Original papers

Fatty liver—an additional and treatable feature of the insulin resistance syndrome

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Summary

To test the hypothesis that fatty liver coexists with other metabolic abnormalities of the insulin resistance syndrome, and responds to their amelioration, we prospectively studied 48 consecutive patients with chronically elevated liver enzymes and clinical, ultrasound and histological findings consistent with fatty infiltration of the liver. Most of the patients were overweight or obese (64\%) with increased waist circumference which closely relates to visceral fat. Only 10\% of the patients had normal glucose tolerance: 44\% had diabetes mellitus, 29\% impaired glucose tolerance, and 17\% were hyperinsulinaemic. The most common dyslipidaemia found was hypertriglyceridaemia and/or low HDL-C (86\%). Dietary intervention and follow-up (median 24 months), supplemented by oral hypoglycaemic or lipid-lowering drugs as needed, resulted not only in weight loss (mean 3.7 kg), decreased fasting blood glucose (\(p<0.005\)) and improvement in serum lipid profile (\(p<0.02\) for both triglycerides or HDL-C) but also in an improvement of serum liver enzymes and ultrasonographic and histological findings consistent with fatty infiltration of the liver. Thus, fatty liver was strongly associated with many features of the insulin resistance syndrome, and follow-up revealed a high potential for reversibility and a benign course.

Introduction

General practitioners and specialists are frequently confronted with a finding of abnormal liver function tests (LFTs) in patients who are either asymptomatic or have vague abdominal discomfort. In 1980, Ludwig and colleagues described the entity of non-alcoholic steatohepatitis (NASH),\textsuperscript{1} characterized by histological findings of fatty changes with lobular hepatitis. The prevalence of NASH in patients who undergo liver biopsy is 1.2\% to 9\%.\textsuperscript{2} However, many more patients with disturbed LFTs and negative results on work-up for other diseases have histological findings of fatty infiltration without inflamma-

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aemia. This variability has led some authors to doubt the importance of the association between NASH and obesity or diabetes. These inconsistencies may be due to incomplete data, since in most of the previous studies, oral glucose tolerance tests (OGTT) and detailed lipid profiles were not included. Also, some common and confounding conditions such as hepatitis C, some degree of alcohol consumption, or increased hepatic iron stores were not properly ruled out in several earlier studies. The natural course of the disease is also unclear, ranging from benign to a progressive course leading to cirrhosis and hepatic failure. A favourable effect of weight loss on the course of the disease has been suggested, but the data are again inconsistent.

The aims of the present study were to perform a comprehensive metabolic work-up, including OGTT and a detailed anthropometric and lipid profile, in patients with ‘primary’ fatty liver, and to evaluate the clinical course, and specifically the effect on the liver, of improvement of the metabolic profile.

Methods
Patients
Patients referred to our Gastroenterology Unit because of chronically (≥ 6 months) elevated liver enzymes, underwent a thorough work-up that included a detailed history of drugs or alcohol consumption, hepatitis B and C serology, determination of caeruloplasmin, copper, α1-antitrypsin, iron and ferritin levels, and autoantibody screening. All consecutive patients who had no data suggesting any specific aetiology (including ethanol consumption < 40 g/week), and whose abdominal US examination was consistent with the diagnosis of fatty infiltration of the liver according to accepted criteria, were referred to the metabolic clinic for further evaluation.

