Human Renal Transplants

I. Glomerular Changes*

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Thickening of the glomerular capillary basement membranes has frequently been
seen by light microscopy in renal allografts transplanted into patients whose immune
responses have been modified by chronic
uremia, ionizing radiation, various immuno
suppressive drugs, or by some combination
of these. In most instances the
changes have been minor,4 5 6 14 18 35 41 42 44 56
When one such kidney was ex
amined electron microscopically, the
basement membrane thickening was found
to be focal and accompanied by fusion of
the epithelial foot processes.58 Although
two patients were reported as showing
more pronounced changes,22 23 it was
Hamburger and his colleagues11-13 15 who
first emphasized the severity of the glo
merular lesions that may occur in long
surviving renal allografts. They described
six patients, three of whom developed
proteinuria and renal insufficiency 3
months to 1 year after transplantation; no
particular clinical signs were present in
the other three patients when they were
diagnosed on biopsy of the graft at 3, 6,
and 31 months respectively. One of these
patients died suddenly from renal infarc
tion 1 month later, while another slowly
deteriorated and perished 10 months after
the onset of proteinuria. The other four pa
tients remained well with normal renal
function 1 month to 2 years 10 months
after diagnosis. Ultrastructurally in the
glomeruli of all these cases there was
"proliferation of endocapillary cells with
subendothelial and intercellular hyaline
deposits."11 Stress is also laid on glomeru
lar lesions in renal transplants in a recent
light microscopy report from another cen
ter.30

Little is known about the incidence,
etiology, and pathogenesis of these glo
merular lesions and the prognosis for the
patient. In this paper an attempt is made
to answer some of these questions on the
basis of the light and electron microscopic
glomerular changes encountered in 50 renal
allografts when they were examined 43 days
to 2 years 3 months after transplantation. As
controls a renal isograft and a donor's re
mainning kidney were studied.

A preliminary account of the changes in
some of these kidneys has already been
given.42 49 59

MATERIALS AND METHODS

Patients with Renal Allografts

Living Donors. The 35 patients from the
University of Colorado Medical Center,
Denver, Colorado, received kidneys from
living donors, and each recipient is denoted
by a number prefixed by the letters LD
(Tables 1 and 2). The same code has been
used previously47 48 59 and further details of
any of the patients can be obtained by
<table>
<thead>
<tr>
<th>Patient</th>
<th>Sex and age</th>
<th>Disease</th>
<th>Donor sex, age, and relationship</th>
<th>No. of rejection episodes</th>
<th>Time biopsy taken after transplant</th>
<th>Renal function at time of biopsy</th>
<th>Uremic proteinuria</th>
<th>Outcome</th>
</tr>
</thead>
<tbody>
<tr>
<td>LD1</td>
<td>M, 14</td>
<td>CPGN</td>
<td>F, 35, mother</td>
<td>1</td>
<td>2 yr.</td>
<td>100/100</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>LD3</td>
<td>M, 22</td>
<td>CMGN</td>
<td>X, 22, dizygotic twin</td>
<td>0</td>
<td>2 yr.</td>
<td>131/130</td>
<td>130</td>
<td>130</td>
</tr>
<tr>
<td>LD15</td>
<td>M, 25</td>
<td>CPGN</td>
<td>M, 24, brother</td>
<td>1</td>
<td>1 yr. 9 mo.</td>
<td>139/139</td>
<td>139</td>
<td>139</td>
</tr>
<tr>
<td>LD18</td>
<td>M, 29</td>
<td>CPGN</td>
<td>F, 44, sister</td>
<td>1</td>
<td>2 yr. 2 mo.</td>
<td>61/61</td>
<td>61</td>
<td>61</td>
</tr>
<tr>
<td>LD21</td>
<td>F, 23</td>
<td>Poly</td>
<td>F, 41, mother</td>
<td>2</td>
<td>2 yr. 1 mo.</td>
<td>65/65</td>
<td>65</td>
<td>65</td>
</tr>
<tr>
<td>LD27</td>
<td>F, 20</td>
<td>CMGN</td>
<td>M, 22, unrelated</td>
<td>2</td>
<td>2 yr.</td>
<td>59/59</td>
<td>59</td>
<td>59</td>
</tr>
<tr>
<td>LD30</td>
<td>M, 41</td>
<td>CMGN</td>
<td>M, 28, unrelated</td>
<td>2</td>
<td>1 yr. 11 mo.</td>
<td>70/70</td>
<td>70</td>
<td>70</td>
</tr>
<tr>
<td>LD35</td>
<td>M, 26</td>
<td>CPyN</td>
<td>F, 41, wife</td>
<td>1</td>
<td>1 yr. 10 mo.</td>
<td>70/70</td>
<td>70</td>
<td>70</td>
</tr>
<tr>
<td>LD40</td>
<td>F, 21</td>
<td>CMGN</td>
<td>F, 57, mother</td>
<td>1</td>
<td>1 yr. 10 mo.</td>
<td>66/66</td>
<td>66</td>
<td>66</td>
</tr>
<tr>
<td>LD41</td>
<td>M, 4</td>
<td>CPGN</td>
<td>F, 37, mother</td>
<td>2</td>
<td>1 yr. 10 mo.</td>
<td>35/35</td>
<td>35</td>
<td>35</td>
</tr>
<tr>
<td>LD45</td>
<td>M, 35</td>
<td>CPGN</td>
<td>F, 29, sister</td>
<td>1</td>
<td>2 yr. 1 mo.</td>
<td>48/48</td>
<td>48</td>
<td>48</td>
</tr>
<tr>
<td>LD47</td>
<td>M, 37</td>
<td>CPGN</td>
<td>M, 27, brother</td>
<td>4</td>
<td>1 yr. 10 mo.</td>
<td>20/20</td>
<td>20</td>
<td>20</td>
</tr>
<tr>
<td>LD48</td>
<td>M, 34</td>
<td>CPGN</td>
<td>F, 29, sister</td>
<td>2</td>
<td>1 yr.</td>
<td>50/50</td>
<td>50</td>
<td>50</td>
</tr>
<tr>
<td>LD51</td>
<td>M, 18</td>
<td>CPGN</td>
<td>F, 50, aunt</td>
<td>1</td>
<td>1 yr. 11 mo.</td>
<td>92/92</td>
<td>92</td>
<td>92</td>
</tr>
<tr>
<td>LD54</td>
<td>M, 21</td>
<td>CPGN</td>
<td>M, 38, unrelated</td>
<td>3</td>
<td>1 yr. 10 mo.</td>
<td>30/30</td>
<td>30</td>
<td>30</td>
</tr>
<tr>
<td>LD55</td>
<td>M, 21</td>
<td>CMGN</td>
<td>M, 49, father</td>
<td>2</td>
<td>1 yr. 11 mo.</td>
<td>92/92</td>
<td>92</td>
<td>92</td>
</tr>
<tr>
<td>LD56</td>
<td>M, 31</td>
<td>CPGN</td>
<td>M, 23, cousin</td>
<td>1</td>
<td>1 yr. 3 mo.</td>
<td>50/50</td>
<td>50</td>
<td>50</td>
</tr>
<tr>
<td>LD63</td>
<td>M, 35</td>
<td>CPGN</td>
<td>M, 33, unrelated</td>
<td>0</td>
<td>1 yr. 10 mo.</td>
<td>65/65</td>
<td>65</td>
<td>65</td>
</tr>
<tr>
<td>LD69</td>
<td>M, 19</td>
<td>CPGN</td>
<td>F, 40, mother</td>
<td>2</td>
<td>2 yr. 6 mo.</td>
<td>20/20</td>
<td>20</td>
<td>20</td>
</tr>
<tr>
<td>M13</td>
<td>M, 28</td>
<td>CPyN</td>
<td>F, 29, cadaver</td>
<td>4</td>
<td>10 mo.</td>
<td>90/90</td>
<td>90</td>
<td>90</td>
</tr>
<tr>
<td>M17</td>
<td>M, 45</td>
<td>CPGN</td>
<td>M, 5, cadaver</td>
<td>3</td>
<td>7 mo.</td>
<td>10/10</td>
<td>10</td>
<td>10</td>
</tr>
<tr>
<td>M18</td>
<td>M, 29</td>
<td>CMGN</td>
<td>M, 37, cadaver</td>
<td>7</td>
<td>4 mo.</td>
<td>70/70</td>
<td>70</td>
<td>70</td>
</tr>
<tr>
<td>M25</td>
<td>M, 37</td>
<td>Poly</td>
<td>M, 41, cadaver</td>
<td>6</td>
<td>1 yr. 7 mo.</td>
<td>73/73</td>
<td>73</td>
<td>73</td>
</tr>
<tr>
<td>E1</td>
<td>F, 21</td>
<td>CPyN</td>
<td>M, 22, cadaver</td>
<td>1</td>
<td>1 yr. 5 mo.</td>
<td>112/112</td>
<td>112</td>
<td>112</td>
</tr>
<tr>
<td>AR</td>
<td>M, 27</td>
<td>CPGN</td>
<td>F, 41, cadaver</td>
<td>2</td>
<td>48 da.</td>
<td>120/120</td>
<td>120</td>
<td>120</td>
</tr>
<tr>
<td>ET</td>
<td>M, 14</td>
<td>CPGN</td>
<td>F, 34, cadaver</td>
<td>5</td>
<td>1 yr.</td>
<td>60/60</td>
<td>60</td>
<td>60</td>
</tr>
<tr>
<td>SL1</td>
<td>M, 30</td>
<td>CPGN</td>
<td>F, 17, cadaver</td>
<td>1</td>
<td>8 mo.</td>
<td>50/50</td>
<td>50</td>
<td>50</td>
</tr>
</tbody>
</table>

