

Operational Tolerance In Living Related Donor Kidney Transplant – A Case Report

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Abstract:

Kidney transplant is the preferred treatment modality for end stage kidney disease (ESKD) patients. Ability to control immune response in the recipient by using lifelong immunosuppression leads to preservation of graft and immune tolerance². Patients with kidney transplant require lifelong immunosuppression to prevent acute rejection. Rarely patients other than fraternal twins, may not require immunosuppression at all and such state is called operational tolerance. Few case reports have been reported in the literature of operational transplant and here we present the first case reported from Pakistan of operational transplant in a female who received kidney transplant donated by her younger sister.

Key Words: Kidney transplant, operational transplant, immunosuppression, acute rejection.

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Introduction

Kidney transplant is the preferred treatment modality for end stage kidney disease (ESKD) patients. The first long-term kidney transplant was done between monozygotic twins in 1954 that did not require immunosuppression and the graft survived for 8 years. Later in 1962, the first transplant between genetically non-related individuals was done with the use of immunosuppression therapy.¹ Ability to control immune response in the recipient by using lifelong immunosuppression leads to preservation of graft and immune tolerance.²

Immune tolerance is the unresponsiveness of immune cells to self-antigens preventing an immune response to antigens produced by the body itself or recognized from a prior encounter. In organ transplantation, immune tolerance leads to long term graft preservation. However, immunosuppressive drugs have their own side effects including infections, risk of malignancies and metabolic disorders.³⁻⁵ Graft dysfunction usually occurs due to non-adherence to the treatment, under dosing or overdosing of the immunosuppressive medications. Cases of preserved graft function without immunosuppression in kidney transplant have been reported pointing towards immune tolerance.⁶⁻⁸ Immune tolerance is defined as a process where immune cells are made unresponsive to the self antigens and here it refers to the process of long-term graft preservation following transplantation. Various studies evaluating immune tolerance have focused on the role played by the mediators of inflammation, T and B lymphocytes, Natural Killer (NK) cells, macrophages, dendritic cells (DCs) and myeloid-derived suppressor cells. The interplay between all these factors and cells determines the outcome of long-term graft survival and at the same time, it might promote an immune tolerance status⁹. Here we discuss a case of kidney transplant from her younger sister done in 1986 and is currently not on any maintenance immunosuppression medication.

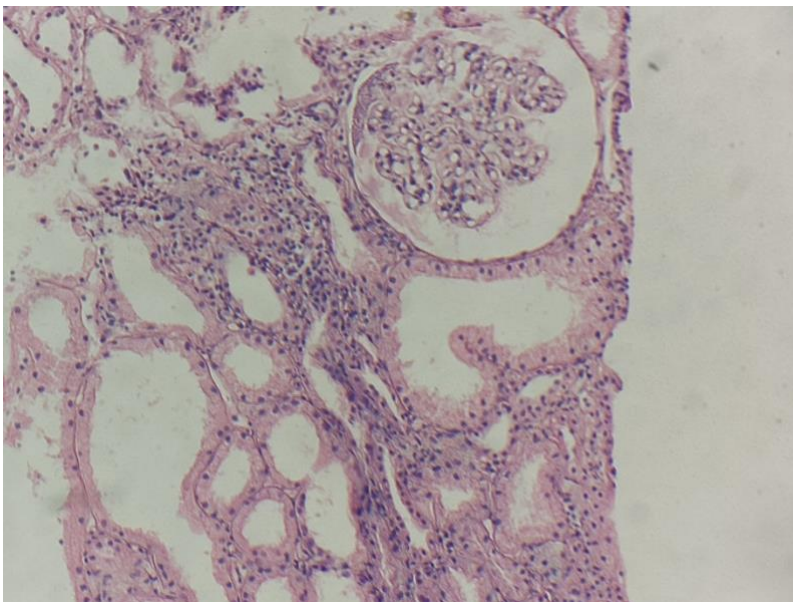
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Case Report