Metabolic evaluation
All patients underwent anthropometric evaluation and at least three fasting blood glucose tests. Weight, height and circumferences at the waist and hip were measured as previously described. Body mass index (BMI; weight (kg)/height²(m)) and waist:hip ratio (WHR) were calculated from the data. All patients not diagnosed as diabetic, based on fasting hyperglycaemia (plasma glucose ≥ 7.0 mmol/l on more than one occasion), underwent OGTT. Following a 12-h fast, a standard 75-g OGTT was administered with determination of glucose and insulin at 0, 60 and 120 min. Insulin levels were determined using a monoclonal antibody (Sanofi Diagnostica Pasteur). The findings of the OGTT were interpreted as normal, impaired glucose tolerance (IGT) or diabetes (NIDDM). (IGT, plasma glucose ≥ 7.8 mmol/l at 2 h but < 11.1 mmol/l; and diabetes ≥ 11.1 mmol/l). Patients who had a normal glucose response but an elevated fasting insulin level, exceeding the 90th percentile of its distribution among patients with normal glucose tolerance, were diagnosed as hyperinsulinaemic (HI). Lipid determinations were performed twice on fasting blood samples in a Boehringer-Hitachi analyzer using Boehringer-Mannheim kits. High-density lipoprotein cholesterol (HDL-C) was determined after precipitation of low-density lipoprotein (LDL) and very-low-density lipoprotein (VLDL) with phosphotungstate. Cholesterol concentration was determined in the supernatant. Lipoprotein abnormalities were defined according to the Lipid Research Clinic data for age and gender, i.e. above the 90th percentile for total cholesterol and triglyceride levels and below the 10th percentile for HDL-C levels.

Pathology
Liver biopsies were randomly performed and reviewed by an experienced pathologist who was blinded to the clinical details. Steatosis, fibrosis and inflammation were graded according to standard histological criteria.

Intervention and follow-up
All patients were referred to a qualified dietician, and were followed by visits every 3–4 weeks. Subjects who had elevated triglyceride levels (above 2.3 mmol/l) and/or low HDL-C (below 0.9 mmol/l), were instructed to follow a ‘high-fat’ diet which provided 20% of total energy as monounsaturated fat, < 10% as saturated fat, 10% as polyunsaturated fat, and 45% as carbohydrate. The other patients were counselled to use the American Heart Association diet which limits fat intake to < 30%, saturated fat < 10%, polyunsaturated fat < 10% and monounsaturated 10–15% of total calories, with 50–60% as carbohydrates. Patients who failed to respond to at least 6 months of dietary therapy alone, received lipid-lowering drugs according to the National cholesterol Education Program guidelines. Patient compliance was monitored by independently administered questionnaires and their weight, LFT and metabolic profiles were tested.

Data analysis
Data were expressed as means ± SD. For comparison of the metabolic data before and after treatment, Student’s t-test was used, or where variables were not normally distributed, the Mann-Whitney test. Multiple regression analysis was performed (SPSS
Fatty liver and insulin resistance

Table 1  Serum liver enzymes of patients with ‘primary’ fatty liver

<table>
<thead>
<tr>
<th>Enzyme</th>
<th>Mean ± SD</th>
<th>Range</th>
<th>Normal range</th>
<th>Patients with abnormality (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>AST</td>
<td>56.6 ± 24.7</td>
<td>15–166</td>
<td>0–37</td>
<td>77</td>
</tr>
<tr>
<td>ALT</td>
<td>90.7 ± 43.9</td>
<td>36–281</td>
<td>0–41</td>
<td>92</td>
</tr>
<tr>
<td>SAP</td>
<td>108 ± 73.5</td>
<td>27–444</td>
<td>53–128</td>
<td>15</td>
</tr>
<tr>
<td>GGT</td>
<td>98 ± 97</td>
<td>16–486</td>
<td>0–48</td>
<td>52</td>
</tr>
</tbody>
</table>

The data presented are the maximal values observed at presentation, in U/l. AST, serum aspartate-aminotransferase; ALT, serum alanine-aminotransferase; GGT, serum gamma-glutamyl transpeptidase; SAP, serum alkaline phosphatase. All patients had normal prothrombin time and normal serum albumin and bilirubin levels.

