CALL FOR PAPERS | Insulin Resistance and the Cardiometabolic Syndrome: Adipose Tissue and Skeletal Muscle Factors

Liver fat, visceral adiposity, and sleep disturbances contribute to the development of insulin resistance and glucose intolerance in nondiabetic dialysis patients

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Departments of 1Nephrology, 2Radiology, 3Biochemistry, 4Biomathematics, School of Medicine, University of Thessaly, Larissa; 5Department of Sport Science, University of Thessaly, Trikala; 6Institute for Human Performance and Rehabilitation CE.RE.TE.TH, Trikala, Greece; and 7Institute of Biomedical Research and Technology, CE.RE.TE.TH, Larissa, Greece

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Liver fat, visceral adiposity, and sleep disturbances contribute to the development of insulin resistance and glucose intolerance in nondiabetic dialysis patients. Am J Physiol Regul Integr Comp Physiol 295: R1721–R1729, 2008. First published October 1, 2008; doi:10.1152/ajpregu.00935.2007.—Hemodialysis patients exhibit insulin resistance (IR) in target organs such as liver, muscles, and adipose tissue. The aim of this study was to identify contributors to IR and to develop a model for predicting glucose intolerance in nondiabetic hemodialysis patients. After a 2-h, 75-g oral glucose tolerance test (OGTT), 34 hemodialysis patients were divided into groups with normal (NGT) and impaired glucose tolerance (IGT). Indices of insulin sensitivity were derived from OGTT data. Measurements included liver and muscle fat infiltration and central adiposity by computed tomography scans, body composition by dual energy X-ray absorptiometer, sleep quality by full polysomnography, and functional capacity and quality of life (QoL) by a battery of exercise tests and questionnaires. Cut-off points, as well as sensitivity and specificity calculations were based on IR (insulin sensitivity index by Matsuda) using a receiver operator characteristics (ROC) curve analysis. Fifteen patients were assigned to the IGT, and 19 subjects to the NGT group. Intrahepatic fat content and visceral adiposity were significantly higher in the IGT group. IR indices strongly correlated with sleep disturbances, visceral adiposity, functional capacity, and QoL. Visceral adiposity, O2 desaturation during sleep, intrahepatic fat content, and QoL score fitted into the model for predicting glucose intolerance. A ROC curve analysis identified an intrahepatic fat content of >3.97% (sensitivity, 100; specificity, 35.7) as the best cutoff point for predicting IR. Visceral and intrahepatic fat content, as well as QoL and sleep seemed to be involved at some point in the development of glucose intolerance in hemodialysis patients. Means of reducing fat depots in the liver and splanchic area might prove promising in combating IR and cardiovascular risk in hemodialysis patients.

Insulin sensitivity index by Matsuda; homeostasis assessment model of insulin resistance; functional capacity; oral glucose insulin sensitivity; quality of life; quantitative insulin-sensitivity check index

Insulin resistance (IR) is generally described as an impaired ability of plasma insulin to adequately promote tissue glucose disposal. It is frequently, but not invariably, accompanied by hyperinsulinemia and an impaired glucose tolerance. In chronic renal failure, IR (37) and glucose intolerance (19) are very common in nondiabetic hemodialysis patients, secondary to renal disease. IR is present in almost all patients with end-stage renal disease; however, only 30–50% will finally manifest an impaired glucose tolerance (37). In addition, IR significantly contributes to the increased cardiovascular morbidity and mortality in these patients (60). Both disorders are considered to be caused by uremia (12), a notion strongly supported by the fact that renal replacement therapy and renal transplantation significantly improve uremia and ameliorate IR and glucose intolerance (31). Nevertheless, the pathogenic mechanisms underlying these abnormalities of glucose metabolism in renal disease and uremia have not been fully elucidated.

Main target organs for insulin action and, therefore, sites responsible for IR, are the liver, skeletal muscles, and adipose tissue (58). Reduced responsiveness to insulin in these tissues would lead to IR and later on to glucose intolerance (47). In individuals with normal renal function, various factors have been implicated in the development of IR including an excess of intrahepatic fat (22), skeletal muscle’s fat infiltration (17, 18), an increased central obesity (16) and visceral adiposity (4), the degree of malnutrition (62), poor physical activity levels (20), and, recently, sleep disorders (45, 49). All of the above factors or symptoms also have been shown to be important determinants of the quality of life (QoL). A strong body of evidence suggests that IR results from the accumulation of intracellular lipid metabolites in target organs as, for example, reported in studies of both humans (30) and knockout animals (42).

