Decrees in Apolipoprotein(a) after Renal Transplantation: Implications for Lipoprotein(a) Metabolism

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Serum concentrations of apolipoprotein(a) [apo(a)], the unique glycoprotein of lipoprotein(a), are increased in patients with end-stage renal failure. We prospectively studied serum apo(a) and other lipoproteins in 20 consecutive patients, ages 46 ± 11 years, before and for six months after successful renal transplantation. All patients received cyclosporine, and no patient was treated for hyperlipidemia. The mean creatinine clearance increased from 7.5 mL/min before transplant surgery to 40.9 mL/min six months afterwards (P < 0.001). Apo(a) decreased from a median of 403 units/L before transplantation to 184 units/L at one week (P < 0.001) and was 170 units/L (P < 0.001) at six months. For the assay used, 1 unit of apo(a) is equivalent to 1 mg of lipoprotein(a). In contrast, from baseline to six months, increases were found for low-density lipoprotein (LDL) cholesterol (P = 0.03), high-density lipoprotein cholesterol (P = 0.06), apo B (P = 0.07), and apo A-I (P = 0.01). The decrease in apo(a) in individual patients was significantly correlated with the increase in creatinine clearance (r = -0.48, P < 0.001). The single patient who developed nephrotic syndrome after renal transplantation had marked increases in apo(a) (693–1595 units/L), apo B, and LDL cholesterol, which paralleled the degree of proteinuria. These findings suggest that abnormal renal function affects the regulation of lipoprotein(a) metabolism.

Cardiovascular disease is an important cause of morbidity and mortality in patients with end-stage renal failure (1, 2) and also after renal transplantation (2, 3). Lipoprotein abnormalities have been documented in patients with chronic renal failure (4), including those undergoing renal dialysis (5), as well as after renal transplantation (6). Recently, increased concentrations of apolipoprotein(a) [apo(a)] have also been reported in patients undergoing hemodialysis for end-stage renal failure (7, 8).

Apo(a), a glycoprotein with structural homology to plasminogen, is linked to apo B, the principal apolipoprotein of low-density lipoproteins (LDL) cholesterol, to form lipoprotein(a) [Lp(a)] (9). Increased concentrations of Lp(a) are associated with an increased risk of coronary artery disease, similar to and synergistic with LDL cholesterol (10, 11), and possibly with an increased risk of thrombosis (9). Lp(a) has been shown to accumulate in atherosclerotic lesions of coronary bypass grafts and native coronary arteries in concentrations that are positively correlated with serum Lp(a) values (12). An association with thrombosis is suggested by the close homology of apo(a) with plasminogen and is supported by in vitro and in vivo evidence (9, 13).

Recent studies indicate that Lp(a) concentrations in apparently normal subjects are related to the apo(a) phenotype, which is determined by a series of autosomal alleles at the apo(a) gene locus (14), and that apo(a) is produced in the liver (15). One of the striking properties of Lp(a) metabolism is the stability of Lp(a) concentrations in individual patients, being correlated in normal subjects with the rate of Lp(a) synthesis rather than with the rate of catabolism (16). Little is known, however, about alterations in Lp(a) concentrations in conditions known to be associated with abnormal lipoprotein metabolism, e.g., renal disease. The mechanism(s) mediating the increase in apo(a) concentrations in patients undergoing renal dialysis are not known, and the effect on apo(a) concentrations of improved renal function after renal transplantation has not been reported.

To examine the effects of renal transplantation on serum apo(a) concentrations in patients with end-stage renal failure, we prospectively studied a consecutive series of patients with end-stage renal failure before and after successful renal transplantation.

Patients and Methods

Patients

At one institution, 26 consecutive adult patients undergoing renal transplantation for end-stage renal failure were initially enrolled in the study. All patients received cadaveric kidney grafts. Six patients underwent graft nephrectomy within one month of transplantation and were excluded from further analysis. Renal transplantation was successful in the remaining 20 patients, and in these patients dialysis was not required six weeks after the surgery.