*CPGN: Chronic proliferative glomerulonephritis; RPpN: rapidly progressive acute glomerulonephritis; CMGN: chronic membranous glomerulonephritis; CPyN: chronic pyelonephritis; Poly: bilateral polyacue disease.

a Cr. Creatinine clearance
b Biopsied during a rejection episode.
### Table 2. Clinical Data on the 25 Patients with Minor Glomerular Changes or Normal Glomeruli

<table>
<thead>
<tr>
<th>Recipient</th>
<th>Donor</th>
<th>Sex and age, and relationship</th>
<th>No. of rejection episodes</th>
<th>Time biopsy taken after transplant</th>
<th>Renal function at time of biopsy Ccr</th>
<th>Urinary protein</th>
<th>Outcome</th>
</tr>
</thead>
<tbody>
<tr>
<td>Patient no.</td>
<td>Sex and age</td>
<td>Disease</td>
<td></td>
<td></td>
<td></td>
<td>ml./min.</td>
<td>mg./100 ml.</td>
</tr>
<tr>
<td>LD2</td>
<td>M, 38</td>
<td>CPGN</td>
<td>F, 32, sister</td>
<td>1</td>
<td>2 yr.</td>
<td>129</td>
<td>0</td>
</tr>
<tr>
<td>LD5</td>
<td>M, 23</td>
<td>CPGN</td>
<td>M, 22, brother</td>
<td>1</td>
<td>1 yr. 11 mo.</td>
<td>130</td>
<td>0</td>
</tr>
<tr>
<td>LD12</td>
<td>M, 49</td>
<td>RPGN</td>
<td>M, 53, brother</td>
<td>2</td>
<td>1 yr. 10 mo.</td>
<td>65</td>
<td>0</td>
</tr>
<tr>
<td>LD13</td>
<td>M, 47</td>
<td>CPGN</td>
<td>F, 35, mother</td>
<td>2</td>
<td>1 yr. 9 mo.</td>
<td>105</td>
<td>0</td>
</tr>
<tr>
<td>LD14</td>
<td>M, 44</td>
<td>CPyN</td>
<td>M, 37, brother</td>
<td>0</td>
<td>1 yr. 9 mo.</td>
<td>102</td>
<td>0</td>
</tr>
<tr>
<td>LD17</td>
<td>F, 15</td>
<td>CPyN</td>
<td>F, 44, mother</td>
<td>1</td>
<td>2 yr. 1 mo.</td>
<td>71</td>
<td>8</td>
</tr>
<tr>
<td>LD25</td>
<td>M, 33</td>
<td>CPGN</td>
<td>M, 29, brother</td>
<td>1</td>
<td>2 yr.</td>
<td>84</td>
<td>5</td>
</tr>
<tr>
<td>LD33</td>
<td>F, 18</td>
<td>CPGN</td>
<td>F, 42, mother</td>
<td>1</td>
<td>1 yr. 11 mo.</td>
<td>184</td>
<td>0</td>
</tr>
<tr>
<td>LD34</td>
<td>F, 6</td>
<td>CPGN</td>
<td>F, 30, mother</td>
<td>1</td>
<td>1 yr. 11 mo.</td>
<td>50</td>
<td>0</td>
</tr>
<tr>
<td>LD37</td>
<td>M, 21</td>
<td>CPGN</td>
<td>F, 43, mother</td>
<td>1</td>
<td>1 yr. 10 mo.</td>
<td>159</td>
<td>22</td>
</tr>
<tr>
<td>LD39</td>
<td>M, 17</td>
<td>CPGN</td>
<td>F, 48, mother</td>
<td>1</td>
<td>1 yr. 10 mo.</td>
<td>87</td>
<td>20</td>
</tr>
<tr>
<td>LD42</td>
<td>F, 19</td>
<td>CPGN</td>
<td>M, 47, father</td>
<td>1</td>
<td>1 yr. 9 mo.</td>
<td>84</td>
<td>0</td>
</tr>
<tr>
<td>LD49</td>
<td>M, 32</td>
<td>CPGN</td>
<td>F, 41, sister</td>
<td>0</td>
<td>1 yr. 10 mo.</td>
<td>70</td>
<td>4</td>
</tr>
<tr>
<td>LD50</td>
<td>F, 16</td>
<td>CPGN</td>
<td>M, 38, father</td>
<td>1</td>
<td>2 yr.</td>
<td>121</td>
<td>2</td>
</tr>
<tr>
<td>LD52</td>
<td>F, 15</td>
<td>CPGN</td>
<td>M, 36, father</td>
<td>0</td>
<td>1 yr. 11 mo.</td>
<td>103</td>
<td>8</td>
</tr>
<tr>
<td>LD53</td>
<td>M, 15</td>
<td>CPGN</td>
<td>M, 33, uncle</td>
<td>1</td>
<td>1 yr. 11 mo.</td>
<td>101</td>
<td>3</td>
</tr>
<tr>
<td>LD55</td>
<td>M, 27</td>
<td>CPGN</td>
<td>F, 23, sister</td>
<td>0</td>
<td>1 yr. 9 mo.</td>
<td>114</td>
<td>4</td>
</tr>
<tr>
<td>M8</td>
<td>M, 24</td>
<td>Hydr</td>
<td>F, 53, cadaver</td>
<td>3</td>
<td>1 yr. 2 mo.</td>
<td>50</td>
<td>100</td>
</tr>
<tr>
<td>M15</td>
<td>M, 22</td>
<td>CPGN</td>
<td>M, 34, cadaver</td>
<td>2</td>
<td>8 mo.</td>
<td>35</td>
<td>80</td>
</tr>
<tr>
<td>M21</td>
<td>M, 37</td>
<td>CPGN</td>
<td>M, 22, cadaver</td>
<td>1</td>
<td>7 mo.</td>
<td>80</td>
<td>80</td>
</tr>
<tr>
<td>M28</td>
<td>M, 25</td>
<td>CPGN</td>
<td>M, 17, cadaver</td>
<td>2</td>
<td>79 da.</td>
<td>45</td>
<td>40</td>
</tr>
<tr>
<td>CBF</td>
<td>M, 25</td>
<td>CPGN</td>
<td>M, 43, cadaver</td>
<td>5</td>
<td>11 mo.</td>
<td>33</td>
<td>0</td>
</tr>
<tr>
<td>EF</td>
<td>M, 17</td>
<td>CPyN</td>
<td>F, 41, cadaver</td>
<td>3</td>
<td>1 yr. 10 mo.</td>
<td>124</td>
<td>0</td>
</tr>
<tr>
<td>IDT2</td>
<td>M, 18</td>
<td>CPGN</td>
<td>M, 18, identical twin</td>
<td>0</td>
<td>2 yr. 3 mo.</td>
<td>115</td>
<td>37</td>
</tr>
</tbody>
</table>

