

Polyomavirus Update 2008

Daniel C. Brennan, MD, FACP

Professor of Medicine

Washington University School of Medicine

Director, Transplant Nephrology

Barnes-Jewish Hospital

St. Louis, Missouri

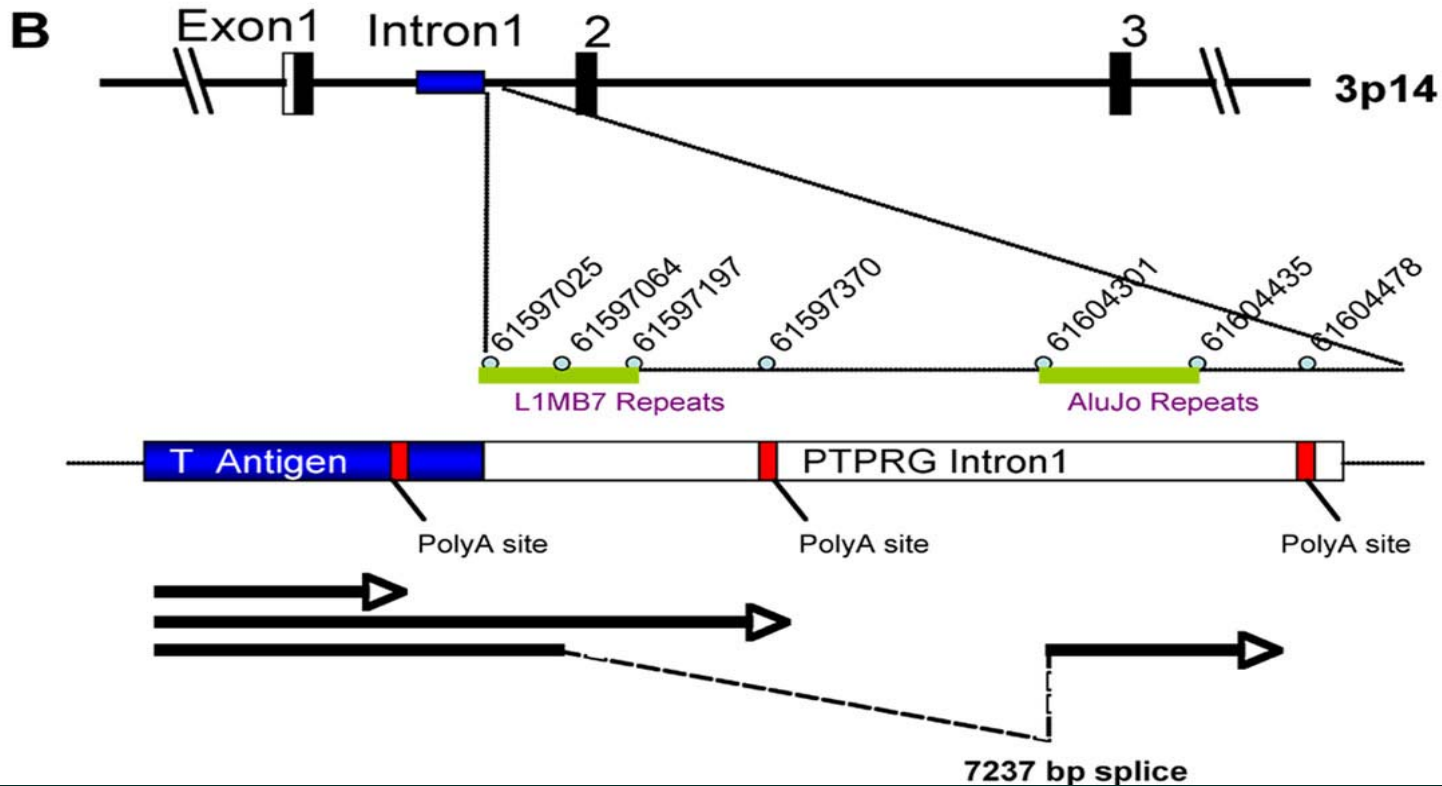
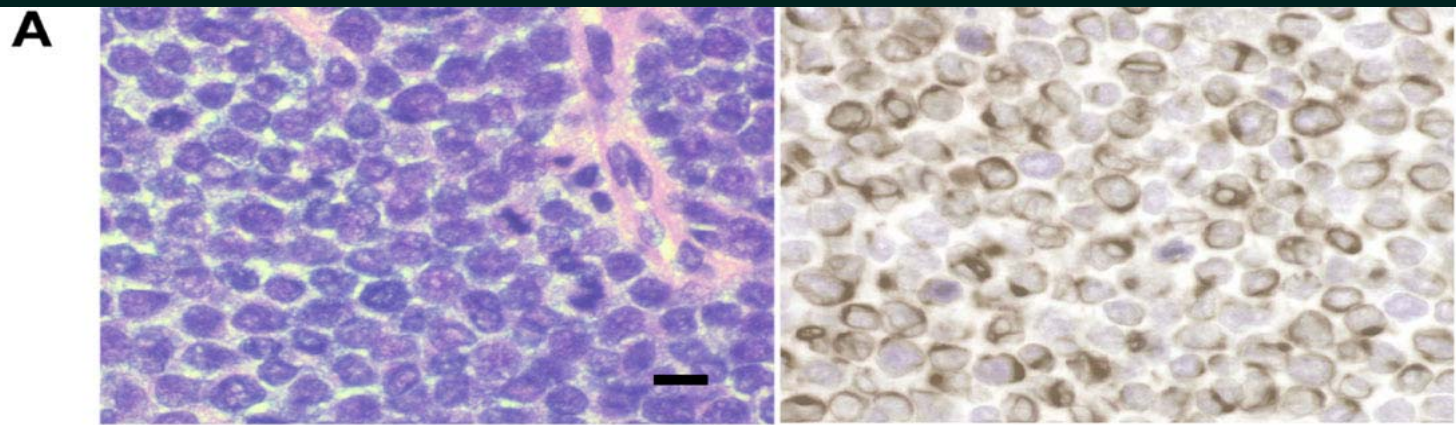
Acknowledgments

- **Washington University**
 - Greg Storch
 - Dan Bohl
 - Monique Gaudreault-Keener
 - Stephanie Torrence
 - Rebecca Schuessler
 - Matt Koch
 - Brent Miller
 - Decha Enkvetchakul
 - Niraj Desai
 - Martin Jendrisak
 - Surendra Shenoy
 - Jeffrey Lowell
 - Karen Hardinger
 - Irfan Agha
- **National Institutes of Health**
 - Caroline Ryschkewitsch
 - Eugene Major
 - Rosalyn Mannon
- **University of Pittsburgh**
 - Parmjeet Randhawa
- **St. Louis University**
 - Mark Schnitzler

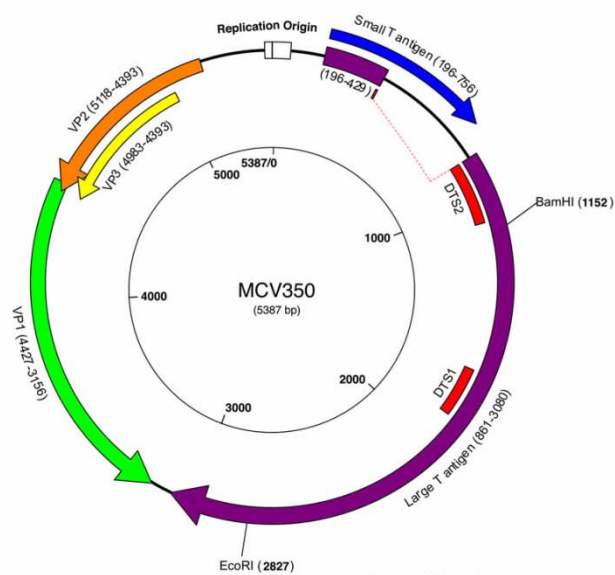
Polyoma Viruses of Human Relevance

- **SV-40 (simian virus-40)**
 - Exposure from polio vaccine
 - Associated with meningiomas and FSGS
- **JC Virus**
 - Polymultifocal leukencephalopathy (PML)
 - in HIV patients
 - Natalizumab and rituximab treated patients
- **BK Virus**
 - BK nephropathy
- **KI and WU respiratory polyomaviruses (2007)**
- **MCV (Merkel Cell Virus) (2008)**

Merkel Cell Tumor Associated with New Polyomavirus

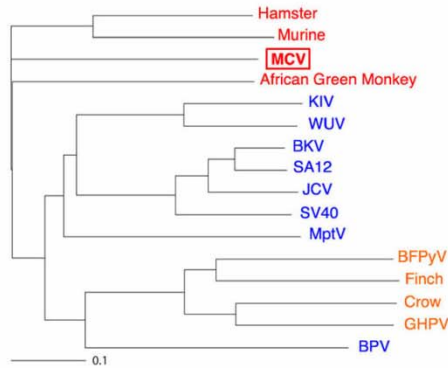


A

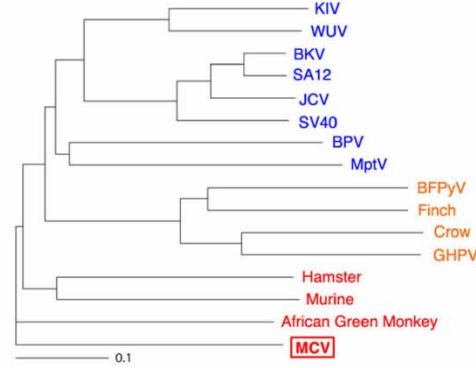


B

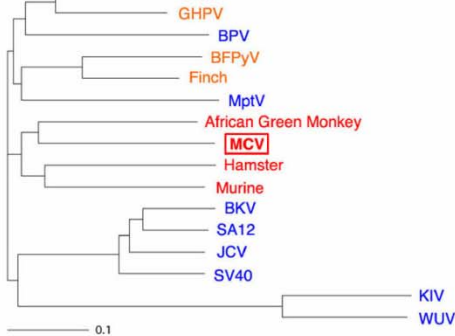
Small T antigen



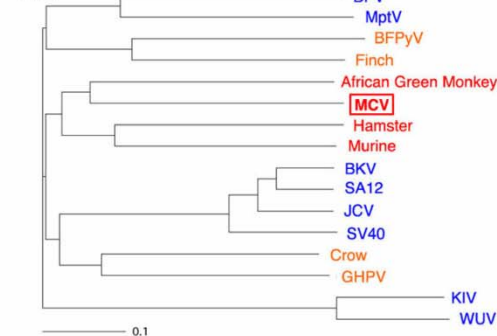
Large T antigen



VP1



VP2



MCV or MCPyV Genome Schematic and Comparison to other Polyomaviruses

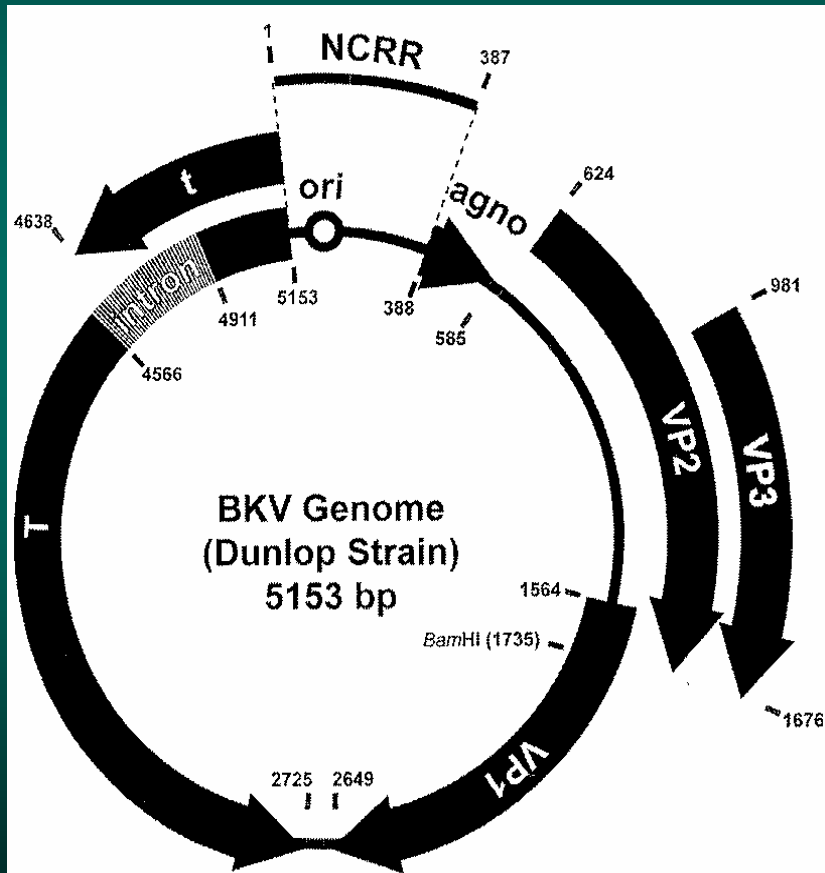
MCV has highest homology to the MuPyV subgroup and is most closely related to AGM PyV. It is more distantly related to known human polyomaviruses and SV40.