Table 2  Metabolic data for 48 patients with fatty liver

<table>
<thead>
<tr>
<th></th>
<th>Mean ± SD</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total cholesterol (mmol/l)</td>
<td>6.6 ± 1.9</td>
<td>4.0–15.5</td>
</tr>
<tr>
<td>Triglyceride (mmol/l)</td>
<td>4.0 ± 3.6</td>
<td>1–22.8</td>
</tr>
<tr>
<td>HDL cholesterol (mmol/l)</td>
<td>0.96 ± 0.2</td>
<td>0.6–1.4</td>
</tr>
<tr>
<td>OGTT* results:</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fasting glucose (mmol/l)</td>
<td>5.7 ± 0.8</td>
<td>4.4–6.4</td>
</tr>
<tr>
<td>2-hours glucose (mmol/l)</td>
<td>7.6 ± 2.0</td>
<td>3.9–10.9</td>
</tr>
<tr>
<td>Fasting insulin (pmol/l)</td>
<td>105 ± 43</td>
<td>37–294</td>
</tr>
<tr>
<td>2 hours insulin (pmol/l)</td>
<td>627 ± 407</td>
<td>168–1776</td>
</tr>
</tbody>
</table>

HDL, high-density lipoprotein; LDL, low-density lipoprotein; OGTT, oral glucose tolerance test.

* For 27 non-diabetic patients.

software version 8) to determine which of the changes in metabolic and anthropometric parameters were associated with the improvement in ALT values. Independent variables were: weight, triglyceride, glucose, cholesterol and HDL cholesterol levels.

Results

Forty-eight consecutive patients who fulfilled the criteria for ‘primary’ fatty liver, were evaluated. There were 27 male and 21 female patients whose mean age was 51.7 ± 13.3 years. The mean BMI was 31.5 ± 11.5 kg/m² and 64% of the patients were obese (BMI ≥ 27.8 kg/m² in males and ≥ 27.3 kg/m² in females). Abdominal obesity (defined in males as a waist/hip ratio > 0.95 and in females as > 0.8), was found in 63% of the patients. Waist circumference was 101 ± 6.7 cm for male and female patients, respectively, and waist/hip ratio was 1 ± 0.1 for both groups. Hypertension (≥ 140/90 mmHg) was present in 58%, but drug therapy, when used, did not include medications known to be associated with fatty liver or dyslipidaemia.

The most consistent abnormality of serum liver enzymes (Table 1), detected in almost all patients, was an elevation of about two-fold in ALT, whereas increased AST levels were noted in 77% of the patients, but to a lesser degree. Elevated GGT levels were found in 52% of the patients, sometimes accompanied by an increased serum alkaline phosphatase in 15%. None of the patients had bilirubinemia, reduced serum albumin levels or prolongation of the prothrombin time. All patients had findings consistent with fatty infiltration by ultrasonography which is a highly accurate tool for the diagnosis of fatty liver. In 16 of the 48 patients the diagnosis was confirmed by a liver biopsy (Table 3). These patients were randomly selected for biopsy and their clinical and laboratory profiles were not significantly different from the remaining cohort (Table 3). The biopsies revealed marked steatosis in all specimens (16/16). Eight patients had inflammatory changes (mostly lobular) and six of those had associated hepatic fibrosis which was grade 3 in two patients only. No biopsy showed liver cirrhosis.

A detailed fasting lipid profile was obtained for all patients (Table 2). Some kind of dyslipidaemia was detected in 90% of the patients. Elevation of triglycerides with or without low HDL-C or elevated total cholesterol levels was found in 73% of the patients. Low HDL-C levels, often with elevated triglyceride or total cholesterol levels, were detected in 68% of the patients. Altogether, hypertriglyceridaemia and/or low HDL-C was found in 41/48 (86%) of cases. The relative distribution of the lipid profile abnormalities is presented in Figure 1. Fasting plasma glucose determinations or OGTT revealed diabetes in 21 cases (44%). In most of them (12/21, 57%), diagnosis of diabetes was not made prior to their presentation for the current evaluation. Of the remaining 27 patients, IGT was found in 14 patients, and a further eight patients who showed an otherwise normal glucose response to the OGTT, manifested clearly elevated fasting insulin levels (Figure 2). Altogether, 90% of the patients had some abnormality in their glucose-insulin metabolism. Only five had a normal response to an OGTT with normal fasting insulin levels (below the 90th percentile). The relative distribution of these findings is presented in Figure 2. Patients with diabetes tended to be more...
### Table 3  Characteristics of 16 patients who underwent liver biopsy

