# Liver Fat Is Increased in Type 2 Diabetic Patients and Underestimated by Serum Alanine Aminotransferase Compared With Equally Obese Nondiabetic Subjects

ANNA KOTRONEN, MB[1,2]
LEENA JUURINEN, MD[1]
ANTTI HAKKARAINEN, BSC[3]
JUKKA WESTERBACKA, MD, PHD[1]
ANJA CORNÉR, MD[1]
ROBERT BERGHOLM, MD, PHD[1,2]
HANNELE YKI-JÄRVINEN, MD, PHD, FRCP[1]

**OBJECTIVE** — The purpose of this study was to determine whether type 2 diabetic patients have more liver fat than age-, sex-, and BMI-matched nondiabetic subjects and whether liver enzymes (serum alanine aminotransferase [S-ALT] and serum aspartate aminotransferase) are similarly related to liver fat in type 2 diabetic patients and normal subjects.

**RESEARCH DESIGN AND METHODS** — Seventy type 2 diabetic patients and 70 nondiabetic subjects matched for BMI, age, and sex were studied. Liver fat ($^1$H-magnetic resonance spectroscopy), body composition (magnetic resonance imaging), and biochemical markers of insulin resistance were measured.

**RESULTS** — The type 2 diabetic patients had, on average, 80% more liver fat and 16% more intra-abdominal fat than the nondiabetic subjects. The difference in liver fat between the two groups remained statistically significant when adjusted for intra-abdominal fat ($P < 0.05$). At any given BMI or waist circumference, the type 2 diabetic patients had more liver fat than the nondiabetic subjects. The difference in liver fat between the groups rose as a function of BMI and waist circumference. Fasting serum insulin ($r = 0.55$, $P < 0.0001$), fasting plasma glucose ($r = 0.29$, $P = 0.0006$), A1C ($r = 0.34$, $P < 0.0001$), fasting serum triglycerides ($r = 0.36$, $P < 0.0001$), and fasting serum HDL cholesterol ($r = -0.31$, $P = 0.0002$) correlated with liver fat similarly in both groups. The slopes of the relationships between S-ALT and liver fat were significantly different ($P = 0.004$). Liver fat content did not differ between the groups at low S-ALT concentrations (10–20 units/l) but was 70–200% higher in type 2 diabetic patients compared with control subjects at S-ALT concentrations of 50–200 units/l.

**CONCLUSIONS** — Type 2 diabetic patients have 80% more liver fat than age-, weight-, and sex-matched nondiabetic subjects. S-ALT underestimates liver fat in type 2 diabetic patients.

*Diabetes Care* **31:165–169, 2008**

It has been estimated that ~70–80% of type 2 diabetic patients have nonalcoholic fatty liver disease (1,2). In addition, 9 of 12 prospective epidemiological studies have shown that elevated serum liver enzyme concentrations predict type 2 diabetes independent of obesity (3). These data thus suggest that liver fat content is increased in patients with type 2 diabetes compared with equally obese nondiabetic subjects. To date, only one study has addressed this question (4). In this study, liver fat was measured qualitatively using the liver-to-spleen attenuation ratio (4). Liver fat was increased in type 2 diabetic patients compared with 10 weight-matched normal subjects (4). The small number of normal subjects, however, prevents any firm conclusions from being drawn.

Clinically, it would be helpful to have a simple measure of liver fat in patients with type 2 diabetes, as liver fat is closely correlated with insulin requirements (5,6) and may be an important parameter to consider when choosing patients for peroxisome proliferator–activated receptor-γ agonist therapy (7,8). Serum alanine aminotransferase (S-ALT) correlates with liver fat content in nondiabetic subjects (9,10), but whether S-ALT relates to liver fat similarly in type 2 diabetic patients has not been examined.

In the present study, we studied a group of 70 type 2 diabetic patients and a group of 70 nondiabetic subjects matched for age, sex, and BMI. Liver fat content was measured using proton magnetic resonance spectroscopy, and body composition was measured using magnetic resonance imaging. In addition, measures of glycemia and insulinemia, serum lipids, and serum liver enzyme concentrations were determined. The data show that in type 2 diabetic patients compared with nondiabetic subjects, liver fat is increased independent of obesity and fat distribution and that S-ALT underestimates the amount of liver fat.