Feng, www.sciencexpress.org/
17 January 2008 / Page 1 /
10.1126/science.1152586

Scope of the Problem: BKN

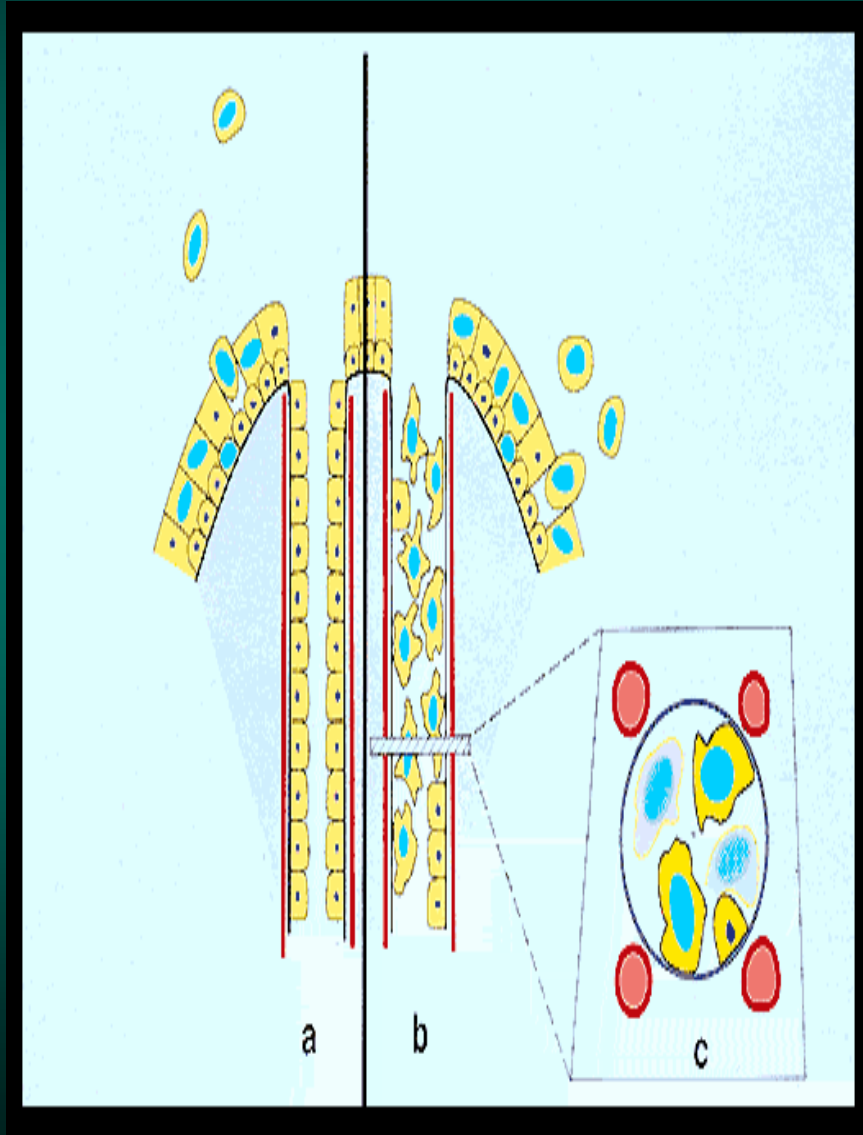
- Emerged after 1995; along with Tacrolimus and MMF
- Occurs in 0-10% of RTR
- Results in allograft dysfunction in 80%
- Causes allograft loss in up to 50%
- Histology mimics common biopsy findings
- No anti-viral treatment currently exists

Polyomavirus Structure



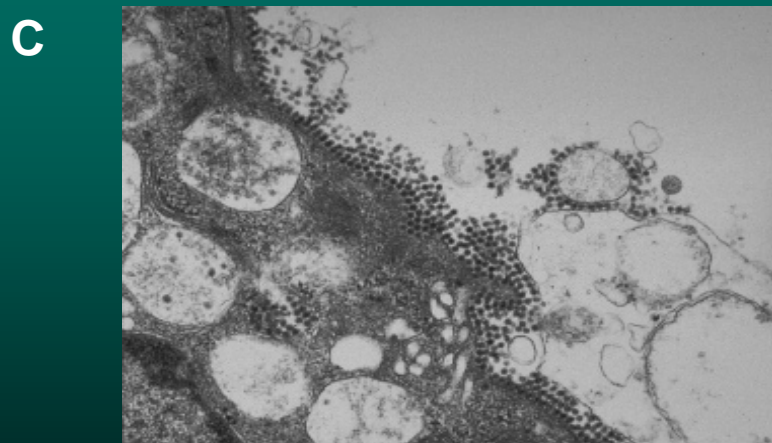
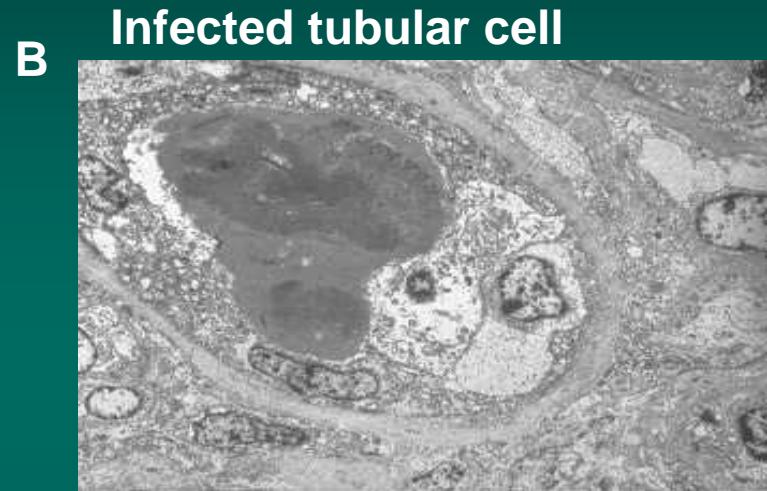
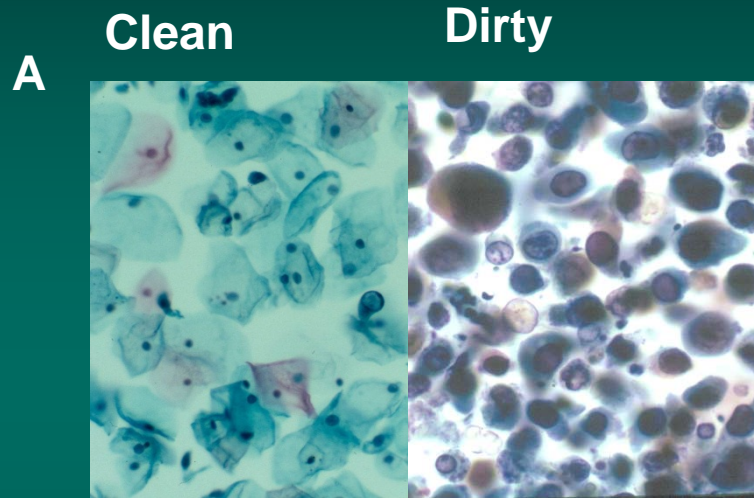
- Circular, double-stranded DNA genome
- ~5200 base pairs
- Early genes (regulatory): large T-antigen and small t-antigen
- Late genes (structural): VP1, VP2, VP3, and agnoprotein
- Non-coding control region (NCCR): contains the origin and transcription factor binding sites

Pathogenesis of BK Nephropathy



- Lysis of tubular cells releases BKV into tubules with bare basement membranes
- Virus particles can leak into the interstitium, from where the virus gains access to capillaries and viremia results
- Somewhere along this pathway, injury and genotype rearrangements may change the virulence characteristics of the virus.

Diagnosis of BKV Nephropathy: Decoy Cells



Viral particles

Decoy cells in “clean” urine occurs in with stable renal function (A, left)

Decoy cells in “dirty” urine occurs with nephropathy (A, right)

EM may show infected decoy cells in the urinary space (B) or viral particles (C)

