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Utilization of a Cell Mediated Immunity Assay to Adjust Immunosuppression Following Heart Transplantation

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University of Nebraska, 2021

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ABSTRACT

While heart transplantation remains the gold standard therapy for end-stage heart failure, complications remain common post-transplant. Infection is a common cause of morbidity and mortality within the first-year post-transplant. ImmuKnow (ViraCor-IBT Laboratories Inc.) is a cell mediated immunity (CMI) assay utilized in transplant populations to monitor the degree of immunosuppression. We aimed to determine whether utilization resulted in lower rates of infection and immunosuppression-related side effects. This was a prospective interventional trial of transplant patients from June 2018-June 2019. CMI was assessed at standard time points and adjustments in tacrolimus were made per set protocol. Outcomes were compared to historical controls. Thirty-one patients were enrolled in the intervention and control groups. There were no differences in average CMI levels between patients with infection versus those without infection. There were no significant differences in the number of patients with infections. Nine patients had bacterial infections within the 1st-year in the interventional group compared to 12 patients in the control group (p=0.6). Nine patients had a viral infection in the interventional group versus eight patients in the control group (p=0.7). There were no differences in rejection episodes between groups. There was no difference between groups in renal function or blood sugar control over one-year follow up. Use of CMI assay post-heart transplant did not result in lower incidence of infection nor in differences in renal function or blood sugar control. Further studies are needed to better evaluate the utility of routine use of this assay to guide immunosuppression following heart transplant.

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WBC	white blood cell
FDA	Food and Drug Administration
CMI	cell mediated immunity
CMV	*cytomegalovirus*
ACR	acute cellular rejection
AMR	antibody mediated rejection
ISHLT	International Society for Heart and Lung Transplantation
GFR	glomerular filtration rate
LVAD	left ventricular assist device
UNOS	United Network for Organ Sharing

CHAPTER 1: INTRODUCTION

While heart transplantation remains the gold standard therapy for the treatment of end-stage heart failure, there are more patients listed for transplantation than organs available.[1] This means that monitoring and management strategies that aim to reduce graft loss and mortality following transplantation are vitally important to help improve optimal allocation of organs and longevity of transplants. Heart transplant recipients require lifelong immunosuppression to help reduce the risk of graft loss from rejection. Immunosuppression must be finely balanced to not only avoid rejection, but also minimize the risk of infectious episodes from over-immunosuppression as well as reduce the development of medication-induced side-effects and complications.[2]

Classically, physicians and providers utilize trough levels of immunosuppressants, laboratory tests assessing white blood cell (WBC) and lymphocyte counts, time since transplantation, renal function, and rejection and infection history to guide specific immunosuppressant use and dosing.[2,3] However, these strategies do not reliably assess the patient's true cellular immune function, with several studies showing immunosuppression doses and common laboratory variables do not strongly correlate with in-vivo cell-mediated immunity.[4-6] To provide better monitoring of immunosuppression, the Food and Drug Administration (FDA) approved the use of ImmuKnow (ViraCor-IBT Laboratories Inc., Lee's Summit, MO.) in 2002 in solid-organ transplant recipients for better assessment of a recipient's cell mediated immunity (CMI). While both retrospective and prospective studies have investigated the utility of assessing CMI to identity risk of rejection and infection across all types

of solid organ transplant recipients[4-11], there is limited data on any prospective protocol utilizing this assay in heart transplant recipients.

We aimed to determine whether utilization of a cell-mediated immune assay in a prospective, protocolized fashion in heart transplant recipients would improve infection and rejection rates as well as reduce immunosuppression-related side effects in real-world clinical practice.

CHAPTER 2: METHODS

Study Design

This was a prospective interventional trial comparing outcomes of adult heart transplant recipients whose immunosuppressive regimen was monitored and adjusted utilizing a cell-mediated immune assay to historical controls utilizing previous standard of care. Outcomes were assessed up to 12 months after transplantation in both the intervention and control groups. Heart transplant recipients in the intervention group were enrolled from June 2018 through June 2019. Patients receiving a heart transplant from January 2015 through December 2015 were utilized as the historical controls. This study was approved by the institutional review board at our institution.