## RESEARCH DESIGN AND METHODS

— A total of 70 nondiabetic subjects and 70 type 2 diabetic patients were recruited by newspaper advertisements and by contacting occupational health services in Helsinki based on the following inclusion criteria: *1*) aged 18–70 years; 2) no known acute or chronic disease based on history, physical examination, and standard laboratory tests (blood counts, serum creatinine, thyroid-stimulating hormone, electrolyte concentrations, and electrocardiogram); 3) alcohol consumption <20 g/day; and

From the [1]Division of Diabetes, University of Helsinki, Helsinki, Finland; the [2]Minerva Medical Research Institute, Helsinki, Finland; and the [3]Helsinki Medical Imaging Centre, Hospital District of Helsinki and Uusimaa, Helsinki, Finland.

Address correspondence and reprint requests to Anna Kotronen, MB, Division of Diabetes, University of Helsinki, Helsinki, Finland, P.O. Box 700, Room C418B, FIN 00029 HUCH, Helsinki, Finland. E-mail: anna.kotronen@helsinki.fi.

Received for publication 29 July 2007 and accepted in revised form 6 October 2007.

Published ahead of print at http://care.diabetesjournals.org on 12 October 2007. DOI: 10.2337/dc07-1463.

**Abbreviations:** S-ALT, serum alanine aminotransferase; S-AST, serum aspartate aminotransferase.

A table elsewhere in this issue shows conventional and Système International (SI) units and conversion factors for many substances.

**Table 1—*Subject characteristics***

| | Nondiabetic subjects | Type 2 diabetic patients | *P* value |
|---|---|---|---|
| *n* (women) | 70 (33) | 70 (33) | |
| Age (years) | 48 ± 1 | 49 ± 1 | NS |
| Body composition | | | |
| BMI (kg/m$^2$) | 31.2 ± 0.6 | 31.6 ± 0.6 | NS |
| Waist (cm) | 105 ± 1 | 107 ± 1 | NS |
| % fat | 30 ± 1 | 31 ± 1 | NS |
| Subcutaneous fat (cm$^3$) | 4,600 ± 270 | 4,400 ± 230 | NS |
| Intra-abdominal fat (cm$^3$) | 1,900 (1,300–2,500) | 2,200 (1,600–3,500) | 0.02 |
| Liver fat (%) | 7.3 (3.0–17.0) | 13.0 (6.0–20.5) | 0.005 |
| Glycemic parameters | | | |
| Fasting plasma glucose (mmol/l) | 5.6 ± 0.1 | 9.3 ± 0.3 | <0.0001 |
| A1C (%) | 5.6 ± 0.1 | 7.6 ± 0.2 | <0.0001 |
| Fasting serum insulin (mU/l) | 8.4 (6.0–12.7) | 12.0 (8.0–18.0) | 0.004 |
| Serum lipids | | | |
| Fasting serum triglycerides (mmol/l) | 1.90 ± 0.17 | 2.55 ± 0.23 | 0.007 |
| Fasting serum HDL cholesterol (mmol/l) | 1.36 ± 0.04 | 1.17 ± 0.04 | 0.0003 |
| Fasting serum LDL cholesterol (mmol/l) | 3.18 ± 0.11 | 2.75 ± 0.11 | 0.0007 |
| Fasting serum free fatty acids (μmol/l) | 650 ± 30 | 780 ± 30 | 0.003 |
| Blood pressure | | | |
| Systolic blood pressure (mmHg) | 136 ± 2 | 135 ± 2 | NS |
| Diastolic blood pressure (mmHg) | 85 ± 1 | 83 ± 1 | NS |
| Liver enzymes | | | |
| S-ALT (units/l) | 31 (23–62) | 38 (28–58) | NS |
| S-AST (units/l) | 29 (22–44) | 29 (24–48) | NS |
| AST/ALT | 0.83 (0.59–1.04) | 0.85 (0.68–1.05) | NS |

Data are means ± SEM or, for non-normally distributed data, median (25th–75th percentiles).