A 68 years old female, known case of Hypertension for last 30 years developed ESKD likely secondary to chronic Glomerulonephritis – native kidney biopsy never done. Patient underwent living related donor kidney transplant in 1986 in Bombay, India. The kidney was donated by her elder sister. Pre-transplant immunological workup including tissue typing and cross matching was not available for our review. Immunosuppression following kidney transplant consisted of oral prednisolone and Azathioprine (AZA). Patient had a stable graft function at discharge with a serum creatinine of 1.0mg/dL. She subsequently lost to follow-up with her transplant team. Immunosuppressive drugs continued until 2006 when she was diagnosed to have Endometrial carcinoma for which she underwent hysterectomy at which point her antiproliferative medication was stopped. She then further stopped taking her steroid as well considering it to be potential risk for her carcinoma recurrence. For the next 14 years, the patient maintained her kidney function off immunosuppression until 2020 when she had an episode of acute gastroenteritis and presented to our emergency with a rise in her serum creatinine to 4.3mg/dl, baseline verbal reported as serum creatinine of 0.9mg/dl previously. She was managed with intravenous hydrocortisone, antibiotics and hydration, and her serum creatinine improved to 2.3mg/dl at discharge. She was also discharged on prednisolone 15 mg daily. Other immunosuppression was considered but not added since she had sepsis and have been doing well without any immunosuppressive medications.

At follow up two weeks later she still had raised serum creatinine of 2.0 mg/dl, doppler ultrasound showed normal sized transplanted kidney in right iliac fossa with normal resistive index for vascular flow. Autoimmune workup was negative. Urine R/E was unremarkable. Transplanted kidney biopsy was done which showed no evidence of rejection, but presence of interstitial inflammation in non-scarred cortical areas (i3) and minimal tubular atrophy (ct1) without interstitial fibrosis Fig 1, Fig 2, Fig 3. Immunohistochemical stains including CD3 and CV40 were negative. C4d staining was minimal (<10%) as shown in Fig 4. Being a unique case, her immunosuppression was discussed in the transplant nephrology group and she was advised to initiate AZA and taper prednisolone to 5 mg daily. She however after 2 weeks, stopped AZA considering it to be unsafe due to her history of endometrial carcinoma and continued with prednisolone at 5 mg daily. Follow-up at 4 weeks after kidney biopsy revealed serum creatinine to be 1.6mg/dl. A follow-up visit at 6 months she had stopped her prednisolone 5 mg daily also, serum creatinine was 1.28mg/dl and bland urine sediment.

At follow-up of 2 years after her presentation, she is not on any immunosuppressive medications and maintaining stable serum creatinine of 1.2mg/dl.



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Figure 1. H&E stain at 100X showing intact glomeruli and tubules (proximal)