Patient Population

Consecutive solitary heart transplant recipients were prospectively monitored for degree of immunosuppression utilizing the cell-immune assay after a change in our program's immunosuppression monitoring policy and algorithm. Multi-organ transplant recipients were excluded from this policy. Historical controls included all solitary heart transplant recipients transplanted during the calendar year of 2015 as this as the last year a cell-mediated immunity

assay was not utilized routinely in a non-protocolized fashion for immunosuppression monitoring.

Cell Mediated Immunity Testing

We utilized the commercially available ImmuKnow (ViraCor-IBT Laboratories Inc., Lee's Summit, MO.) assay which has been approved by the U.S. FDA to measure and monitor underlying immune response in solid-organ transplant recipients receiving immunosuppressive therapy. This assay measures cell-mediated immunity via quantification of ATP concentration released from CD4+ T-lymphocytes. This ATP release is triggered via mitogenic stimulation and cells are then lysed to release intracellular ATP. Based on previous studies in solid organ transplant populations, levels of ATP activity of 225 ng/mL or lower indicate a low cellular immune response, levels of 525 ng/mL or higher indicate a strong cellular immune response, and levels of 226 ng/mL to 524 ng/mL suggest a moderate cellular immune response.

Cell-mediated immunity was assessed as part of the pre-transplant evaluation process and repeated every six months as part of waitlist management. Following heart transplantation, an immune function assay was assessed on post-operative day 1 and with endomyocardial biopsy collection and/or outpatient visits at weeks 1, 2, 3, 4, 6, and 8, and months 3, 6, 9, and 12.

Immunosuppression and Intervention

Immunosuppression was administered via induction therapy and a triple-drug regimen according to our institutions standard practice. Induction therapy consisted of basiliximab 20mg 1 hour prior to incision and on post-operative day 4 and 1 g methylprednisolone 1 hour prior to incision. Post-operative steroid tapering was as follows: methylprednisolone 125mg every 8 hours for six doses followed by prednisone 60mg daily for 2 days, 50mg daily for 2 days, 40mg

daily for 2 days, 30mg daily for 2 days, and then a maintenance dose of 20mg daily. Prednisone is gradually decreased and stopped on a patient-by-patient basis over subsequent 6 months. Methylprednisolone doses were converted into an equivalent prednisone dose for direct comparison over time between the two groups. Mycophenolate mofetil is initiated on post-operative day 1 at 2000mg per day in recipients 50 years of age or older and 3000mg per day in recipients younger than 50 years of age.

Tacrolimus was initiated by post-operative day 5 once stable renal function and good urine output achieved. In historical controls, doses were adjusted in 0.5 to 1mg increments to achieve a target trough level of 10-12ng/mL early post-operatively and gradually adjusted over time depending on patient's glomerular filtration rate and time since transplant as well as rejection and infection history, generally targeting a trough level of 6-12ng/mL depending on a combination of these factors. In the intervention group, tacrolimus was also initiated by post-operative day 5 again when renal function was stable, but cell-mediated immunity was utilized to adjust tacrolimus dose based off the algorithm shown in Table 1. Immune response levels between 130 and 450ng/mL of ATP were utilized as the target range as these levels have been shown in several previous trials as thresholds for increased risks of infection or rejection and have also been used as target goals in a prospective interventional trial in liver transplant recipients.[12]

Time since transplant and ImmuKnow ranges	Goal FK level in patients with GFR > 40	Goal FK level in patients with GFR < 40
0-3 Months		
ImmuKnow <130	**8-10 ng/ml**	**6-8 ng/ml**
ImmuKnow 130-450	**10-12 ng/ml**	**8-10 ng/ml**
ImmuKnow > 450	**10-12 ng/ml**	**10-12 ng/ml**
3-12 Months		
ImmuKnow <130	**6-8 ng/ml**	**4-6 ng/ml**
ImmuKnow 130-450	**8-10 ng/ml**	**6-8 ng/ml**
ImmuKnow > 450	**10-12 ng/ml**	**8-10 ng/ml**
> 12 Months		
ImmuKnow <130	**4-6 ng/ml**	**4-6 ng/ml**
ImmuKnow 130-450	**6-8 ng/ml**	**6-8 ng/ml**
ImmuKnow > 450	**8-10 ng/ml**	**8-10 ng/ml**

Table 1. Tacrolimus adjustment protocol utilizing cellular immune assay in conjunction with renal function and time post-transplant.