*4*) no evidence of hepatitis A, B, or C or of autoimmune hepatitis or clinical signs or symptoms of inborn errors of metabolism or history of use of toxins or drugs known to induce hepatitis. Exclusion criteria included proliferative retinopathy and use of antihypertensive agents that could possibly influence glucose metabolism (β-blockers and thiazides) or of thiazolidinediones. A total of 13 nondiabetic subjects and 32 type 2 diabetic patients received antihypertensive medications (ACE inhibitors or calcium channel blockers). A total of 31 type 2 diabetic patients were treated with diet alone, 17 were treated with metformin, and 22 were treated with a combination of metformin and insulin. Five nondiabetic subjects and 22 type 2 diabetic patients were receiving statins. For the insulin-treated patients, additional inclusion criteria included stable body weight and insulin dose for at least 6 months. Data for nondiabetic subjects (9) and for 46 of the type 2 diabetic patients participating in treatment studies (5,6,11) have been reported previously. For the latter patients, only baseline data are included. The study protocol was approved by the ethics committee of the Helsinki University Central Hospital, and each participant provided written informed consent.

### Liver fat content (proton spectroscopy)

Liver fat content was measured by proton magnetic resonance spectroscopy as described previously (5). This measurement has been validated against histologically determined lipid content (12) and against estimates of fatty degeneration or infiltration by X-ray computer-assisted tomography (5). All spectra were analyzed by a physicist in a blinded fashion. The reproducibility of repeated measurements of liver fat in nondiabetic subjects studied

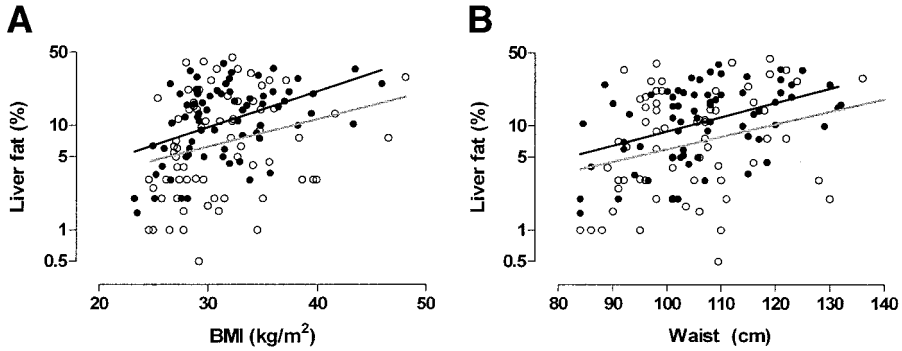


**Figure 1—***The relationships between liver fat and body composition. Liver fat content correlates with BMI* (A) *{*r *= 0.45,* P *< 0.0001 for type 2 diabetic patients (regression equation: liver fat percent [LFAT%] =* $10^{[-0.0524\pm0.27+0.034\pm0.01\times BMI]}$*);* r *= 0.26,* P *= 0.029 for nondiabetic subjects (LFAT% =* $10^{[0.003\pm0.37+0.0264\pm0.01\times BMI]}$*)}; and waist circumference* (B) *[*r *= 0.45,* P *= 0.0001 for type 2 diabetic patients (LFAT% =* $10^{[-0.42\pm0.36+0.014\pm0.003\times waist]}$*);* r *= 0.29,* P *= 0.017 for nondiabetic subjects (LFAT% =* $10^{[-0.42\pm0.52+0.012\pm0.005\times waist]}$*)]. Open circles and gray lines denote nondiabetic subjects, and filled circles and black lines denote type 2 diabetic patients.*