In the past, we have shown that hemodialysis patients suffer from muscle atrophy (54) and weakness (25, 26), exhibit increased levels of intramuscular fat content (26, 57), and have reduced functional capacity and low daily physical activity levels (24). Moreover, hemodialysis patients also suffer from sleep disorders (46, 55) and show increased levels of visceral...
adiposity and central obesity (43, 56). It seems, therefore, that factors involved in the development of IR and glucose intolerance in nonrenal failure populations, are present and might even be compounded under uremic conditions. The relationship between all of these contributing factors and their role in the development of IR and glucose intolerance in hemodialysis patients has not yet been clearly ascertained. Notably, there are no data available assessing intrahepatic fat content in relation to IR and glucose intolerance in hemodialysis patients.

The aims of the present study were 1) to measure intrahepatic, intramuscular, visceral, and total body fat content to examine the possible role of the adipose tissue in the development of IR and glucose intolerance in a group of non-diabetic hemodialysis patients; 2) to evaluate sleep disorders, nutritional status, muscle size, and physical activity levels to investigate how parameters related to the QoL may predispose to IR; 3) to assess the differences in the above parameters between the hemodialysis patients with normal and the hemodialysis patients with disturbed glucose tolerance; and 4) to develop a model for predicting glucose intolerance in stable non-diabetic hemodialysis patients.

METHODS

Subjects. Hemodialysis patients were recruited from the dialysis unit at the University General Hospital of Larissa, Greece. Entry criteria included being on chronic hemodialysis for 6 mo or more with dialysis delivery (KT/V) > 1.1. Patients were excluded if they had diabetes mellitus as well as reasons for being in a catabolic state, such as hyperthyroidism, active vasculitis, malignancies, HIV and opportunistic infections, or inflammations that required intravenous antibiotics within 3 mo prior to enrollment, since those conditions are known to affect muscle size, body composition, and functionality. The causes of renal failure in the participants were glomerulonephritis (10), polycystic kidneys (9), renovascular disease (4), hypertension (5), and unknown reasons (6). None of our patients were alcoholic or consuming more than 5 units of alcohol per week. All patients gave informed consent prior to study participation. The study was approved by the Ethics Committee on Human Research at the University Hospital of Larissa, Greece.

Study design. Patients participated in an all-night in-hospital polysomnographic session for the assessment of sleep quality and quantity, followed the next morning by a computed tomography (CT) scan, oral glucose tolerance test (OGTT), body composition assessment [dual energy X-ray absorptiometry (DEXA)], and a series of functional tests, before their dialysis session. Routine monthly laboratory results were also recorded. A single-pool Kt/V was calculated from pre- and postdialysis blood urea nitrogen measurements using the Daugirdas II equation (10).

IR. Glucose tolerance was assessed using a 75-g, 2-h OGTT (2) by measuring the area under the glucose curve (AUCGlucose) using a trapezoidal integration. Insulin sensitivity indices were calculated using the insulin sensitivity index by Matsuda et al. (IS(Matsuda)) (39), the oral glucose insulin sensitivity (OGIS) index (38), the quantitative insulin-sensitivity check index (QUICKI) (29) as well as the homeostasis assessment model of IR (HOMA-IR) (14) (Table 1). Glycosylated hemoglobin (Hba1c) was also measured using in hospital routine clinical laboratory procedures.

Body composition. Whole body, regional fat, and lean body mass were measured by a DEXA system (Lunar model DPX Madison, WI). Post hoc regional analysis was performed as previously described (36). The waist-to-hip ratio (WHR) was calculated as waist circumference at midway between the iliac crest and the lowest margins of the ribs over the hip circumference at the maximum circumference of buttocks. An average of three readings of each measurement was taken for the calculation of the WHR. Visceral and subcutaneous adipose tissue of the abdominal areas at the level of L4-L5 lumbar area, was assessed by analyzing images collected by a CT system (model Tomoscan SR5000; Philips) (35).

Fat infiltration. Intrahepatic fat accumulation was assessed by the same CT system at the level of T12-L1 vertebrae using the ratio of mean liver-to-spleen attenuation (11). Muscle cross-sectional area and intramuscular fat (extramyocellular lipids) accumulation were assessed by images collected using the same CT system by collecting six images 2-cm apart at the larger girth of the right thigh for each patient as previously described (32).

Image analysis of the CT slices was performed using a customized software program written in Interactive Data Language (Research Systems, Boulder, CO). This software, based on variations in signal intensity, allowed for the quantification of fat (visceral and subcutaneous adipose tissue), muscle, liver, and miscellaneous components (connective tissue, fascia, intramuscular or intrahepatic fat) expressing the data as area (cm²) and percentage of tissue composition (%fat, %muscle, or %liver, and %miscellaneous) (26, 56).