<table>
<thead>
<tr>
<th>Patient</th>
<th>BMI (kg/m²)</th>
<th>AST (U/l)</th>
<th>ALT (U/l)</th>
<th>SAP (U/l)</th>
<th>GGT (U/l)</th>
<th>Glucose tolerance</th>
<th>T.Chol. (mmol/l)</th>
<th>TG (mmol/l)</th>
<th>HDL (mmol/l)</th>
<th>Liver biopsy</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Steatosis</td>
</tr>
<tr>
<td>1</td>
<td>30</td>
<td>111</td>
<td>147</td>
<td>79</td>
<td>323</td>
<td>DM</td>
<td>6.21</td>
<td>4.78</td>
<td>0.91</td>
<td>++</td>
</tr>
<tr>
<td>2</td>
<td>26</td>
<td>27</td>
<td>40</td>
<td>300</td>
<td>107</td>
<td>DM</td>
<td>8.33</td>
<td>3.42</td>
<td>1.24</td>
<td>+++</td>
</tr>
<tr>
<td>3</td>
<td>36</td>
<td>44</td>
<td>76</td>
<td>64</td>
<td>NA</td>
<td>DM</td>
<td>8.46</td>
<td>5.01</td>
<td>1.14</td>
<td>+++</td>
</tr>
<tr>
<td>4</td>
<td>24</td>
<td>42</td>
<td>149</td>
<td>98</td>
<td>65</td>
<td>HI</td>
<td>10.40</td>
<td>4.73</td>
<td>0.88</td>
<td>+++</td>
</tr>
<tr>
<td>5</td>
<td>32</td>
<td>44</td>
<td>71</td>
<td>107</td>
<td>24</td>
<td>IGT</td>
<td>7.09</td>
<td>2.41</td>
<td>0.70</td>
<td>+++</td>
</tr>
<tr>
<td>6</td>
<td>30</td>
<td>53</td>
<td>85</td>
<td>95</td>
<td>133</td>
<td>IGT</td>
<td>4.60</td>
<td>4.45</td>
<td>0.72</td>
<td>+++</td>
</tr>
<tr>
<td>7</td>
<td>26</td>
<td>105</td>
<td>176</td>
<td>61</td>
<td>56</td>
<td>DM</td>
<td>5.22</td>
<td>1.87</td>
<td>0.70</td>
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</tr>
<tr>
<td>8</td>
<td>37</td>
<td>148</td>
<td>140</td>
<td>75</td>
<td>103</td>
<td>DM</td>
<td>9.23</td>
<td>6.88</td>
<td>0.72</td>
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</tr>
<tr>
<td>9</td>
<td>25</td>
<td>65</td>
<td>118</td>
<td>124</td>
<td>114</td>
<td>HI</td>
<td>4.78</td>
<td>4.51</td>
<td>0.72</td>
<td>+++</td>
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<tr>
<td>10</td>
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<td>203</td>
<td>317</td>
<td>DM</td>
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<td>1.50</td>
<td>0.75</td>
<td>+++</td>
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<tr>
<td>11</td>
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<td>56</td>
<td>114</td>
<td>99</td>
<td>40</td>
<td>DM</td>
<td>5.53</td>
<td>4.79</td>
<td>0.70</td>
<td>+++</td>
</tr>
<tr>
<td>12</td>
<td>28</td>
<td>33</td>
<td>37</td>
<td>62</td>
<td>76</td>
<td>IGT</td>
<td>5.69</td>
<td>4.83</td>
<td>0.98</td>
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</tr>
<tr>
<td>13</td>
<td>26</td>
<td>150</td>
<td>281</td>
<td>131</td>
<td>182</td>
<td>HI</td>
<td>6.36</td>
<td>3.06</td>
<td>1.22</td>
<td>+++</td>
</tr>
<tr>
<td>14</td>
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<td>2.78</td>
<td>1.45</td>
<td>+++</td>
</tr>
<tr>
<td>15</td>
<td>20</td>
<td>56</td>
<td>102</td>
<td>444</td>
<td>486</td>
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<td>7.03</td>
<td>4.02</td>
<td>1.03</td>
<td>+++</td>
</tr>
<tr>
<td>16</td>
<td>25</td>
<td>65</td>
<td>91</td>
<td>258</td>
<td>192</td>
<td>DM</td>
<td>4.29</td>
<td>2.69</td>
<td>0.98</td>
<td>+++</td>
</tr>
<tr>
<td>Mean</td>
<td>29</td>
<td>67</td>
<td>106</td>
<td>148</td>
<td>151</td>
<td></td>
<td>6.54</td>
<td>3.86</td>
<td>1.01</td>
<td>++</td>
</tr>
</tbody>
</table>