Ancillary Histologic Diagnostic Techniques In The Diagnosis Of BKV Nephropathy

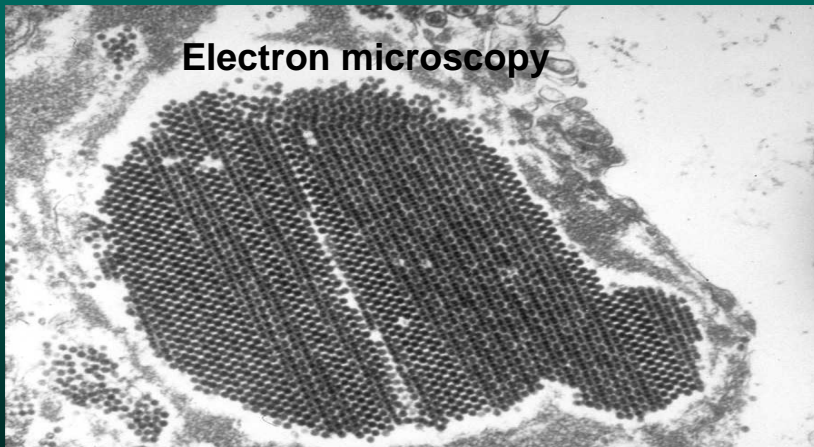
A



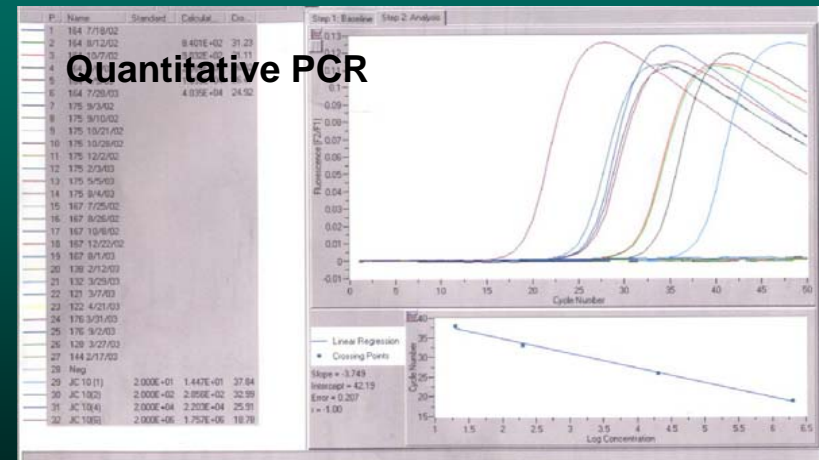
B



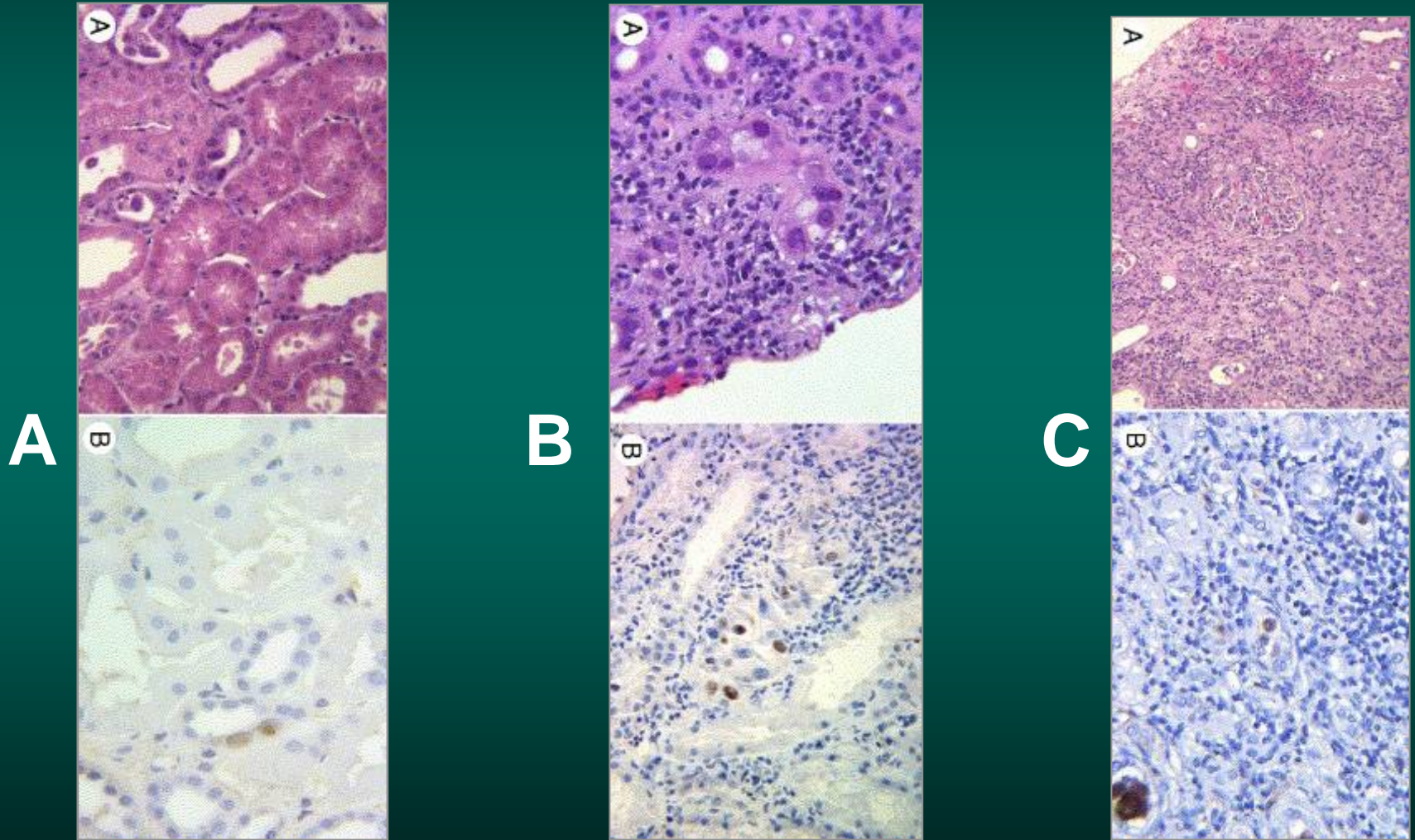
C



D

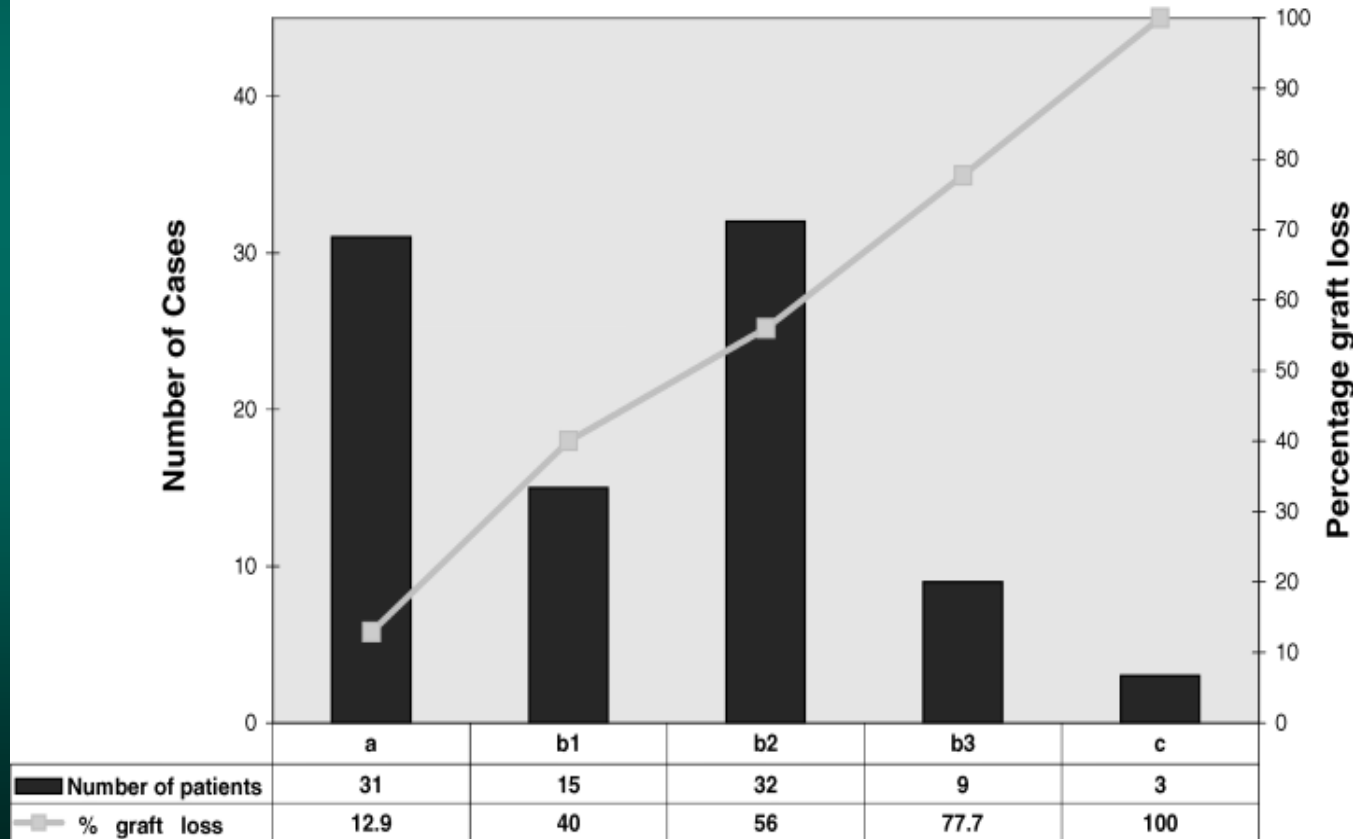


Three Histological Patterns of BK Nephropathy

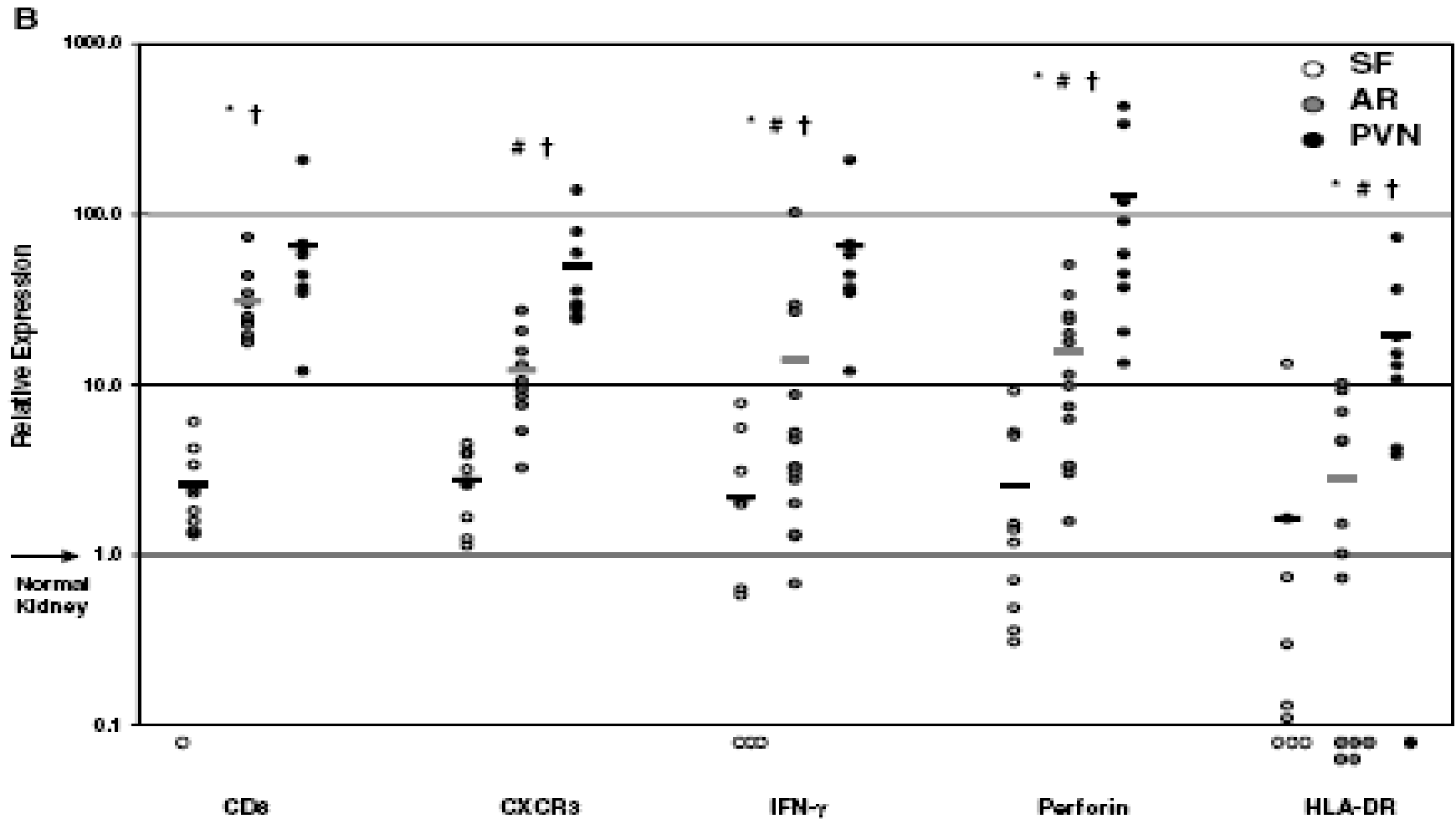


Histology Predicts Outcome

HISTOLOGICAL PATTERN IN INDEX BIOPSY
CORRELATION WITH GRAFT LOSS

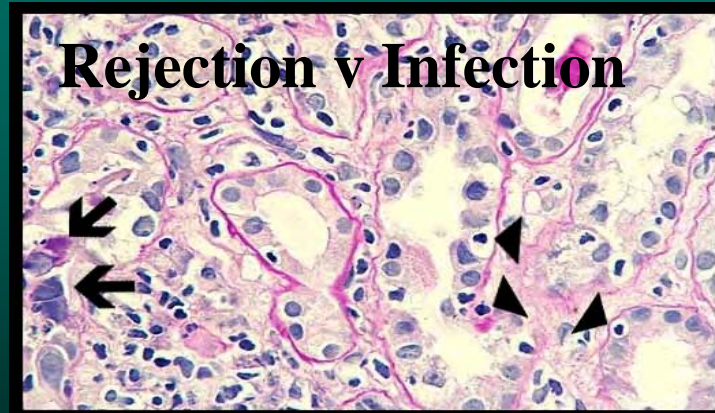
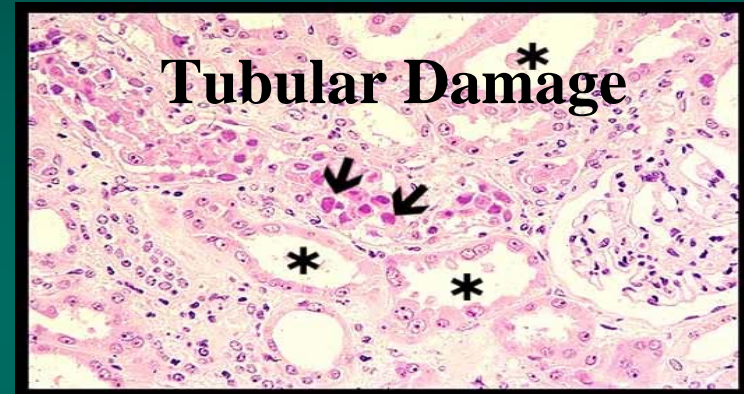
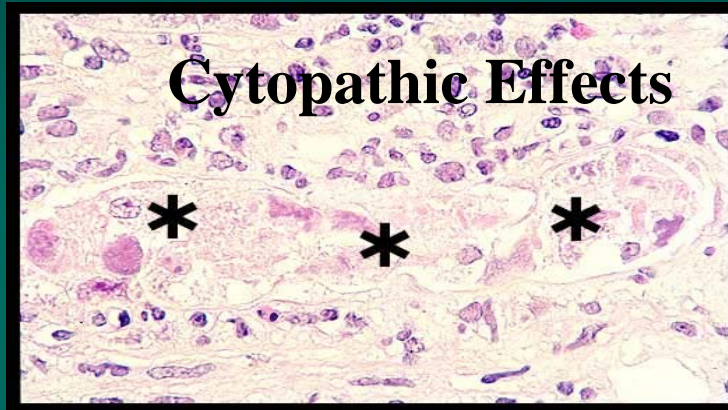


BKV is Th1 Pro-Fibrotic



BKN increases expression of markers of activation, costimulation, inflammation, cell death, cytotoxicity, fibrosis, and epithelial-mesenchymal transformation to as similar or greater degree than acute rejection

Histopathology Diagnostic Quandary



BKN Simulating Vascular Rejection

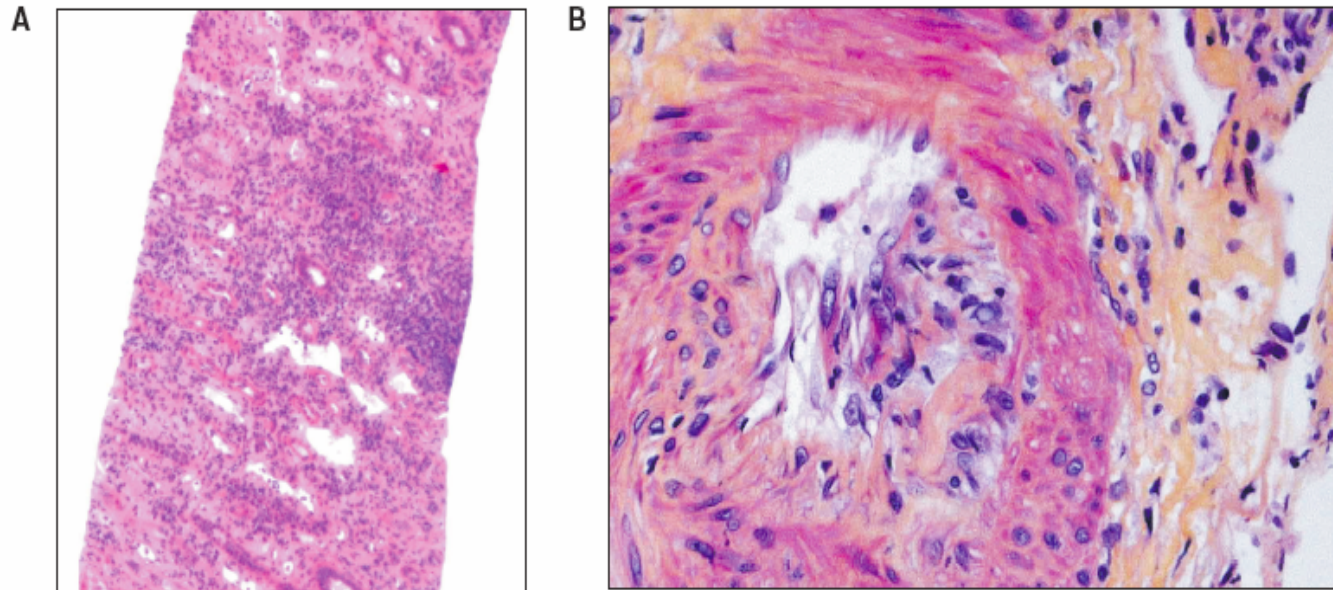
