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1 **Fat accumulates preferentially in the right rather than the left liver lobe in**
2 **non-diabetic subjects**

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12

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1 ABSTRACT

2 **Aims:** To examine the distribution of liver fat (LFAT) in non-diabetic subjects
3 and test whether the fat in the right as compared to the left lobe correlates
4 better with components of the metabolic syndrome or not.

5 **Methods:** In this cross sectional study, we determined LFAT by ¹H-MRS in the
6 right lobe (LFAT%^{MRS}), and by MRI (LFAT%^{MRI}) in four regions of interest
7 (ROIs1-4, two in the right and two in the left lobe) in 97 non-diabetic subjects
8 (age range 22-74yrs, BMI 18-41 kg/m²) and compared the accuracy of LFAT^{MRI}
9 in the different ROIs in diagnosing non-alcoholic fatty liver disease (NAFLD)
10 using areas under the receiver operator characteristic (AUROC) curves.

11 **Results:** 38% of the subjects had NAFLD (LFAT%^{MRS}). LFAT%^{MRI} was
12 significantly higher in the right (5.7±0.5%) than the left (5.1±0.4%) lobe
13 (p<0.02).The AUROC for LFAT%^{MRI} in the right lobe for diagnosing NAFLD
14 was significantly better than that in the left lobe. The relationships between
15 several metabolic parameters and LFAT%^{MRI} in the left lobe were significantly
16 worse than those for LFAT%^{MRS} while there was no difference between
17 LFAT%^{MRS} and right lobe ROIs.

18 **Conclusions:** Liver right lobe contains more fat and correlates better with
19 components of the metabolic syndrome than the left in non-diabetic subjects.

20

21 **Key words:** metabolic syndrome, insulin, obesity

1. Introduction

2 Studies performed in dogs over 100 years ago showed that blood flow from the
3 splenic vein streamlined to the left lobe of the liver, while blood from the
4 superior mesenteric vein was directed to the right lobe[1, 2]. Similarly, studies
5 in humans have shown that blood from the superior mesenteric vein, which
6 drains the right colon, ileum and jejunum, is directed to the right lobe while
7 blood from the spleen and left colon drain to the left lobe[3]. In obese subjects,
8 the rate of visceral lipolysis is increased compared to non-obese subjects and
9 can account for up to 50% of free fatty acids (FFA) delivery to the liver[4]. Many
10 obese subjects accumulate fat in the liver due to non-alcoholic causes (NAFLD)
11 and therefore possibly an increased flux of FFA from visceral fat to the superior
12 mesenteric vein. Studies using Doppler ultrasonography in healthy volunteers
13 have shown that in response to a meal, intrahepatic portal vein blood flow
14 increases more in the right than the left lobe [5]. FFA delivery from both the
15 meal and from visceral lipolysis might preferentially increase liver fat content
16 more in the right than the left lobe. Metabolic consequences of hepatic insulin
17 resistance in NAFLD such as hyperinsulinemia and hypertriglyceridemia could
18 therefore also be better correlated with fat in the right than the left lobe.

19

20 Few data are available examining fat distribution in the human liver. In an
21 autopsy study, Merat et al [6] analyzed three different parts of the liver (2×2×2
22 cm each) and found steatosis to be unevenly distributed ($\kappa = 0.64$).

1 Larson et al [7] and Merriman et al [8] took liver biopsies from the right and left
2 lobes of morbidly obese patients and found low variability for steatosis
3 between right and left lobes. However, multiple liver biopsies can not be used
4 to assess heterogeneity of liver fat because of risks of bleeding.

5

6 Magnetic resonance imaging (MRI) methods enable accurate assessment of
7 liver steatosis[9, 10].Until now, only one study has been published where
8 heterogeneity of liver fat was analyzed in hospitalized patients with type 2
9 diabetes, who were using oral hypoglycaemic agents or insulin, and the right
10 lobe was found to contain more fat than the left lobe[11]. However, the
11 pathways leading to NAFLD may differ between type 2 diabetic patients and
12 non-diabetic subjects[12], and the drugs, especially insulin and glitazones
13 could affect the routes which regulate intrahepatic fat content[13][14].
14 Regional variation in liver fat content in non-diabetic subjects has not been
15 examined.

16

17 The liver is the source of glucose and (VLDL) triglycerides after an overnight
18 fast, two of the key components of the metabolic syndrome. The liver, once
19 fatty, becomes resistant to the normal actions of insulin to inhibit the production
20 of glucose and VLDL[15]. This leads to hyperglycemia and stimulation of
21 insulin secretion as well as hypertriglyceridemia[16]. All components of the
22 metabolic syndrome are significantly correlated with liver fat content [15].

1 However, if there is, as discussed above, more fat in the right than the left lobe,
2 the former might be better correlated with features of insulin resistance than
3 the latter. Indeed, identification of the region, which best correlates with
4 features of insulin resistance, could be considered as the region which
5 physiologically defines the most relevant area of fat accumulation in the liver,
6 and provides the area which enables the most accurate diagnosis of NAFLD.

7

8 In the present study, we hypothesized that the right lobe of the human liver
9 would contain more fat than the left lobe in non-diabetic subjects, and that fat
10 in the right as compared to the left lobe might correlate better with components
11 of the metabolic syndrome. To this end, we quantified liver fat by MRI in 4
12 different regions of interest (ROIs) in 97 non-diabetic subjects, and also
13 measured liver fat by proton magnetic resonance spectroscopy (^1H -MRS), the
14 golden standard for measurement of liver fat content.

1 **2. Methods**

2 **2.1. Subjects**

3 We analyzed data from all non-diabetic subjects studied since August 2007
4 when we added an in-phase (IP) and out-of-phase (OP) imaging sequence to
5 our MRI protocol to enable monitoring of spatial distribution of fat in the liver.

6 This sequence covers the whole liver and allows to quantitate the intensity of
7 the liver. The group comprised of 97 subjects (56women, 41men), who met the
8 following inclusion criteria: i) age 20 to 75 years, ii) no known acute or chronic
9 disease other than obesity, hypertension or NAFLD. iii) no evidence of
10 advanced fibrosis as determined using the NAFLD fibrosis score[17].