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*a* CPGN, Chronic proliferative glomerulonephritis; RPGN, rapidly progressive acute glomerulonephritis; CPyN, chronic pyelonephritis; CMGN, chronic membranous glomerulonephritis; and Hydr, hydronephrosis.

*b* Ccr, Creatinine clearance.

* Biopsied during a rejection episode.
FIGS. 1 AND 2
referring to these publications. The patient LDBS received a renal transplant at the Peter Bent Brigham Hospital, Boston, Massachusetts, and the allograft was eventually removed at Denver. In all but one of the patients, splenectomy and bilateral nephrectomy were performed at or before the time of transplantation: the exception was LD2 who retained his right kidney. The thymus was removed from 12 of the patients, four before and eight from 250 to 520 days after transplantation.

Cadaveric Donors. The seven patients from the Royal Victoria Hospital, Montreal, Canada, the six patients from St. Mary's Hospital, London, England, and the one patient from the John J. Cochran Veterans Hospital, St. Louis, Missouri, all received kidneys from cadavers (Tables 1 and 2). The Montreal recipients are denoted by a number prefixed by the letter M, the London patients by their initials, and the St. Louis patient by SL1. The St. Mary's patients were subjected to splenectomy and bilateral nephrectomy at the time of transplantation; the others were not. Thymectomy was not performed.

In 48 of these cases the tissue was obtained at open biopsy; in two it was collected at autopsy. All the patients had been receiving daily azathioprine; 36 were receiving 2.5 to 25 mg. per day of prednisone; and 26 were being treated with one or more of the antihypertensive drugs chlorothiazide, hydralazine, reserpine, and methyldopa.

CONTROL PATIENTS

Renal Isograft (IDT2). This patient had undergone bilateral nephrectomy, but retained his spleen and thymus. At the time of open biopsy he was receiving no azathioprine or antihypertensive drugs.

Remaining Kidney from a Donor. A 32-year-old man was killed in an accident 2 years 4 months after donating a kidney to his brother (LD15). His remaining hypertrophied kidney (weight 300 gm.) was used as a control in this study.

LYMPHOCYTE TYPING

In 36 of the 50 patients retrospective typing of the lymphocytes of the donor and the recipient was undertaken. Blood lymphocytes obtained from the patients and from their donors were tested against a panel of 65 to 121 different cytotoxic antisera in the presence of rabbit complement.82 Those antisera tending to act alike had been previously classed together by a computer factor analysis program into groups. Each individual's lymphocytes were then "typed" with respect to the presence or absence of seven major factors by regression analysis.83 Incompatibilities between donors and recipients were then noted.

GLOMERULAR PERMEABILITY STUDIES

In those patients with moderate to heavy proteinuria, the selectivity of the protein loss was measured by Dr. J. S. Cameron of Guy's Hospital, London, England, who used a modification of the method first described by Soothill as mentioned by Joachim, Cameron, Schwartz, and Becker.89 The urinary clearance of five to seven plasma proteins, ranging in molecular weight from 40,000 to 1,300,000, was assessed by immunochemical means, and the clearance of each protein was then expressed as a percentage of the clearance of transferrin. The regression line of log clearance upon log molecular weight was calculated and the angle of this line, called \( \theta \), was taken as the index of selectivity.
Figs. 3 and 4
Tissue Processing

Tissue for light microscopy was fixed in 10 per cent neutral formalin or buffered formal saline, embedded in paraffin wax, and serially sectioned. The stains used routinely were hematoxylin and eosin, periodic acid-Schiff reagent (PAS), Weigert's for elastic tissue, picric-Mallory 3, Martius scarlet blue, Mallory's phosphotungstic acid-hematoxylin, and methyl green-pyronin. Other special stains were used when indicated. For electron microscopy small fragments of tissue were fixed in Palade's buffered osmium tetroxide (pH 7.4). After fixation at 4°C for 1 1/2 to 2 hours, the tissues were washed and then dehydrated in a graded series of ethanol solutions and embedded in an epoxy resin, usually Epon 812 but sometimes Araldite. Sections 0.5 μ thick, were cut on a Huxley ultramicrotome with glass knives and then stained with azur II. After identification of suitable glomeruli under the light microscope, ultrathin sections were cut and mounted both on naked and carbon-coated copper grids. After treatment with lead hydroxide, lead citrate, phosphotungstic acid, or a combination of lead hydroxide and uranyl acetate, the ultrathin sections were examined in a Siemens Elmiskop IA or an AEI 6B electron microscope operating at 80 kv. Some tissue from each biopsy was used for immunofluorescent study, the results of which will be reported separately.

