Polyomavirus Infection of Renal Allograft Recipients: From Latent Infection to Manifest Disease

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Abstract. Polyomavirus (PV) exceptionally causes a morphologically manifest renal allograft infection. Five such cases were encountered in this study, and were followed between 40 and 330 d during persistent PV renal allograft infection. Transplant (Tx) control groups without PV graft infection were analyzed for comparison. Tissue and urine samples were evaluated by light microscopy, immunohistochemistry, electron microscopy, and PCR. The initial diagnosis of PV infection with the BK strain was made in biopsies 9 ± 2 mo (mean ± SD) post-Tx after prior rejection episodes and rescue therapy with tacrolimus. All subsequent biopsies showed persistent PV infection. Intranuclear viral inclusion bodies in epithelial cells along the entire nephron and the transitional cell layer were histologic hallmarks of infection. Affected tubular cells were enlarged and often necrotic. In two patients, small glomerular crescents were found. In 54% of biopsies, infection was associated with pronounced inflammation, which had features of cellular rejection. All patients were excreting PV-infected cells in the urine. PV infection was associated with 40% graft loss (2 of 5) and a serum creatinine of 484 ± 326 μmol/L (mean ± SD; 11 mo post-Tx). Tx control groups showed PV-infected cells in the urine in 5%. Control subjects had fewer rejection episodes (P < 0.05) and stable graft function (P = 0.01). It is concluded that a manifest renal allograft infection with PV (BK strain) can persist in heavily immunosuppressed patients with recurrent rejection episodes. PV mainly affects tubular cells and causes necrosis, a major reason for functional deterioration. A biopsy is required for diagnosis. Urine cytology can serve as an adjunct diagnostic tool.

Cytomegalovirus (CMV) is the virus most frequently identified in kidney transplant recipients (1–3). An infection caused by polyomavirus (PV) is exceptional (2,3). This rarity is also underscored by the observation that we had not detected morphologic evidence of a PV infection in renal allografts among 616 kidney transplant recipients managed at our institution between January 1985 and December 1995. PV is a nonenveloped, double-stranded DNA virus. Two strains are associated with disease in humans: BK virus and JC virus (4). In immunocompetent individuals, PV has no great clinical significance, although 60 to 80% of adults are serologically positive (4–8). In healthy individuals, PV resides in a latent state in the kidney (4,7,9–11) and can be activated without functional impairment or ill effects (12–14). Morphologic evidence of viral activation is the presence of PV-infected cells in the urine (intranuclear inclusion cells, or so-called “decoy cells”) (12,14).

In immunocompromised patients, PV can cause a morphologically manifest renal infection with cytopathic signs and functional impairment, a condition termed PV disease. In native and transplanted kidneys, PV (BK virus strain) is found in areas of interstitial nephritis (15–19). In addition, renal allograft recipients were reported to suffer from PV (BK virus)-associated ureteral stenosis, and bone marrow transplant recipients from hemorrhagic cystitis (20–22). However, in the latter group a causative pathobiologic association is disputed by some (12,23–25). A pathogenetic significance is well established for JC virus, which causes multifocal leukoencephalopathy, mainly in AIDS patients (4,7,26).

Although, PV disease is exceptionally rare, we encountered five cases in renal transplant recipients within 8 mo. Because detailed clinical and histologic information was available from all patients before and after manifestation of PV disease, we had a unique opportunity to characterize typical changes.

Materials and Methods

Histology and Urine Cytology

Morphologic changes in tissue samples (25 biopsies and two graft nephrectomies) from five patients who contracted PV disease were studied. PV disease is defined as a morphologically manifest renal infection with cytopathic signs accompanied by varying degrees of interstitial inflammatory cell infiltrates and functional impairment. Thirteen tissue samples (11 biopsies and two graft nephrectomies) were obtained during
the course of persistent PV disease (up to 330 d after the initial diagnosis). Fourteen biopsies were taken before the initial manifestation, i.e., before the first histologic diagnosis of PV infection (including negative workup by immunohistochemistry [IHC]; see below).