11 Exclusion criteria included i) diabetes , ii) autoimmune liver disease (past
12 medical history); iii) viral liver disease (positive for HBsAg or HCVAb) , iv)
13 drug-induced liver disease (past medical and drug use history), v) excessive
14 use of alcohol (more than 20g/day for men, more than 10g/day in women[18]),
15 vi) pregnancy or lactation. All protocols were in accordance with the Helsinki
16 Declaration of 1975 and approved by the ethics committee of the Helsinki
17 University Central Hospital, and each subject provided written informed
18 consent.

19

20 Eligible subjects were studied after an overnight fast. At this visit, body weight
21 and height, waist and hip circumferences were measured, % body fat and
22 blood pressure were recorded, and blood samples were taken for

1 measurement of biochemical parameters as detailed below. On a second
2 occasion, ^1H -MRS and MRI studies were performed to quantitate liver fat (vide
3 infra).

4

5 **2.2. % Liver fat (^1H -MRS)**

6 Localised single voxel ($2 \times 2 \times 2 \text{ cm}^3$) proton spectra were acquired using a 1.5-T
7 whole-body system (Siemens Magnetom Avanto, Erlangen, Germany), which
8 consisted of a combination of whole-body and loop surface coils for
9 radiofrequency transmitting and signal receiving. T1- and T2-weighted
10 high-resolution MR images were used for localisation of the voxel of interest
11 within the right lobe of the liver. ^1H -MRS measurements of the liver fat were
12 performed in the middle of the right lobe at a location that was individually
13 determined for each subject by a single operator; vascular structures and
14 subcutaneous fat tissue were avoided when selecting the voxel. Single voxel
15 spectra were recorded using the point resolved spectroscopy (PRESS), with
16 an echo time of 30 msec, a repetition time of 3000 msec, 1024 data points
17 over 1000 kHz spectral width with 16 averages. A short echo time and long
18 repetition time were chosen to ensure a fully relaxed water signal, which was
19 used as an internal standard. Chemical shifts were measured relative to water
20 at 4.70 ppm. The methylene signal, which represents intracellular triglyceride,
21 was measured at 1.3 ppm. Signal intensities were quantified by using a jMRUI
22 v3.0 analysis program [19] with an advanced method for accurate, robust and

1 efficient spectral fitting (AMARES)[20]. Signal intensities were corrected for T2
2 relaxation as previously described[21]. Spectroscopic intracellular triglyceride
3 content (LFAT) was expressed as a ratio of the area under the methylene peak
4 to that under the methylene and water peaks. LFAT% was converted from
5 signal ratios to volume fractions by applying the method validated by Longo et
6 al [22] and using experimentally determined values by Szczepaniak et al.
7 [23](x 100 = % liver fat). This measurement has been validated against
8 histologically determined lipid content [24] and against estimates of fatty
9 degeneration or infiltration by X-ray computer-assisted tomography by us [25]
10 and others[26]. All spectra were analysed by a physicist who was unaware of
11 any of the clinical data. The reproducibility of repeated measurements of LFAT
12 in non-diabetic subjects studied on two occasions in our laboratory is 11%.
13 NAFLD was defined as in the population-based Dallas Heart Study, as a
14 LFAT%> 5.55%[27].

15

16 **2.3. Liver fat fraction (MRI)**

17 A body coil was used to obtain coronal scout images of the upper abdomen. A
18 stack of transaxial IP and OP T1-weighted dual-echo fast spoiled-gradient
19 recalled images were obtained in two breath holds using following imaging
20 parameters: 103 msec repetition time, 2.12 msec (OP)/4.8 msec (IP) echo time,
21 80° flip angle, 24 slices (x2), 1 cm slice thickness, 512 x 448 matrix and 5 dm x
22 4.37 dm field of view, the same matrix and field was used for all patients.

1 Trans-axial abdominal scans encompassing the entire liver were acquired in
2 two breath-holds using the magnet mentioned above. Each 2D slice was
3 displayed on the video screen. Using the Image J 1.46r software to analyze the
4 intensity of the liver, four ROIs (1-2 cm in diameter) were obtained in the liver
5 above the portal vein (two in the right lobe and two in the left lobe, Fig.1). ROIs
6 1-4 were located in liver segments II, IV, VIII, VII, respectively. The standard
7 deviation of the signal intensity measurements within each ROI was kept to
8 less than 10%. The ROIs included areas of parenchyma that did not contain
9 vessels or artifacts, and the ROIs were in corresponding locations in the paired
10 IP and OP MR images. The signal intensity of the spleen was measured as the
11 mean intensity from the three 1 to 2 cm ROIs (Fig.1). LFATFr^{MRI} was quantified
12 on T1-weighted dual-echo gradient-echo MR images as the percentage of
13 relative signal intensity loss of the liver on OP images, with the following
14 formula: $(SI_{in} - SI_{out}) / SI_{in} \times 100$, where SI is the mean liver signal intensity
15 divided by the mean spleen signal intensity, SI_{in} is IP signal intensity, and SI_{out}
16 is OP signal intensity [10]. LFATFr^{MRI} was converted to ¹H-MRS % units (% liver
17 fat by MRI, LFAT%^{MRI}) using the equation relating MRI and ¹H-MRS
18 measurements (see results).

19

20 **2.4. Intra-abdominal fat and subcutaneous fat (MRI)**

21 A series 16 of T1-weighted axial images with selective fat excitation were
22 acquired from a region extending from 8 cm above to 8 cm below the L4/5

1 lumbar intervertebral disks (16 slices, field of view 438×500 mm², slice
2 thickness 1cm, breath-hold repetition time 91 msec, echo time 5.24 msec).
3 Intra-abdominal and subcutaneous fat areas for each slice were determined
4 using a region growing method in SliceOmatic v4.3 image analysis program
5 (Tomovision, Montreal, Canada). The areas were multiplied by the slice
6 thickness and the results were expressed as total volumes of Intra-abdominal
7 and subcutaneous adipose tissues[25].