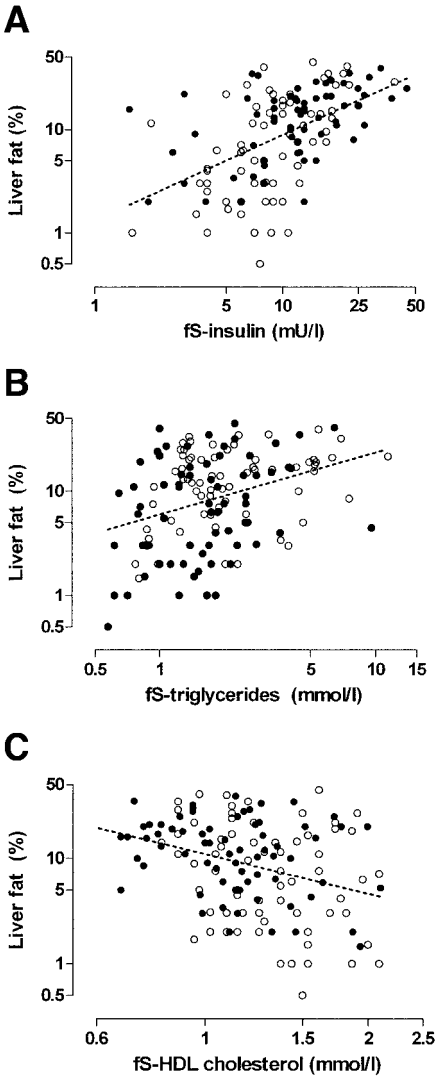


**Figure 2—***The associations between liver fat and fasting serum (fS) insulin concentrations and serum lipids. Liver fat content is related to fasting serum insulin* (A) ($r = 0.55$, $P < 0.0001$ *for both nondiabetic subjects and type 2 diabetic patients), fasting serum triglycerides* (B) ($r = 0.36$, $P < 0.0001$ *for both nondiabetic subjects and type 2 diabetic patients), and fasting serum HDL cholesterol* (C) ($r = -0.31$, $P = 0.0002$ *for both nondiabetic subjects and type 2 diabetic patients). Dashed lines denote both nondiabetic subjects and type 2 diabetic patients. Open circles denote nondiabetic subjects, and filled circles denote type 2 diabetic patients.*

on two occasions in our laboratory is 11% (13).

### Measurements of body composition

Intra-abdominal and subcutaneous fat content were determined by magnetic resonance imaging as described previously (5). Percent body fat was determined using bioelectrical impedance spectroscopy (14). Waist circumference was measured midway between the spina iliaca superior and the lower rib margin (15).

### Analytical procedures

Plasma glucose, A1C, serum free insulin, fasting serum C-peptide, fasting serum HDL cholesterol, fasting serum triglyceride, fasting serum LDL, fasting serum free fatty acids, serum aspartate aminotransferase (S-AST), and S-ALT concentrations were measured as described previously (9).

### Statistical analyses

Non-normally distributed data were used after logarithmic (base 10) transformation. If distributed normally, data are shown as means ± SEM, whereas non-normally distributed data are shown as median and the interquartile range (25th–75th percentiles). An unpaired Student's *t* test was used to compare mean values between the groups. ANCOVA was used to compare slopes and intercepts of regression lines for the nondiabetic subjects and the type 2 diabetic patients and for comparison of liver fat in the type 2 diabetic patients treated with diet, with metformin, or with insulin-metformin combination therapy. If neither the slopes nor the intercepts differed between the groups, a common regression equation was calculated. Correlation analyses were performed using Spearman's nonparametric rank correlation coefficient. ANCOVA was used to adjust for intra-abdominal fat content. Calculations were performed using GraphPad Prism (version 4.00 for Windows; GraphPad Software, San Diego, CA), and SPSS 14.0 for Windows (SPSS, Chicago, IL). $P < 0.05$ was considered statistically significant.

## RESULTS

— Nondiabetic subjects and type 2 diabetic patients were equally obese and similar with respect to age and sex (Table 1). The type 2 diabetic patients had higher fasting serum insulin, fasting serum triglyceride, and fasting serum free fatty acid concentrations and lower fasting serum HDL cholesterol concentrations that the nondiabetic subjects. The type 2 diabetic patients had ~16% more intra-abdominal and ~80% more liver fat than the nondiabetic subjects. The difference in liver fat remained significant even after adjustment for intra-abdominal fat content ($P < 0.05$). The type 2 diabetic patients treated with diet, metformin, or the combination of insulin and metformin differed slightly with respect to age (45 ± 2 [mean ± SEM], 55 ± 2, and 50 ± 2 years for diet, metformin, and insulin and metformin, respectively; $P = 0.003$ for diet versus metformin), whereas BMI (32.0 ± 0.7, 30.2 ± 1.4, and 32.2 ± 1.0 kg/m$^2$; NS), liver fat content (median 15.5% [interquartile range 7.0–21.0], 9.0% [4.2–19.5], and 14.0% [7.0–21.3]; NS), and S-ALT concentrations (42 [29–69], 31 [21–46], and 38 units/l [26–52] NS) were similar between the groups.