RESULTS

Contralateral Donor Kidney

By light microscopy the only unusual glomerular feature of the remaining donor kidney was hypertrophy. Ultrastructurally the visceral epithelial cells contained several cytosomes and cytosegregosomes, and there were rare, tiny, focal areas of fusion of the foot processes (Fig. 1). These changes in fine structure were regarded as being within normal limits.

Renal Isograft

Apart from hypertrophy the glomeruli appeared to be normal by light microscopy. Ultrastructurally, small focal areas of fusion of the visceral epithelial foot processes were more common and larger than in the control donor kidney (Fig. 2). The epithelial cytoplasm that abutted against the capillary basement membranes contained aggregates of a dense, finely granular material. These areas of density were more pronounced than in normal glomeruli. The capillary basement membrane was of normal thickness in most places, the width being 3300 Å (mean of 250 measurements; range, 1600 to 5200 Å; standard deviation, 540). There were, however, several small subendothelial thickenings and irregularities. These were mostly of the same density as the normal lamina densa; a few were less dense. The endothelial cells were normal and the capillary lumina were patent and contained many red cells and an occasional platelet. There was a slight increase of mesangial matrix.

Renal Allografts

Biopsied During Rejection. Four of the 50 patients (LD27, LD54, AR, and CB) were biopsied during a rejection episode. This was a clinical diagnosis based upon the occurrence shortly before the biopsy of oliguria and a fall in creatinine clearance in a patient previously doing well. In two of the cases there was also an outpouring of lymphoid and tubular cells in the urine. The rejection episode was rapidly reversed in one of the patients (CB) with

Fig. 3. Biopsy of a renal allograft taken during a rejection episode which was later reversed by treatment with return of normal function to the graft (patient CB). A glomerular capillary loop (cap') is occluded by a large aggregate of intact platelets together with two erythrocytes (rbc). The absence of much interdigitation of pseudopods suggests that the platelets are only loosely adherent. An adjacent capillary loop (cap") is unobstructed. There is fusion of epithelial (ep) foot processes (arrow). ×7680.

Fig. 4. Same renal allograft as was shown in Figure 3. A glomerular capillary loop (cap) is almost occluded by aggregated platelets. At the periphery of the platelet mass, there are many bulbous pseudopods (ps). The platelet lysosomes and plasma membranes are mostly intact. There are a few fragments of fibrin (arrows). end, Endothelial cell. ×7500.
restoration of normal function, but the other three patients failed to respond to treatment and their renal allografts had ultimately to be removed.

Reversible Rejection. By light microscopy, in the case where intensified treatment with steroids and azathioprine produced recovery of renal function, there were pale, homogeneous masses occluding the capillary lumina in several of the glomeruli. There were also a few small areas in which the glomeruli showed periglomerular fibrosis and some increase of mesangial matrix. In the electron microscope the affected capillary loops were found to be blocked by aggregated masses of platelets (Fig. 3). At the periphery of the clumps many of the platelets had bulbous pseudopods which were pale and lacked lysosomes and mitochondria (Fig. 4). In most of the aggregates the platelet lysosomes and plasma membranes were still intact and there was not much interdigitation of pseudopods, which suggested that the platelets were only loosely adherent. Tiny amounts of fibrin were present in several of the clumps. In many of the capillaries there were small, focal, subendothelial accumulations of amorphous material on the basement membrane. A few platelets had inserted pseudopods between adjacent endothelial cells and were in contact with this altered membrane (Fig. 5). Both the endothelial and epithelial cells were enlarged, and their content of cytosomes, cytosomes, multivesicular bodies, and ribosomes was increased. The epithelial cells possessed microvilli, and there were areas of fusion of the foot processes.

Irreversible Rejection. In two patients these were second rejection episodes, beginning 1 month and 2 years respectively after transplantation; in the third patient a third rejection episode began at 1 year 10 months. Under light microscopy the majority of the glomeruli in each transplant showed thickening of the capillary basement membranes by PAS-positive material. Deposits of similar material were present in the mesangium of many tufts. A few of the glomeruli were hypercellular, either focally or diffusely. Several of the glomerular capillaries contained trapped neutrophils, small plugs of amorphous material, and fragments of phosphotungstic acid-hematoxylin-positive fibrin. A few showed adhesions between tuft and capsule. Rare cellular capsular crescents were present. Ultrastructurally the epithelial cells were enlarged and their cytoplasm contained increased numbers of ribosomes, cytosomes, cytosomes, and multivesicular bodies. The Golgi bodies were enlarged. Fusion of foot processes, although focal, was extensive (Fig. 6), and that part of the epithelial cytoplasm in contact with the capillary basement membranes was dark and granular. Almost all the capillary basement membranes were greatly thickened by subendothelial accumulations of an amorphous, finely granular material which had an electron density similar to normal lamina densa (Figs. 6 to 8). The lamina interna rara was lost, and there were some extensions of the amorphous material between endothelial cells. A few oval, much denser deposits lay in the lamina densa (Figs. 6 and 8). In some areas cytoplasmic fragments and organelles, together with fragments of fibrin and lipid particles, were embedded in the subendothelial deposit (Figs. 6 and 8). In a few places larger fibrin masses, which showed the characteristic periodicity of about 200 Å, were incorporated into the thickened wall and covered by a thin layer of endothelium (Figs. 6 and 7). The endothelial cells were greatly enlarged and contained increased numbers of free ribosomes, cytosomes, multivesicular bodies, and vacuoles. The amount of rough endo-

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Fig. 5. Same renal allograft as was shown in Figures 3 and 4. A glomerular capillary loop is obstructed by an aggregation of intact platelets (pl) and rare fibrin fragments (fine arrows). One platelet has inserted a pseudopod (broad arrow) between two of the endothelial cells lining the capillary and is making contact with the basement membrane. Focal deposits of homogeneous material (z) are present on the subendothelial aspect of this membrane and there is fusion of some of the epithelial foot processes (ep). end, Endothelial cell. ×10,300.
plasmic reticulum in these cells was greater than normal, and the Golgi bodies and centrolioles were prominent (Fig. 7). The capillary lumina were narrowed and contained erythrocytes, lymphoid cells, platelets, and occasional neutrophil polymorphonuclear leukocytes. The platelets, several of which showed loss of lysosomes and rupture of cell membranes, were usually in clumps intermixed with fibrin (Fig. 9). Neutrophils were found in association with the platelet-fibrin aggregates, completely occluding some capillary loops. Occasionally there appeared to be loss of the plasma membranes between the endothelial cells and the polymorphonuclear leukocytes (Fig. 9). The mesangial matrix was increased in amount and in several glomeruli contained small strands of fibrin. There were increased numbers of lysosomes, cytosegresomes, and ribosomes in the mesangial cells.

**Biopsied When Not in Rejection.** Forty-six of the 50 patients were biopsied at a time when there was no clinical evidence of rejection. In 24 of the renal allografts, light microscopy of ordinary sections 4 \( \mu \) thick showed that the glomerular capillary basement membranes were thickened. The other 22 kidneys appeared normal.