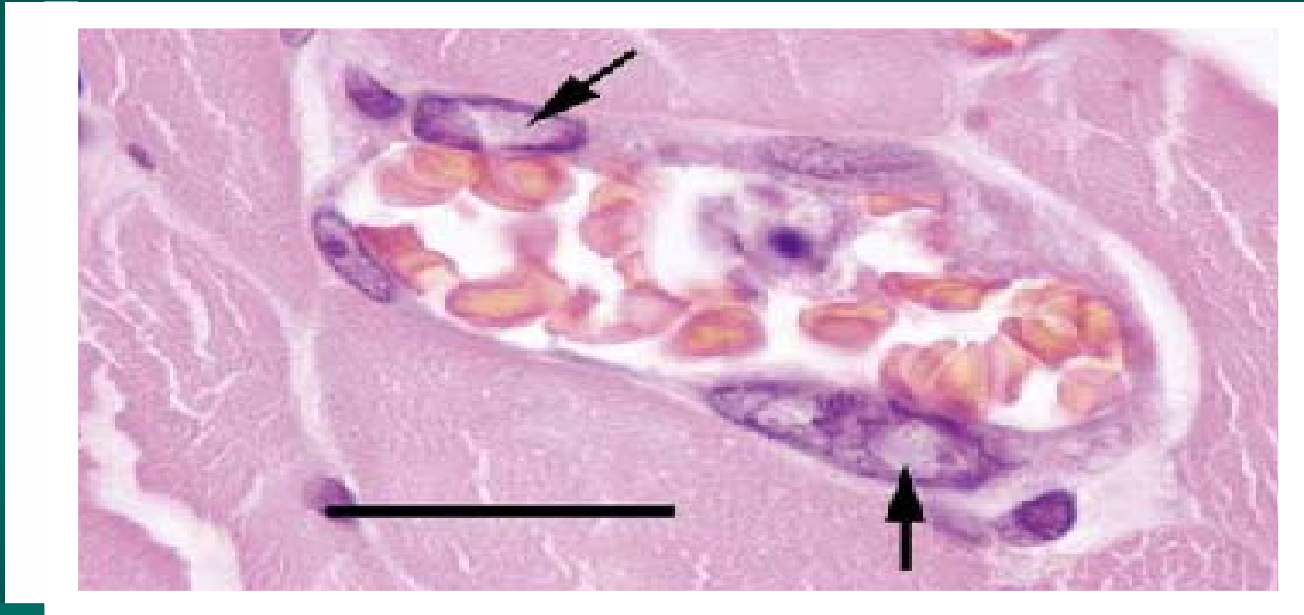


Figure 4: Renal biopsy 3. (A) Intense interstitial inflammation with lymphocytic infiltration and tubulitis. There were no clear viral inclusions. H&E. Original magnification, ×30. (B) In this somewhat tangential cut, subendothelial lymphocytic infiltration classic of vascular rejection is seen. Graded as acute vascular rejection, Banff 1997, type IIA. HPS stain. Original magnification, ×250.

Immunosuppression for BK Masquerading as Rejection: Vasculopathy and Death

Deltoid muscle specimen showing small blood vessels with enlarged, hyperchromatic endothelial cells



Arrows point to intranuclear inclusions

Potential Clinical Risk Factors for BKV Nephropathy

Donor factors

- Deceased-donor
- Active BK or CMV
- Donor seropositivity
- Absence of HLA-C7

Recipient factors

- Higher age
- Male gender
- Negative recipient BK antibody status
- Steroid pulses
- Prior acute rejection episodes