For comparison, 108 renal allograft biopsies from patients lacking any morphologic evidence of PV disease were evaluated (including negative workup by IHC; see below). All of the 133 biopsies were performed due to deterioration of graft function at similar time intervals posttransplantation. Thirteen distinct histologic criteria were analyzed and semiquantitatively graded on a scale of 1 (minimal change) to 4 (marked change): extent of mononuclear cell infiltrate in the interstitium and tubulitis; vascular rejection (VR) with necrosis; transplant endarteritis; sclerosing VR (“chronic vascular rejection”); transplant glomerulopathy; transplant glomerulonephritis; extent of interstitial fibrosis; ischemic tubular damage; cyclosphere-induced tubular damage; toxic arteriolopathy; arteriolosclerosis; glomerulonephritis (immune complex-mediated); and pyelonephritis. A diagnosis of acute renal allograft rejection was based on the criteria of the Cooperative Clinical Trials in Transplantation classification scheme (27,28). The histologic evaluation was performed according to standard protocols by light microscopy (hematoxylin and eosin, periodic acid-Schiff, trichrome, methenamine silver stains) accompanied by IHC and electron microscopy. For routine IHC, a standard panel of antibodies was used (directed against IgG, IgM, IgA, complement factors C3, C4, C5b–9, C1q, and fibrin).

Voided urine samples were analyzed at time of biopsies as a standard patient management procedure. Urine cytology reports were retrospectively evaluated from 483 renal transplant recipients (including five patients with PV disease) managed in Basel between 1985 and 1997. The presence of PV-infected intranuclear inclusion cells, so-called “decoy cells,” as a morphologic marker for viral replication was semiquantitatively recorded as absent, scant (1 to 4 per 10 high-power fields [HPF]), or abundant (>5 per 10 HPF). From all patients with abundant decoy cell excretion (>5 per 10 HPF in at least one urine sample), renal allograft biopsies were (re-)examined (including IHC; see below) to search for cytopathic signs of a PV graft infection.

**Immunohistochemical Detection of Viruses**

PV was detected by indirect IHC on urine cytology, and tissue samples (fresh frozen and formalin-fixed). Several mouse monoclonal antibodies (mAb) were used to detect different PV strains: (1) mAb detecting both BK and JC strains (Readysysteme, Bad Zurzach, Switzerland); (2) mAb detecting JC strain (Readysysteme); (3) mAb detecting the SV40 large T antigen (Calbiochem/Oncogene Research Products, Cambridge, MA). The latter antibody reacts with primate (SV40) and human PV (BK and JC strains) and works after formalin fixation, using the microwave technique for antigen retrieval (Tris-HCl buffer, pH 10.5, 15 min at 100°C, followed by overnight incubation with the primary antibody at 4°C, dilution 1:3200). All staining procedures were performed according to a previously described protocol with the avidin-biotin-peroxidase technique (29). DAB (diaminobenzidine tetrahydrochloride) and AEC (3- amino,9-ethylcarbazole) were used as chromogens. All biopsies were also analyzed for the presence of CMV, Epstein–Barr virus (EBV), herpes simplex virus (HSV) and adenosine antigens (CMV [IEA]; mouse monoclonal antibody, Biosoft, Vairilhes, France; CMV [EA] and EBV [LMP, ZEBRA, EBNA]; mouse monoclonal antibodies, Dako, Glostrup, Denmark; HSV [types 1 and 2]; rabbit polyclonal antibody, Dako, Glostrup, Denmark; adenovirus: mouse monoclonal antibody, Chemicon, Temecula, CA).