### Relationships between body composition and liver fat content

Liver fat content was significantly correlated with BMI and waist circumference in the type 2 diabetic patients and in the nondiabetic subjects (Fig. 1). At any given BMI or waist circumference, the type 2 diabetic patients had significantly more liver fat than the nondiabetic subjects. The type of antihyperglycemic therapy had no impact on these relationships (data not shown). At a BMI of 25 kg/m$^2$, the type 2 diabetic patients had ~40% more liver fat than the nondiabetic subjects. This difference rose gradually as a function of BMI. For example, at a BMI of 40 kg/m$^2$, the type 2 diabetic patients had 80% more liver fat than the nondiabetic subjects. A similar phenomenon was observed when waist was plotted against liver fat. For example, at waist circumferences of 85 and 140 cm, liver fat content was 50 and 90% higher in the type 2 diabetic patients than in the nondiabetic subjects, respectively. Intra-abdominal fat was similarly related to liver fat in both nondiabetic subjects and type 2 diabetic patients ($r = 0.45$, $P < 0.0001$). The volume of subcutaneous fat did not correlate with liver fat content in either group.

### Relationships between insulin, glycemia, lipids, and liver fat content

Liver fat content was similarly related to fasting serum insulin ($r = 0.55$, $P < 0.0001$), C-peptide ($r = 0.40$, $P < 0.0001$), A1C ($r = 0.34$, $P < 0.0001$), fasting plasma glucose ($r = 0.29$, $P = 0.0006$), fasting serum triglyceride ($r = 0.36$, $P < 0.0001$), and fasting serum HDL cholesterol ($r = -0.31$, $P = 0.0002$) concentrations in the nondiabetic subjects and the type 2 diabetic patients (Fig. 2).

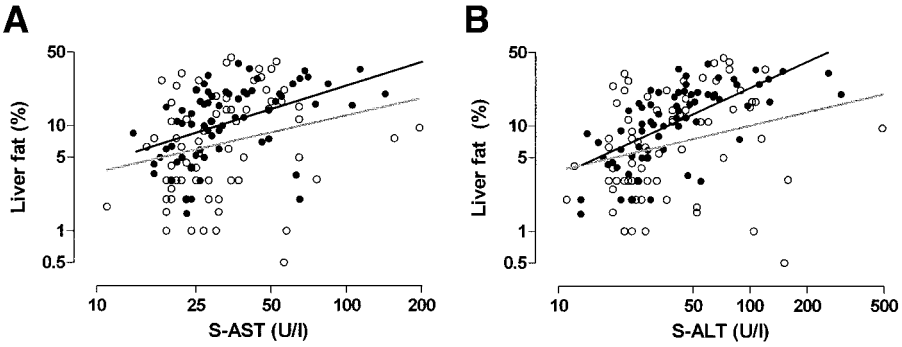


**Figure 3—***The relationships between liver fat content and liver enzymes. Liver fat content associates with S-AST* (A) *{*r *= 0.49,* P *< 0.0001 for type 2 diabetic patients (liver fat % [LFAT%] =* $10^{[-0.11\pm0.25+0.75\pm0.16\times\log(\text{S-AST})]}$*);* r *= 0.24,* r *= 0.043 for nondiabetic subjects (LFAT% =* $10^{[0.01\pm0.40+0.54\pm0.26\times\log(\text{S-AST})]}$*)}; and S-ALT* (B) *[*r *= 0.66,* P *< 0.0001 for type 2 diabetic patients (LFAT% =* $10^{[-0.30\pm0.19+0.83\pm0.12\times\log(\text{S-ALT})]}$*);* r *= 0.26,* P *= 0.027 for nondiabetic subjects (LFAT% =* $10^{[0.15\pm0.3+0.43\pm0.19\times\log(\text{S-ALT})]}$*)] concentrations. Open circles and gray lines denote nondiabetic subjects, and filled circles and black lines denote type 2 diabetic patients.*