The study population was 10 men and 10 women, mean age 46 (range 24–58) years. All patients were undergoing dialysis before transplantation, seven by chronic ambulatory peritoneal dialysis and 13 by hemodialysis. The mean duration of dialysis was 26 (range 1–120) months. The etiology of renal failure, as determined by renal biopsy, was glomerulonephritis (n = 8), analgesic nephropathy (n = 4), reflux nephropathy (n = 4), polycystic disease (n = 2), diabetes mellitus (n = 1),
and amyloid nephropathy (n = 1). After transplantation, all patients were treated with a combination immunosuppressive regimen of daily prednisone, azathioprine, and cyclosporine. The mean doses of immunosuppressive drugs at six months post-transplantation were prednisone 11 mg/day, azathioprine 98 mg/day, and cyclosporine 338 mg/day. No patient received lipid-lowering medication (determined by physician preference) or thiazide diuretics during the study period. Beta-adrenergic blocking drugs were given to six patients before transplantation and to nine patients afterwards; the latter were receiving metoprolol (n = 4), atenolol (n = 3), or labetalol (n = 2). Four patients had clinical or angiographic evidence of vascular disease: two had ischemic heart disease, one had peripheral vascular disease, and one had cerebrovascular disease.

Methods

We collected blood samples after a 6 to 12-h preoperative fast before the transplant surgery and then after a 12-h overnight fast one week, one month, three months, and six months after transplantation. Plasma cholesterol, triglyceride, high-density-lipoproteins (HDL) cholesterol, and creatinine were measured by standard enzymatic and colorimetric methods. LDL cholesterol was determined by the Friedewald formula (17). Creatinine clearance was calculated from plasma creatinine by the method of Cockroft and Gault (18). We measured serum apo(a), apo A-I, and apo B-100 by RIA (Pharmacia Diagnostics, Uppsala, Sweden). Apo(a) values are expressed as units per liter with 1 unit of apo(a) being equivalent to 1 mg of Lp(a). The intra- and interassay coefficients of variation for the serum apo(a) assay in our laboratory were 4.8% and 5.7%, respectively, at a concentration of 103 units/L (19) and for apo B were 4.5% and 6.1%. For apo A-I, however, CVs were 5.3% and 6.8%.

The distributions of apo(a), triglyceride, HDL cholesterol, and apo A-I were markedly skewed, and we therefore applied logarithmic transformation to produce approximately normal distribution before statistical analysis. We used Student's paired t-test to compare pre- and post-transplant values and used Student's unpaired t-test and the chi-square test with correction for continuity to compare differences between groups at baseline and after transplantation. We used Spearman's rank correlation coefficient to evaluate the degree of association between variables. Statistical significance was defined as two-tailed, P < 0.05. Analyses were performed with the SPIDA software package. Apo(a) values are reported as median (range) unless otherwise indicated, and other values as mean (SD).

Results

Lipoprotein concentrations and creatinine clearance before and after transplantation are shown in Table 1. Apo(a) decreased significantly between baseline and one week after surgery (P < 0.001) and again between one week and one month (P = 0.002). We noted a small increase from one to six months (P = 0.006), which remained significant (P = 0.01) if the single patient with the nephrotic syndrome after transplant, discussed below, was excluded from the analysis. Overall, apo(a) concentration decreased markedly from a median of 403 to 170 units/L (P < 0.001), i.e., equivalent to a reduction from 403 to 170 mg/L Lp(a). The mean apo(a) concentration decreased from 547 to 312 units/L, and the mean percentage reduction in apo(a) concentration between baseline and six months was 39% (95% confidence interval, 16%–63%). Apo(a) declined after transplantation in 17 of the 20 patients (85%) and was >300 units/L in only five patients (25%) at six months, compared with 11 patients (55%) at baseline.

A single patient in the series developed nephrotic syndrome after transplantation; no other patient had >2 g/day proteinuria. This 48-year-old man with membrano-proliferative glomerulonephritis had undergone hemodialysis for 18 months before transplantation. Nephrotic syndrome developed in the first month after surgery and was shown by renal biopsy to be due to recurrent glomerulonephritis. The development of proteinuria was associated with marked increases in apo(a) (Figure 1), total and LDL cholesterol, and apo B (Table 2). Two other patients without significant proteinuria (<0.2 g/day) had modest increases in apo(a) during the study period—from 590 to 635 units/L in one patient and from 130 to 181 units/L in the other.

There was no significant correlation between apo(a) and proteinuria (Figure 2), total and LDL cholesterol, and apo B.