Functional capacity. Functional capacity was assessed by a battery of tests including the North Staffordshire Royal Infirmary (NSRI) walk test, the time to perform five sit-to-stand cycles, the number of sit-to-stand cycles achieved in 60 s, and the time to walk a distance of 6.06 meters (20 ft) at normal (slow walk) and fast pace. These tests are described in detail elsewhere (56).

Nutritional status. The nutritional status of the patients was assessed using the seven-point subjective global assessment scale. The validity and reliability of this method of nutritional assessment has been previously reported in dialysis patients (15).

Polysomnography. Polysomnograms (Somnoscreen; Somnomedics, Randersacker, Germany) were collected overnight at the Sleep Disorders Laboratory of University General Hospital of Larissa, as previously reported (9). Sleep stages and arousals were determined using standard criteria (59a). Obstructive apnea was defined as the presence of chest/abdominal wall motion in the absence of airflow for at least 10 s in duration. Hypopnea was defined as: 1) a reduction in airflow signal amplitude of at least 50% compared with baseline, 2) the presence of chest/abdominal wall motion, and 3) oxygen desaturation of hemoglobin by 4% or with an arousal. The respiratory disturbance index was equal to the sum of the number of hyponeas and obstructive and mixed apneas (apneas with both central and obstructive components) per hour of sleep. The arousal index was defined as the total number of arousals in sleep, divided by the total sleep time. The oxygen desaturation index was defined as the average of desaturation events per hours of recording. Desaturation events were defined as O₂ saturation < 90% for > 15 s. Sleep stages and arousals were determined using standard criteria (59a).

Questionnaires. The questionnaire materials were completed with the interview method by experienced personnel (G. K. Sakkas, C. D. Giannaki, C. Karatzafieri). QoL was assessed by using the Short Form Questionnaire 36 (SF36 QoL) adjusted and validated in dialysis patients (40). The Zung Self-Rating Depression Scale was used to assess levels of depression. This questionnaire is very sensitive to the early signs of depression and has been previously used in hemodialysis patients (41).

Biochemical assessment. Biochemical examination was performed at the ClinLab of the University Hospital under routine procedures (including glucose, insulin, C-reactive protein, parathyroid hormone, vitamin D, blood urea nitrogen). Monthly laboratory results were recorded for all hemodialysis patients participating in our study.

Statistical analysis. The Spearman Rank Correlation Test was used to assess the relationship between the IS(Matsuda) and the examined variables. Moreover, to compare the two groups (normal glucose tolerant vs. impaired glucose tolerant) for differences in the fat accumulation (visceral, intrahepatic, intramuscular, total body), body composition, sleep quality and quantity, as well as functional capacity and QoL, unpaired t-tests for continuous normally distributed vari-
TABLE 1. Patient characteristics presented as pool data and divided in NGT and IGT groups

<table>
<thead>
<tr>
<th>Variables</th>
<th>Pool Data</th>
<th>Spearman Rank Correlation to ISI Matsuda</th>
<th>NGT</th>
<th>IGT</th>
<th>P Values*</th>
</tr>
</thead>
<tbody>
<tr>
<td>N</td>
<td>34</td>
<td>N/A</td>
<td>19</td>
<td>15</td>
<td>0.43</td>
</tr>
<tr>
<td>Female/Male</td>
<td>11/23</td>
<td>N/A</td>
<td>7/12</td>
<td>4/11</td>
<td>0.05</td>
</tr>
<tr>
<td>Age, yr.*</td>
<td>52.4±16.1</td>
<td>r = -0.514, P = 0.01</td>
<td>47.4±18.1</td>
<td>58.4±10.7</td>
<td>0.05</td>
</tr>
<tr>
<td>Range</td>
<td>18–74</td>
<td></td>
<td>18–71</td>
<td>40–74</td>
<td></td>
</tr>
<tr>
<td>BMI, kg/m²</td>
<td>26.1±4.1</td>
<td>r = -0.527, P = 0.01</td>
<td>25.8±3.9</td>
<td>26.4±3.9</td>
<td>0.66</td>
</tr>
<tr>
<td>KTV*</td>
<td>1.3±0.5</td>
<td>r = 0.13, P = 0.45</td>
<td>1.1±0.3</td>
<td>1.4±0.5</td>
<td>0.05</td>
</tr>
<tr>
<td>Years in Dialysis</td>
<td>2.5±1.5</td>
<td>r = -0.006, P = 0.70</td>
<td>2.2±1.2</td>
<td>2.9±1.8</td>
<td>0.20</td>
</tr>
<tr>
<td>SGA, A/B/C</td>
<td>29/50</td>
<td>r = 0.175, P = 0.48</td>
<td>13/60</td>
<td>14/10</td>
<td>0.45*</td>
</tr>
</tbody>
</table>

