Kidney and heart allograft transplantations are often life-saving therapies for the end-stage disease of these organs. However, the newly transplanted organ is susceptible to hyperacute, acute, and chronic rejection. The introduction of the crossmatch test (mixing donor leukocytes with recipient’s serum) in the 1960s eliminated nearly all cases of hyperacute rejection. Similarly, the successful development in the 1980s-1990s of effective immunosuppressive treatments minimized problems with acute rejection. Despite this progress, almost half of kidney or heart allograft patients develop irreversible changes called chronic rejection. In particular, cardiac allograft vasculopathy (CAV) accounts for 32% of deaths within 5 years; however, CAV is also present in 53% of surviving patients at 10 years after transplantation. Similarly, chronic allograft nephropathy (CAN) is detectable in 45% of kidney transplants 8 years after transplantation. Although the diagnoses of CAV and CAN have been extensively described in the Banff pathological classifications, the exact mechanisms are poorly understood. Most importantly, both CAV and CAN lack any effective therapies.

What exactly is chronic allograft rejection? This is a slowly acting or even subclinical inflammatory process that affects graft blood vessel walls to cause local smooth muscle cell (SMC) proliferation. This expansive SMC proliferation eventually occludes blood flow through the graft and leads to graft failure.

Caitlin Baum BSc, an MD/PhD student in my lab, and I have been working on explaining some important aspects of the chronic rejection process. We propose that a unique cytokine, interleukin-21 (IL-21), is intimately involved in the chronic rejection process. Indeed, IL-21 is the latest addition to the entire family of very important cytokines (IL-2, -4, -7, -9, -15, and -21) all of which use a common-y chain (yc) receptor in addition to a private α chain receptor that is unique for each cytokine. All of these cytokines are required for the full function of T and B cells, which
also are necessary for different phases of allograft rejection (Figure 1). It is well established that IL-2, IL-7, and IL-15 signal through Janus tyrosine kinase 3 (Jak3) and signal transducer and activator of transcription 5 (Stat5) molecules for the most optimal T cell response resulting in an acute allograft rejection.

The work performed in my lab suggests that IL-21, signaling through Jak3 and Stat3, is necessary for chronic allograft rejection. Our recent publication already showed that IL-21 is necessary for T-cell survival and proliferation using a unique model of IL-2-deprived culture conditions. Other authors’ published work further revealed that IL-21 is a growth factor inducing cell division/proliferation of multiple immune cells and that IL-21 is critical in the development of chronic auto-immunities and chronic anti-viral responses. Based on these preliminary data, we plan to discern the exact role of IL-21 in chronic rejection and, in particular, the effects of IL-21 on T and B cells. Further work is planned to investigate whether IL-21 affects the activation and proliferation of endothelial cells and smooth muscle cells. Based on this experimental study, we want to propose a novel therapeutic intervention with IL-21 receptor fusion protein (IL-21R-Fc) to prevent chronic rejection.

Over the last two years work in my lab has generated results suggesting that IL-21 regulated chronic immune responses during cardiac allograft rejection. Indeed, the survival of heart allografts was evaluated in wild-type mice and in IL-21 knockout mice (IL21-/-) lacking a functional IL-21 cytokine. The results confirmed that heart allografts were protected from developing chronic rejection in IL21-/- mice (survival more than 100 days) but were susceptible to rejection in wild-type recipients (56.3 ± 22.9 days; n=10; p<0.001). The protection from chronic rejection in IL-21-/- recipients was further confirmed with reductions in pathological scores showing reduced smooth muscle cell proliferation. The IL-21Rfc therapeutic agent also protected against chronic rejection.

The work shows that IL-21 is involved in regulating chronic rejection process. This raises hope for using agents similar to IL-21Rfc fusion protein for preventing chronic rejection changes. An important benefit to IL-21 inhibition is that this pathway is unique to chronic immune responses; therefore, any blockade should not render the patient immunocompromised. Further experiments will shed more light on the exact mechanism of chronic rejection.

Clinical Research Snippets

Anand B. Mutgi, MD
Sadik A. Khuder, PhD

Dietary recommendations for fat intake by various medical societies have varied over the last several decades. Recently a consensus has emerged, recommending decreased intake of Trans-fat and saturated-fat and increased intake of poly-unsaturated fat (canola, sunflower oil) and mono-unsaturated fats (olive oil, peanut oil). However, the impact and magnitude of this recommendation is unclear despite New York City banning Trans fats. This month we review a study that examined the associations of specific dietary fats and mortality.

Investigators used data from 2 very large cohort studies, the Nurses’ Health Study (NHS) and the Health Professionals Follow-up Study (HPFS). The NHS data were...
obtained from 83,349 registered Female nurses aged 30 to 55 years in 1976, who responded to the semi-quantitative food frequency questionnaire (SFFQ) in 1980. The HPFS data were obtained from 51,529 Male health care professionals aged 40 to 75 years in 1986. Both cohorts were free of cardiovascular disease, cancer, and types 1 and 2 diabetes at baseline and were followed through biennial mailed questionnaires that inquired about lifestyle risk factors and other exposures of interest, as well as newly diagnosed diseases. Dietary information was collected with SFFQs and this included fat consumption from different sources during the preceding year. Analysis was conducted to assess the association of cardiovascular disease, mortality and total mortality controlling for other risk factors.

Higher intakes of polyunsaturated fatty acids and monounsaturated fatty acids and lower intakes of saturated fatty acids and trans-fatty acids were associated with lower mortality (21%, 11% respectively).

Replacing saturated fat with total carbohydrates yielded minimal health benefits. Replacing saturated fat with monounsaturated fat and/or polyunsaturated fat was associated with a significantly lower risk for total and cause-specific mortality due to several major chronic diseases. Replacing just 5% of saturated fat reduced mortality by 13 to 27%. Intake of linoleic acid, the most abundant Omega-6, showed strong inverse associations with total and cause specific mortality.

Based on the above observational (cohort) data we feel that replacement of saturated fats with canola oil (polyunsaturated fat) or olive, peanut oil (monounsaturated fat) should continue to be a key message in dietary recommendations. The study also supported the elimination of trans-fatty acids (partially hydrogenated oils like margarine).

**New Clinical Trials**

Prospective Observational Trial to Evaluate the Correlation of T-SPOT® response to CMV Disease and T cell Mediated Acute Graft Rejection.  
Dr. Ortiz - Surgery

A151216: Adjuvant Lung Cancer Enrichment Marker Identification and Sequencing Trial (ALCHEMIST).  
Dr. Skeel - Medicine

A081105: Randomized Double Blind Placebo Controlled Study of Erlotinib or Placebo in Patients with Completely Resected Epidermal Growth Factor Receptor (EGFR) Mutant Non-Small Cell Lung Cancer (NSCLC).  
Dr. Skeel - Medicine

E4512: A Phase 3 Double-Blind Trial for Surgically Resected Early Stage Non-Small Cell Lung Cancer: Crizotinib versus Placebo for Patients with Tumors Harboring the Anaplastic Lymphoma Kinase (ALK) Fusion Protein.  
Dr. Skeel - Medicine

Adjuvant Nivolumab in Resected Lung Cancers (ANVIL) - A Randomized Phase 3 Study of Nivolumab After Surgical Resection and Adjuvant Chemotherapy in Non-Small Cell Lung Cancers.  
Dr. Skeel - Medicine

**IRB Corner**

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