### Relationships between liver enzymes and liver fat content

S-AST and S-ALT correlated with liver fat content in both nondiabetic subjects and type 2 diabetic patients. The slopes of the regression lines between S-ALT concentrations and liver fat content differed significantly between the nondiabetic subjects and the type 2 diabetic patients ($P = 0.004$). For any given S-ALT, the type 2 diabetic patients had more liver fat than the nondiabetic subjects (Fig. 3). Liver fat content did not differ between the groups at low S-ALT concentrations (10–20 units/l) but was 70, 125, and 200% higher in type 2 diabetic patients compared with control subjects at S-ALT concentrations of 50, 100, and 200 units/l. At normal S-AST concentrations (15–45 units/l), liver fat content was 30–70% higher in the type 2 diabetic patients than in the nondiabetic subjects. This increase rose gradually with increasing S-AST concentrations and averaged 80 and 130% at S-AST concentrations of 60 and 100 units/l. Serum liver enzyme concentrations did not differ between the subjects who were using statins compared with those who were not (S-AST median 29 units/l [interquartile range 22–40] and 29 units/l [23–46], NS; S-ALT 35 [21–51] and 33 units/l [24–64], NS). The relationship between liver enzymes and liver fat content did not differ between subjects who were and were not using statins in either group (data not shown).

**CONCLUSIONS** — In the present study, we used state-of-the-art methodology for quantification of liver fat in a large group of type 2 diabetic patients and carefully age- and sex-matched equally obese nondiabetic subjects. Liver fat content was on average 80% higher in the type 2 diabetic patients than in the nondiabetic subjects. This difference was not influenced by the type of antihyperglycemic treatment. Somewhat unexpectedly, S-ALT and S-AST were not related to liver fat similarly in the diabetic and nondiabetic subjects. Both S-ALT and S-AST underestimated liver fat content in type 2 diabetic patients. This underestimation became more pronounced at increasing concentrations of both S-ALT and S-AST.

In many previous studies, liver fat content, measured by proton magnetic resonance spectroscopy, has been found to exceed the upper limit of normal liver fat content (5.56%) (16) in type 2 diabetic patients. However, except for one study that included 10 nondiabetic control subjects (4), in these studies there was either no control group (5,6,11,17–19) or the control subjects were not matched for body weight (20,21). It has thus remained unclear whether the increase in liver fat content has just been a consequence of obesity in type 2 diabetic patients. The present study suggests that type 2 diabetic patients have more liver fat at any given BMI than nondiabetic subjects and that the difference in liver fat content between the groups increases with increasing obesity. In keeping with Kelley et al. (4), intra-abdominal fat was similarly related to liver fat in both type 2 diabetic patients and nondiabetic subjects, and the type 2 diabetic patients had more intra-abdominal fat than the nondiabetic subjects.

The higher liver fat content in the type 2 diabetic patients could contribute to diabetic dyslipidemia. The fatty liver overproduces VLDL particles in both nondiabetic subjects and type 2 diabetic patients, which, in the face of unchanged VLDL clearance, increases serum triglyceride concentrations (21). Hypertriglyceridemia in turn leads to low HDL cholesterol concentrations (22). The similar relationship between triglycerides and HDL cholesterol concentrations and liver fat content in the diabetic patients and nondiabetic subjects (Fig. 2) combined with the kinetic study (21) suggests that excess liver fat indeed contributes to diabetic dyslipidemia.

An intriguing observation in the present study was that the type 2 diabetic patients had 40–200% more liver fat at the same S-ALT and S-AST concentrations than the nondiabetic subjects (Table 2). Both enzymes thus underestimate liver fat in type 2 diabetes, and for any given amount of liver fat, S-ALT and S-AST are lower in type 2 diabetic patients than in normal subjects. Both S-ALT and S-AST increase in response to hepatocyte damage until hepatocytes are lost, and cirrhosis develops (23,24). To what extent the lower enzyme levels reflect a difference in hepatocellular damage cannot be determined from the present study.

It is important to develop tools to diagnose a fatty liver in type 2 diabetic patients because nonalcoholic steatohepatitis is more common in type 2 dia-

**Table 2—*Liver fat content at increasing S-ALT and S-AST concentrations in nondiabetic subjects and type 2 diabetic patients***

| | Liver fat content | |
|---|---|---|
| | Nondiabetic subjects | Type 2 diabetic patients |
| S-ALT | | |
| 20 units/l | 5.1 | 6.0 |
| 40 units/l | 6.9 | 10.7 |
| 60 units/l | 8.2 | 15.0 |
| 80 units/l | 9.3 | 19.0 |
| 100 units/l | 10.2 | 22.9 |
| S-AST | | |
| 20 units/l | 5.2 | 7.3 |
| 40 units/l | 7.5 | 12.3 |
| 60 units/l | 9.3 | 16.7 |
| 80 units/l | 10.9 | 20.8 |
| 100 units/l | 12.3 | 24.5 |

Data are %.

betic patients than in nondiabetic subjects (25,26) and can progress to cirrhosis and liver failure (27). The present data suggest that if assessed using S-ALT or S-AST, hepatic steatosis is underestimated in type 2 diabetic patients compared with equally obese nondiabetic subjects. Thus, there is a need to develop new serum markers of steatosis to complement S-AST and S-ALT, which have been used in the clinic for almost 50 years (28).