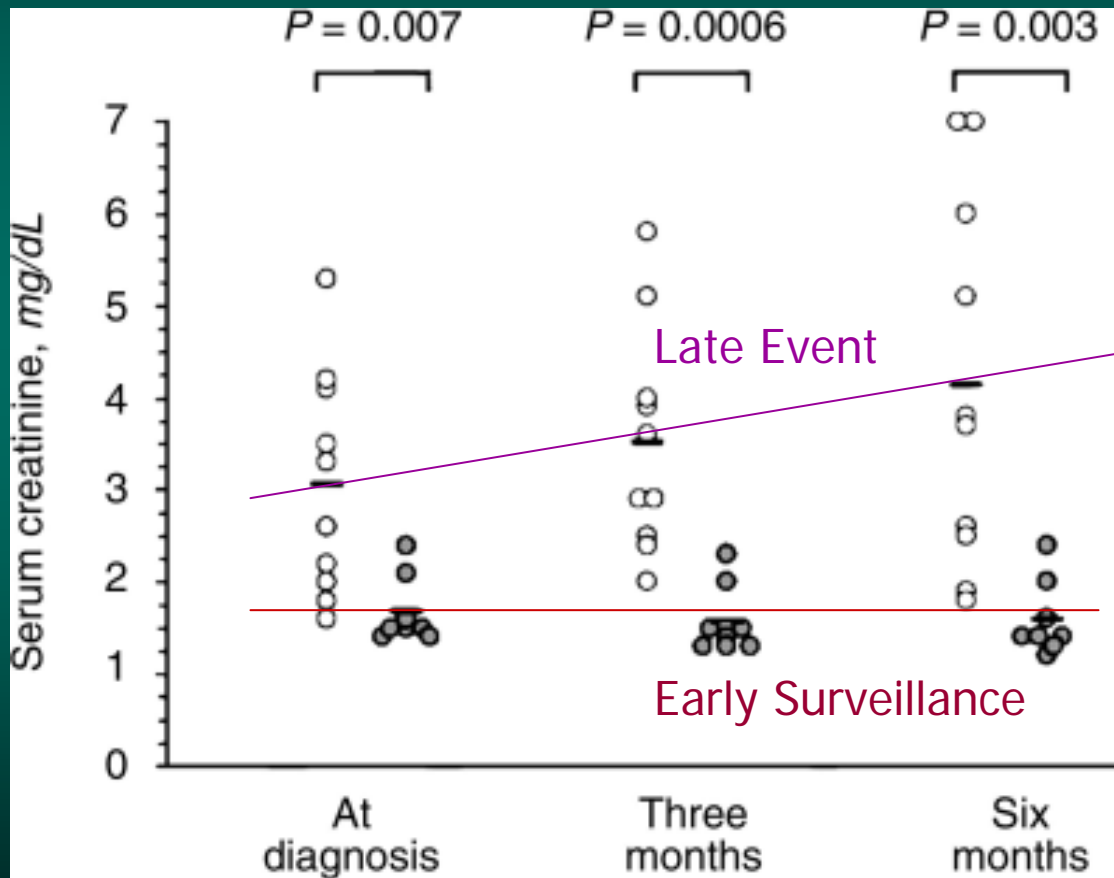
Transplant Factors

- Procurement injury
- Cold ischemia time
- Delayed graft function
- Ureteral stents
- Intensive immunosuppression with Tacrolimus \pm MMF, SRL
- Treatment of rejection with lymphocyte depleting agents or steroids
- Drug toxicity
- Increased HLA mismatches

Potential Viral Risk Factors for Progression of BKV Infections

- High viral titers in urine > 1 billion copies/mL
- High viral titers in blood >10,000 copies/mL
- Viral subtype
- Synergistic viral infection with CMV
- Poor antibody response in recipients
- Poor cellular immune response
- DNA mutations in VP1, NCCR, and T-Ag

Factors Associated with Worse Outcome



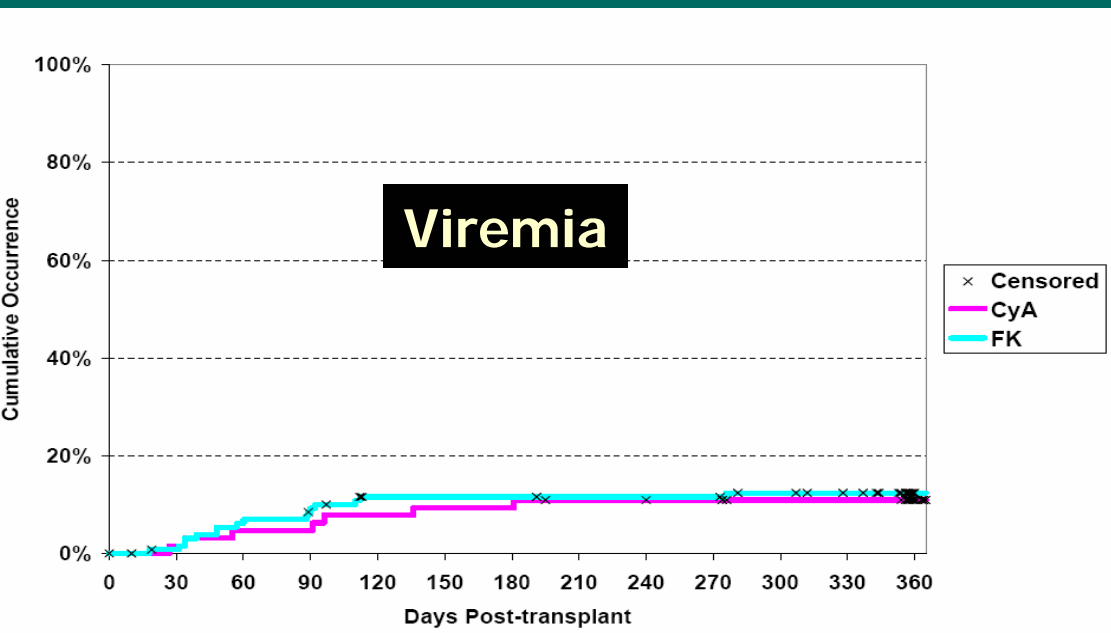
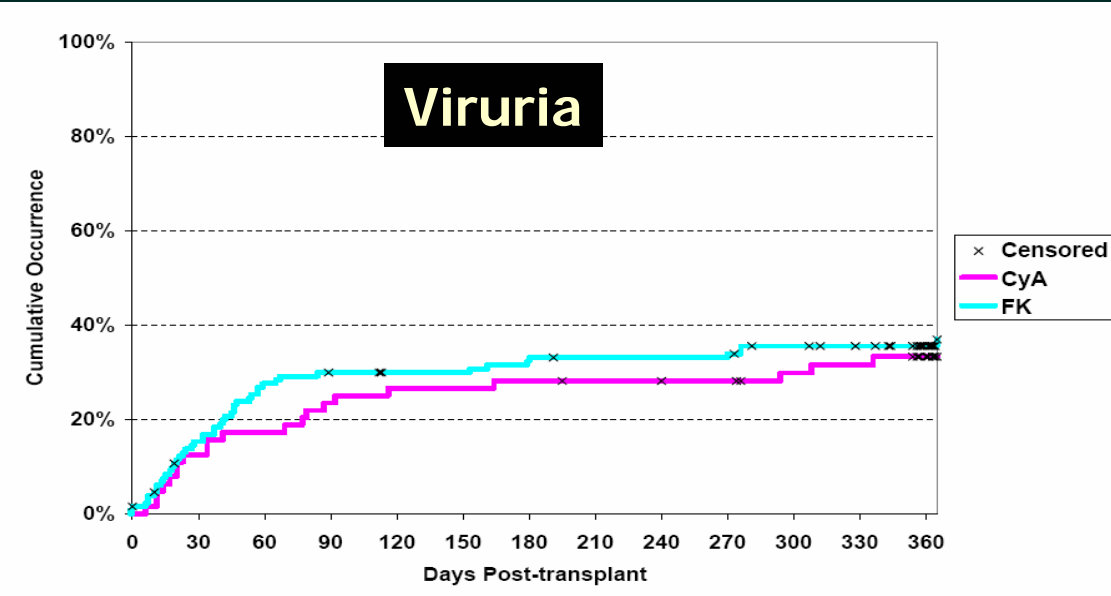
Worse Outcome

Event Biopsy
(73 vs 0%, p=0.004)

CMV D+/R-
(71 vs 9%, p=0.01)

No Ab induction
(82 vs 17%, p=0.01)

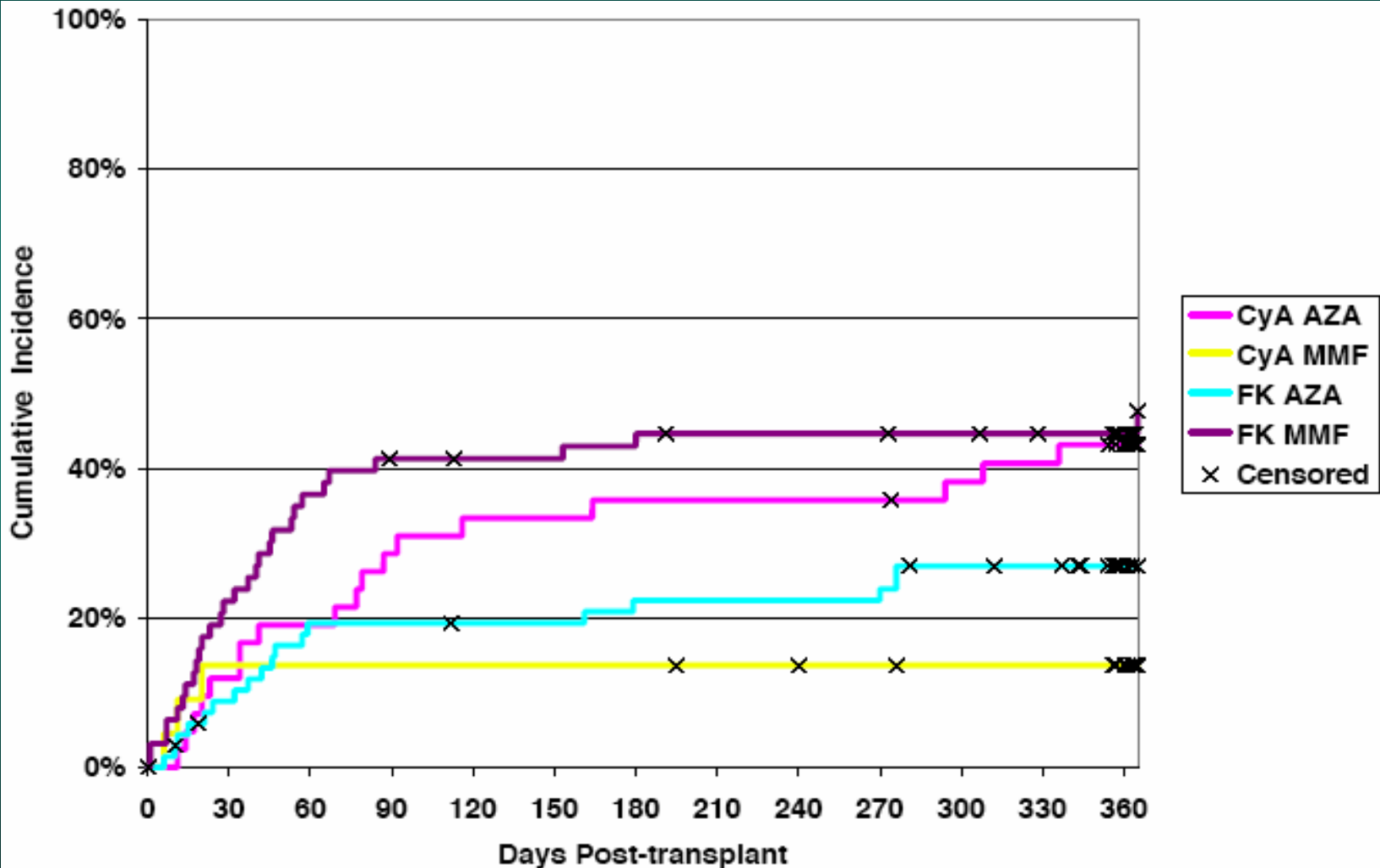
No improvement
of BKN on bx
(100 vs 40%, p=0.04)