Glucose disposal assessment

- Fasting plasma glucose, mg/dl: 91.9±12.7; P = 0.01
- Plasma glucose at 120 min, mg/dl: 140.9±48.0; P = 0.01
- Fasting plasma insulin, μU/ml: 9.3±9.5; P = 0.01
- HbA1c, %: 5.1±0.5; P = 0.12
- AUCGlucose (0-120), mg·dl⁻¹·min⁻¹: 146.3±31.7; P = 0.01
- ISI Matsuda Index: 9.2±7.0; N/A
- HOMA-IR: 2.2±2.3; N/A
- QUICKI: 0.35±0.0; N/A
- OGIS, ml·min·m²: 431±64; P = 0.01
- Hypertension prevalence: 7 (20%); N/A
- Cardiovascular disease: 4 (12%); N/A
- Ankle brachial index: 1.2±0.2; N/A
- Body Composition (DEXA): LBM/height, kg/m: 27.8±4.5; r = 0.135, P = 0.68
- Body fat, %: 26.1±12.0; r = -0.212, P = 0.41
- Trunk fat/height, kg/m: 5.6±3.0; r = -0.492, P = 0.05
- WHR: 1.0±0.1; r = -0.58, P = 0.01

Data are mean ± SD. NGT, the normal glucose tolerance group; IGT, the impaired glucose tolerance group; BMI, body mass index; KTV, dialysis efficiency; ISI, insulin sensitivity index; SGA, subjective global assessment; HbA1c, glycosylated hemoglobin; AUC, area under the OGTT curve; G0/I0, fasting glucose-to-fasting insulin ratio; HOMA-IR, homeostasis model assessment for insulin resistance; QUICKI, quantitative insulin-sensitivity check index; OGIS, oral glucose insulin sensitivity; GITT, oral glucose tolerance test; LBM, lean body mass; Trunk fat, fat in kg calculated from DEXA analysis; WHR, waist-to-hip ratio. *Age and KTV were included as a covariate in the analysis. The Spearman Rank correlation test was used to assess the relation between the ISI Matsuda index and the examined variables. An unpaired t-test was used for assessing the differences between the two groups.
cholecalciferol as standard care up to three times weekly (∼3 µg/wk).

None of the above parameters were correlated significantly with the ISIMatsuda index.

Body composition. Parameters of body composition and post hoc regional analysis of DEXA images did not differ statistically between the NGT and IGT groups. Trunk fat assessed by DEXA and WHR correlated significantly with ISIMatsuda index (Table 1).

Visceral adiposity assessed by CT was found to be increased in IGT patients compared with NGT patients, while the size of thigh muscles did not differ between the two groups (Table 2).

Fat infiltration. Although content of the intrahepatic lipids was 10-fold increased in the IGT group compared with the NGT group, the thigh intramuscular lipid content (known also as extramyocellular lipids) did not differ between the two groups. The ISIMatsuda index did not correlate significantly with extramyocellular lipids, while there was a week correlation with intrahepatic lipids content (Table 2).

Functional capacity and QoL. In the pool data, the ISIMatsuda correlated significantly with the NSRI functional test time (Table 2). Moreover, the ISIMatsuda correlated significantly with the depression score and the QoL Total and Vitality scores (Table 2). Functional capacity, assessed by a battery of five tests did not differ between the two patient groups. The QoL score was reduced by 18% in the IGT patients compared with their NGT counterparts (Table 2).

Polysonmography. The majority of the parameters examined during the sleep assessment did not differ between the two groups (Table 2). However, the %oxygen saturation during sleep was (P = 0.04) lower in the IGT group compared with the NGT group. The ISIMatsuda correlated significantly with %oxygen saturation in the pool data set (Table 2).

Probability model and ROC analysis. The probability model for the prediction of the glucose intolerance of the hemodialysis patients identified four parameters to fit the model significantly: %oxygen saturation during sleep, visceral adipose tissue over total adipose tissue, QoL and %intrahepatic lipids (Fig. 1; Table 3). The area under the ROC curve of the intrahepatic fat content and the AUCGlucose 0–120 value were significantly larger than the area under the diagonal reference line (Fig. 2). The cutoff points for the diagnosis of IR were > 3.97% for intrahepatic fat (sensitivity 100, specificity 35.7) and > 181.37 mg·dl⁻¹·min⁻¹ for AUCGlucose 0–120 (sensitivity 100, specificity 66.7) as shown in Table 4.