**PCR Studies**

The genome detection of polyomavirus JC strain and BK strain used a modified semi-nested format based on a previous report (30). The outer primers 5'-AAG TCT TTA GGG TCT TCT AC-3' and 5'-GTG CCA ACC TAT GGA ACA GA-3' amplified a fragment of 176 bp common to both viruses. The inner primer pair combined the common 5'-AAG TCT TTA GGG TCT TCT AC-3' with either the BK-specific primer 5'-GAG TCC TGG TGG AGT TCC-3' to obtain a BK fragment of 149 bp, or the JC-specific primer 5'-GAA TCC TGG TGG AAT ACA-3' yielding a JC fragment of 146 bp. Both were confirmed by dideoxy sequencing (H.H.H., personal observation). DNA was prepared by protease K digestion and purification on silica spin columns (QiAmp, Qiagen, Basel, Switzerland) from the following sources: one nephrectomy from a patient with manifest PV disease and voided urine from five patients with PV disease.

A total of 0.5 μg of DNA was used per reaction. Amplification was performed after preincubation (37°C, 4 min) for uracyl-N-glycosylase deamination followed by 94°C at 30 s, 50°C at 45 s, and 72°C at 60 s (30 cycles, for both the outer and the inner amplification). Ten-microliter aliquots of the inner PCR product were separated on NuSiVee:Agoarese (3%-1%) electrophoresis gels. The threshold of detection was determined to be five genomes. Two parallel runs were analyzed for each result, and if equivocal, repeated. The results were interpreted as positive (2 of 2 runs positive); weakly positive (1 of 2 runs positive, viral load close to detection threshold); negative (2 of 2 runs negative).

**Results**

### PV Disease

**Patients.** Five patients (mean age at transplantation, 46 ± 2 yr; range, 27 to 57; four men and one woman) showed a morphologically manifest renal allograft infection due to polyomavirus (BK strain). PV infection became manifest and was histologically diagnosed several months after transplantation (9 ± 2 mo, mean ± SD). Twelve patients showed no evidence of PV infection (n = 14) lacked cytopathic signs of viral infection by light microscopy or IHC. Three patients had received a cadaver kidney, and two received organs from living unrelated donors. All of the five patients had a complicated posttransplantation course with recurrent rejection episodes. Before a manifest infection was diagnosed, immunosuppression had been switched in all patients from cyclosporine to high-dose tacrolimus rescue therapy for 5 ± 3 mo (mean ± SD) (Figure 1). Patients were kept on tacrolimus; three patients were switched back to cyclosporine 98 ± 46 d (mean ± SD; range, 49 to 140 d) after the initial diagnosis of PV disease (Figure 1).

**Cytopathic Changes.** PV disease was diagnosed histologically. The morphologic characterization was based on light microscopy, immunohistochemistry and electron microscopy. The diagnostic hallmark was the detection of intranuclear viral inclusions, which were exclusively found in epithelial cells (with the exception of podocytes). All tissue samples (n = 13) from patients with PV disease (n = 5) showed viral inclusions. Three types of intranuclear inclusion bodies were seen by light microscopy with similar frequencies along the entire nephron (Figure 2): (1) a homogeneous ground-glass appearance; (2) a central eosinophilic granular appearance with a (mostly) incomplete halo; (3) a more homogeneous, finely granular pattern. Cells with viral changes were often (but not always) enlarged with polymorphic nuclei. Frequently, tubular cells...
were rounded-up, necrotic, and extruded from the epithelial cell layer into tubular lumens causing marked denudation of basement membranes (Figure 3). Although cytopathic signs were seen along the entire nephron, they were often abundant in distal tubular segments and collecting ducts. Sporadically, infected cells were noted in the parietal epithelium lining Bowman’s capsule (Figure 4). By IHC, PV (BK strain) was demonstrated (Figure 5; nuclear staining profile: AB detecting SV40 - positive; AB detecting BK/JC - positive; AB detecting JC - negative). Positive staining was also noted in tubular cells inconspicuous by light microscopy. IHC did not reveal PV in nonepithelial cells and podocytes. By electron microscopy, intranuclear viral particles were granular, sometimes arranged as crystalloids (Figure 6). They measured between 35 and 40 nm in diameter, a size characteristic for PV (4,7,12,18). Viral particles were occasionally detected adherent to the external surface of cell membranes (Figure 7). In the cytoplasm, viral particles were only rarely noted by electron microscopy (Figure 7) or IHC. Ultrastructurally, PV was not seen in nonepithelial cells and podocytes. In three biopsies (two patients), small glomerular crescents with inclusion-bearing cells were seen (Figure 8). Crescent formation affected between 10 and 20% of glomeruli. There was no evidence of an immune complex-mediated glomerulonephritis. In the renal pelvis and ureters (two nephrectomies), viral inclusion bodies were identified in superficial transitional cells by light microscopy and additionally in the basal cell layer by IHC. Transitional cells almost exclusively showed intranuclear inclusions of the ground-glass type (in contrast to nephrons), closely resembling decoy cells in urine cytology (Figure 9).