**Renal Allografts with Glomerular Capillary Basement Membrane Thickening Obvious in 4-\( \mu \) Sections (Group A).** In the majority (63 per cent) of the 24 renal allografts, the PAS-positive thickening of the capillary walls seen by light microscopy was focal within the glomerular tufts and associated with large mesangial deposits of similar material producing a picture resembling lobular glomerulonephritis. Less frequently this was a diffuse process resembling chronic membranous glomerulonephritis. These changes were sometimes accompanied by localized areas of increased tuft cellularity (21 per cent), and adhesions between tuft and Bowman's capsule (37 per cent). A few glomeruli contained cribriform areas in the tuft resembling those seen in radiation nephritis. Capsular crescents and lobular lesions containing trapped erythrocytes and polymorphonuclear leukocytes were rare except in two cases: RT where the former lesions were so common as to give the picture of a rapidly progressive acute glomerulonephritis, and M17 where the latter lesions were common. Examination of plastic-embedded sections 0.5 \( \mu \) thick revealed another feature which distinguished the renal allografts of M17 and RT from the other 22 kidneys in this group. Only these two kidneys showed large hemispheric nodules, or "humps," on the epithelial side of the capillary basement membrane.

The electron microscopic appearances of the glomeruli are summarized in Table 3. In all cases the capillary basement membrane was altered by subendothelial accumulations of amorphous material similar to that seen in the three transplants that were biopsied during irreversible rejection (Fig. 10). The deposits obliterated the lamina interna rara, but the lamina densa was generally normal. The density and compactness of the deposits varied; in many capillary loops, rarefied areas were present (Figs. 11 and 12). In a few of the tufts it was difficult to distinguish deposit from lamina densa, and the over-all effect was of a greatly thickened basement membrane (Fig. 13). In
Figs. 8 and 9
TABLE 3. **glomeruler Ultrastructural Changes in 50 Renal Allografts from Patients Treated with Azathioprine and Prednisone**

<table>
<thead>
<tr>
<th>Type of change</th>
<th>Kidneys showing various ultrastructural features*</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Group A</td>
</tr>
<tr>
<td>Visceral epithelial cells</td>
<td></td>
</tr>
<tr>
<td>Enlarged</td>
<td>75</td>
</tr>
<tr>
<td>Fusion of foot processes</td>
<td>100</td>
</tr>
<tr>
<td>Microvilli</td>
<td>50</td>
</tr>
<tr>
<td>Increased number of organelles</td>
<td>67</td>
</tr>
<tr>
<td>Enlarged Golgi bodies</td>
<td>67</td>
</tr>
<tr>
<td>Capillary basement membranes</td>
<td></td>
</tr>
<tr>
<td>Subendothelial deposits</td>
<td>100</td>
</tr>
<tr>
<td>Subepithelial &quot;humps&quot;</td>
<td>8</td>
</tr>
<tr>
<td>Deposits in lamina densa</td>
<td>17</td>
</tr>
<tr>
<td>Extension of deposit between endothelial cells</td>
<td>96</td>
</tr>
<tr>
<td>Incorporation of cell fragments in deposit</td>
<td>88</td>
</tr>
<tr>
<td>Endothelial cells</td>
<td></td>
</tr>
<tr>
<td>Increased numbers</td>
<td>21</td>
</tr>
<tr>
<td>Enlarged</td>
<td>75</td>
</tr>
<tr>
<td>Increased number of organelles</td>
<td>71</td>
</tr>
<tr>
<td>Enlarged Golgi bodies</td>
<td>33</td>
</tr>
<tr>
<td>Capillary lumina</td>
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</tr>
<tr>
<td>Narrowed</td>
<td>71</td>
</tr>
<tr>
<td>Obstructed</td>
<td>29</td>
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<tr>
<td>Containing impacted polymorphonuclear leukocytes</td>
<td>17</td>
</tr>
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<td>Containing monocytes</td>
<td>42</td>
</tr>
<tr>
<td>Containing lymphoid cells</td>
<td>54</td>
</tr>
<tr>
<td>Mesangium</td>
<td></td>
</tr>
<tr>
<td>Increased numbers of cells</td>
<td>42</td>
</tr>
<tr>
<td>Increased amount of matrix</td>
<td>92</td>
</tr>
<tr>
<td>Increased number of organelles in cells</td>
<td>63</td>
</tr>
</tbody>
</table>

* The 46 kidneys in groups A and B were biopsied at a time when the patient was not in a clinical rejection episode. Group A consists of 24 renal allografts which by light microscopy showed obvious glomerular capillary basement membrane thickening. Group B is composed of 22 renal allografts with normal glomerular capillary basement membranes by light microscopy. The whole series comprises 50 renal allografts and includes four which were examined during a clinical rejection episode.

37 per cent of the allografts, the deposits affected all the capillaries examined, but in the majority the change was focal, and normal loops were present. The deposits bulged into the capillary lumina and were covered by an intact endothelial layer (Figs. 11, 14, and 15). The material also extended between endothelial cells (Fig. 14). Really dense deposits in the lamina densa or in the subendothelial layer were

**Fig. 8.** Enlargement of part of Figure 6. The lamina densa (ld) contains an oval, dense deposit (d). Between the endothelial cell (end) and the lamina densa there is an accumulation of cell fragments (cf), organelles (or), and fibrin (f) embedded in an amorphous, finely granular material. The cytoplasm of the endothelial cell contains increased numbers of ribosomes (arrow). ep, Epithelial cell. ×17,700.