8

9 **2.5. Other measurements and analytical procedures**

10 Waist circumference was measured midway between the spinailiaca superior
11 and the lower rib margin, and hip circumference at the level of the greater
12 trochanters [28]. Body weight was recorded to the nearest 0.1 kg using a
13 calibrated weighting scale (Soehnle, Monilaite-Dayton, Finland) with subjects
14 barefoot and wearing light indoor clothing. Body height was recorded to the
15 nearest 0.5 cm using a ruler attached to the scale. % body fat was determined
16 by bioimpedance plethysmography as previously described [29].

17

18 Blood pressure was measured in the sitting position after 10 to 15 minutes of
19 rest using a random-zero sphygmomanometer (Erka, Germany). Fasting
20 plasma glucose (FPG) concentrations were measured in duplicate using the
21 glucose oxidase method using Beckman Glucose Analyzer II (Beckman
22 Instruments, Fullerton, CA)[30]. Fasting serum (fS) free insulin concentrations

1 were measured with the Auto-DELFLIA kit (Wallac, Turku, Finland).
2 Glycosylated hemoglobin A_{1c} (HbA_{1c}) was measured by high performance
3 liquid chromatography using the fully automated Glycosylated Haemoglobin
4 Analyzer System (BioRad, Richmond, CA)[31]. Serum (S) total cholesterol,
5 high density lipoprotein (HDL) cholesterol and triglyceride concentrations were
6 measured with the enzymatic kits from Roche Diagnostics using an
7 auto-analyser (Roche Diagnostics Hitachi, Hitachi Ltd., Tokyo, Japan). The
8 concentrations of low density lipoprotein (LDL) cholesterol were calculated
9 using the Friedewald formula[32]. Serum albumin, blood counts, alanine
10 aminotransferase (ALT), aspartate aminotransferase (AST) and
11 gamma-glutamyltransferase (GGT) activities were determined as
12 recommended by the European Committee for Clinical Laboratory Standards.
13 The NAFLD fibrosis score was calculated as described by Angulo et al [17]
14 and as recommended by the American College of Gastroenterology[33].
15 Serum hepatitis B virus surface antigen (HBsAg) and hepatitis C virus antibody
16 (HCV-Ab) were tested by the Enhanced Electrochemiluminescence method.
17
18 Homeostasis model assessment-insulin resistance (HOMA-IR) was calculated
19 as described by Matthews et al [34].

20

21 **2.6. Statistical analyses**

1 Normal distribution of data was analyzed using the Kolmogorov-Smirnov test.
2 Non-normally distributed data were used after logarithmic (base 10)
3 transformation. Normally distributed data are shown as mean \pm SE, whereas
4 non-normally distributed data are shown as median followed by the 25% and
5 75% percentiles. The unpaired Student's t-test and the Mann-Whitney test
6 were used to compare mean values of normally and non-normally distributed
7 data. Pearson's correlation coefficient was calculated for normally and
8 Spearman's rank correlation coefficient for non-normally distributed data. Lin's
9 concordance coefficient, which combines measurements of precision and
10 accuracy, was computed to determine whether the observed data deviate
11 significantly from the line of perfect agreement[35]. Multiple linear regression
12 analysis was performed to set an equation to get the LFAT%^{MRI} from
13 LFATFr^{MRI} using LFAT%^{MRS} as reference standard and test which part of
14 LFAT%^{MRI} was the independent predictor of LFAT%^{MRS}. Correlation
15 coefficients were compared using the Hotelling-Williams test by R 2.13.1
16 statistical package(R software, R Foundation for Statistical Computing, Vienna,
17 Austria). Receiver operating characteristic (ROC) curves were set up by SPSS
18 20.0 for Windows (SPSS, Chicago, IL) and compared using the Medcalc
19 12.7.1.0 (MedCalc Software, Mariakerke, Belgium). Values of area under
20 receiver operating characteristic (AUROC) equalling 1 represent a perfect test;
21 between 0.9 and 1 were considered as excellent; between 0.8 and 0.9 good;
22 between 0.7 and 0.8 fair; between 0.6 and 0.7 poor; and 0.5 a worthless test.

- 1 Other calculations were made using SPSS 20.0 for Windows and GraphPad
- 2 Prism version 5.00 for Windows (GraphPad Software, San Diego, CA). A
- 3 p-value of less than 0.05 was considered statistically significant.

1 **3. Results**

2 **3.1. Characteristics of the study subjects**

3 Anthropometric and biochemical characteristics of the subjects are
4 summarized in Table 1. 38% of the subjects had NAFLD as measured by
5 LFAT%^{MRS}. None of the subjects had evidence of fibrosis (NAFLD fibrosis
6 score < 0.676 [17]). The LFAT%^{MRS} averaged 5.8±0.6% (1.9±0.2% for
7 non-NAFLD, 12.0±0.7% for NAFLD, p<0.001).

8

9 **3.2. Regional variation in liver fat**

10 The equation relating LFAT%^{MRS} (right lobe) and LFATFr^{MRI} (right lobe) was as
11 follows: $LFAT\%^{MRS} = (2.837 \pm 0.374) + (0.198 \pm 0.013) \times LFATFr^{MRI}$ (r=0.837,
12 p<0.001, Fig. 2). LFAT%^{MRI} in ROIs 1, 2, 3 and 4 averaged: 4.8±0.5%,
13 5.5±0.5%, 5.8±0.5% and 5.7±0.5%. The LFAT%^{MRI} was significantly lower in
14 ROI 1 than in ROI 2 (p<0.02), ROI 3 (p<0.001) and ROI 4 (p<0.02, Fig.3).
15 LFAT%^{MRI} was significantly higher in the right (5.7±0.5%) than the left
16 (5.1±0.4%) lobe (p<0.02, Fig.3).