CMV, EBV, HSV, or adenovirus were not found. By standard immunohistochemistry using a routine panel of antibodies directed against immunoglobulins and complement factors, a specific staining pattern was not present.

**Identification of BK Virus by PCR (Tissue, Urine).** One nephrectomy specimen (Figure 10) and urine samples from five patients were studied by PCR. All examined samples revealed abundant amplicons of 149 bp indicative of BK virus genomes. JC virus was not detected.

**PV Infection and Interstitial Inflammation.** In seven biopsies (7 of 13; 54%), PV infection of tubular epithelial cells was associated with widespread inflammatory cell infiltrates in
Figure 2. Types of intranuclear viral inclusion bodies. (a) Homogeneous ground-glass configuration (long arrow) and eosinophilic granular appearance with an incomplete halo (thick arrow) encountered in the same tubular cross section (hematoxylin and eosin [H&E] stain, ×400); (b) Homogenous finely granular pattern (arrow; periodic acid-Schiff [PAS] stain, ×400).

Figure 3. Tubules with infected cells. (a) An infected cell is rounded up and extruded into the lumen (arrow). The interstitium shows only minimal inflammation (PAS stain, ×251). (b) Necrotic cells with cytopathic signs sloughed off into the lumen. The tubular basement membrane is denuded (arrow, H&E stain, ×160) .

Figure 4. Intranuclear inclusion bodies in parietal epithelial cells lining Bowman’s capsule (arrow, H&E stain, ×160).

Figure 5. Polyomavirus detected by immunohistochemistry. Individual tubular epithelial cells show a positive staining reaction (formalin-fixed and paraffin-embedded tissue, antibody detecting the SV40 large T antigen, ×160).
the interstitium and tubulitis. Inflammatory infiltrates were predominately composed of lymphocytes and histiocytes in combination with some polymorphonuclear leukocytes and plasma cells. Focally, plasma cells were abundant. In four biopsies (4 of 13; 31%), the infiltrate was sparse with minute tubulitis (≤1 nonatrophic tubular cross section involved). The infiltrating cells were randomly distributed and not primarily associated with virally affected tubules (Figure 11a). Mononuclear cell infiltrates and tubulitis were often noted in areas lacking viral inclusion bodies. Some infected tubules, especially in the medulla, did not show an adjacent inflammatory response (Figure 11b). In two biopsies (2 of 13; 15%), there was negligible interstitial inflammation.

**Morphology of Persistent PV Disease.** After the initial histologic diagnosis of PV disease had been made, eight subsequent tissue samples were examined during the course of persistent graft infection (range, 40 to 330 d postdiagnosis; six biopsies and two graft nephrectomies; five patients). All tissue samples showed the three types of viral inclusion bodies, severe focal tubular injury, and varying degrees of inflammatory cell infiltrates (see above). Chronic changes increased: (1) marked interstitial fibrosis (three patients; six samples); (2) marked arteriolosclerosis (one patient, one sample); (3) marked “cyclosporine-type” arteriolopathy (two patients, two samples); (4) intimal fibrosis (“chronic vascular rejection;” two patients, two samples).