Occurrence of BK Viruria and Viremia by Immunosuppressive Regimen

Brennan, AJT, 2005; 5: 582-94

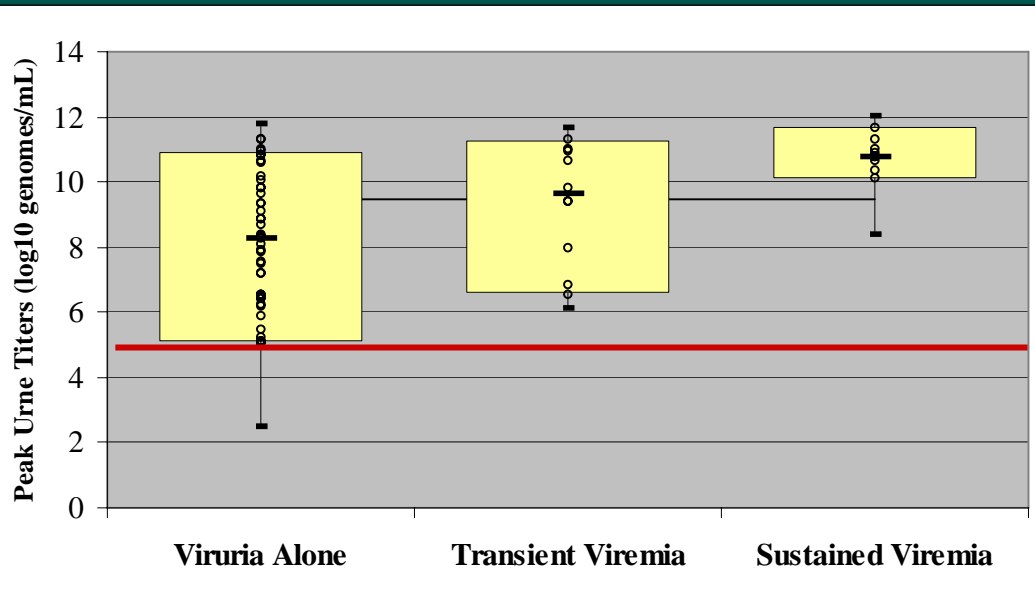
BK Viruria is Lowest with CyA-MMF



BK & Thymoglobulin

- **12 patients (Haploidentical LRA) did not receive Thymoglobulin**
- **BK Viruria was detected in 5 (42%) of these patients**
- **No difference in the incidence of viruria or viremia for different regimens if these patients were excluded**
- **Also no difference in incidence of viruria or viremia for patients who received or not received Thymoglobulin**

BKV in Urine at Level $> 9.5 \log_{10}$ is Associated with Sustained Viremia



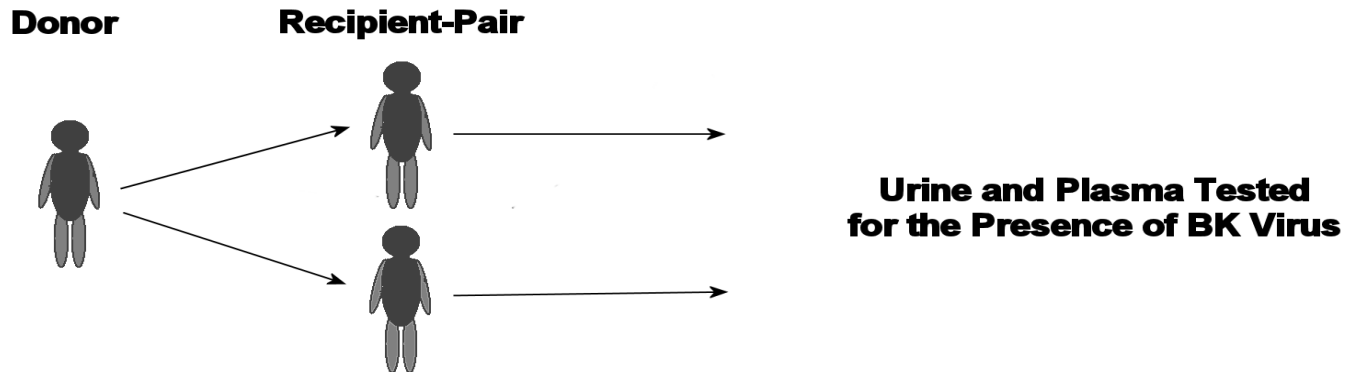
Viremia	No.	Peak Urine Level	
		< 9.5 (n=40)	> 9.5 (n=30)
None	47	33	14
Transient	12	6	6
Sustained	11	1	10

Peak urine level of > 9.5 is associated with a 3-fold increased risk of viremia and a 13.2-fold increased risk of sustained viremia.

But PPV $< 20\%$ for sustained viremia.

BK Virus Infection in Recipient-Pairs

A



Recipient-Pair BKV Status	BKV Infection in Recipients		Recipient-Pairs
	Recipient A	Recipient B	
Concordant	None	None	10
	Viruria	Viruria	6
Discordant	None	Viruria	3
	None	Viremia	1

One deceased donor can donate a single kidney to two different recipients A or B (a recipient-pair). Of the 20 recipient-pairs in the study, 16 had concordant infections: 6 with viruria, and 4 had discordant infections. $P=0.02$, $\kappa = 0.588$ (95% CI 0.23-0.95, $p=0.007$).

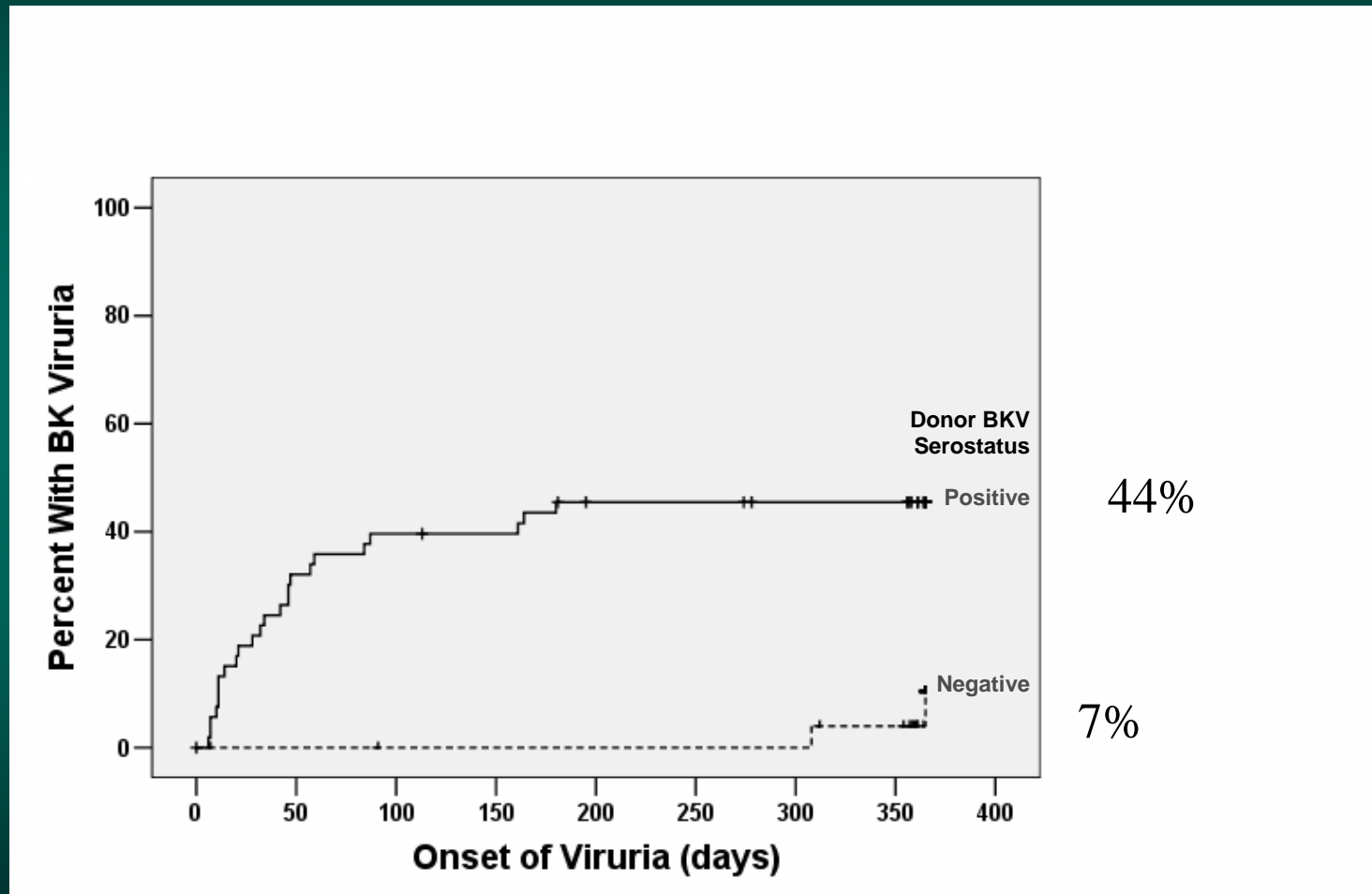
Comparison of 396 bp Sequence Including the NCCR Among The Six Recipient-pairs With Viruria Showed Identity

B

Recipient Pairs	1	58	135	171	215	228	341	381	394	395
1	A	C	G	G	A		C	D	GG	
2	A	C	G	G	A		C	T	AA	
3	C	T	G	A	T		A	T	AA	
4	C	T	C	G	A		C	T	AA	
5	C	T	C	G	A		C	T	AA	
6	C	T	C	G	A		C	T	AA	
WW	C	T	G	A	T		A	T	AA	

Each pair had identical sequences. Numbering based on BKV strain WW (NCBI M1587). The position of nucleotide substitutions are noted along the top. D=deletion.

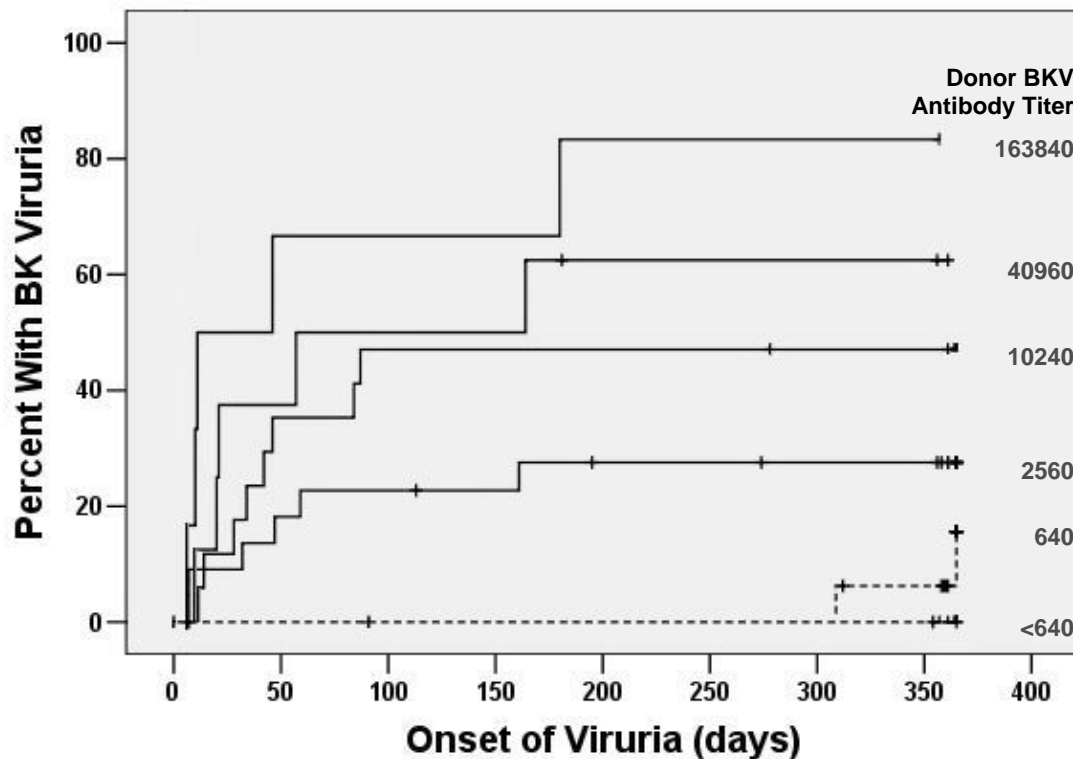
BK Viruria in the Recipient is Related to the Donor BK-Serostatus