17

18 The correlation coefficients (Pearson) between LFAT%^{MRS} and LFATFr^{MRI} in
19 ROIs were: r=0.738, p<0.001 for ROI 1, r=0.785, p<0.001 for ROI 2, r=0.825,
20 p<0.001 for ROI 3, r=0.828, p<0.001 for ROI 4; r=0.837, p<0.001 for the right
21 lobe and r=0.781, p<0.001 for the left lobe. Lin's concordance coefficients
22 between LFAT%^{MRS} and LFAT%^{MRI} in ROIs were: rho(c)=0.704, p<0.001 for

1 ROI 1, $\rho(c)=0.763$, $p<0.001$ for ROI 2, $\rho(c)=0.808$, $p<0.001$ for ROI 3, and
2 $\rho(c)=0.820$, $p<0.001$ for ROI 4, $\rho(c)=0.824$, $p<0.001$ for the right lobe,
3 $\rho(c)=0.74$, $p<0.001$ for the left lobe.

4

5 Multiple linear regression analysis showed $\text{LFAT}\%^{\text{MRI}}$ in the right but not the
6 left lobe was an independent predictor of $\text{LFAT}\%^{\text{MRS}}$ ($\beta=0.160$, $p<0.001$).

7

8 We analyzed ROC curves to determine how use of different ROIs influence the
9 diagnosis of NAFLD. $\text{LFAT}\%^{\text{MRI}}$ in ROI 3 showed excellent accuracy in
10 diagnosing NAFLD (AUROC = 0.936 ± 0.023 , $p<0.001$, sensitivity 94.6%,
11 specificity 83.3%). The AUROC for ROI 1 was good (AUROC = 0.879 ± 0.034 ,
12 $p<0.001$, sensitivity 70.3%, specificity 90.0%) but significantly lower than that
13 of AUROC for ROI 3 ($p<0.02$; Table 2). Likewise, the AUROC was significantly
14 higher using $\text{LFAT}\%^{\text{MRI}}$ in the right (AUROC = 0.934 ± 0.025 , $p<0.001$,
15 sensitivity 86.5%, specificity 90.0%) than in the left lobe
16 (AUROC = 0.894 ± 0.032 , $p<0.001$, sensitivity 73.0%, specificity 91.7%) lobe
17 ($p<0.05$, Table 2, Fig. 4).

18

19 **3.3. Metabolic parameters and liver fat in different ROIs**

20 The relationships between metabolic parameters and $\text{LFAT}\%^{\text{MRS}}$ and
21 $\text{LFAT}\%^{\text{MRI}}$ in the different ROIs are shown in Table 3. The correlation
22 coefficients between BMI, waist and hip circumference, body fat,

1 intra-abdominal and subcutaneous fat, S-AST, S-ALT and LFAT%^{MRS} were
2 significantly higher than those between these metabolic features and
3 LFAT%^{MRI} in ROI 1 (Fig. 5, Table 3). The correlation coefficient between
4 HOMA-IR, S-AST, S-ALT and LFAT%^{MRI} in ROI 2 was significantly lower than
5 those between LFAT%^{MRS} and these metabolic features (Fig. 5, Table 3). The
6 correlation coefficients between the metabolic parameters and LFAT%^{MRI} in
7 ROIs 3 and 4 did not differ from those relating the respective parameters and
8 LFAT%^{MRS} (Fig. 5, Table 3).

1 **4. Discussion**

2 In the present study of 97 non-diabetic subjects, we found the right lobe of the
3 liver to contain significantly more fat than the left lobe. Measurement of liver fat
4 in the right lobe perhaps provided a significantly more accurate diagnosis of
5 NAFLD than that in the left lobe. Since liver fat was not homogenously
6 distributed, we examined, which region in the liver was most closely
7 associated with metabolic abnormalities associated with NAFLD. The
8 correlation coefficients for measures of obesity, liver enzymes and HOMA-IR
9 were best associated with fat in the right lobe of the liver. The right lobe could
10 therefore be recommended as the location for quantification of liver fat in
11 diagnosing NAFLD.

12

13 Compared to $^1\text{H-MRS}$, MRI is available on most MR units and can be
14 performed easily in routine examinations [36]. Previous studies have shown
15 that MRI-based measurements of the liver fat are closely correlated with those
16 obtained by $^1\text{H-MRS}$ [9, 37]. These studies included patients with type 2
17 diabetes ($n=33$) and those with a fatty liver ($n=12$), and found the correlation
18 coefficient between MRI and $^1\text{H-MRS}$ methods to be 0.959 [9] and 0.950 [37].
19 The correlation coefficient found in the present study in 97 subjects (0.837) is
20 in line with these data.

21

22 Distribution of liver fat has been analysed using MRI or MRS in only one

1 study[11]. In the study of Captan et al in hospitalized type 2 diabetic patients,
2 liver fat averaged 8.9% in the right and 7.7% in the left lobe ($p < 0.001$). The
3 present data in non-diabetic subjects showed a similar difference in the fat
4 content of the liver lobes as Captan et al[11].

5

6 The present study does not provide mechanistic insights as to why liver fat
7 content is significantly higher in the right than the left lobe. The most probable
8 explanation is provided by previous data showing that portal blood flow is
9 streamline rather than turbulent. Portal blood flow conveyed via the superior
10 mesenteric vein drains visceral fat and supplies the right lobe of the liver while
11 blood from the spleen flows into the left lobe[1, 2]. Fat does not accumulate in
12 the spleen in obesity[38, 39]. In keeping with these data on blood flow and on
13 the increased visceral lipolysis in obesity[4], we found waist circumference and
14 visceral fat to correlate better with fat in the right than the left lobe. Dauzat et al
15 measured the hemodynamic effects of a meal on the splanchnic and hepatic
16 circulation in 30 healthy volunteers using Doppler ultrasonography[5].
17 Intrahepatic portal blood flow was shown to increase significantly more in the
18 right than the left lobe [5]. Thus, the right lobe may receive more substrates for
19 hepatic lipogenesis such as fatty acids, glucose and amino acid also
20 postprandially. Direct measurement of blood flow distribution in the liver of
21 patients with NAFLD combined with quantification of the contribution of
22 different pathways (lipolysis, de novo lipogenesis, the spillover pathways,

1 uptake of chylomicron remnants postprandially) would be needed to provide
2 mechanistic insights for the present data.