**Acknowledgments**— This study was supported by research grants from the Academy of Finland, the Sigrid Juselius Foundation, the Medical Society of Finland, and Novo Nordisk Foundation. This work is part of the project Hepatic and Adipose Tissue and Functions in the Metabolic Syndrome (www.hepadip.org), which is supported by the European Commission as an Integrated Project under the 6th Framework Programme (Contract LSHM-CT-2005-018734).

We gratefully acknowledge Katja Tuominen, Mia Urjansson, and Pentti Pölönen for excellent technical assistance and Anna-Maija Häkkinen, Leena Ryysy, Mirja Tiikkainen, Satu Vehkavaara, and Anneli Seppälä-Lindroos for their contributions.

**References**

1. Angulo P: Nonalcoholic fatty liver disease. *N Engl J Med* 346:1221–1231, 2002
2. Targher G, Bertolini L, Padovani R, Rodella S, Tessari R, Zenari L, Day C, Arcaro G: Prevalence of nonalcoholic fatty liver disease and its association with cardiovascular disease among type 2 diabetic patients. *Diabetes Care* 30:1212–1218, 2007
3. Kotronen A, Yki-Jarvinen H: Fatty liver: a novel component of the metabolic syndrome. *Arterioscler Thromb Vasc Biol.* In press
4. Kelley DE, McKolanis TM, Hegazi RA, Kuller LH, Kalhan SC: Fatty liver in type 2 diabetes mellitus: relation to regional adiposity, fatty acids, and insulin resistance. *Am J Physiol* 285:E906–E916, 2003
5. Ryysy L, Hakkinen AM, Goto T, Vehkavaara S, Westerbacka J, Halavaara J, Yki-Jarvinen H: Hepatic fat content and insulin action on free fatty acids and glucose metabolism rather than insulin absorption are associated with insulin requirements during insulin therapy in type 2 diabetic patients. *Diabetes* 49:749–758, 2000
6. Juurinen L, Tiikkainen M, Hakkinen AM, Hakkarainen A, Yki-Jarvinen H: Effects of insulin therapy on liver fat content and hepatic insulin sensitivity in patients with type 2 diabetes. *Am J Physiol* 292:E829–E835, 2007
7. Yki-Jarvinen H: Thiazolidinediones. *N Engl J Med* 351:1106–1118, 2004
8. Belfort R, Harrison SA, Brown K, Darland C, Finch J, Hardies J, Balas B, Gastaldelli A, Tio F, Pulcini J, Berria R, Ma JZ, Dwivedi S, Havranek R, Fincke C, DeFronzo R, Bannayan GA, Schenker S, Cusi K: A placebo-controlled trial of pioglitazone in subjects with nonalcoholic steatohepatitis. *N Engl J Med* 355:2297–2307, 2006
9. Kotronen A, Westerbacka J, Bergholm R, Pietilainen KH, Yki-Jarvinen H: Liver fat in the metabolic syndrome. *J Clin Endocrinol Metab* 92:3490–3497, 2007
10. Westerbacka J, Corner A, Tiikkainen M, Tamminen M, Vehkavaara S, Hakkinen AM, Fredriksson J, Yki-Jarvinen H: Women and men have similar amounts of liver and intra-abdominal fat, despite more subcutaneous fat in women: implications for sex differences in markers of cardiovascular risk. *Diabetologia* 47: 1360–1369, 2004
11. Tiikkainen M, Hakkinen AM, Korsheninnikova E, Nyman T, Makimattila S, Yki-Jarvinen H: Effects of rosiglitazone and metformin on liver fat content, hepatic insulin resistance, and gene expression in adipose tissue in patients with type 2 diabetes. *Diabetes* 53:2169–2176, 2004
12. Thomsen C, Becker U, Winkler K, Christoffersen P, Jensen M, Henriksen O: Quantification of liver fat using magnetic resonance spectroscopy. *Magn Reson Imaging* 12:487–495, 1994