Rejection and Infection

Monitoring for rejection was performed utilizing right ventricular endomyocardial biopsies as per our program's standard monitoring schedule at weeks 1-4, 6, 8, and 12 and at 12 months. Gene-expression testing via AlloMap profile (CareDx, Inc., Brisbane, CA) was utilized for rejection surveillance starting at 4 months with abnormal scores followed up via endomyocardial biopsy. Rejection was defined as acute cellular rejection (ACR)[13] and antibody mediated rejection (AMR)[13,14] utilizing International Society for Heart and Lung Transplantation (ISHLT) definitions.

We monitored for infection utilizing clinical assessment via history and physical exam along with laboratory, microbiologic, and imaging studies as clinically indicated. Infection was defined as signs/symptoms of infection with concurrent microbiologic and/or imaging findings consistent with infection that required initiation or escalation of antimicrobial therapy for treatment.

Infection prophylaxis was utilized following heart transplantation and at our center includes use of trimethoprim/sulfamethoxazole 60mg/800mg thrice weekly for the first 12

months following transplantation as *Pneumocystis carinii* and *Toxoplasmosis gondii* prophylaxis. Valganciclovir or valacyclovir are utilized depending on donor and recipient cytomegalovirus (CMV) status for the first 3 months following transplant for CMV prophylaxis.

Outcomes

The primary outcome of our study was the rate of infection and rejection episodes in the intervention and control groups. Secondary outcomes included one-year survival, development or progression of post-transplant renal dysfunction (as assessed by need for dialysis, creatinine level, and glomerular filtration rate (GFR) as measured by the formula: 175 x $(S_{cr})^{-1.154}$ x $(Age)^{-0.203}$ x (0.742 if female) x (1.212 if African American), development or worsening of diabetes (worsening hemoglobin A1c), dose of tacrolimus, and addition of proliferation signal inhibitor.

Statistical Analysis

Baseline continuous variables are presented as mean ± standard deviation and categorical variables are presented as number and percentage. Baseline characteristics were compared between the two groups (CMI versus non-CMI) using the independent groups t-test and nonparametric Mann-Whitney U test for continuous variables and Chi-square test for categorical variables. Time-to-event analyses were performed utilizing the Kaplan-Meier method and compared between the two groups using the Logrank test. Linear mixed modeling with repeated measures over time was used to compare renal function (creatinine and glomerular filtration rate [GFR]), markers of diabetes control (blood sugar and hemoglobin A1c), FK trough level, and doses of immunosuppressive agents between the two groups over the course of study follow up. All statistical analyses were performed using SAS version 9.4 (SAS Institute Inc., Cary, NC).

CHAPTER 3: RESULTS

Patient Characteristics

A total of 33 patients underwent heart transplantation between June 2018 and June 2019. Two patients underwent dual organ transplantation (heart-kidney) and were thus excluded from this protocol leaving a total of 31 patients in the intervention group. A total of 33 patients underwent heart transplantation during calendar year 2015. Two patients underwent dual organ transplantation (heart-kidney) and were thus excluded from the analysis leaving a total of 31 patients in the control group. Patients in the intervention group had an average age of 54.3 (±13.2) year, 9 (29%) were female sex, 14 (45.2%) had heart failure related to an ischemic cardiomyopathy, and 14 (45.2%) had been supported by a left ventricular assist device (LVAD) at the time of transplantation. Compared to the control group, patients in the intervention group had lower rates of previous smoking (p=0.007), lower rates of kidney dysfunction as evidenced by lower creatinine (p=0.007) and higher GFR (p=0.01), and lower total bilirubin (p=0.02) at the time of transplantation. Full baseline characteristics are shown in Table 2.