3

4 The present data are to our knowledge the first to relate metabolic features of
5 NAFLD to fat content of different regions of the liver. Given the heterogeneous
6 distribution of liver fat, such analysis is of interest as it helps to identify the
7 region of the liver, which should be used for diagnosis of NAFLD. We found the
8 correlation coefficients between LFAT%^{MRI} in ROI3 and ROI4 and measures of
9 both subcutaneous and visceral obesity to be as good as those between these
10 measures and LFAT%^{MRS}. The correlation coefficient between LFAT%^{MRI} in
11 ROI1 was worse than that between the measures of obesity and LFAT%^{MRS}.
12 These data suggest that obesity increases liver fat especially in the right lobe
13 of the liver. Liver enzymes, a simple but not ideal marker of NAFLD and
14 non-alcoholic steatohepatitis (NASH)[40], were also better correlated with liver
15 fat in the right lobe. Finally, HOMA-IR, a product of fasting insulin and glucose,
16 was significantly worse correlated with LFAT%^{MRI} in ROI 2 but not the other
17 ROIs than with LFAT%^{MRS}. These data suggest that fat accumulates in the
18 right region of the liver and that this may correlate better with insulin resistance
19 and metabolic abnormalities than the left .

20

21 One limitation of our study was only addressed to evaluate the relative
22 distribution of liver fat into the right and the left lobe and its association with

1 metabolic abnormalities, but not addressed to explore the relationship between
2 the liver fat distribution and the severity of liver damage. Accordingly, no
3 insights about this relationship should be provided. Another shortcoming was
4 that we did not offer much mechanism as to heterogeneity of liver fat that
5 should be further studied in the future.

6

7 **5. Conclusion**

8 In conclusion, liver fat is heterogeneously distributed in non-diabetic subjects,
9 right lobe containing more fat than the left. When measured by MRI as in the
10 present study, quantification of fat in the right as compared to the left lobe
11 perhaps allows a more accurate diagnosis of NAFLD (Fig. 4). Metabolic
12 abnormalities including obesity, liver enzymes and insulin resistance are also
13 closer associated with fat in the right than in the left lobe (Table 3, Fig. 5).
14 Quantification of liver fat in the right lobe may become of increasing clinical
15 interest by ultrasound techniques or in conjunction with abdominal MRI
16 performed for other indications. The study results are preliminary and need to
17 be reproduced in a larger sample size and validated.

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6

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8 analyses and interpretation of the data, drafting and critical revision of the
9 manuscript; AH: research design and conduction; YZ: statistical analyses; NL:
10 research design and conduction, critical revision of the manuscript; HYJ:
11 research design and conduction, analysis and interpretation of the data,
12 drafting and critical revision of the manuscript, study supervision.

13

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15

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Table 1. Anthropometric and biochemical characteristics of the subjects.

	Total (n=97)	Non-NAFLD (n=60)	NAFLD (n=37)
Age (year)	51±1	49±2	54±2*
Weight (kg)	83.6±1.8	77.9±2.0	92.2±2.9***
Body mass index (kg/m ²)	28.3±0.5	26.6±0.6	31.0±0.8***
Waist circumference (cm)	94.9±1.6	89.9±1.9	102.7±2.2***
Hip circumference (cm)	104.7±1.1	101.3±1.2	110.1±1.8***
Waist-to-hip-ratio	0.90±0.01	0.88±0.01	0.93±0.01**
Fasting plasma glucose (mmol/l)	5.4±0.1	5.2±0.1	5.8±0.1***
fS-Insulin (mU/l)	6.1 (3.5-12.5)	4.6 (2.7-7.1)	12.0 (6.7-14.7)***
HOMA-IR	1.42 (0.84-3.09)	0.98 (0.60-1.70)	2.87 (1.68-3.74)***
fS-Triglycerides (mmol/l)	1.10 (0.74-1.57)	0.89 (0.70-1.29)	1.36 (0.93-1.86)**
fS-HDL cholesterol (mmol/l)	1.44±0.04	1.57±0.06	1.26±0.05**
fS-LDL cholesterol (mmol/l)	3.07±0.08	3.07±0.10	3.06±0.15
S-ALT (U/L)	29 (22-44)	24 (17-32)	45 (35-66)
S-AST (U/L)	30 (25-35)	26 (23-31)	35 (30-43)
S-GGT (U/L)	24 (16-44)	19 (13-28)	36 (24-60)
NAFLD fibrosis score	-2.19±1.31	-2.36±1.17	-1.98±1.47
NAFLD fibrosis score%†	0	0	0

Abbreviations: NAFLD, non-alcoholic fatty liver disease; fS, fasting serum; HOMA-IR,

homeostasis model assessment-insulin resistance; S, serum; ALT, alanine aminotransferase; AST, aspartate aminotransferase; GGT gamma-glutamyltransferase; HDL, high density lipoprotein; LDL low density lipoprotein. Data are shown as mean \pm SE or median (25% percentile, 75% percentile). * $p < 0.05$, ** $p < 0.02$, *** $p < 0.001$ for vs. non-NAFLD. †Patients with advanced fibrosis (NAFLD fibrosis score > 0.676).

Table 2. ROC curves to diagnose the NAFLD using LFAT%^{MRI} in different ROIs

	AUROC	SE	p value for AUROC	Sensitivity %	Specificity %	PPV %	NPV %	Youden index
ROI 1	0.879	0.034	<0.001	70.3	90.0	81.3	83.0	0.603
ROI 2	0.894	0.032	<0.001	83.8	78.3	70.5	88.7	0.621
ROI 3	0.936**††	0.023	<0.001	94.6	83.3	77.8	96.2	0.779
ROI 4	0.930	0.029	<0.001	89.2	90.0	84.6	93.1	0.792
Right lobe	0.934*†§	0.025	<0.001	86.5	90.0	84.2	91.5	0.765
Left lobe	0.894‡‡	0.032	<0.001	73.0	91.7	84.4	84.6	0.647

Abbreviations: ROC, receiver operating characteristic; NAFLD, non-alcoholic fatty liver disease; ROI, region of interest; AUROC, area under the receiver operating characteristic; PPV, positive predictive value; NPV, negative predictive value.