DISCUSSION

Our results demonstrated that the excess of intrahepatic fat deposition in hemodialysis patients is an important contributor to the development of IR and glucose intolerance. Forty-four percent of the nonidiabetic hemodialysis patients examined in this study were found to have abnormal glucose tolerance. Four parameters, the increased fat deposition in the liver, the

Table 2. Polysomnographic evidence, CT analysis, functional capacity, and quality of life parameters presented as pool data (both groups) and divided as NGT and IGT groups

<table>
<thead>
<tr>
<th>Variables</th>
<th>Pool Data</th>
<th>Spearman Rank Correlation With ISI Matsuuda</th>
<th>NGT</th>
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<th>P Values*</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Polysonmography</strong></td>
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<tr>
<td>Sleep duration, min</td>
<td>303.4±78.2</td>
<td>r = 0.274, P = 0.15</td>
<td>315.2±86.8</td>
<td>289.1±66.5</td>
<td>0.40</td>
</tr>
<tr>
<td>Sleep efficiency, %</td>
<td>69.1±18.8</td>
<td>r = 0.203, P = 0.28</td>
<td>72.1±20.5</td>
<td>65.9±16.7</td>
<td>0.19</td>
</tr>
<tr>
<td>RDI</td>
<td>11.5±18.3</td>
<td>r = -0.362, P = 0.04</td>
<td>15.2±23.1</td>
<td>7.1±8.3</td>
<td>0.13</td>
</tr>
<tr>
<td>Arousal index, events/h</td>
<td>31.7±22.7</td>
<td>r = -0.051, P = 0.77</td>
<td>30.1±23.8</td>
<td>33.6±21.9</td>
<td>0.85</td>
</tr>
<tr>
<td>%O2 saturation during sleep</td>
<td>94.1±2.1</td>
<td>r = 0.551, P = 0.01</td>
<td>94.8±1.9</td>
<td>93.2±2.1</td>
<td>0.04</td>
</tr>
<tr>
<td>ODI, events/h</td>
<td>19.9±27.2</td>
<td>r = -0.437, P = 0.02</td>
<td>21.9±33.7</td>
<td>17.6±18.3</td>
<td>0.58</td>
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<tr>
<td><strong>CT Analysis</strong></td>
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<tr>
<td>Abdominal CSA, cm²</td>
<td>629.4±152.8</td>
<td>r = -0.666, P = 0.01</td>
<td>620.7±170.5</td>
<td>640.4±150.4</td>
<td>0.67</td>
</tr>
<tr>
<td>VAT/TAT</td>
<td>0.4±0.1</td>
<td>r = -0.429, P = 0.02</td>
<td>0.3±0.1</td>
<td>0.4±0.1</td>
<td>0.01</td>
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<tr>
<td>VAT/SAT</td>
<td>0.6±0.3</td>
<td>r = -0.373, P = 0.04</td>
<td>0.5±0.2</td>
<td>0.8±0.4</td>
<td>0.01</td>
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<tr>
<td>Thigh Muscle Fat Deposition (%)</td>
<td>17.3±6.8</td>
<td>r = 0.031, P = 0.88</td>
<td>16.9±6.3</td>
<td>17.6±7.7</td>
<td>0.98</td>
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<tr>
<td>Thigh muscle CSA, cm²</td>
<td>99.6±24.4</td>
<td>r = 0.071, P = 0.68</td>
<td>99.8±25.8</td>
<td>99.2±23.7</td>
<td>0.95</td>
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<tr>
<td>Intrahepatic fat deposition, %</td>
<td>3.2±5.8</td>
<td>r = -0.350, P = 0.06</td>
<td>0.6±0.9</td>
<td>6.7±7.8</td>
<td>0.01</td>
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<tr>
<td><strong>Functional Tests</strong></td>
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<tr>
<td>Normal walk, s</td>
<td>6.2±1.6</td>
<td>r = -0.167, P = 0.35</td>
<td>6.4±1.7</td>
<td>6.0±1.6</td>
<td>0.52</td>
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<tr>
<td>Fast walk, s</td>
<td>4.2±1.4</td>
<td>r = -0.194, P = 0.28</td>
<td>3.9±0.9</td>
<td>4.4±1.3</td>
<td>0.23</td>
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<tr>
<td>Sit to stand 5 reps, s</td>
<td>10.1±3.7</td>
<td>r = -0.436, P = 0.01</td>
<td>9.7±3.1</td>
<td>10.6±3.4</td>
<td>0.37</td>
</tr>
<tr>
<td>Sit to stand 60+ reps</td>
<td>29.9±9.2</td>
<td>r = 0.292, P = 0.12</td>
<td>29.2±10.1</td>
<td>29.7±8.2</td>
<td>0.95</td>
</tr>
<tr>
<td>NSRI walk test, s</td>
<td>81.7±34.1</td>
<td>r = -0.522, P = 0.01</td>
<td>79.9±36.6</td>
<td>84.1±31.5</td>
<td>0.73</td>
</tr>
<tr>
<td><strong>Questionnaires</strong></td>
<td></td>
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<tr>
<td>Zung depression scale</td>
<td>36.9±7.1</td>
<td>r = -0.500, P = 0.01</td>
<td>35.2±7.4</td>
<td>38.7±6.5</td>
<td>0.21</td>
</tr>
<tr>
<td>SF36 total score</td>
<td>71.6±17.8</td>
<td>r = 0.317, P = 0.08</td>
<td>78.1±12.9</td>
<td>63.7±20.2</td>
<td>0.01</td>
</tr>
<tr>
<td>SF36 physical function</td>
<td>69.8±30.9</td>
<td>r = 0.430, P = 0.01</td>
<td>68.1±34.5</td>
<td>72.0±26.9</td>
<td>0.72</td>
</tr>
<tr>
<td>SF36 vitality</td>
<td>71.9±18.2</td>
<td>r = 0.432, P = 0.01</td>
<td>79.7±13.8</td>
<td>62.6±19.8</td>
<td>0.01</td>
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<tr>
<td>SF36 mental health</td>
<td>71.9±19.1</td>
<td>r = 0.237, P = 0.19</td>
<td>78.7±13.9</td>
<td>63.8±21.7</td>
<td>0.03</td>
</tr>
</tbody>
</table>