Table 1. Lipoprotein Concentrations before and during the Six Months after Renal Transplantation

<table>
<thead>
<tr>
<th>Time after transplantation</th>
<th>Apo(a), units/L</th>
<th>Cholesterol, mmol/L</th>
<th>Triglyceride, mmol/L</th>
<th>LDL cholesterol, mmol/L</th>
<th>HDL cholesterol, mmol/L</th>
<th>Apo A-I, g/L</th>
<th>Apo B, g/L</th>
<th>Creatinine clearance, mL/min</th>
</tr>
</thead>
<tbody>
<tr>
<td>Baseline</td>
<td>403 (43–1407)*</td>
<td>6.02 (1.34)</td>
<td>2.39 (0.90)</td>
<td>3.59 (1.27)</td>
<td>1.35 (0.85)</td>
<td>1.13 (0.35)</td>
<td>1.01 (0.36)</td>
<td>7.5 (3.1)</td>
</tr>
<tr>
<td>One week</td>
<td>184 (37–1042)</td>
<td>4.89 (0.93)</td>
<td>2.46 (1.29)</td>
<td>2.77 (0.88)</td>
<td>1.04 (0.50)</td>
<td>0.80 (0.19)</td>
<td>1.05 (0.16)</td>
<td>21.8 (27.0)</td>
</tr>
<tr>
<td>One month</td>
<td>137 (25–717)</td>
<td>6.14 (1.50)</td>
<td>1.83 (1.02)</td>
<td>3.73 (1.22)</td>
<td>1.61 (0.59)</td>
<td>1.08 (0.21)</td>
<td>1.00 (0.29)</td>
<td>31.4 (13.3)</td>
</tr>
<tr>
<td>Three months</td>
<td>138 (21–1182)</td>
<td>6.66 (1.13)</td>
<td>2.30 (0.97)</td>
<td>4.18 (1.12)</td>
<td>1.43 (0.55)</td>
<td>1.17 (0.26)</td>
<td>1.12 (0.28)</td>
<td>38.8 (8.7)</td>
</tr>
<tr>
<td>Six months</td>
<td>170 (19–1595)*</td>
<td>6.66 (1.35)</td>
<td>2.18 (0.91)</td>
<td>4.30 (1.18)*</td>
<td>1.44 (0.59)</td>
<td>1.25 (0.27)*</td>
<td>1.21 (0.28)</td>
<td>40.9 (8.9)*</td>
</tr>
</tbody>
</table>

* Values for apo(a) are median (range); all other values are mean (SD).

b P < 0.001 for comparison of values at six months and baseline.

P < 0.001.
concentrations during the study period and any other lipid fraction. We assessed the correlation between changes in renal function and apo(a) concentrations from the percentage changes in creatinine clearance and in apo(a) concentrations in each patient during the pre- and post-transplant periods. We found a highly significant relationship ($r = -0.48$, $P < 0.001$).

We compared patients with clinically significantly increased concentrations of apo(a) before transplantation, defined as $> 300$ units/L apo(a), i.e., $> 300$ mg/L Lp(a) ($10, 11$), and the remaining patients (median 856, range 351–1407 units/L, $n = 11$ vs 183 (43–278) units/L, $n = 9$). We found no significant difference between the two groups for any other lipid fraction, patient's age (mean 46 vs 47 years), body weight (70 vs 64 kg), duration of dialysis (28 vs 24 months), or creatinine clearance (8.7 vs 6.5 mL/min). The single patient with amyloid nephropathy had the highest baseline apo(a) concentration, 1407 units/L. Apo(a) concentrations in the four patients with vascular disease, 135 (137–1407) units/L, were not significantly different from those in the other patients, 522 (43–1150) units/L.

When we compared the concentrations of pre-transplant lipoproteins in the seven patients with chronic ambulatory peritoneal dialysis and in the 13 patients undergoing hemodialysis, we noted significant increases in the former for total cholesterol [6.90 (1.54) vs 5.55 (0.98) mmol/L, $P = 0.03$], LDL cholesterol [4.46 (1.57) vs 3.13 (0.81) mmol/L, $P = 0.02$], and apo B [1.29 (0.40) vs 0.84 (0.24) g/L, $P = 0.01$]. Apo(a) was 666 (155–1407) units/L in the peritoneal dialysis patients and 351 (43–1150) units/L in the hemodialysis patients ($P > 0.05$). Creatinine clearance was similar in the peritoneal dialysis [7.0 (1.7) mL/min] and hemodialysis [7.8 (0.6) mL/min] groups.

When we compared concentrations of other lipid fractions at baseline and six months post-transplant, LDL cholesterol ($P = 0.03$) and apo A-I ($P = 0.01$) were significantly increased. We also found nonsignificant increases for total cholesterol, HDL cholesterol, and apo B, whereas triglyceride decreased slightly. The LDL/HDL ratio was not significantly different at baseline and at six months (3.29 vs 3.34), nor was the apo B/apo A-I ratio (0.99 vs 1.00).