*p<0.05, **p<0.02 for vs. ROI 1; †p<0.05, ††p<0.02 for vs. ROI 2; ‡‡p<0.02 for vs. ROI 3; §p<0.05 for vs. left lobe.

Table 3. Correlation coefficients between metabolic parameters and LFAT%^{MRS} or LFAT%^{MRI} in different ROIs

	LFAT% ^{MRS}	LFAT% ^{MRI}						
		Mean (ROI 1-4)	ROI 1	ROI 2	ROI 3	ROI 4	Left lobe	Right lobe
Body mass index	0.577***	0.441***	0.366***†	0.412***	0.469***	0.429***	0.404***	0.454***
Waist circumference	0.624***	0.503***	0.417***†	0.488***	0.545***	0.494***	0.465***	0.525***
Hip circumference	0.539***	0.400***	0.297**†	0.391***	0.446***	0.401***	0.344***†	0.428***
Waist-to-hip-ratio	0.470***	0.404***	0.386***	0.378***	0.419***	0.378***	0.400***	0.403***
Fasting plasma glucose	0.467***	0.352***	0.269**	0.297**	0.383***	0.404***	0.298**	0.396***
HBA1c	0.445***	0.477***	0.424***	0.464***	0.482***	0.491***	0.449***	0.493***
fS-insulin	0.615***	0.518***	0.468***	0.450***	0.512***	0.537***	0.477***	0.534***
HOMA-IR	0.618***	0.543***	0.457***	0.447***†	0.516***	0.543***	0.469***	0.539***

fS-triglyceride	0.547***	0.431***	0.387***	0.384***	0.429***	0.422***	0.406***	0.427***
fS-HDL cholesterol	-0.391***	-0.377***	-0.398***	-0.337***	-0.335***	-0.360***	-0.381***	-0.355***
S-AST	0.556***	0.409***	0.332***†	0.356***†	0.444***	0.438***	0.364***†	0.440***
S-ALT	0.708***	0.574***	0.504***††	0.502***††	0.594***	0.604***	0.527***†	0.604***
S-GGT	0.548***	0.446***	0.411***	0.409***	0.473***	0.440***	0.434***	0.454***
Body fat (%)	0.348***	0.249**	0.108†	0.221*	0.310**	0.286**	0.167	0.308**
Intra-abdominal fat	0.668***	0.598***	0.511***†	0.579***	0.614***	0.574***	0.554***	0.604***
Subcutaneous fat	0.521***	0.427***	0.295**†	0.406***	0.485***	0.453***	0.343**	0.479***

Abbreviations: LFAT%^{MRS}, % liver fat by proton magnetic resonance spectroscopy; LFAT%^{MRI}, % liver fat by magnetic resonance imaging; ROI, region of interest; fS, fasting serum; HBA1c, glycosylated hemoglobin A_{1c}; HDL, high density lipoprotein; S, serum; AST, aspartate aminotransferase; ALT alanine aminotransferase; GGT, gamma-glutamyltransferase. *p<0.05, **p<0.02, ***p<0.001 for r-values; Hotelling-Williams test to compare r-values for the relationship between LFAT%^{MRS} or LFAT%^{MRI} in different ROIs and given metabolic parameters, †p<0.05, ††p<0.05 for LFAT%^{MRI} in different ROIs vs. LFAT%^{MRS}.

FIGURES

Fig. 1. Positions of region of interests (ROIs) 1-4 in the liver and 3 ROIs in the spleen on T1-weighted dual-echo gradient-echo sequence. ROIs 3, 4 were in the right and ROIs 1, 2 in the left hepatic lobe. ROIs 1-4 belong to liver segment II, IV, VIII, VII, respectively.