**SD** 5 40 64 106 130 1.76 1.40 0.21

NA, not available; IGT, impaired glucose tolerance test; HI, hyperinsulinaemia; N, normal glucose tolerance; TG, triglyceride; L, lobular; P, portal; SD, standard deviation; AST, aspartate aminotransferase; ALT, alanine aminotransferase; SAP, serum alkaline phosphatase; GGT, gamma-glutamyl transpeptidase; T.Chol., total cholesterol; HDL, high-density lipoprotein.
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Mean triglyceride levels were reduced from 4.0 ± 3.6 to 2.4 ± 0.9 mmol/l (p < 0.002). Total cholesterol was reduced from 6.6 ± 1.9 to 5.7 ± 1.1 mmol/l (p < 0.005), and HDL-C levels increased from 0.96 ± 0.2 to 1.1 ± 0.2 mmol/l (p < 0.002).

During the follow-up period, liver enzyme levels were reduced to some degree in all but two patients (46/48, 96%). In 25 patients (52%) they were reduced back to the normal range. The two patients who did not improve their LFTs were a 48-year-old obese male with diabetes (diagnosed by fasting hyperglycaemia) and dyslipidaemia with liver biopsy showing fatty infiltration (+2), and a 55-year-old obese woman with IGT and dyslipidaemia who refused a liver biopsy. Both patients did not lose weight and did not receive hypoglycaemic or lipid-lowering medications. None of the patients developed symptoms or laboratory features suggestive of liver function deterioration. The improvement in liver enzymes was not associated with initial anthropometric or metabolic findings. The six patients who had fibrosis in their liver biopsies had a similar response as the patients who had only steatosis. Regression analysis revealed that the independent variables that were associated with improvement of ALT were the changes in HDL cholesterol levels (p < 0.01) and weight (p = 0.05).

Discussion

Our patients with ‘primary’ fatty liver had clinical features compatible with the insulin resistance syndrome.

These include glucose intolerance or hyperinsulinaemia (90%), a characteristic dyslipidaemia (elevated triglycerides and/or low HDL-C (86%), hypertension (58%), and obesity (64%). The pattern of fat accumulation in our patients with fatty liver was characteristic of the insulin resistance syndrome: their waist circumference was significantly increased and values > 95 cm for men and > 80 cm for women relate very closely to visceral fat, hyperinsulinaemia and insulin resistance. The uniformity of the metabolic profile in this study can be explained by the careful metabolic evaluation. For example, in 12/21 diabetic patients (57%), diabetes had not been diagnosed prior to the metabolic evaluation in this study. It was recently suggested that patients with NASH who are euglycaemic should be followed closely for the development of diabetes, but this was based on anecdotal experience, never before on a prospective study of consecutive patients presenting with NASH. Low HDL-C and/or hypertriglyceridaemia (mostly modest), which was found in the majority of our patients, is also a typical finding in patients with diabetes or insulin resistance. Previous studies...
of patients with NASH or fatty liver have not included
detailed lipid profiles, nor was OGTT performed, and therefore the true prevalence of this dyslipidaemia may have been underestimated.