Data are mean ± SD. Polysomnographic evidence, CT analysis, functional capacity and quality of life parameters presented as pool data (both groups) and divided as NGT and IGT groups. RDI, Respiratory disturbance index (the sum of the number of hypopneas, obstructive and mixed apneas (apneas with both central and obstructive components), per hour of sleep); Arousal index, defined as the total number of arousals in sleep, divided by the total sleep time; ODI, oxygen desaturation index (the total number of desaturation events per hour of recording); CSA, cross-sectional area; VAT, visceral adipose tissue; TAT, total adipose tissue; SAT, subcutaneous adipose tissue; NSRI, North Staffordshire Royal Infirmary; SF36, sort form 36 quality of life questioner. An unpaired t-test was used for assessing the differences between the two groups. Spearman Rank correlation test was used to assess the relation between ISI matsuda index and the examined variables.
creased visceral adiposity, the low oxygen blood saturation during sleep, and the low overall QoL appeared to be involved in the development of glucose intolerance in hemodialysis patients. The level of intrahepatic fat content appeared to be a sensitive indicator for predicting IR in nondiabetic stable hemodialysis patients.

In this study, we have selected nine parameters that, according to the literature, significantly contribute to the development of IR and glucose intolerance in nonrenal failure populations, and we tested them in a model as predictors for glucose intolerance in our hemodialysis population. Intrahepatic fat content, visceral adiposity, oxygen blood saturation during sleep, and QoL fit statistically in the probability model (Fig. 1). Moreover, the intrahepatic fat content (apart from the obvious effect on the AUCGlucose) came out as a very sensitive index in establishing a cutoff threshold in the subsequent ROC analysis (Table 4).

The suggested prediction model can be useful at two levels: first, it could help to identify hemodialysis patients at high risk for developing type 2 diabetes mellitus (Fig. 1), and, second, it could help to improve the design of the potential rehabilitation approaches for reversing or reducing the glucose intolerance in hemodialysis patients.

The 3.97% of intrahepatic fat content cutoff threshold found in this study, could be helpful for the identification of IR state in hemodialysis patients. According to this threshold, 30% of our hemodialysis patients are insulin resistant, and the majority of them are also glucose intolerant (in agreement with the AUCGlucose delineation of IR). Interestingly, the intrahepatic fat threshold for IR development determined in this study is very close to the intrahepatic fat threshold established for the diagnosis of nonalcoholic fatty liver disease (cutoff > 5.0% intrahepatic fat) in obese patients with normal renal function (8, 50). This is the first study in hemodialysis patients examining their intrahepatic fat content. Notably, 18% of the patients examined were found to have intrahepatic lipid content above the 5% threshold for the diagnosis of nonalcoholic fatty liver disease. Unfortunately, our patients were not further examined by an independent specialist to establish a nonalcoholic fatty liver disease diagnosis.

Although, the sensitivity for all the parameters examined by the ROC analysis was 100%, the intrahepatic lipids threshold showed lower specificity (35.7%) compared with AUC for the glucose values (66.7%). Both analyses, however, showed very similar area under the ROC curve (0.97 and 0.98 for intrahepatic fat and OGTT, respectively) making them equally significant for the assessment of IR.