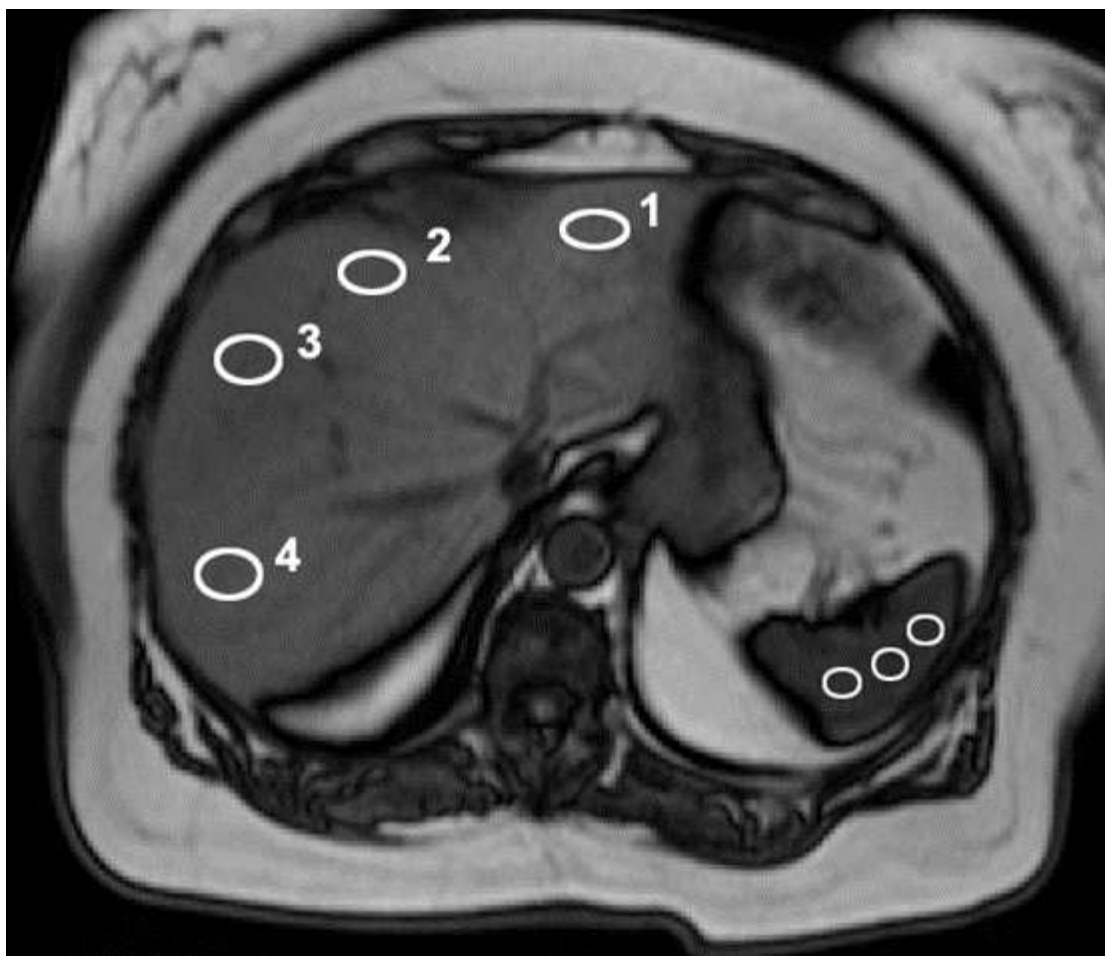


Fig. 2. Correlation between % liver fat by proton magnetic resonance spectroscopy (LFAT%^{MRS}) and liver fat fraction by magnetic resonance imaging (LFATFr^{MRI}). LFATFr^{MRI} in the right lobe was closely correlated with LFAT%^{MRS} ($r=0.837$, $p<0.001$).

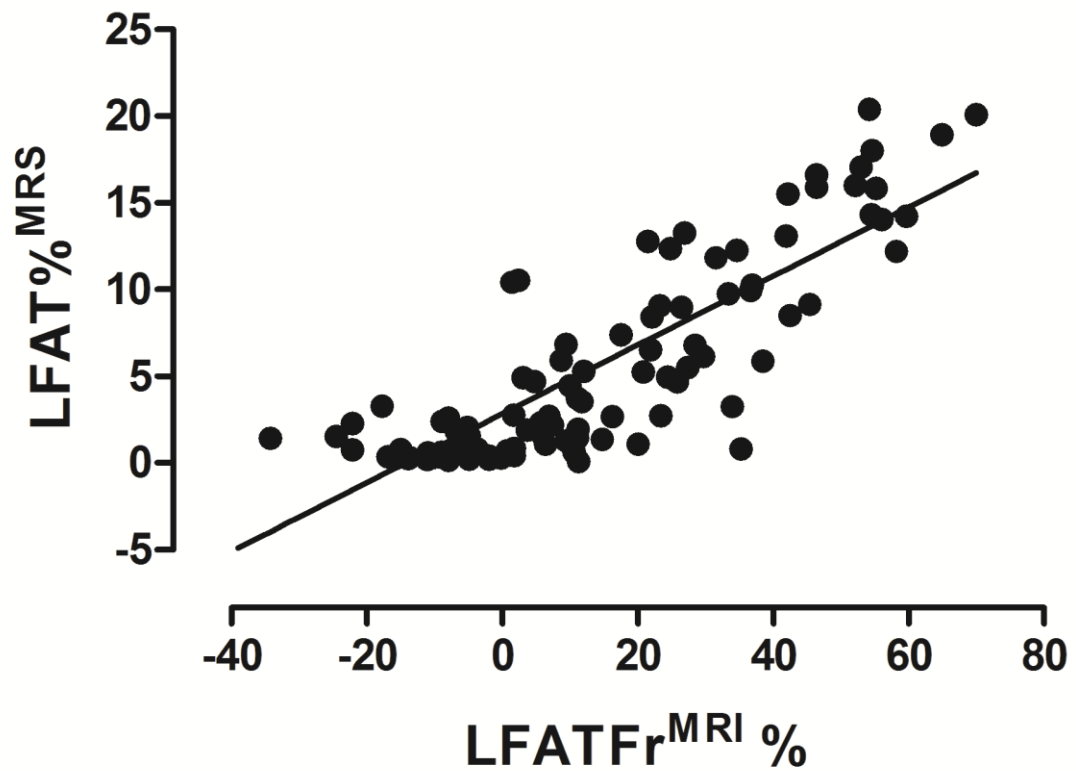


Fig. 3. % liver fat by magnetic resonance imaging (LFAT%^{MRI}) in different region of interests (ROIs). Data are shown as mean \pm SE. ** $p < 0.02$, *** $p < 0.001$.