The pathogenesis of hepatic steatosis remains unclear. Several of the recognized associated conditions, such as intestinal bypass surgery, steroid treatment, and total parenteral nutrition, can cause a metabolic shift favouring lipogenesis rather than lipolysis, and lead to steatosis. Increased levels of free fatty acids are known to occur in patients with NIDDM or insulin resistance, because of the reduced ability of insulin to suppress lipolysis. Resistance to the suppressive effect of insulin on the production of VLDL in the liver is another postulated defect in insulin-resistant states. Insulin resistance is commonly associated with obesity, but can exist independently, as encountered in one of our cases, a lean female with Turner’s syndrome, a syndrome known to be associated with insulin resistance as an early defect. This patient had marked steatosis, and demonstrates that obesity is not a prerequisite. More substantial support for the role of insulin resistance in the pathogenesis of fatty liver comes from two previous studies that demonstrate a clear association between fatty liver and insulin levels in both adults and children with fatty liver. ALT levels correlated significantly with fasting insulin levels, suggesting that hyperinsulinaemia is an important contributor to the development of fatty liver, more crucial to the pathogenesis than anthropomorphic data, blood glucose or serum lipids.

Recently, a ‘two-hit’ hypothesis for the development of steatohepatitis has been proposed. The first hit produces steatosis, the second, any source of oxidative stress that increases lipid peroxidation and subsequently leads to increased liver damage. Factors which can produce a ‘second hit’ such as iron overload, alcohol, and drugs were conspicuously absent in our study group. For example, only two of our patients had (minimally) elevated iron levels, in contrast to 58% in Bacon et al. It was easier to exclude the effects of alcohol in our patients, since the prevalence of alcoholism and even social drinking in Israel remains very low. The careful exclusion of these factors may explain the remarkably benign clinical course.

None of the 48 patients developed signs suggestive of advanced liver disease, and in all but two patients, liver enzyme levels were significantly reduced during follow-up, becoming normal in half of the patients. These results are in accordance with several previous studies that showed a similarly benign clinical course, and in contrast with others who noted a progressive course leading to cirrhosis. These inconsistencies may also be due to variations in the extent and severity of the initial liver pathology. In one series, one third of the patients had significant fibrosis or cirrhosis, in contrast to another study where biopsies revealed only steatosis in almost all patients. In our group, underlying conditions were carefully excluded and the patients (according to the representative liver histology obtained in a third) had only steatosis, some inflammation (in half) and low-grade fibrosis. Previous studies in which the natural course of the disease is described were not aimed at examining the effect of therapeutic interventions.

However, there are data suggesting that diet-induced weight reduction leads to biochemical and histological improvement. As the present study was designed to evaluate the effect on the liver of improvement of the metabolic profile, each patient was referred to a dietician for individualized counselling and follow-up. The results showed that dietary intervention was highly effective: an average moderate weight reduction of 3.7 kg was associated with a marked improvement of the metabolic parameters and a decrease, often normalization, of liver enzymes. This is in contrast to previous work showing that only a weight reduction of at least 10% could correct abnormal hepatic test results. Furthermore, our observations on the five patients treated with statins whose LFTs likewise improved, imply that the use of statins in patients with dyslipidaemia and disturbed LFTs due to fatty liver is appropriate.

In conclusion, most of our 48 consecutive patients with ‘primary’ fatty liver had a characteristic clinical picture compatible with the insulin resistance syndrome including obesity with visceral fat deposition, hypertension, glucose intolerance and typical dyslipidaemia. Moreover, simple measures aimed at improving metabolic profile, such as weight reduction and the use of anti-diabetic or lipid-lowering drugs, were associated with marked reduction or normalization of liver enzyme levels in most patients, confirming the association, and suggesting that the natural course of this entity is benign and treatable.

Acknowledgements

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References

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