In this study, intrahepatic fat, visceral adiposity, sleep, and QoL appeared to be good predictors of the development of glucose intolerance. It is conceivable, therefore, to hypothesize that by targeting these important contributors, we might be able to improve insulin sensitivity and glucose intolerance in hemodialysis patients. Studies in patients with no kidney disease have shown that weight reduction by diet and/or exercise

Table 3. Spearman’s correlation coefficients between the four parameters used in the probability models

<table>
<thead>
<tr>
<th>Variables</th>
<th>VAT/TAT</th>
<th>SF36 QoL</th>
<th>Intrahepatic Fat</th>
<th>% O₂ Saturation</th>
</tr>
</thead>
<tbody>
<tr>
<td>VAT/TAT</td>
<td>1.000</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>SF36 QoL</td>
<td>-0.05, P = 0.77</td>
<td>1.000</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Intrahepatic fat</td>
<td>-0.06, P = 0.71</td>
<td>-0.16, P = 0.07</td>
<td>1.000</td>
<td></td>
</tr>
<tr>
<td>% O₂ saturation</td>
<td>-0.33, P = 0.08</td>
<td>-0.11, P = 0.55</td>
<td>-0.22, P = 0.23</td>
<td>1.000</td>
</tr>
</tbody>
</table>

QoL, quality of life.
ISI-Matsuda and OGIS indices were sensitive enough to detect glucose intolerance (51, 61). Very often, exercise training has been used as a means of improving glucose tolerance and insulin sensitivity (6, 48). It is well known that hemodialysis patients suffer from IR and glucose intolerance (51, 61). Very often, exercise training has been used as a means of improving glucose tolerance and insulin sensitivity (6, 48). It remains to be seen whether interventions, such as exercise training and diet alterations, could reduce visceral and intrahepatic fat content in hemodialysis patients and thus improve IR and prevent or defer the development of glucose intolerance.

It is well known that hemodialysis patients suffer from IR that seems to be directly related to their uremic state (33). Various indices of IR have been used for assessing the insulin action in hemodialysis patients and for this reason we have included in our study the five most accepted indices (ISI-Matsuda, OGIS, HOMA-IR, QUICKI, G0/I0). Our data showed that only ISI-Matsuda and OGIS indices were sensitive enough to detect differences between the NGT and IGT groups. In addition, although the HbA1c is not considered to be the most accurate index for long-term glycemic control in hemodialysis patients, due to the shortened red blood cell survival in uremia (27), it was found to be statistically different between the two groups, while being within the accepted normal range (4.5–6.3%). Bearing in mind the small sample size of our study, it seems that even when fasting glucose and insulin, as well as glycosylated hemoglobin, are within the normal range, implying good glycemic control, a significant number of hemodialysis patients remain undiagnosed for IR, having thus a significant risk for developing cardiovascular events (60) and/or type 2 diabetes in the coming years (21).

Strong correlations were found between ISI-Matsuda (and OGIS) and indices of central adiposity, sleep quality, functional capacity, and QoL, implying that disturbances in insulin action are closely related to those parameters in this patient population. However, although diet, muscle mass, and intramuscular fat content, are recognized as important contributors to IR development in nonrenal populations, in our study, none of those correlated significantly with any of the insulin sensitivity indices nor did they differ significantly between the two groups. In accordance with the notion that physical activity promotes euglycemia, in our study, the NSRI test score was correlated significantly with ISI-Matsuda (6, 48).

We have divided our patient population into two groups according to their OGTT postload glucose levels. Although, by definition, none of our patients suffered from diabetic nephropathy or diabetes, there were significant differences between the two groups, closely related to the pathophysiology of IR (58). More specifically, visceral adiposity was 31% higher in the IGT group compared with the NGT group, similar to what others have found in nonrenal failure glucose-intolerant patients (5, 16). Moreover, the IGT group exhibited increased intrahepatic fat accumulation by 10-fold, even exceeding the levels for the diagnosis of nonalcoholic fatty liver disease. This is in agreement with the general notion that IR and disturbances in liver metabolism are concomitant (30). In addition, the IGT group had significantly lower oxygen saturation during sleep, implying an increased hypoxic stress during sleep (1, 53). Sleep disorders have been linked to IR in nonrenal failure patients (23); however, this link has not been studied in hemodialysis patients. In the present study, the respiratory disturbance index did not correlate with ISI-Matsuda; neither did it differ significantly between the two groups. This result is somehow unexpected since hemodialysis patients with sleep apnea syndrome have reduced functional capacity and altered muscle composition (55), both predisposing to impaired glucose tolerance. It is possible that the longer the patients suffer from sleep disorders, the more insulin resistant they become; however, no such data are yet available for this population of hemodialysis patients.