At-Risk	81	62	57	56	51	51	49	47	29
With Viruria	0	17	21	21	24	24	24	25	26

N=81; P<0.001

BK Viruria is Ordered with BK Donor Titer



Recipients At-Risk For BKV Infections By Donor BKV Antibody Titer

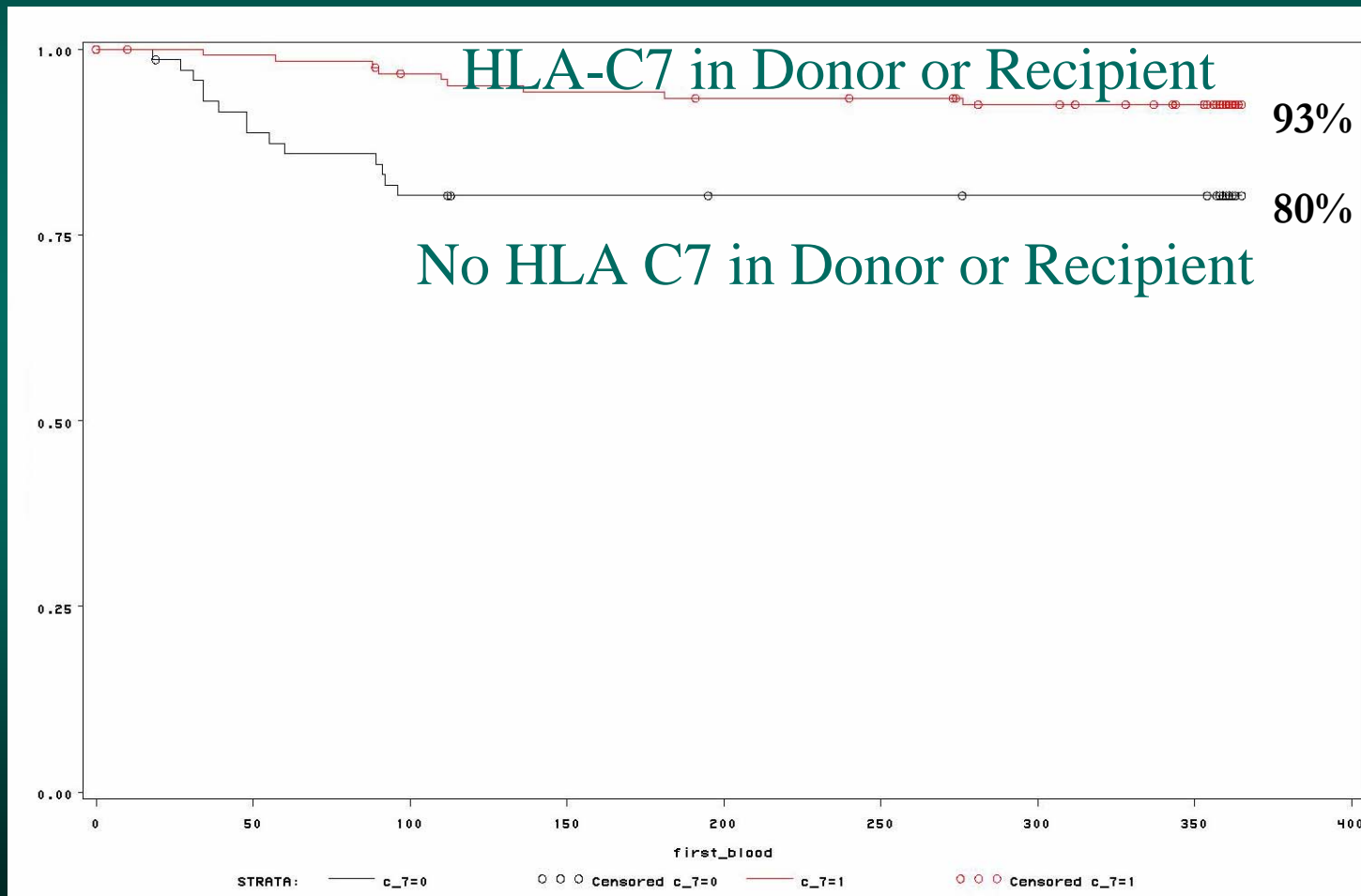
<640	10	10	9	9	9	9	9	9	5
640	17	16	16	16	16	16	16	14	10
2560	22	18	17	16	14	14	13	13	8
10240	18	11	9	9	9	9	8	8	6
40960	8	5	4	4	2	2	2	2	0
163830	6	2	2	2	1	1	1	1	0

N=81; P<0.001

Bohl, AJT 2005: 5:2213-21

Absence of HLA-C7 in the Donor or the Recipient is Associated with the Development of BK-Viremia

Fraction of recipients free of BK viremia

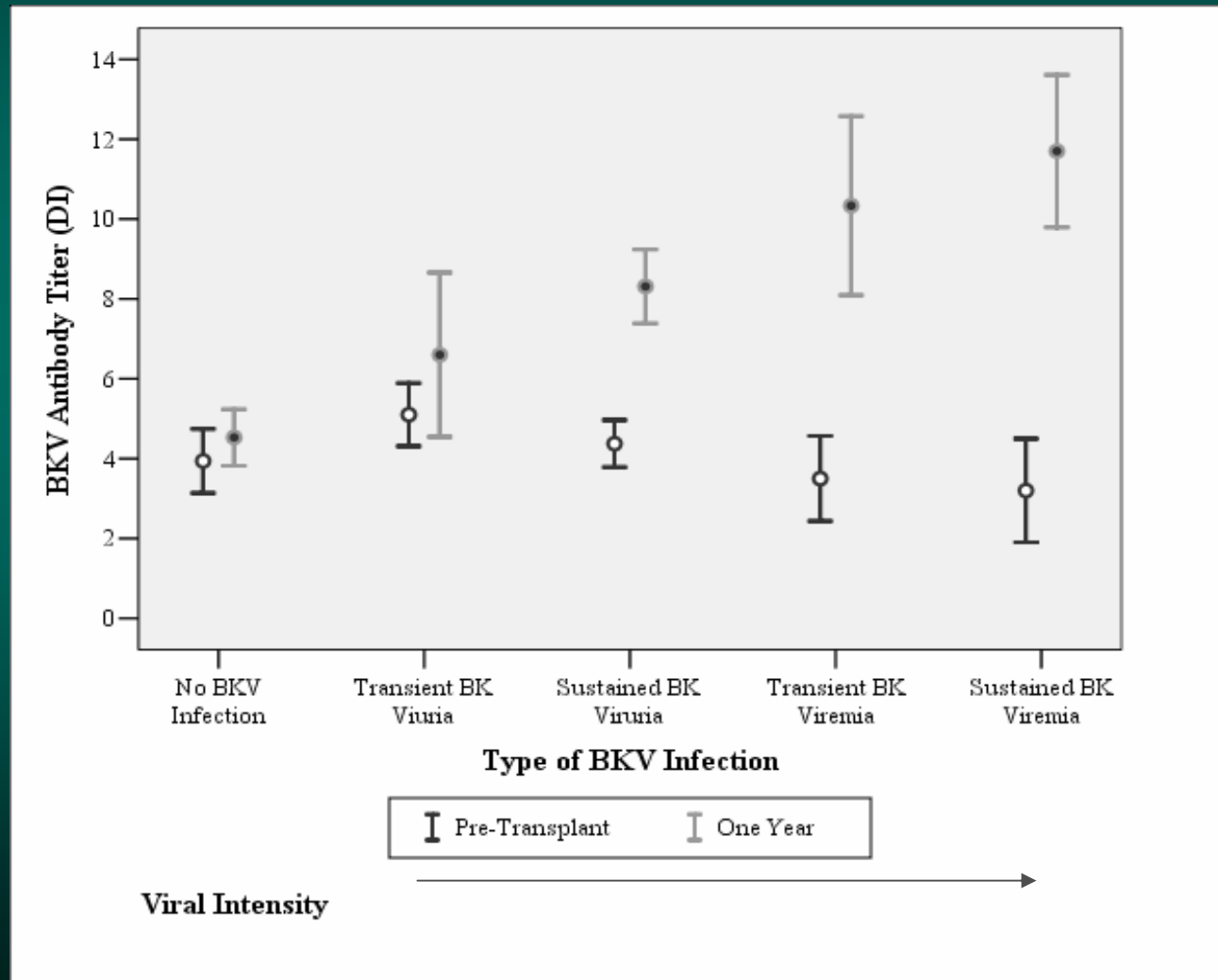


P=0.005

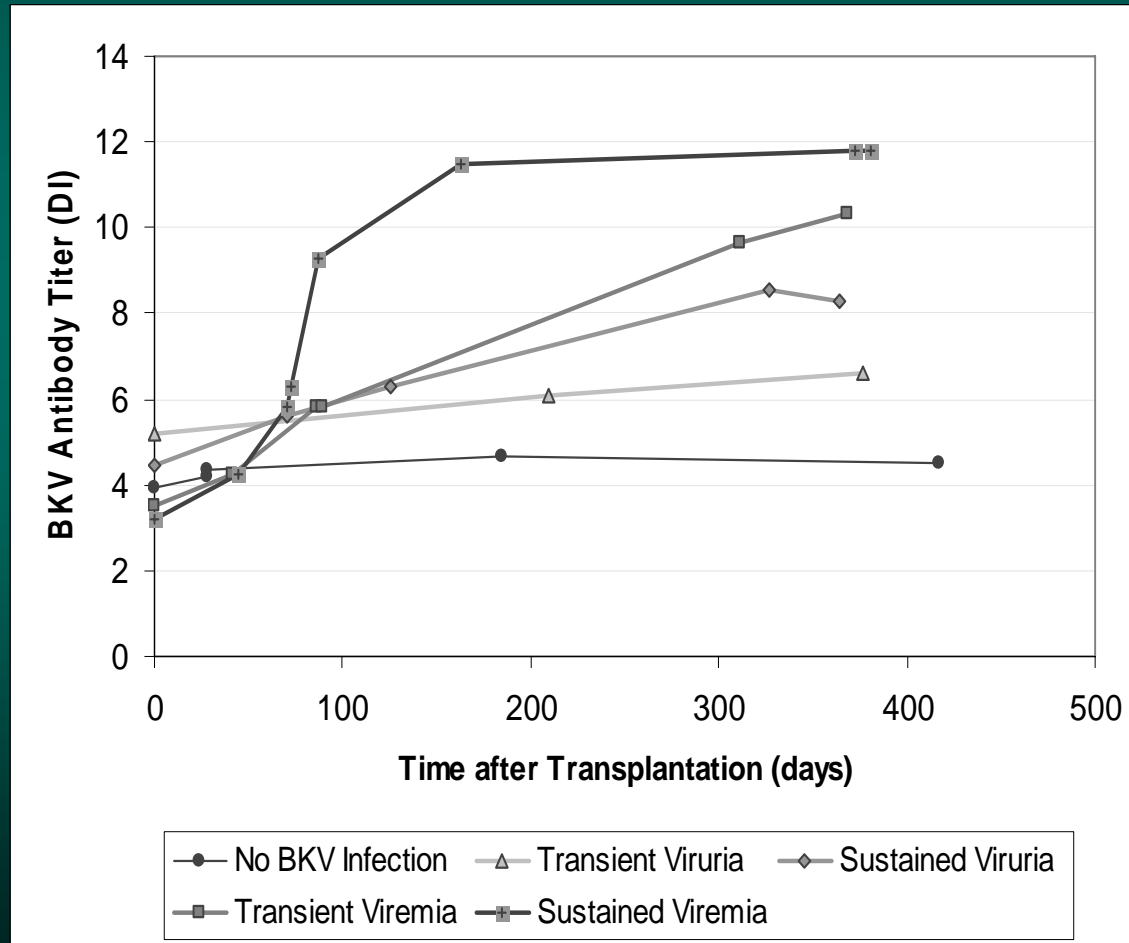
Time after transplant (days)