Characteristic	Intervention Group (N=31)	Control Group (N=31)	p-value
Age, yr (SD)	54.3 (13.2)	54.7 (14.8)	0.9
Female Sex, N (%)	9 (29)	7 (22.6)	0.6
White/Caucasian, N (%)	27 (87.1)	29 (93.5)	0.4
BMI, kg/m^2 (SD)	28.8 (4.8)	28.7 (4.9)	0.9
Ischemic Cardiomyopathy, N (%)	14 (45.2)	15 (48.4)	0.8
Support by LVAD, N (%)	14 (45.2)	21 (67.7)	0.07
Smoking History, N (%)			**0.007**
Current	0 (0)	0 (0)	
Former	16 (51.6)	26 (83.9)	
Never	15 (48.4)	5 (16.1)	
Diabetes Mellitus, N (%)	7 (22.6)	7 (22.6)	1
Hypertension, N (%)	17 (54.8)	15 (48.4)	0.6
Hyperlipidemia, N (%)	23 (74.2)	22 (71)	0.8
Chronic Kidney Disease, N (%)	8 (25.8)	10 (32.3)	0.6
Laboratory Values			
Hemoglobin, g/dL (SD)	11.5 (2.6)	11.3 (2.4)	0.7
Platelet Count, cells/uL (SD)	215,935.5 (80,007.9)	230,354.8 (61,599.5)	0.4
Sodium, mmol/L (SD)	137.6 (3.5)	137 (3.1)	0.4
BUN, mg/dL (SD)	20.8 (8.3)	21.5 (10.4)	0.8
Creatinine, mg/dL (SD)	**1.1 (0.3)**	**1.4 (0.5)**	**0.007**
Glomerular Filtration Rate, mL/min/1.73m^2 (SD)	**80.2 (30)**	**62.7 (21.5)**	**0.01**
Total Bilirubin, mg/dL (SD)	**0.7 (0.4)**	**1 (0.5)**	**0.02**
Albumin, g/dL (SD)	3.9 (0.6)	3.8 (0.8)	0.8
Ejection Fraction, % (SD)	30 (15.3)	26.3 (15.4)	0.4
Right Atrial Pressure, mmHg (SD)	10.9 (6.4)	9.2 (5.5)	0.3
Wedge Pressure, mmHg (SD)	14.6 (8.9)	12.8 (6.6)	0.4
Cardiac Index, L/min/m2 (SD)	2.4 (0.5)	2.6 (0.7)	0.1
Peak VO2, mL/kg (SD)	13.8 (3.4)	14.8 (5.1)	0.5
VE/VCO2, slope (SD)	35.8 (5.7)	34.8 (7.1)	0.6

Table 2. Baseline characteristics of study participants.

CMI Levels

Average CMI levels and standard deviations obtained in the treatment group at standard time points are displayed in Table 3 as well as graphically to visualize trend in Figure 1.

Time Post-Transplant	CMI Level ng/mL (SD)
Day 1	283.8 (241.2)
1 Week	366.1 (210.6)
2 Weeks	326.6 (188.2)
3 Weeks	320 (260.5)
4 Weeks	442.6 (280)
6 Weeks	375.3 (211.3)
8 Weeks	390.6 (204.8)
3 Months	348.1 (190.8)
6 Months	247.5 (120.8)
9 Months	329.4 (177.5)
1 Year	297.3 (177)

Table 3. Average CMI levels collected at standard time points post-transplant

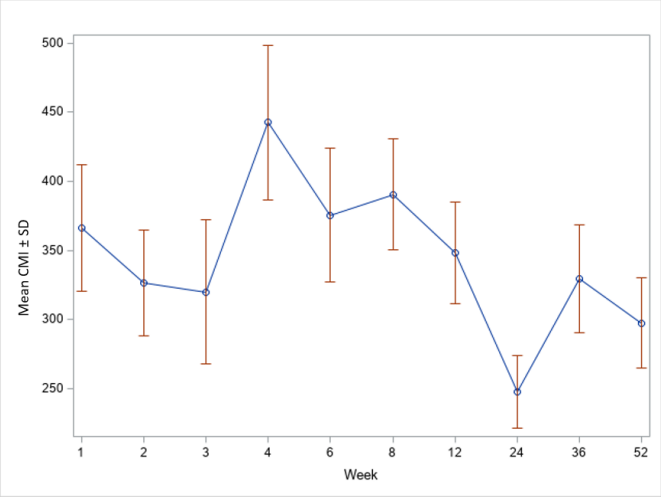

Figure 1. Graphical representation in variation of average CMI level with SD over time post-transplant.