Overall, we found in this small group of patients on hemodialysis that they share common contributors in the development of IR and glucose intolerance with patients with normal kidney function. Whether the pathogenic mechanism of IR in these cases involves only uremia or other additional factors, i.e., inflammation, similar to other chronic diseases, remains to be investigated.

### Table 4. AUC values produced from the ROC curve analysis

<table>
<thead>
<tr>
<th>Variables</th>
<th>NGT vs. IGT</th>
<th>Cutoff Threshold</th>
<th>Sensitivity, %</th>
<th>Specificity, %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Intrahepatic fat, %</td>
<td>0.97 (0.935–1)</td>
<td>3.97</td>
<td>100</td>
<td>35.7</td>
</tr>
<tr>
<td>Glucose_{AUC}, mg·dl⁻¹·min⁻¹</td>
<td>0.99 (0.957–1)</td>
<td>181.37</td>
<td>100</td>
<td>66.7</td>
</tr>
</tbody>
</table>

Data are presented as AUC (95% CI). ROC, receiver operator characteristics.
Contrary to other studies, which linked IR to a sedentary lifestyle and to poor physical activity levels in nonrenal failure populations (7, 20), in our study no differences in functional capacity were found between the two groups. This possibly could be masked under the already low fitness levels that hemodialysis patients experience (24).

Although the hemodialysis patients’ QoL is very poor compared with other populations (44), in our study, the hemodialysis patients in the IGT group reported even poorer QoL scores compared with the NGT group. In our study, vitality and mental health stood out as the components that mainly influenced the total score of the SF36 questionnaire (Table 2). As QoL components can both affect and be affected by disease symptoms, we are not able to say whether QoL could affect insulin action or the other way around. Our observations though are in agreement with other studies in nonrenal failure patients (52), which show reduced QoL in patients with IR and metabolic syndrome. Still it should be noted that our hemodialysis patients come from a more or less uniform background as they all receive state disability support, they receive free medical treatment, they live with their families, they are coming from the same agricultural area, and they share similar cultural and religious beliefs.

Low-grade systemic inflammation is considered a potentially important candidate in the pathogenesis of IR in end-stage renal disease patients (28, 63). In our study, there were no differences in C-reactive protein levels between the two groups nor was there a significant correlation with any of the insulin sensitivity indices in the pool data. It is possible that a higher body mass index and/or a higher percentage of total body fat could be required for this factor to gain strength in contributing to the prediction of IR in hemodialysis patients.

It has been reported that IR is closely related to muscle wasting (33, 59) and malnutrition (13) in nondiabetic hemodialysis patients. In our study, no such a relationship was found between $\text{ISI}_\text{Matsuda}$ and total lean body mass (by DEXA) or thigh muscle cross-sectional area (by CT) or nutritional status, with the exception of WHR, which correlated significantly with $\text{ISI}_\text{Matsuda}$ in the pool data. As WHR reflects total central adiposity, it seems that it can still be useful during outpatients screening for IR in hemodialysis patients.

In our study, two important weaknesses have to be acknowledged. At first, a sample size of 34 hemodialysis patients is not sufficient enough to support strong statements for the prediction of IR and glucose intolerance. Second, since the gold standard technique for the assessment of IR is the hyperinsulinemic euglycemic clamp, the OGTT-derived values of insulin sensitivity should be interpreted with caution. It is possible that the calculated indices of IR could be affected in an unknown manner by other factors related to either the dialysis procedure or the nature of the renal diseases.

**Perspective and Significance**

We have shown for the first time that the intrahepatic fat content could be an important contributor for IR and glucose intolerance in hemodialysis patients. One out of three nondiabetic hemodialysis patients had abnormal glucose tolerance while 18% had high liver fat content. Health parameters that affect QoL, such as sleep status and central adiposity, but also the QoL score per se should also be considered as important players for glucose intolerance. Patients with end-stage renal disease and a “normal” body mass index may at the same time have fatty liver and increased visceral fat depots. Future research on interventions for better diet, increased exercise, and overall improved physical activity, in an effort to preserve a fat-free body composition may be the next steps for improving treatment in this patient group. Consideration of all these data into a model to predict glucose intolerance could help minimize the number of hemodialysis patients that remain undiagnosed with IR and improve prognosis. Furthermore, development of cheaper methods than the CT scan or MRI and spectroscopy to determine liver fat content could have a considerable impact on health costs.

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