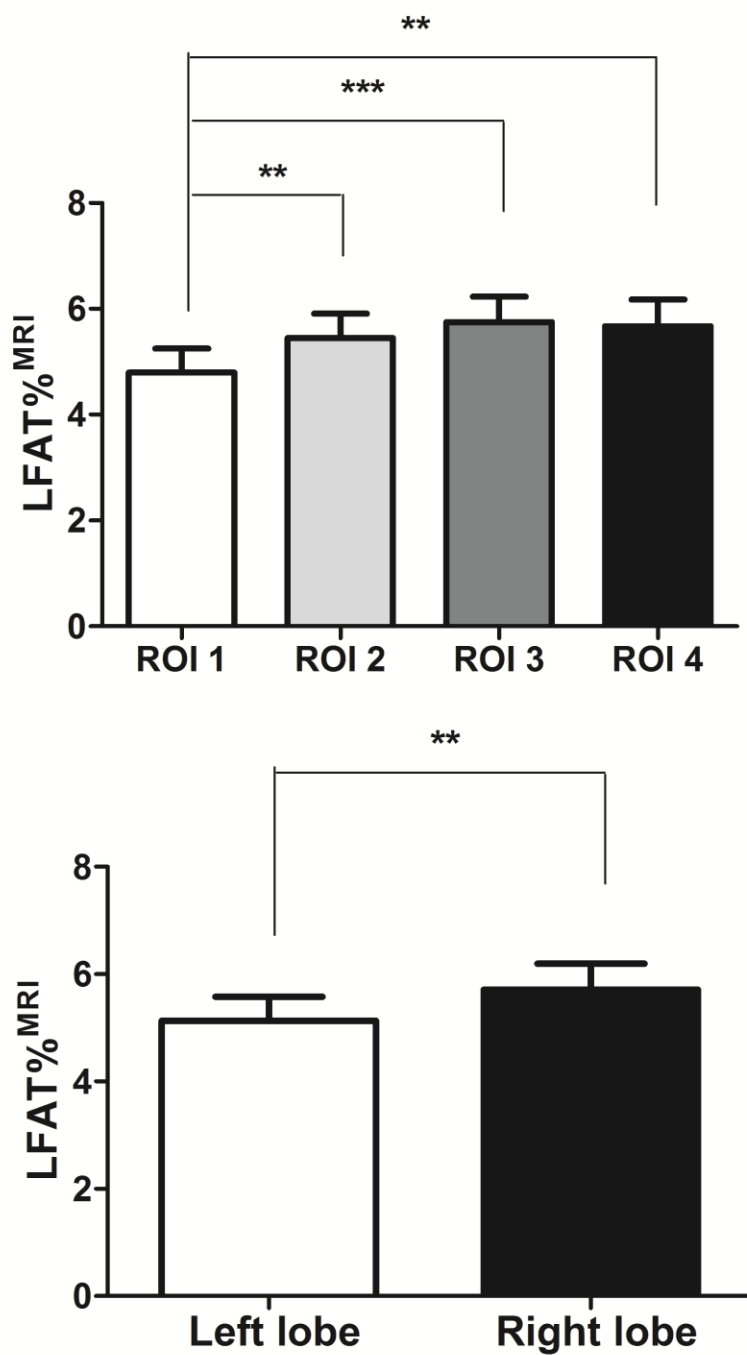


Fig. 4. Receiver operating characteristic (ROC) curves to diagnose the non-alcoholic fatty liver disease using % liver fat by magnetic resonance imaging (LFAT%^{MRI}) in the right and left lobe. Area under receiver operating characteristic (AUROC) was significantly higher using LFAT%^{MRI} in the right (AUROC = 0.934±0.025, p< 0.001, sensitivity 86.5%, specificity 90.0%) rather than in the left (AUROC = 0.894±0.032, p<0.001, sensitivity 73.0%, specificity 91.7%) lobe.

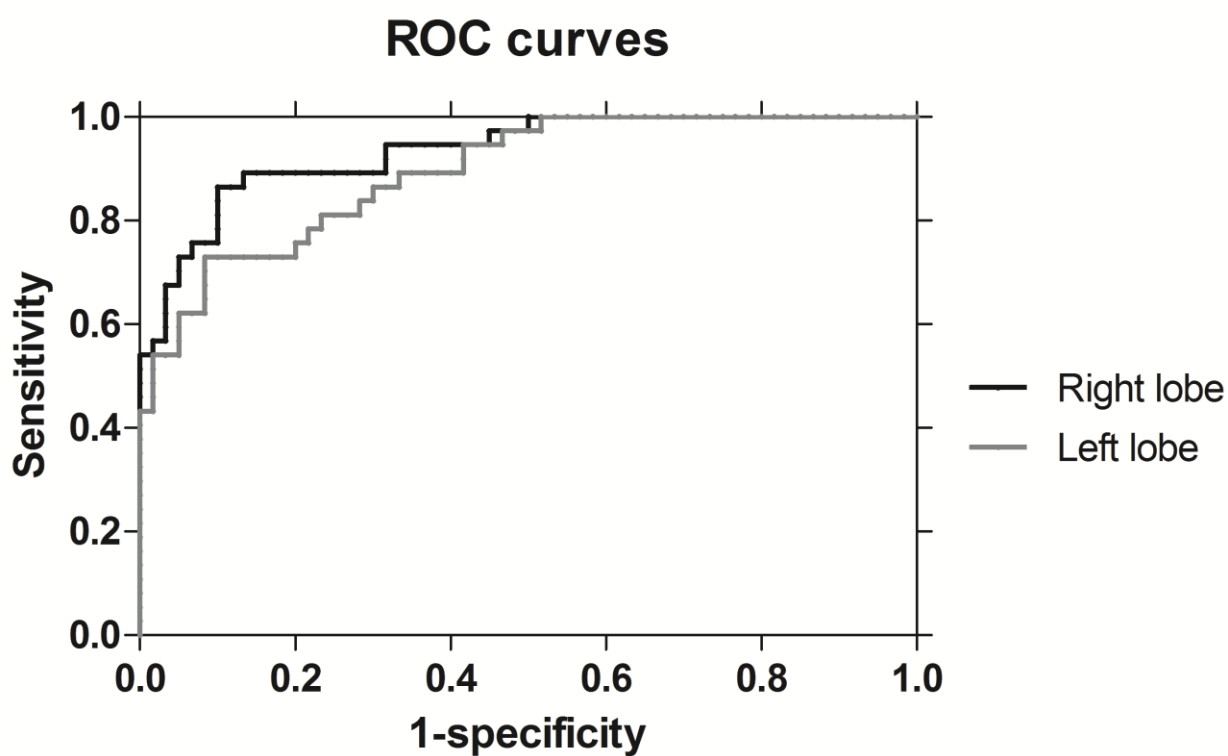


Fig. 5. The relationships between metabolic parameters and proton magnetic resonance spectroscopy (LFAT%^{MRS}) or % liver fat by magnetic resonance imaging (LFAT%^{MRI}) in different region of interests (ROIs). The Hotelling-Williams test was used to compare r-values for the relationship between LFAT%^{MRS} (white bar) or LFAT%^{MRI} in different ROIs (grey or black bars) and given metabolic parameters, *p<0.05, **p<0.02. FPG, fasting plasma glucose; HOMA-IR, Homeostasis model assessment-insulin resistance; BMI, body mass index; S-ALT, serum alanine aminotransferase.