Average CMI levels collected at standard time points were compared between patients in the treatment group who had at least one infection within the first-year post-transplant (N=12) and those patients who did not experience any study-defined infections within the first year (N=19). These results are displayed in Table 4. There were no statistically significant differences in CMI levels at any time point post-transplant between patients with and without at least one infectious episode within the first year.

	Infection N=12	**No infection N=19**	**p-value**
Average CMI Day 1, ng/mL (SD)	125.6 (101.7)	317.1 (275.0)	0.07
Average CMI Week 1, ng/mL (SD)	314.5 (245.4)	426.0 (167.4)	0.2
Average CMI Week 2, ng/mL (SD)	290.8 (108.1)	358.6 (218.8)	0.4
Average CMI Week 3, ng/mL (SD)	278.0 (262.5)	356.5 (269.0)	0.5
Average CMI Week 4, ng/mL (SD)	495.2 (365.0)	420.1 (233.8)	0.5
Average CMI Month 6, ng/mL (SD)	550.7 (410.2)	347.3 (158.6)	0.14
Average CMI Month 2, ng/mL (SD)	385.6 (225.1)	408.9 (195.9)	0.8
Average CMI Month 3, ng/mL (SD)	373.1 (218.8)	336.0 (179.4)	0.6
Average CMI Month 6, ng/mL (SD)	214.9 (58.3)	271.5 (144.1)	0.3
Average CMI Month 9, ng/mL (SD)	347.6 (132.9)	321.6 (213.3)	0.8
Average CMI Year 1, ng/mL (SD)	348.0 (109.2)	268.6 (207.1)	0.3

Table 4. Comparison of average CMI levels within the treatment group in those with and without an infectious episode.

Survival

There was no difference in 1-year survival between the two groups with 93.5% survival in the intervention group and 93.5% survival in the control group. Two deaths occurred within the first year in the intervention group, one from primary graft dysfunction and the second from infectious cause, specifically pseudomonas peritonitis. Two deaths also occurred within the first year in the control group, both deaths were from primary graft dysfunction.

Infectious Episodes

There was no difference in the number of bacterial infections between the two groups. In the intervention group, a total of 9 patients (29%) had a bacterial infection within the first year compared to 12 patients (38.7%) in the control group (p=0.3). The intervention group sustained 14 bacterial infections within the first year compared to 16 bacterial infections in the control group (p=0.6). Bacterial infections in the intervention group included: 3 episodes of *clostridium difficile* colitis, 2 urinary tract infections, 2 episodes of mediastinitis, 1 empyema, 1 episode of peritonitis, 1 episode of upper extremity cellulitis, and 4 episodes access-site infections (utilized for temporary mechanical circulatory support post-transplant). For full comparisons of infectious rates between groups see Table 5.

Viral infections were likewise similar between the groups. In the intervention group, a total of 9 patients (29%) had a viral infection within the first year compared to 8 patients (25.8%) in the controls (p=0.7). There were a similar number of total viral infections, 11, in both the intervention and control groups (p=1). Viral infections in the intervention group included: 6 episodes of CMV, 2 episodes of Norovirus, 2 episodes of respiratory syncytial virus, 1 episode of herpes zoster, and 1 reactivation of a herpes simplex genital infection. Full comparisons are presented in Table 5.

	Intervention Group N=31	Control Group N=31	p-value
Patients with bacterial infection, N (%)	9	12	0.58
Total number of bacterial infections, N	14	16	
Patients with clostridium difficile infection, N (%)	3	5	0.34
Patients with viral infection, N (%)	9	8	0.69
Total number of viral infections, N	11	11	
Patients with CMV infection, N (%)	5	8	0.25
Patients with ACR, N (%)	6	3	0.92
Patients with AMR, N (%)	3	3	0.65

Table 5. Comparison of number of infectious and rejection episodes between groups.

Transplant Rejection

Rejection episodes were similar between the two groups. In the intervention group, 6 patients (19.4%) experienced an episode of ACR compared to 3 patients (9.7%) with ACR in the control group (p=0.92). There were 3 episodes of AMR each within the intervention (9.7%) and control groups (9.7%) (p=1). See Table 5 for comparisons.