In one patient, PV-associated crescent formation was observed in two subsequent biopsies (50 d and 330 d after the initial diagnosis of PV disease). The percentage of crescents remained unchanged over time (<20%); however, they became increasingly fibrotic with pronounced layering of the basement membrane of Bowman’s capsule associated by an inflammatory cell infiltrate (Figure 12).

The gross appearance of two graft nephrectomies revealed kidneys slightly reduced in size with a smooth, focally finely granular outer surface. The parenchyma was homogenously ochre brown; the corticomedullary junction was ill-defined.

**Decoy Cells in the Urine.** Urine samples from all patients with persistent PV disease revealed decoy cells with ground-glass type intranuclear inclusions (Figure 9) positive for PV by IHC and electron microscopy. Decoy cells were typically excreted in large numbers. Small numbers (<5 per 10 HPF) were only sporadically noted during the course of disease. The excretion of decoy cells vanished in one patient after transplant nephrectomy (starting 2 d postoperatively; follow-up: 4 mo). In two patients tested, urine samples revealed large numbers of decoy cells 10 and 5 mo before manifestation of PV disease when the corresponding biopsies lacked cytopathic signs (see above).

**Differences between PV Disease Group and Control Subjects**

Decoy cell excretion was a typical finding in patients with PV disease. To evaluate the specificity of decoy cell excretion, urine cytologies from 483 renal allograft recipients (including five patients with PV disease) were analyzed. Abundant decoy

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*Figure 6. Electron micrographs showing intranuclear viral inclusion bodies. (A) Dense granular viral particles in a transitional cell (×3800). (B) A tubular cell with loosely arranged granular viral particles (×5600). (Inset) A crystalloid arrangement of polyomavirus with particles measuring between 35 and 40 nm (×97,000).*
cells were found in 28 recipients (6%) and scant decoy cells in 72 (15%). PV disease was associated with abundant decoy cell excretion in five of 28 (18%) recipients; twenty-three of twenty-eight (82%) were free of disease (no cytopathic changes in graft biopsies by light microscopy or IHC).

Tissue samples from patients with and without PV disease ($n = 27$ versus $n = 108$) differed in only two histologic parameters from 13 analyzed ($P < 0.05$, Fisher exact test) (Table 1): Interstitial cellular rejection and transplant glomerulitis were more frequently found in patients with PV disease (60 and 29%) than without (35 and 4%, respectively). Interstitial rejection and transplant glomerulitis were particularly frequent in the disease group before PV infection became manifest.

**Outcome**

Renal function in patients with PV disease deteriorated quite rapidly. Two patients (2 of 5; 40%) lost their grafts within 40 and 113 d postdiagnosis of infection. The serum creatinine level of the remaining three patients was $484 \pm 326 \mu$mol/L (mean $\pm$ SD; 11 mo posttransplantation). In comparison, the 23 transplant recipients with abundant decoy cells in urine
cytology but without PV disease had a mean serum creatinine level of 207 ± 177 μmol/L (mean ± SD; 57 mo posttransplantation; correlation with disease group \( P = 0.01 \), t test). Two of the latter 23 patients (9%) had lost their grafts (correlation with disease group \( P < 0.01 \), \( \chi^2 \) test).

**Discussion**

Renal allograft recipients require permanent immunosuppression and, therefore, are at an increased risk for infections. Morphologic evidence of viral renal allograft disease is exceptional, in particular a manifest PV infection (2,18). Although we did not see a single patient with PV disease among 616 renal allograft recipients between 1985 and 1995, we suddenly encountered a cluster of five patients.