We also assessed the correlation between cumulative immunosuppressant dose adjusted for body weight (mg/kg) and lipoprotein concentrations during the post-transplant period. Azathioprine does in individual patients varied little during the study and were not included in the analysis. Cumulative prednisone dose was significantly correlated with total cholesterol ($r = 0.45$, $P < 0.001$), LDL cholesterol ($r = 0.49$, $P < 0.001$), HDL cholesterol ($r = 0.23$, $P = 0.04$), apo A-I ($r = 0.41$, $P < 0.001$), and apo B ($r = 0.24$, $P = 0.04$). Cumulative cyclosporine dose was significantly correlated with total cholesterol ($r = 0.48$, $P < 0.001$), LDL cholesterol, ($r = 0.46$, $P < 0.001$), HDL cholesterol ($r = 0.23$, $P = 0.04$), and apo A-I ($r = 0.50$, $P < 0.001$). There was a nonsignificant correlation between cyclosporine and apo B and no correlation between either prednisone or cyclosporine and apo(a). Pre- and post-transplant concentrations of apo(a) and other lipoproteins in patients receiving beta-adrenergic blocking drugs and in the remaining patients were not significantly different.

**Discussion**

Since Lindner et al. (1) reported in 1974 that cardiovascular disease is a major cause of death in chronic dialysis patients, the possible relationship between the lipoprotein abnormalities associated with chronic renal failure and atherogenesis has been studied extensively (20). It has proved difficult to explain the increased cardiovascular risk by contributions from abnormal lipid concentrations and other recognized risk factors (21). The present study was prompted by reports of marked increases of apo(a) concentrations in patients with end-stage renal failure (7, 8). Parsy et al. (8) found a mean apo(a) concentration of 440 mg/L compared with 120 mg/L in controls, and Parra et al. (7) reported a mean concentration of 378 mg/L compared with 125 mg/L. Although we did not use controls, we confirmed that apo(a) is markedly increased in end-stage renal failure, with a median in our patients of 403 units/L, equivalent to 403 mg/L Lp(a). An increase >300 mg/L is associated with a twofold increase in coronary risk (11), and we found this value in 55% of our patients. Concentrations of this order were found in ~20% of patients without coronary artery disease (22) and in 30% of patients with angiographically proven coronary disease (11). It is interesting that the highest baseline apo(a) concentration, 1407 mg/L, was in a patient with amyloid nephropathy, because Karádi et al. (23) have also reported very high concentrations in one patient.

The major finding of the present study was a large reduction in apo(a) concentrations after renal transplantation, with a decline to values slightly higher than in the normal population. We saw this reduction in 17 of the 20 patients; moreover, it was apparent within one month after transplantation and was maintained throughout the six-month study period. The early decline in apo(a) concentrations was particularly striking.
because Lp(a) values in normal subjects are reported to increase for as long as one month after surgical procedures (24). We also found a significant correlation between the change in renal function and apo(a) values but no correlation between apo(a) and apo B or apo A-I. These findings are consistent with a relationship between abnormal renal function and the regulation of Lp(a) metabolism.

The kidney itself has not been reported to participate directly in apo(a) metabolism. However, hepatic production of apo(a) correlates with apo(a) concentrations in normal subjects (16), and it is possible that this is stimulated by the metabolic derangements of end-stage renal failure. Catabolism of apo(a) could also be impaired in renal failure. Neary and Gowland (25) reported increased concentrations of free apo A-I (molecular mass 45,000 Da) in patients with renal failure and suggested that free apo A-I was removed from blood by glomerular filtration and tubular catabolism. The large size of apo(a), 400–700,000 Da (14), precludes the possibility of glomerular excretion of intact apo(a). However, an accumulation of apo(a) fragments in renal failure or tubular catabolism of apo(a) cannot be excluded.

Factors in the post-transplant state other than improved renal function, e.g., immunosuppressive drug therapy, could also contribute to the decline in concentrations of apo(a). However, we found no correlation between immunosuppressants dosage and apo(a) concentrations; also, the high concentrations pre-transplant cannot be due to immunosuppressive drug therapy. Moreover, cyclosporine is associated with increases in apo B and LDL cholesterol (26), in contrast with the decline in apo(a) that we observed.

Of particular interest was the dramatic and parallel increase of apo(a) and proteinuria in the one patient who developed nephrotic syndrome, because this event may provide insight into the mechanism of the initial increase in concentrations of apo(a). It is relevant that the only patient in whom nephrotic syndrome developed after transplantation was also the only patient in whom there was a marked increase in apo(a) (Figure 1), and that this occurred in association with an increase in the other apo B-containing lipoproteins. Increased serum apo(a) in patients with proteinuria and normal plasma creatinine has been recently reported (23). However, a direct temporal relationship between the development of proteinuria and apo(a) increase has not been demonstrated previously. Joven et al. (27) recently reported that the increase in apo B-containing lipoproteins associated with nephrotic syndrome was mediated by a reversible increase in apo B production, with a normal catabolic rate. It is possible that a similar mechanism is responsible for the increased apo(a) in nephrotic syndrome.