Bohl, AJT 2005; 5:2213-21

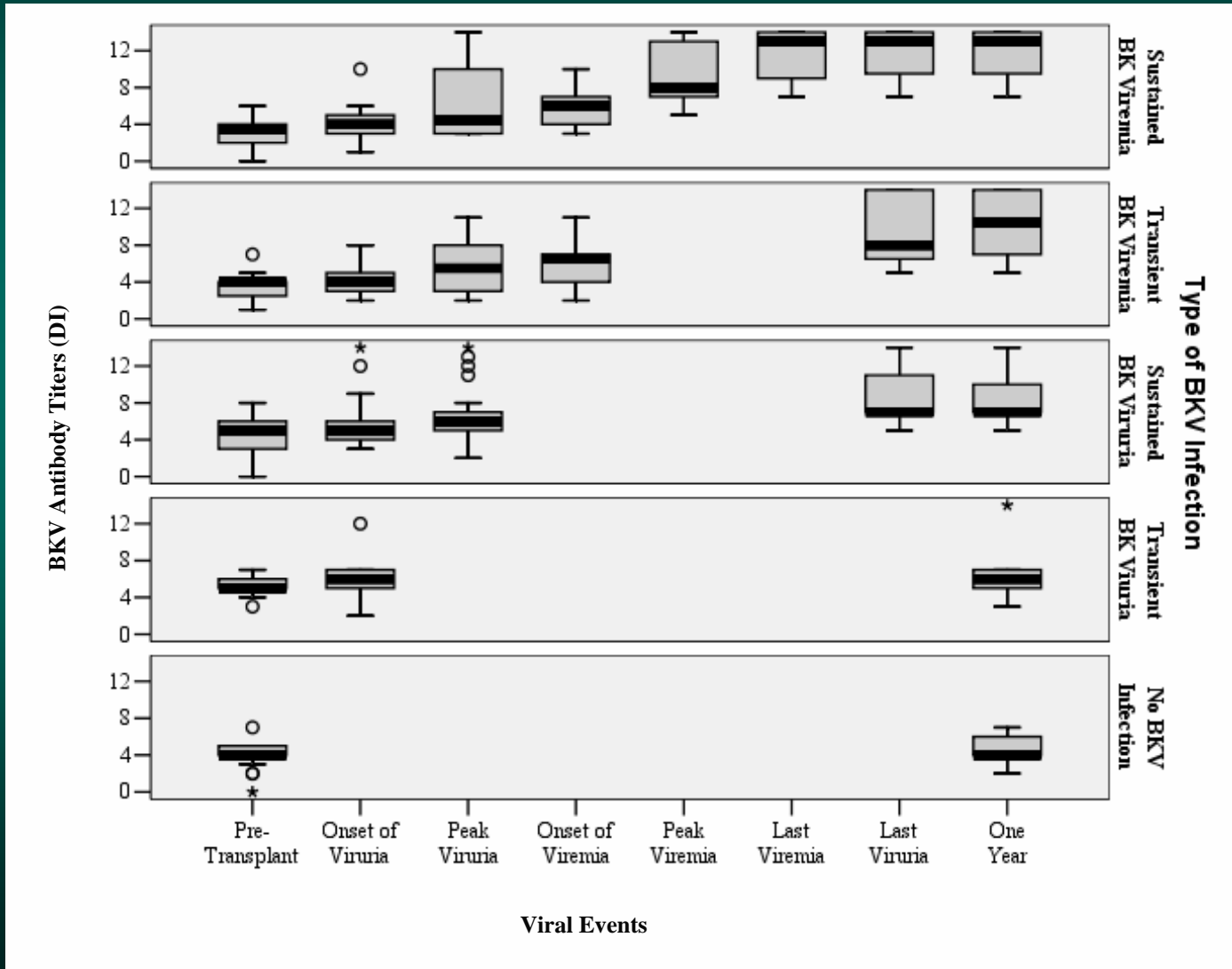
BKV Antibody Levels Pre-Transplant and at 1-Year Post-Transplant per Intensity of BKV Infection



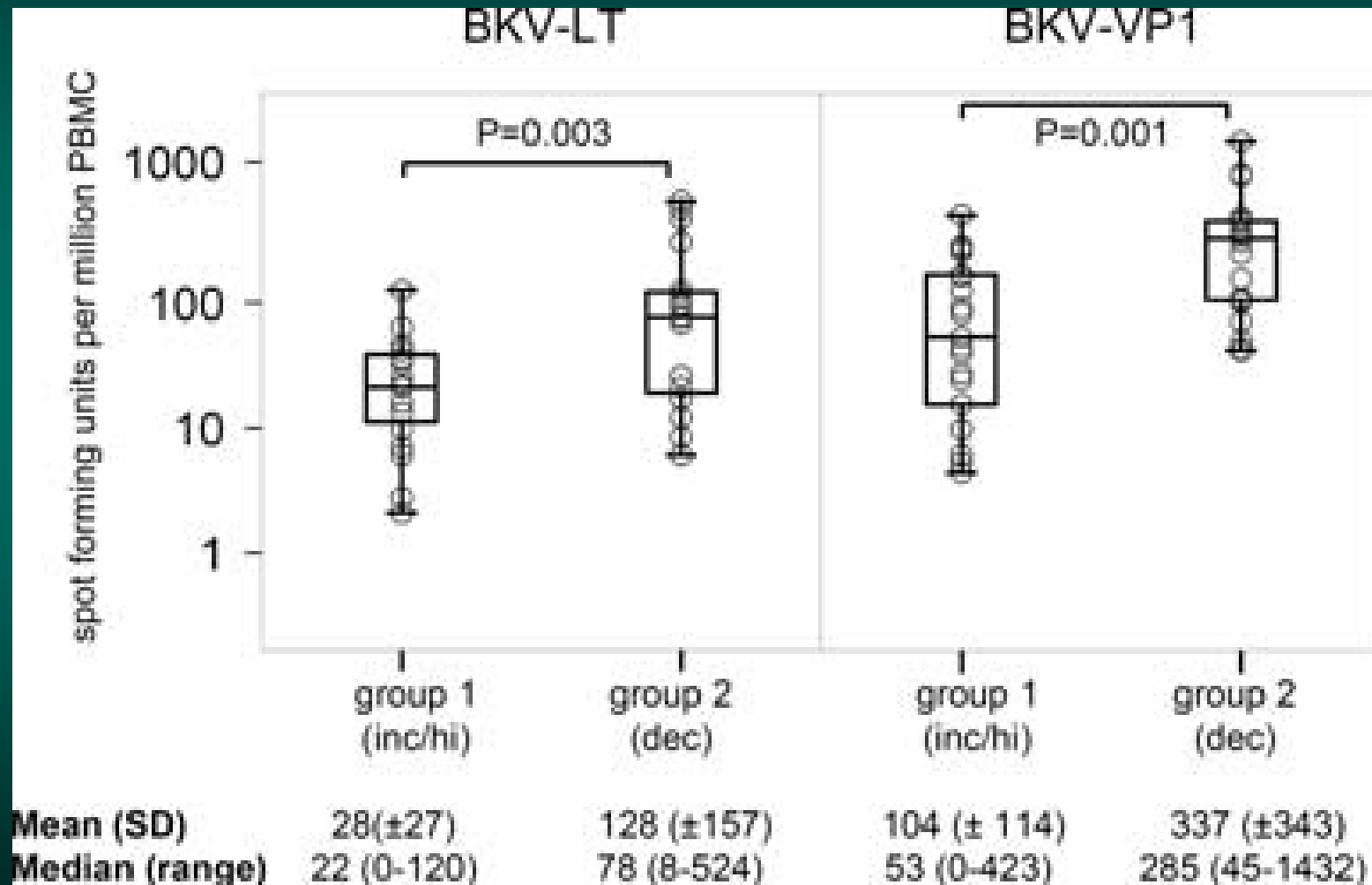
Post-transplant BKV Antibody Response Related to Time After Transplant



Post-transplant BKV Antibody Response Related to Virologic Events



The Cellular Response May be Important for Viral Clearance



Treatment of BKVN

- **Prevention:**
 - **Immediate withdrawal of the antimetabolite upon detection of viremia**
 - **Protocol tapering of maintenance immunosuppression**
- **Treatment**
 - **Decrease immunosuppression**
 - Calcineurin
 - antimetabolite
 - **Possible adjunctive treatment**
 - Cidofovir
 - Leflunomide
 - Quinolone antibiotics
 - Intravenous immunoglobulin

Brennan DC et al. *Am J Transplant* 2005;5:582-594; Bressollette-Bodin C et al. *Am J Transplant* 2005;5:1926-1933; Hirsch HH et al. *Transplantation* 2005;79:1277-1286; Vasudev B et al. *Kidney Int* 2005;68:1834-1839; Leung AY et al. *Clin Infect Dis* 2005;40:528-537; Williams JW et al. *N Engl J Med* 2005;352:1157-1158; Sener A et al. *Transplantation* 2006;81:117-120; Randhawa PS. *Clin Infect Dis* 2005;41(9):1366-1367; author reply 1367.

Our Screening Protocol

BLOOD BK-PCR MONTHS 1 to 6, 9 and 12, and UPON RENAL DYSFUNCTION

Only two patients developed BKV viremia by day 30

One patient developed BKV nephropathy between months 3-6

BLOOD BK-PCR⁺

Increased serum creatinine

Normal serum creatinine

Test Urine BKV PCR?

Biopsy

Decrease Immunosuppression and Monitor q 2 weeks until clear

Rejection + BKVN

BKVN alone

No BKVN

Increased creatinine

Normal creatinine

1) Rx rejection with IVIG & consider 2-6

2) Decrease immunosuppression

3) Quinolone or cidofovir?

4) Leflunomide to replace the antimetabolite?

5) Monitor creatinine

6) Monitor BK q 2 weeks until clear. Consider increasing immunosuppression after 6-12 weeks.

Repeat biopsy

Follow-up

Management of BK-nephropathy after Return to Dialysis

- Transplant and native nephrectomy?
 - No, ignores other uroepithelium and sites
- Assess BK-viruria if urine output
- Assess BK-viremia if anuric
 - Transplant when BK cleared?
- Transplant nephrectomy and simultaneous living donor kidney transplant?¹
- Immunosuppression?
- Post transplant monitoring?

¹Womer K et al. American Journal of Transplantation 2006;5

Conclusions

- BK infection occurs commonly and early after renal transplantation
- Donor antibody titer is the biggest risk
- Lack of donor or recipient HLA-C7 was associated with BK infection
- Viruria precedes viremia
- Urine or Blood PCR are excellent screening tools for BK
- No difference in BK infection with FK vs CyA or MMF vs Aza

- Preemptive d/c of the antimetabolite prevents progression from viremia to nephropathy, safely
- Other strategies have not been evaluated prospectively
- No treatment exists, Cidofovir, Leflunomide, and IgG are used
- The quinolones may have a role in treatment