PV renal allograft disease can only be diagnosed histologically. Severe tubular injury with cellular enlargement, frank epithelial necrosis, denudation of tubular basement membranes, and distinct intranuclear inclusion bodies in epithelial cells including transitional cells suggest an infection with PV. Infected parietal epithelial cells occasionally form glomerular crescents, a highly exceptional finding in the absence of an underlying glomerular tuft pathology (glomerulonephritis). The morphology of a PV infection is typical, however, not pathognomonic, since adenovirus and HSV may cause a similar pattern. Diagnostic confirmation is easily achieved by immunohistochemistry and electron microscopy, which shows viral particles of approximately 40 to 50 nm typical for PV. The detection of urinary decoy cells is helpful in identifying PV activation and replication. However, decoy cells alone do not necessarily indicate PV disease (31,32). The false positive rate in our series was high (82%; sensitivity: 100%), underscoring the necessity of a renal biopsy for a definitive diagnostic workup.

PV renal allograft disease was associated with deteriorating graft function. Two factors contributed to functional impairment: tubular necrosis induced by PV infection and sclerosing allograft nephropathy. PV infection caused frank tubular necrosis and, thus, altered renal function significantly. Sloughed necrotic epithelial cells formed intraluminal casts leaving behind denuded tubular basement membranes. Denuded areas of basement membranes permit leaking of fluid into the interstitial compartment, which is associated with functional impair-

**Figure 8.** Glomerular crescent. Intranuclear viral inclusion (arrow) in a small cellular crescent (asterisk: glomerular tuft; PAS stain, ×400).

**Figure 9.** Transitional cells infected by PV compared with “decoy cells” in urine cytology. (A) Low power view of the urothelium showing cytopathic signs in superficial transitional cell layers (arrow, H&E stain, ×62) pictured in Panel B with high magnification (arrow, H&E stain, ×400). (C) Immunohistochemistry reveals a positive staining pattern not only in superficial transitional cells with cytopathic signs, but also in occasional basal cells that were unremarkable by light microscopy. The lamina propria is without inflammation (formalin-fixed and paraffin-embedded tissue, antibody detecting the SV40 large T antigen, ×160). (D) Urine cytology with a PV-infected cell, so-called “decoy cell,” showing a ground-glass type intranuclear inclusion body (arrow, Papanicolaou stain, ×400).
ment (33). In addition, tubular casts may cause an obstructive component. Such pathways are not unique to PV disease but are well described in other forms of acute tubular necrosis (33). Because PV never cleared from the kidneys, tubular injury did not heal, and therefore functional impairment persisted. In addition, over time recurrent rejection episodes with transplant endarteritis led to interstitial and intimal fibrosis (i.e., “chronic allograft rejection”). However, the prevalence of chronic changes did not differ significantly from control subjects, emphasizing that tubular necrosis is a main factor for functional deterioration.

There is controversy about whether the interstitial inflamma-

Figure 10. PCR detection of BK virus genomes in a nephrectomy specimen from a patient with PV disease. Tissue samples from the renal cortex and medulla were macroscopically separated followed by DNA extraction. Agarose gel showing a 149-bp amplicon of BK virus genomes (arrow) in the renal cortex (lanes 12, 13, 17, and 18) and medulla (lanes 7 and 8). Lanes 1 and 22, size standards; lane 2, human DNA control; lanes 3 and 4, external positive BK DNA controls; lanes 5, 6, 9, 11, 14, 16, 19, and 21, negative H2O controls; lanes 7 and 8, renal medulla; lane 10, renal medulla with external positive BK DNA control; lanes 12, 13, 17, and 18, renal cortex; lanes 15 and 20, renal cortex with external positive BK DNA control. With excess target, the semi-nested PCR technique gives rise to a double band. The human control DNA preparation from peripheral blood mononuclear cells (lane 2) consistently yields an unspecific band of 130 bp.

Figure 11. Inflammation and viral inclusion body. (a) There is diffuse interstitial inflammation associated with tubulitis, features suggestive of cellular rejection. Only one intranuclear viral inclusion is present (arrow, H&E stain, ×80); (b) Cytopathic signs in the medulla. Low power view of the renal medulla showing scattered inclusion bodies in tubular cells (arrowhead). There is no interstitial inflammatory response (H&E stain, ×25).