The mechanism of Lp(a) catabolism in normal subjects and the effects of lipid-lowering drug therapy on Lp(a) are also consistent with an alteration in the rates of production being the major determinant of changes in Lp(a) concentration. Lp(a) is a weaker ligand for the LDL receptor than LDL (16, 28). Although in vivo studies differ regarding the role of the LDL-receptor pathway in Lp(a) clearance (28, 29), Lp(a) concentrations are not affected by agents that lower LDL cholesterol by up-regulation of LDL receptors, e.g., cholestyramine (30) and hydroxymethylglutaryl-CoA reductase inhibitors (31). This suggests that LDL receptors do not play a major physiological role in Lp(a) catabolism. In support of the greater contribution of production, Carlson et al. (32) reported that nicotinic acid resulted in a marked reduction in Lp(a) concentrations, which was highly correlated (r = 0.88) with the reduction in LDL cholesterol. They argued that this correlation arose from inhibition of the synthesis of apo B, which is common to the two lipoproteins. In the present study, pre-transplant concentrations of apo B, LDL cholesterol, and apo(a) were higher in patients treated with peritoneal dialysis than in those managed by hemodialysis. Sniderman et al. (33) showed that these differences for apo B and LDL cholesterol are due to increased apo B production with peritoneal dialysis. Although the higher apo(a) in patients treated with peritoneal dialysis in the present series was not statistically significant, the trend is consistent with an increase in apo(a) due to increased production.

The relatively small number of patients studied limits our analysis of the complex factors in the post-renal-transplant state, which may influence apo(a) concentrations. However, the prospective nature of the study, with each patient as his or her own control, strengthens the validity of the major finding of a large reduction in

<table>
<thead>
<tr>
<th></th>
<th>Baseline</th>
<th>One week</th>
<th>One month</th>
<th>Three months</th>
<th>Six months</th>
</tr>
</thead>
<tbody>
<tr>
<td>Apo(a), units/L</td>
<td>693</td>
<td>411</td>
<td>395</td>
<td>1182</td>
<td>1595</td>
</tr>
<tr>
<td>Proteinuria, g/day</td>
<td>0</td>
<td>1.8</td>
<td>6.3</td>
<td>7.1</td>
<td>13.6</td>
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<tr>
<td>Creatinine clearance, mL/min</td>
<td>8</td>
<td>8</td>
<td>33</td>
<td>37</td>
<td>44</td>
</tr>
<tr>
<td>Cholesterol, mmol/L</td>
<td>4.9</td>
<td>3.7</td>
<td>5.2</td>
<td>8.3</td>
<td>10.3</td>
</tr>
<tr>
<td>Triglyceride, mmol/L</td>
<td>2.4</td>
<td>0.8</td>
<td>1.0</td>
<td>1.8</td>
<td>4.7</td>
</tr>
<tr>
<td>LDL cholesterol, mmol/L</td>
<td>2.6</td>
<td>2.1</td>
<td>3.4</td>
<td>5.7</td>
<td>7.3</td>
</tr>
<tr>
<td>HDL cholesterol, mmol/L</td>
<td>1.2</td>
<td>1.2</td>
<td>1.3</td>
<td>1.8</td>
<td>0.9</td>
</tr>
<tr>
<td>Apo A-I, g/L</td>
<td>1.13</td>
<td>1.13</td>
<td>0.88</td>
<td>1.23</td>
<td>1.12</td>
</tr>
<tr>
<td>Apo B, g/L</td>
<td>0.73</td>
<td>0.74</td>
<td>0.90</td>
<td>1.42</td>
<td>2.00</td>
</tr>
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</table>
apo(a) concentrations after transplantation. These results are consistent with a relationship between improved renal function after transplantation and a reduction in previously increased apo(a) values. Increased hepatic production of Lp(a) appears to be the most likely mechanism of the increased apo(a) concentrations associated with renal failure, although the evidence is indirect. Our findings have implications for the regulation of Lp(a) metabolism and for the etiology of vascular disease in patients with abnormal renal function. Further investigation with determination of apo(a) phenotypes and apo(a) turnover studies in patients with renal disease is required.

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References