**Fig. 9.** Renal allograft biopsied during a rejection episode which later proved irreversible (patient LD27). A glomerular capillary loop is obstructed by an aggregate of platelets (pl) and fibrin (f), and by a neutrophil polymorphonuclear leukocyte (poly). There appears to be breakdown of the plasma membranes separating the polymorphonuclear leukocyte and the endothelium lining the capillary at the point marked with an arrow. There is fusion of the epithelial foot processes (ep). mc, Mesangial cell. ×7180.
Fig. 10. Biopsy of a renal allograft 1 year 10 months after transplantation (LD41). The basement membranes (bm) of three glomerular capillary loops (cap) are diffusely thickened by a subendothelial accumulation of amorphous material. The epithelial cells (ep) are enlarged and possess microvilli (m). Rbc, Erythrocyte; us, urinary space; end, endothelial cell. ×6100.
unusual (Fig. 15). Sometimes it was as though the lamina densa had been split and a broad band of loosely arranged material inserted between the two layers (Fig. 16). Fragments of cell cytoplasm and isolated organelles were commonly incorporated in the subendothelial accumulations. In a few allografts the cell fragments were large and, in serial sections, some could be shown to be extensions from mesangial cells; others appeared to be endothelial, and a few resembled platelets (Fig. 17). Subepithelial deposits were rare being present in only three of the 24 kidneys; they were always combined with subendothelial collections. In one patient (SL1) they consisted of small foci with prominent rarefied areas, but in the other two kidneys (M17 and RT) there were dense hemispheric humps on the epithelial side of the capillary basement membranes (Figs. 18 and 19). In almost all of the kidneys the mesangial matrix was increased (Figs. 13 and 20), and in 42 per cent there was hyperplasia of the mesangial cells. The endothelial cells were enlarged in 75 per cent of the transplants; in the five most severely affected kidneys, they were also increased in number. Their cytoplasm was voluminous and was the cause of narrowing of many and obliteration of a few of the capillary lumina (Fig. 13). A few kidneys showed capillary obstruction as a result of impaction and disintegration of neutrophil polymorphonuclear leukocytes; this was particularly a feature of the two kidneys with subepithelial humps (Figs. 21 and 22). Monocytes and lymphoid cells were frequently found in groups filling the capillary lumina. Platelets were sometimes present, but not clumped together. The visceral epithelial cells were enlarged in most of the transplants (Fig. 23); occasionally trimucleate forms were encountered (Fig. 24). In all cases there were areas in which the epithelial foot processes were fused producing a continuous sheet of cytoplasm over the capillary basement membranes (Fig. 25), but only in a few allografts was this change widespread. Epithelial microvilli (Fig. 10) were present in several of the kidneys. In many of the transplants the epithelial, endothelial, and mesangial cells possessed increased numbers of cytosomes, cytosomes, and free ribosomes, increased amounts of rough endoplasmic reticulum, and enlarged Golgi bodies. Multivesicular bodies were more common than usual in the endothelial and epithelial cells.

Renal Allografts with Normal Glomerular Capillary Basement Membranes by Light Microscopy (Group B). Of the 22 renal allografts that appeared normal on light microscopy, all were abnormal on electron microscopy in that they showed fusion of their epithelial foot processes (Table 3). In many this was of slight degree and, although more than was seen in the control donor kidney, it was comparable to the amount of change found in the renal isograft. Only 13 (59 per cent) of these kidneys had normal capillary basement membranes; the others all showed small, focal, subendothelial deposits of the same nature as were encountered on a much larger scale in group A (Fig. 26). An appreciable number of the epithelial cells were enlarged; these, together with some of the endothelial and mesangial cells, showed increased numbers of ribosomes, cytosomes, and cytosegresomes. Monocytes and lymphoid cells were more common in the capillaries of these transplants than in those of the isograft or the control donor kidney. Several of the kidneys showed more mesangial matrix than normal.

Correlation of Glomerular Changes with Degree of Compatibility of Donor and Recipient as Shown by Lymphocyte Typing

In 36 of the 50 patients retrospective typing of the lymphocytes of donor and recipient was undertaken. It will be seen from Table 4 that in general the allografts with normal glomeruli came from patients who were regarded as being compatible with their donors by lymphocyte typing, while the majority of grafts with glomerular capillary basement membrane thickening came from patients who were incompatible with their donors in one or more major lymphocyte groups. This association of lymphocyte incompatibility with glomerular damage in the renal graft was statistically significant ($p = 0.02$).
ASSOCIATION OF GLOMERULAR DAMAGE WITH ALTERATIONS IN RENAL FUNCTION

The mean creatinine clearance in the 27 patients with marked glomerular lesions in their allografts was 65.4 ml per minute with a range of 4 to 139 ml per minute, compared with a mean of 92.8 ml per minute with a range of 33 to 184 ml per minute for the 23 patients without obvious glomerular lesions (Tables 1 and 2). The urine produced by the allografts with glomerular capillary basement membrane thickening contained more protein (mean 84 mg per 100 ml; range 0 to 350 mg per 100 ml) than that excreted by the transplant with normal glomeruli (mean 16.7 mg per 100 ml; range 0 to 100 mg per 100 ml). The patients with proteinuria over 100 mg per 100 ml showed moderately unselective protein loss with an index of selectivity (θ) in no instance lower than 62.4°. There was no obvious correlation between this value and the degree or type of ultrastructural change present in the glomerular capillary basement membranes.

DISCUSSION

Our findings fully justify the stress that Hamburger and associates11-13, 15 have placed upon the glomerular changes that may occur in long-surviving renal allografts. Marked lesions were present in many of the glomeruli in 27 of the 50 allogeneic kidneys we examined. In the majority the most striking abnormality was an accumulation of amorphous material on the subendothelial aspect of the capillary basement membranes. When present in appreciable amounts the deposit was easily detectable by light microscopy as a

<table>
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<tr>
<th>Table 4. Association of Lymphocyte Incompatibility between Donor and Recipient, with Presence of Marked Glomerular Damage in the Renal Allografts</th>
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<tbody>
<tr>
<td>Histopathology</td>
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<tr>
<td></td>
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<tr>
<td>Compatible</td>
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<tr>
<td>Allografts showing obvious glomerular capillary basement membrane thickening under light microscopy</td>
</tr>
<tr>
<td>Allografts without significant glomerular capillary basement membrane changes</td>
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<tr>
<td>Total</td>
</tr>
</tbody>
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*χ² = 5.45; p = 0.02.

PAS-positive thickening of the glomerular capillary wall. This change was accompanied by increased amounts of basement membrane-like material in the mesangium, by hypertrophy of the epithelial and endothelial cells, by fusion of some of the epithelial foot processes, and sometimes by hyperplasia of the mesangial cells.

At present the incidence of these glomerular lesions is unknown. Some indication is given by considering the first Denver series of renal allografts in which 70 kidneys were transplanted into 64 patients. In the following 1½ years, 34 of these kidneys were examined, either after

Fig. 11. Renal allograft 1 year 9 months after transplantation (LD18). Segment of glomerular capillary wall in which the basement membrane is thickened by a subendothelial deposit of mixed density and compactness. The inner border of the lamina densa is indicated by arrows. The lamina interna rara has disappeared. ep, Capillary lumen; end, endothelial cell; ep, epithelial cell. ×13,800.

Fig. 12. Part of a glomerular capillary loop from the same renal allograft as was shown in Figure 10. The subendothelial amorphous deposit (dep) varies in compactness; dense areas (arrow) are intermixed with paler areas. ua, Urinary space. ×9200.

Fig. 13. Renal allograft 1 year 10 months after transplantation (LD40). In this kidney it is difficult to distinguish deposit from lamina densa, and the over-all effect is of greatly thickened capillary basement membranes. One capillary (arrow) is occluded by enlarged endothelial cells (end). The amount of mesangial matrix (mm) is increased. ms, Mesangial cell. ×3680.
Figs. 14 to 16
surgical removal or at autopsy; only two showed glomerular capillary basement membrane thickening. However, 28 of these kidneys had functioned for less than 3 months. Of the 35 long-surviving patients, 34 were biopsied at about 2 years after transplantation and are reported in detail in the present paper. Eighteen biopsies showed glomerular basement membrane changes obvious on light microscopy, an incidence of 53 per cent, and a further six biopsies, 17 per cent, which appeared normal on light microscopy showed minor subendothelial deposits on electron microscopy.

