fat and biological and genetic markers needs to be determined. Similarly, the relative accuracy of the MRI measurement is affected by the use of images obtained by using multislice techniques. Typically, 10 or more slices are acquired at a time, covering an area of the body of 40 cm or more. We have shown previously that there can be significant image distortion when images are acquired at increasing distances from the isocenter of the magnet (3). For this reason, we acquired all images individually at the isocenter of the magnet.

The whole body MRI approach provides an accurate measurement of the volume of adipose tissue, which, in turn, can be rigorously validated against gold standards of volume. However, converting these results into quantities of triglycerides does require the use of certain assumptions. These relate to the composition of the tissue detected as “fat” in the MRI images. Some previous MRI studies of body fat content have referred to the results loosely as “adipose tissue” without specifying what percentage of this adipose tissue actually

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**Fig. 6.** Bland and Altman (4) plots of differences between measurements using impedance (A) and anthropometry (B) vs. means for MRI.
corresponds to triglycerides within the cells (24, 26, 18). Others have used values of adipocyte composition in the existing literature to assign the corresponding volume detected by MRI to triglycerides (1, 9). We have illustrated here that these assumptions can significantly affect the absolute levels of body fat content ascribed to a given subject. This is an important factor when MRI results are compared with other techniques. However, the use of these assumptions in the interpretation of the MRI results should not significantly limit the application of MRI to studies of the effects on health and disease of body fat content and distribution. This is because these assumptions do not alter the intersubject and intraindividual differences in total and compartmental triglycerides in human subjects.

As expected, total internal fat increased with increasing subcutaneous fat; however, a significant number of lean subjects had similar or higher percentages of internal fat than did some obese individuals. This, in part, explains the relatively weak correlation that we found between total subcutaneous and internal body fat. A better correlation was found between visceral and subcutaneous adipose tissue. This may be because the visceral fat measurement does not include any contribution from the nonvisceral internal fat. It is therefore possible that the deposition of visceral and abdominal subcutaneous adipose tissue may be related in some way, whereas the deposition of nonvisceral adipose tissue is independent of one another.

Considerable work has been carried out to develop noninvasive techniques that can accurately determine body fat content in human subjects. Similarly, substantial work has gone into investigating the possible environmental and genetic determinants of body fat distribution. However, the relationship between different body fat depots during a period of weight gain is not fully understood. Our study has shown significant individual variations in total internal and visceral fat, which demonstrates the difficulties in predicting internal fat content (visceral and nonvisceral) from techniques that are unable to measure regional fat distribution directly. It is also important to note that this is also a significant problem for MRI studies, which use limited data acquisition, including single-slice techniques. This is emphasized by the fact that in many volunteers we found that there was more nonvisceral internal fat than visceral fat. The physiological function of the nonvisceral internal fat depot has not been fully determined. However, its accurate measurement may be important, particularly in light of the possible relationship between muscle triglycerides and non-insulin-dependent diabetes mellitus. It may therefore be pertinent to measure both visceral and nonvisceral internal fat in future studies of body fat content.

Webster et al. (27) hypothesized that, when a person puts on weight, the added tissue has a constant ratio of fat to fat-free mass, with ~75% fat and ~25% fat-free mass, so the percent body fat will approach 75% at infinite weight but will not exceed it. The percent body fat of several of the PWS subjects, measured by MRI (by using model A), was close to this theoretical maximum body fat content. It is unclear whether this effect is peculiar to subjects with PWS or is a general finding. It is interesting to note, however, that these volunteers with the same percent body fat had very different body weights. The plateau at a slightly lower maximum percent body fat observed in this study may represent a mechanical upper limit for a mobile subject. Indeed, one of the most obese volunteers scanned in this study had a body fat distribution. However, the use of these assumptions in the interpretation of the MRI images of her lower legs. To our knowledge, such structure has not been previously reported and may be associated with the need to provide mechanical support for the excess adipose tissue.

There are a number of difficulties in comparing different techniques directly with each other as they all make inherent assumptions, which can create bias. Most techniques for measuring body fat or lean tissue content need to assume a constant density for both fat (0.9 kg/l) and the fat-free (1.1 kg/l) body components. This may be particularly inaccurate for the fat-free body components as the water in the fat %. Although the standard methods correlated relatively well with the direct MRI measurement, the percent body fat observed in this study may represent a lower absolute percentage than the MRI measurements. The plateau at a slightly lower maximum percent body fat observed in this study may represent a mechanical upper limit for a mobile subject. Indeed, one of the most obese volunteers scanned in this study had a lower maximum percent body fat than the MRI images of her lower legs. To our knowledge, such structure has not been previously reported and may be associated with the need to provide mechanical support for the excess adipose tissue.

Conventional indirect techniques, including underwater weighing, body water dilution, impedance, and anthropometry, measure body fat content by empirically determined relationships on the basis of population averaging. A critical difference between MRI and the other techniques is that MRI volume measurement is amenable to absolute calibration. This leaves only tissue distribution and tissue content as possible sources of measurement errors in different individuals. MRI-based studies are likely to be less affected by individual variability and may therefore achieve higher statistical power for a given sample size.

In conclusion, MRI gives a fast, reliable, and nonbiased measurement of body fat content (subcutaneous and internal) in subjects encompassing a wide variety of body shapes and sizes. It has allowed accurate assessment of the relationship between internal and subcutaneous fat and has shown great variation in internal fat content within the subject population studied. The technique described herein will help to determine accurately the impact of genetic and environmental factors on different body fat compartments.
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