The transcriptome of Antibody-Mediated Rejection in Human Renal Allografts

Banu Sis[1,2], Luis Hidalgo[1], Jeff Reeve[1], Thomas F. Mueller[1], and Philip F. Halloran[1]

[1]Medicine, Division of Nephrology and Transplantation Immunology, [2]Laboratory Medicine and Pathology, University of Alberta, Edmonton, Alberta, Canada.

The pathogenesis of antibody-mediated rejection (ABMR), and the effects of ABMR on the transcriptome are currently unknown. Using Affymetrix microarrays, we analyzed gene expression in 177 renal allograft biopsies for cause. We previously identified "pathogenesis based transcript sets" (PBTs) associated with cytotoxic T cells (CATs), macrophage activation (IMATs), and gamma interferon effects (GRITs). Geometric means of CATs, GRITs, and IMATs were significantly higher in C4d diffuse+ biopsies compared to C4d- and C4d focal+ biopsies. Moreover, both ABMR and T cell-mediated rejection (TCMR) biopsies showed increased expression of CATs, GRITs, and IMATs compared to biopsies without rejection ($p<.05$). We hypothesized that alloantibody acting on the microcirculation alters endothelial genes. We identified a literature based endothelial gene set, which is differentially expressed in endothelial cells when compared to non-endothelial cells (n=118 unique genes). Of this list, 16 unique "endothelial cell-associated transcripts" (ENDAT) were differentially increased in ABMR vs.TCMR ($p<.05$). These genes included established endothelial markers such as VWF, PECAM1, SELE, CD34, and cadherin 5. ENDAT geomeans significantly correlated with pathologic features of acute and chronic ABMR: C4d deposition, peritubular capillaritis, glomerulitis (g), glomerular double contours (cg), and peritubular capillary basement membrane multilayering (PTCBMML) ($p<.05$). Using an unsupervised class comparison method, we identified 220 transcripts that are significantly different between ABMR and TCMR. The genes that are selectively increased in ABMR included many ENDATs (7 of 17) ($p<.05$, FDR ≤.05). Thus ABMR creates extensive inflammation in the allograft, as measured by the PBTs, which is quantitatively similar to TCMR. However, increased expression of endothelial genes provides a diagnostic feature of ABMR in renal allografts that distinguishes ABMR from TCMR.

Table 1. Endothelial cell-associated transcripts differentiate ABMR from TCMR.

	Normals	C4d diffuse+	C4d focal+	C4d -	ABMR	TCMR
n	8	17	6	148	15 [a]	22
Endothelial cell-associated transcripts	1.0 ± 0.0	1.1 ± 0.0**	0.9 ± 0.8	1.0 ± 0.8	1.1 ± 0.0**	1.0 ± 0.1
CTL associated-transcripts	1.0 ± 0.0	1.6 ± 0.3*	1.1 ± 0.2	1.3 ± 0.4	1.6 ± 0.3	2.0 ± 0.6*
Ifng induced transcripts	1.0 ± 0.0	2.2 ± 0.4**	1.3 ± 0.4	1.6 ± 0.5	2.1 ± 0.4	2.4 ± 0.6
Macrophage activation-associated transcripts	1.0 ± 0.0	2.2 ± 0.4**	1.3 ± 0.4	1.6 ± 0.5	2.1 ± 0.4	2.4 ± 0.7

Correlation coefficients between ENDATs and pathologic features

	C4d deposition	Peritubular capillaritis	g	cg	PTCBMML	t
Endothelial cell-associated transcripts	.376**	.252*	.248**	.261**	.266*	.139

For gene sets, numbers indicate geometric mean ± SD. Normals represent normal cortical tissues from native nephrectomies with cancer. [a] mixed ABMR and TCMR cases were excluded. **p<.001, *p<.05