In most patients the glomerular lesions were accompanied by mild proteinuria (Table 1) which usually commenced 6 months or longer after renal transplantation. In a few cases the protein loss was moderately heavy and unselective in pattern, but this was not associated with edema. Microscopic hematuria and hypertension were not features of the syndrome in our patients, but were described by Hamburger et al. in three of his patients. One of the French cases (patient 7) also developed hypergammaglobulinemia, splenomegaly, and marked hyperplasia of the Kupffer cells of the liver. In several of our patients there were no accompanying signs or symptoms, and the diagnosis was made only after microscopic examination of a biopsy of the graft. Generally the glomerular filtration rate of these patients was impaired, but there was much individual variation and the values were greatly influenced by the presence or absence of accompanying vascular lesions.

The etiology of the glomerular changes is undetermined. It is tempting to think that they result from the production by the host of antibody which becomes deposited on the glomerular capillary basement membranes of the graft. In support of this hypothesis is the finding of a close correlation between incompatibility of host and donor as shown by lymphocyte typing and the occurrence of severe glomerular lesions. The development of splenomegaly and hypergammaglobulinemia in the patients of Hamburger et al. together with reports of rises in the immunoglobulin G levels of other patients and of dogs with renal allografts suggest increased antibody formation by the recipient. Also, subendothelial accumulations of amorphous material, much like those seen in our cases, can be produced in the glomeruli of experimental animals by the injection of nephrotoxic serum, and they occur in the glomeruli of patients suffering from acute glomerulonephritis. The childhood form of lipoid nephrosis, and lupus nephritis. In all these situations the deposits have been shown to contain γ-globulin. However, the fact that similar lesions can be induced in animals by the administration of Freund's adjuvant, synthetic polysaccharides, and ethionine and carbon tetrachloride, indicates a need for caution in regarding these collections of γ-globulin as necessarily indicating immune injury.

In our patients we have no evidence to suggest that drug-induced injury played any part in the production of the renal changes. It is known that azathioprine does not damage renal autografts when administered for 1 year in doses comparable to those used in this study, and although hydrocortisone-like steroids can

**FIG. 14.** Renal allograft 1 year 10 months after transplantation (LD63). Small segment of glomerular capillary wall. A band of amorphous material extends between two endothelial cells (arrow). ep, Epithelial cell; end, endothelial cell; cap, capillary lumen. ×11,400.

**FIG. 15.** Part of a glomerular capillary wall from a renal allograft biopsied 2 years after transplantation (LD48). In the amorphous subendothelial deposit there is a roughly spherical focus (arrow) which is denser than the lamina densa (ld). ×18,800.

**FIG. 16.** Biopsy of a renal allograft 1 year 11 months after transplantation (LD51). The basement membrane (bm) is thickened by a broad band of loosely arranged amorphous material which appears to be sandwiched between two layers of lamina densa. Scattered organelles are incorporated in the deposit. An epithelial cell contains a large cytosome (cy). us, Urinary space. ×7700.
produce glomerular lesions,\textsuperscript{33} ultrastructurally these are distinctive and bear little resemblance to the changes we have described. Furthermore, a few of our patients with glomerular changes had not been treated with steroids, and Hamburger and associates' patient 5 had not even received azathioprine.\textsuperscript{12} Glomerular basement membrane thickening has also been encountered in the renal allografts of 36 per cent of a group of dogs which survived beyond 70 days because of treatment with various immunosuppressive agents.\textsuperscript{39} Several of these animals did not receive azathioprine and none were given steroids. It is significant that the glomerular changes were most pronounced in an animal in which the renal allograft functioned for 11/2 years after all treatment had been withdrawn.\textsuperscript{39}

The possibility that the glomerular lesions were the result of the recipient's original disease has to be considered. Of the 27 patients who later developed pronounced basement membrane deposits in their allografts, two suffered from bilateral polycystic disease of their own kidneys and three from chronic pyelonephritis (Table 1). In these five cases it seems improbable that the initial disease contributed in any way to the changes that later occurred in the transplant. The other 22 patients all suffered from glomerulonephritis and could conceivably have subsequently developed this same condition in their allografts. Although experimentally it has not yet proved possible to transmit nephrotoxic serum nephritis to renal allografts,\textsuperscript{55} this very complication has been reported in five renal isografts\textsuperscript{30} with the production of glomerular capillary basement membrane thickening and capsular crescent formation.\textsuperscript{37} If this were the explanation of the glomerular findings in our cases, it is surprising that the lesions were subendothelial in five of the allografts which had been transplanted into patients whose original disease was the diffuse membranous form of glomerulonephritis, a condition which would have been expected to produce predominantly subepithelial deposits. Because of these findings we think that transmission of glomerulonephritis from the recipient does not explain most of the cases of glomerular damage that are being encountered in long-surviving transplants and that production by the host of antibody directed specifically against the graft is more likely. Two of the 27 patients, however, were in a different category. Not only did their transplants show distinctive, large subepithelial humps, but both these patients were suffering from active glomerulonephritis at the time of renal transplantation. Subepithelial humps are characteristic of poststreptococcal glomerulonephritis,\textsuperscript{17, 21} and as both patients developed their nephritis after a streptococcal infection, we feel that in these two cases it is very probable that the changes in the transplant were induced in some way by the recipient's original disease. In at least two other renal allografts, a similar course of events has been postulated.\textsuperscript{16, 38}

The prognosis for patients with glomerular lesions in their transplants depends upon the reversibility or otherwise of the subendothelial deposits. It is known

![Fig. 17. Biopsy of renal allograft 2 years 1 month after transplantation (LD45). Small segment of glomerular capillary wall in which the basement membrane is thickened by a subendothelial deposit containing large cell fragments (cf). Most of the trapped cells appear to be mesangial or endothelial, but a few (cf) might be platelets. The inner limit of the lamina densa is marked by arrows. as, Urinary space; end, endothelial cell; cap, capillary lumen; rbc, erythrocyte. ×13,500.](image)

![Fig. 18. Part of a glomerular capillary loop from a renal allograft 1 year after transplantation into a patient suffering from rapidly progressive acute glomerulonephritis (patient RT). There is a dense hemispheric hump (hu) on the epithelial side of the thickened basement membrane (bm). The foot processes of the epithelial cell (ep) are fused over the deposit, and the adjacent epithelial cytoplasm is dark and granular (arrow). ×17,000.](image)

![Fig. 19. Renal allograft 7 months after transplantation. There is a typical subepithelial hump (hu) on the thickened glomerular capillary basement membrane (bm). The epithelial cytoplasm surrounding the deposit is dark and granular (arrow). ×19,200.](image)
that accumulations of similar material resolve in acute glomerulonephritis, probably by phagocytosis by endothelial and mesangial cells.\textsuperscript{12} However, although two of the three patients with renal allografts, first described by Hamburger et al. in 1963,\textsuperscript{13} later lost their proteinuria and were well 1 year 6 months and 2 years 10 months later, abnormalities persisted in the fine structure of the glomeruli in their transplants.\textsuperscript{14} The third patient developed progressive renal insufficiency and died 10 months later. Among our 27 patients (Table 11), four (15 per cent) have died since biopsy; one of these was a case in which the allograft may have been affected by the recipient's original poststreptococcal glomerulonephritis. In two of the other three patients, progression of the glomerular lesion played a major part in causing the patient's death. A further five renal allografts (18.5 per cent) have had to be removed at periods of up to 7 months after biopsy because of progressive renal failure. Five patients (18.5 per cent) have a creatinine clearance below 50 ml. per minute, and 13 (48 per cent) have a clearance above this value 11 months to 2 years after biopsy. By contrast, none of the renal allografts with minimal glomerular changes have had to be removed in the period since biopsy; there have been no deaths in this group, and 20 (87 per cent) of the patients have a creatinine clearance above 50 ml. per minute. (Table 2). Although it is far too early to be dogmatic, these findings suggest that the prognosis for patients with marked glomerular capillary basement membrane thickening in their transplants may be worse than was first thought.