Comorbidities and Immunosuppression

Renal function was similar between groups as assessed by changes over time in creatinine (p=0.81) or GFR (p=0.69). There were no significant differences over time in markers of blood sugar control measured via blood sugar (p=0.24) and hemoglobin A1c (p=0.82) between the two groups. We observed no difference over time in regards to trough tacrolimus level (p=0.19) or average dose of immunosuppressive agents between study groups – prednisone (p=0.94), mycophenolate mofetil (p=0.09), and tacrolimus (p=0.52). Average values/levels/doses at study enrollment and other key time points including at one-year follow up are presented in Table 6.

Variable	Pre-Transplant Baseline		3 Months Post-Transplant		6 Months Post-Transplant		1 Year Post-Transplant	
	Treatment Group (N=31)	Control Group (N=31)	Treatment Group (N=31)	Control Group (N=31)	Treatment Group (N=31)	Control Group (N=31)	Treatment Group (N=31)	Control Group (N=31)
Creatinine, mg/dL (SD)	1.1 (0.3)	1.4 (0.5)	1.3 (0.4)	1.4 (0.3)	1.5 (0.6)	1.5 (0.6)	1.4 (0.5)	1.4 (0.3)
Glomerular Filtration Rate, mL/min/1.73 m^2 (SD)	80.2 (30.1)	62.7 (21.5)	62.9 (22.9)	56.9 (16.8)	57.8 (24.4)	56.7 (20.6)	61.2 (22.1)	57.8 (15.5)
Blood Sugar, mg/dL (SD)	122.8 (36.8)	137.5 (41.6)	115.6 (44.8)	121.1 (32.9)	109 (32.7)	112.7 (25.9)	117.7 (43.8)	112.4 (27.7)
Hemoglobin A1c, % (SD)	5.9 (0.7)	5.5 (0.4)					5.9 (0.9)	5.7 (0.8)
FK level, ng/mL (SD)			9.7 (2.9)	9.5 (2.6)	8 (2.6)	8.3 (2.7)	6.9 (3.6)	7.7 (2.4)
Tacrolimus dose, mg/day (SD)			7.1 (3.2)	6 (3.2)	6.2 (3)	5.4 (3.8)	4.4 (3)	5.2 (3.4)
Mycophenolate Mofetil dose, mg/day (SD)			2222 (624.1)	2422.1 (618)	1775.7 (843.9)	2085 (737.7)	1778.1 (793.2)	1655.2 (850.7)
Prednisone dose, mg/day (SD)			9.6 (6.3)	8.1 (3.6)	3.8 (1.3)	5.3 (3)	5 (0)	4.4 (1.3)

Table 6. Comparison of renal function, blood sugar control, FK level, and immunosuppressive doses at key time points between treatment and control groups.

CHAPTER 4: DISCUSSION

This study was the first to prospectively evaluate the utilization of a cell-mediated immunity assay to guide tacrolimus dosing in a protocolized fashion following heart transplantation. No difference in 1-year survival between the two groups was observed. We did not find any difference in rates of total bacterial or viral infections, nor in specific *clostridium difficile* or CMV infections. Utilizing this protocol, we did not find any difference in tacrolimus dosing between groups, nor were there any differences between the two groups regarding changes in renal function or markers of glucose control.

At baseline, patients in the intervention group were less likely to have been former smokers, had better renal function, lower total bilirubin, and there was a trend toward fewer of them being supported with LVAD at the time of transplantation compared to the control group. This likely represents the change in the United Network of Organ Sharing (UNOS) criteria for heart transplant allocation that occurred in October 2018, shortly after our protocol was initiated in June 2018. Several recent studies have shown similar findings, with fewer patients being supported with durable LVAD support and having a lesser-degree of extra-cardiac end-organ dysfunction at the time heart transplantation.[15-18]

Currently, monitoring for degree of immunosuppression and risk of infection, especially within the first-year post-transplant, is performed utilizing close assessment of trough immunosuppressant levels, appropriate immunosuppressant doses, routine laboratory markers like WBC and lymphocyte count, as well as knowledge of both the recipient's and donor's infectious history (e.g., CMV positivity). While multiple factors are utilized to assess risk currently, these factors do not provide good insight into the actual degree of immunosuppression achieved. Several previous studies have shown that trough levels of

immunosuppressants[4-6] and WBC and absolute lymphocyte counts[6] poorly correlate with CMI as measured by the ImmuKnow assay.

