Dr. Megan Sykes

We will test the potential as a biomarker of a high throughput T cell receptor (TCR)-based sequencing method that we have developed for identifying and tracking a large set of donor-specific alloreactive TCR sequences in human transplant recipients. We have used this method to demonstrate a role for clonal deletion in the tolerance achieved in patients who received combined kidney (KT) and bone marrow transplants (BMT) from HLA-mismatched donors and achieve only temporary chimerism. We have further validated the method by demonstrating the marked enrichment of donor-specific TCR sequences in rejecting intestinal allograft mucosal biopsies. In preliminary studies, we have shown that the urine of renal allograft recipients undergoing rejection contains TCR DNA from donor-reactive T cell clones and have obtained evidence that the presence of these clones early post-transplant may predict rejection within the first year. Our method is unique insofar as it specifically tracks T cells that are specific for the donor and therefore may specifically distinguish rejection activity from other types of inflammation. We now propose a pilot study to investigate the potential of urinary donor-specific TCRs as a predictor of graft dysfunction in the first year and as a non-invasive marker for subclinical graft rejection that could be used to supplement or replace the use of surveillance biopsies and permit better tailoring of immunosuppressive therapy to the individual patient. Specifically, we aim to:

1) **Correlate donor-reactive TCR sequencing of PBMC and urine at 1 year with surveillance allograft histology.** If donor-reactive TCR sequences in urine correlate with the presence of biopsy-determined T-cell mediated rejection, future studies of will determine the potential of donor-reactive TCR sequence tracking as a non-invasive biomarker of subclinical rejection. We will also assess the potential of this approach to predict future graft dysfunction.
2) **Investigate the detection of donor-specific TCRs in the urine early post-transplant as a predictor of graft rejection in the first year post-transplant.** Preliminary data shows a strong association between the detection of donor-specific CD4 TCR DNA in the urine 30-40 days post-transplant and the development of rejection during the first year. If validated in a larger cohort of subjects, this non-invasive assay may be used to guide the personalization of immunosuppression in the future.

Results of these studies will guide future prospective cohort studies to determine the predictive and diagnostic potential of this novel biomarker approach.