13. Sutinen J, Hakkinen AM, Westerbacka J, Seppala-Lindroos A, Vehkavaara S, Halavaara J, Jarvinen A, Ristola M, Yki-Jarvinen H: Increased fat accumulation in the liver in HIV-infected patients with antiretroviral therapy-associated lipodystrophy. *AIDS* 16:2183–2193, 2002
14. Lukaski HC, Johnson PE, Bolonchuk WW, Lykken GI: Assessment of fat-free mass using bioelectrical impedance measurements of the human body. *Am J Clin Nutr* 41:810–817, 1985
15. Marti B, Tuomilehto J, Salomaa V, Kartovaara L, Korhonen HJ, Pietinen P: Body fat distribution in the Finnish population: environmental determinants and predictive power for cardiovascular risk factor levels. *J Epidemiol Community Health* 45: 131–137, 1991
16. Szczepaniak LS, Nurenberg P, Leonard D, Browning JD, Reingold JS, Grundy S, Hobbs HH, Dobbins RL: Magnetic resonance spectroscopy to measure hepatic triglyceride content: prevalence of hepatic steatosis in the general population. *Am J Physiol* 288:E462–E468, 2005
17. Bajaj M, Suraamornkul S, Pratipanawatr T, Hardies LJ, Pratipanawatr W, Glass L, Cersosimo E, Miyazaki Y, DeFronzo RA: Pioglitazone reduces hepatic fat content and augments splanchnic glucose uptake in patients with type 2 diabetes. *Diabetes* 52:1364–1370, 2003
18. Carey DG, Cowin GJ, Galloway GJ, Jones NP, Richards JC, Biswas N, Doddrell DM: Effect of rosiglitazone on insulin sensitivity and body composition in type 2 diabetic patients [corrected]. *Obes Res* 10:1008–1015, 2002
19. Mayerson AB, Hundal RS, Dufour S, Lebon V, Befroy D, Cline GW, Enocksson S, Inzucchi SE, Shulman GI, Petersen KF: The effects of rosiglitazone on insulin sensitivity, lipolysis, and hepatic and skeletal muscle triglyceride content in patients with type 2 diabetes. *Diabetes* 51:797–802, 2002
20. Petersen KF, Dufour S, Befroy D, Lehrke M, Hendler RE, Shulman GI: Reversal of nonalcoholic hepatic steatosis, hepatic insulin resistance, and hyperglycemia by moderate weight reduction in patients with type 2 diabetes. *Diabetes* 54: 603–608, 2005
21. Adiels M, Taskinen MR, Packard C, Caslake MJ, Soro-Paavonen A, Westerbacka J, Vehkavaara S, Hakkinen A, Olofsson SO, Yki-Jarvinen H, Boren J: Overproduction of large VLDL particles is driven by increased liver fat content in man. *Diabetologia* 49:755–765, 2006
22. Nikkila EA, Taskinen MR: Relation between H.D.L.-cholesterol levels and triglyceride metabolism. *Lancet* 2:892, 1978
23. Ladue JS, Wroblewski F: Serum glutamic pyruvic transaminase SGP-T in hepatic disease: a preliminary report. *Ann Intern Med* 45:801–811, 1956
24. Corbin IR, Ryner LN, Singh H, Minuk GY: Quantitative hepatic phosphorus-31 magnetic resonance spectroscopy in compensated and decompensated cirrhosis. *Am J Physiol* 287:G379–G384, 2004
25. Ekstedt M, Franzen LE, Mathiesen UL, Thorelius L, Holmqvist M, Bodemar G, Kechagias S: Long-term follow-up of patients with NAFLD and elevated liver enzymes. *Hepatology* 44:865–873, 2006
26. Haukeland JW, Konopski Z, Linnestad P, Azimy S, Marit Loberg E, Haaland T, Birkeland K, Bjoro K: Abnormal glucose tolerance is a predictor of steatohepatitis and fibrosis in patients with non-alcoholic fatty liver disease. *Scand J Gastroenterol* 40:1469–1477, 2005
27. Farrell GC, Larter CZ: Nonalcoholic fatty liver disease: from steatosis to cirrhosis. *Hepatology* 43:S99–S112, 2006
28. Karmen A, Wroblewski F, Ladue JS: Transaminase activity in human blood. *J Clin Invest* 34:126–131, 1955