We found that cell mediated immunity levels rise slightly over the course of the first 4 weeks post-transplant with a slow decline over the remainder of the year which is a similar trend to that observed by Rossano and colleagues in a pediatric population.[6] Average levels remained within the moderate range of cellular immune response over the course of the first-year post-transplant. While a trend was present CMI levels immediately post-transplant between those who would later develop an infectious episode compared to those without, overall there were no statistically significant differences in CMI levels collected at standard time points. While several previous studies in heart transplant patients have shown that CMI levels are lower during or just prior to infectious episode compared to steady state or during rejection,[5,9] and early meta-analysis including various solid-organ transplants found that routine CMI levels have a relatively low accuracy in identifying infectious risk.[7]

As has been reported previously,[19] we found bacterial infections to be the most common type of infection diagnosed within the first year following transplantation. While previous retrospective reports are conflicting on whether low levels of cell-mediated immunity are[5,9] or are not predictive of infectious events,[6,11] our study did not show that prospective utilization of a cell-mediated immunity assay in a protocolized fashion to adjust tacrolimus dosing led to a significant reduction in rates or total number of bacterial or viral infections. This is contradictory to the only other known study to prospectively use a cell-mediated immunity assay to guide immunosuppression following a solid organ transplant, where Ravaioli and colleagues randomized adult patients following liver transplantation to serial immune function testing compared to standard of practice and found that patients in the intervention group had lower incidence of bacterial and fungal infections.[12] The difference in findings is most likely

related to variations in organ-specific management as well as the smaller sample size in our population.

Similar to previous studies in the heart transplant population[5,6,9] as well as other solid-organ transplant populations,[7,12] we did not find that utilization of serial cellular immune function testing resulted in differences in rates of ACR or AMR. Currently, there are alternative non-invasive tests available to evaluate for the risk or presence of allograft rejection, including gene expression profiling[20] and donor-derived cell-free DNA,[21] that are much more accurate and clinically useful.

No significant differences in doses of standard immunosuppressives, including tacrolimus, were present in our study. This is in contrast to a similar study conducted in liver transplant recipients where Ravaioli and colleagues showed that patients who had their tacrolimus doses adjusted based off CMI levels had lower median tacrolimus doses and trough levels within the first 3 months and lower median doses between 6 and 12 months compared with the standard of care group.[12] We also did not observe any changes over time in markers of renal function or diabetes control between the two groups. This most likely is a direct reflection of the absence of similar doses of immunosuppressive agents, as doses of tacrolimus and corticosteroids would have the biggest impact on these markers.

Limitations

While our study offers significant insight into the use of a cell-mediated immunity assay to adjust immunosuppression in heart transplant recipients, our study is not without limitations. First, this was a single-center study and may not be reflective of the population or clinical practice of other centers. Second, this study was not randomized and utilized historical controls as the comparison group, which can result in inherent biases related to changes in practice

habits over time. However, immunosuppression and infectious prophylaxis protocols did not differ significantly between the two time periods. Third, adjustment of tacrolimus dosing based on CMI results were at the discretion of the treating cardiologist and clinical pharmacist and may have not been performed consistently between individual patients. Fourth, tacrolimus was chosen as the immunosuppression agent of choice for adjustment based on CMI values due the ability to follow trough levels when making appropriate adjustments along with the long-term complications associated with tacrolimus use. It is possible that adjustments of mycophenolate mofetil dosing based on CMI levels may be more important than adjustment of tacrolimus dosing.

Conclusion

Utilization of a cell-mediated immunity assay at routine time points following heart transplantation to make protocol-driven adjustments to tacrolimus doses did not result in lower rates of infection or rejection, nor did it result in reduced doses of immunosuppressants or improved renal function or glycemic control compared to historical controls. While assessment of CMI may be clinically beneficial in high-risk patients with active or recent infection to help guide management, routine use in clinically stable patients at standard time periods following heart transplantation do not appear to be beneficial. A larger, multicenter, randomized controlled trial may be needed to fully assess the utility of routine use of a cellular immune function assay to adjust immunosuppression following heart transplantation, but may be technically challenging given variations in clinical practice post-transplant across institutions.

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