Figure 12. Glomerular crescent in a follow-up biopsy 330 d after the initial diagnosis of PV disease. The basement membrane of Bowman’s capsule is seen twice as an inner and outer circular line (arrow) associated with cellular aggregates forming a crescent. The remaining glomerular tuft is seen in the center (asterisk). Same patient as pictured in Figure 8. (PAS stain, ×125).
Cellular rejection was more common in our patients with PV. The kidney could render tubular cells susceptible to viral replication. Recurrent rejection with long-lasting tubular inflammation is suggested by the predominance of viral inclusions in distal membranes. Manifest PV disease. Manifest renal allograft infection can occur before the disease becomes manifest and disappear in all patients with persistent PV disease; they were found, randomly affecting tubules with and without cytopathic signs. This interpretation is supported by the observation that tubulitis was seen in areas lacking cytopathic changes, thus making a diagnosis of virally induced nephritis less likely. We diagnosed interstitial cellular rejection in 54% of tissue samples during the course of PV disease. Virally induced nephritis may coexist, but seems to be of minor significance.

What are the steps involved in PV activation and manifestation of disease? It seems surprising that PV does not affect renal transplant recipients as often, since approximately 60 to 80% of healthy adults are serologically positive for PV (4–8). Under normal immunosurveillance, PV is latent in the kidneys (4,7,9–11) and, therefore, renal allografts carry the risk for viral transmission (8,34) and manifestation of PV disease. Despite the high prevalence of latent virus, PV activation is uncommon, even in immunosuppressed renal allograft recipients. We found abundant decoy cells as a morphologic marker of viral activation and replication (12,14,32) in only 6% of 483 kidney transplant recipients, and manifest renal allograft disease was only seen in 1%. Decoy cells were found in all of our patients with persistent PV disease; they were present before the disease became manifest and disappeared after transplant nephrectomy. Activation of latent virus and replication with excretion of decoy cells likely represents an initial, still balanced, and reversible step along the way to manifest PV disease. Manifest renal allograft infection can develop in an ascending manner via cell-to-cell spread, which is suggested by the predominance of viral inclusions in distal tubular segments and viral particles adherent to outer cell membranes. Recent rejection with long-lasting tubular injury could render tubular cells susceptible to viral replication. Cellular rejection was more common in our patients with PV disease before the infection became manifest than in the control group. Tubular injury has been demonstrated as a promoting factor for a morphologically manifest PV renal infection in mice (35).

A major factor involved in the manifestation of PV disease is high-dose immunosuppression. Therapeutic rescue attempts with tacrolimus, a new immunosuppressive drug, seem particularly important since all of our patients were switched from cyclosporine to tacrolimus months before manifestation of infection (36). Tacrolimus as a possible risk factor has been suggested in the past—all eight previously reported patients with PV allograft disease were on a tacrolimus-based immunosuppressive regimen (17–19). However, tacrolimus cannot be the only factor, because we recently diagnosed PV disease in a patient on cyclosporine and mycophenolate mofetil, not treated with tacrolimus (consultation practice M.J.M.). One previous report (19) described (histologic) clearance of PV from the graft and stabilization of renal function after stopping tacrolimus and reintroducing cyclosporine. We could not confirm this finding. In three of our patients who were switched back to cyclosporine, the virus persisted and renal function deteriorated (36).

In summary, we propose the following evolution and profile of PV disease of renal allografts: (1) reactivation of latent BK virus with replication (decoy cells in urine); (2) long-lasting tubular injury, such as recurrent “therapy-resistant” rejection episodes; and (3) therapeutic rescue attempts with high-dose immunosuppression, frequently tacrolimus-based.

A definitive diagnosis of PV disease requires a renal biopsy, ideally triggered by the detection of large numbers of decoy cells in the urine. PV disease is a rare but serious cause for transplant failure, mainly due to virally induced severe tubular injury.

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