In this paper further light is also shed upon the mechanism of renal shutdown during clinical episodes of allograft rejection in patients receiving immunosuppressive therapy. The glomerular capillaries of four transplants biopsied during such an episode were found to be obstructed by platelet aggregates, and there was also a subendothelial deposit of amorphous material (which proved to be \textgamma-globulin on immunofluorescence) on the basement membranes. Some platelets were also found to have inserted pseudopods between the endothelial cells onto the altered capillary basement membranes. Platelet thrombi were also present in the glomerular capillaries of Hamburger and associates' patient 17 during a rejection episode which later responded to treatment. A repeat biopsy of this kidney 4 months later no longer contained platelet aggregates.\textsuperscript{13} As it is known that platelet aggregation can be induced by contact with antigen-antibody complexes,\textsuperscript{1,26} a possible explanation for these findings is that the arterioles and capillaries of the allografts became coated with antibody and that this layer induced clumping of platelets. Experimentally, it has been shown that platelet aggregates can profoundly affect renal blood flow,\textsuperscript{16} and scattered throughout a transplanted kidney they would explain the alterations in water, electrolyte, and creatinine excretion that occur during a rejection episode.\textsuperscript{45} In \textit{vitro} adenosine diphosphate-induced platelet aggregation can be quickly inhibited by prednisone,\textsuperscript{42} and this may explain why prompt treatment with steroids has been so successful in inducing a diuresis from renal allografts that have shortly before suddenly ceased to function. When aggregation is followed by platelet damage and the appearance of fibrin, then the process is probably irreversible, and our morphologic findings suggest that in rejection episodes resistant to treatment a mixture of amorphous material, probably antibody, together with platelet fragments and strands of fibrin become covered by a new layer of endothelium and incorporated.

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\textbf{Fig. 20.} Biopsy of renal allograft 1 year 5 months after transplantation (patient EN). The mesangial matrix (\textit{mam}) is increased in amount and the mesangial cells (\textit{mc}) are conspicuous. The capillary walls are thickened by subendothelial deposits of amorphous material (\textit{z}). \textit{rbc}, Erythrocyte; \textit{cap}, capillary lumen; \textit{end}, endothelial cell. \textit{X}5600.

\textbf{Fig. 21.} Same renal allograft as was shown in Figure 19. A glomerular capillary (\textit{cap'}) is obstructed by three impacted and disintegrating neutrophil polymorphonuclear leukocytes (\textit{p}). Amorphous deposit is present between the polymorphonuclear leukocytes and on the basement membrane (\textit{arrow}). An adjacent capillary (\textit{cap''}) is unobstructed. \textit{ep}, Epithelial cell. \textit{X}5300.
into the glomerular capillary wall. Radioactive labeling of platelets in patients with renal allografts has produced further evidence compatible with this hypothesis. Just before a rejection episode, there is a fall in the number of circulating platelets accompanied by evidence of their sequestration in the transplant. In those cases where the rejection yields to treatment, the platelet count returns to normal; in patients with irreversible rejection there is no evidence that the platelets ever leave the graft.26

**SUMMARY**

The light and electron microscopic appearances of the glomeruli are described in 50 renal allografts which were examined 43 days to 2 years 3 months after transplantation into patients receiving immunosuppressive treatment. A renal isograft and a donor's remaining kidney were used as controls.

The glomeruli were hypertrophied in all the kidneys. The control donor kidney showed no other significant changes. There were small focal areas of fusion of the visceral epithelial foot processes and a slight increase of mesangial matrix in the renal isograft.

In the renal allografts the epithelial and endothelial cells were hypertrophied, and there was more fusion of the epithelial foot processes than was found in the renal isograft. Subendothelial accumulations of amorphous material were present on the glomerular capillary basement membranes of 37 of the allografts. In 27 of the kidneys these deposits were large and the basement membrane thickening was obvious under light microscopy; in 10 they were small and produced no thickening visible in ordinary sections. Increased amounts of basement membrane-like material were present in the mesangium of those kidneys with subendothelial deposits, and there was sometimes hyperplasia of the mesangial cells.

These changes in the renal allografts were usually associated with mild proteinuria and impairment of glomerular filtration rate. Occasionally the proteinuria was more severe and unselective in type. A nephrotic syndrome was not produced. There was a close association between the occurrence of marked glomerular deposits and incompatibility of host and donor as shown by lymphocyte typing. Evidence is produced that these lesions are not drug-induced and in the majority are unlikely to have been caused by the patient's original disease. It is suggested that they are caused by deposition of circulating antibody directed specifically against the graft.

Two of the transplants were exceptions, in that they showed additional, distinctive subepithelial humps, and in these cases it was considered probable that there had been transmission of the recipient's active glomerulonephritis to the allograft.

Four of the renal allografts which were biopsied during a clinical rejection episode contained platelet aggregates in many of the glomerular capillaries. The ischemia caused by this clumping of platelets may explain the alterations in water, electrolyte, and creatinine concentration that are found in patients undergoing acute rejection.

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REFERENCES


Fig. 24. Renal allograft previously shown in Figures 10 and 12. A trinucleate visceral epithelial cell is shown. Its cytoplasm contains many ribosomes and much rough endoplasmic reticulum. The adjacent capillary basement membrane is greatly thickened (arrows) by a subendothelial deposit. a, Nucleus; us, urinray space. ×4040.

Fig. 25. Biopsy of renal allograft 1 year 10 months after transplantation (LD60). There is fusion of most of the foot processes (arrows) of a glomerular epithelial cell (ep). Another epithelial cell contains a cytosome (cy). In this area the capillary basement membranes are normal. cap, Capillary lumen. ×7150.

Fig. 26. Renal allograft 1 year 10 months after transplantation (patient KF). Under light microscopy the glomerular capillary basement membranes appeared normal. In this electron micrograph small, focal, subendothelial deposits can be seen (arrows), which are of the same nature as those encountered on a larger scale in allografts which had thickened capillary basement membranes by light microscopy. end, Endothelial cell. ×7550.