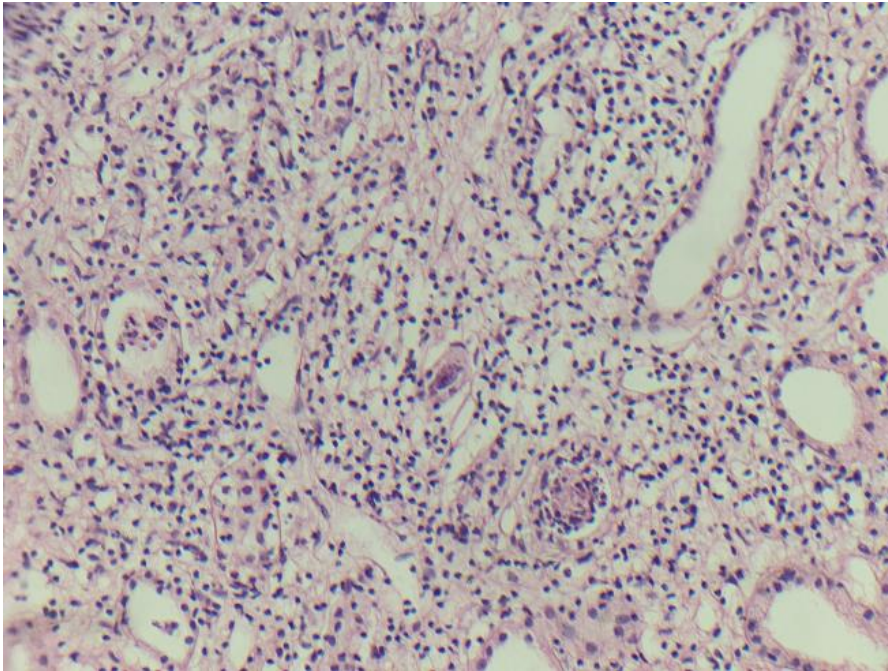


Figure 2. H&E stain at 200X showing chronic interstitial inflammation

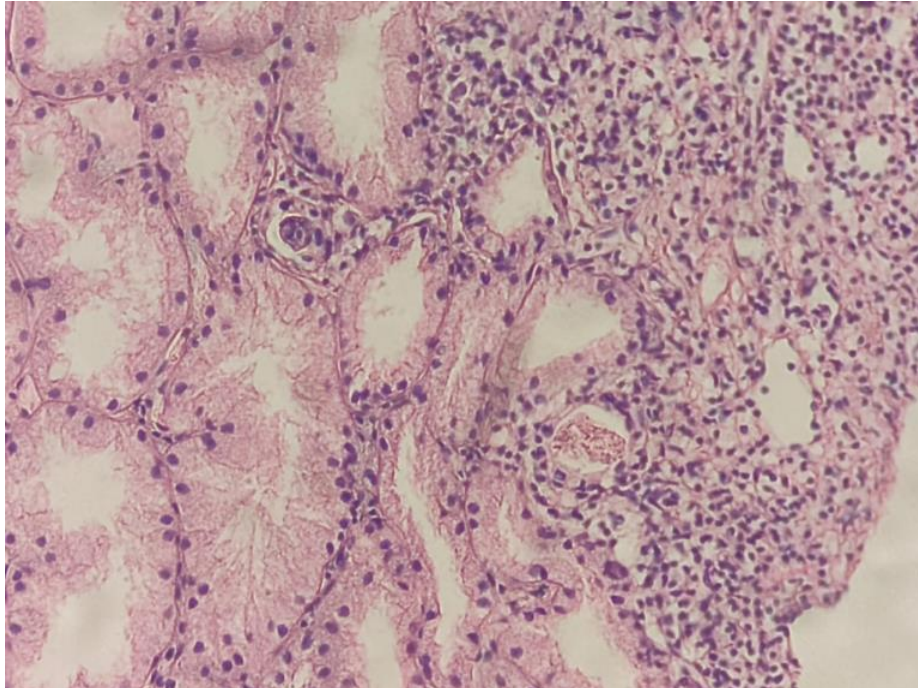


Figure 3. H&E stain at 400X showing cortical interstitial inflammation in cortical areas

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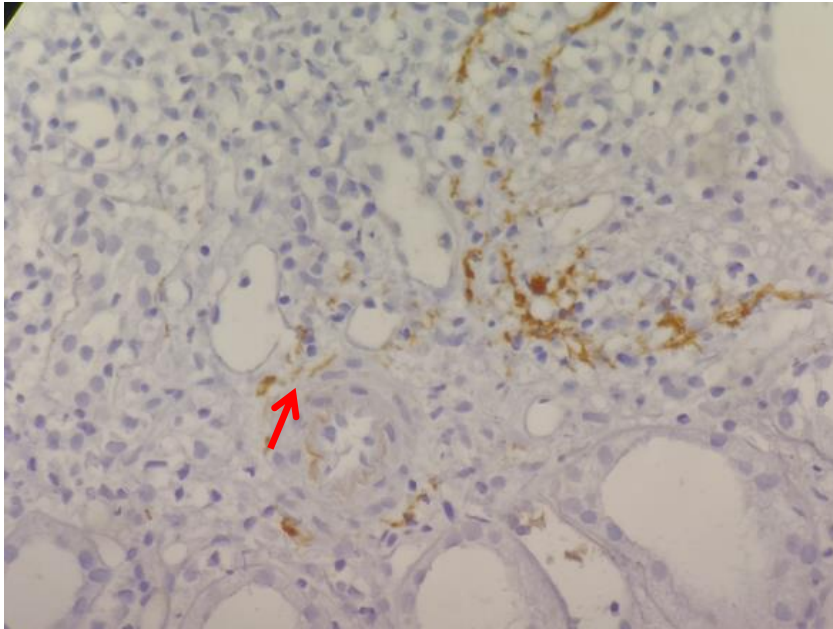


Figure 4. Minimal C4D staining of the allograft biopsy

Discussion

Our above described case was an example of operational tolerance, where patient self-discontinued the immunosuppressive medications for 14 years without any adverse outcomes. Acute Kidney injury is a risk factor for graft dysfunction and graft loss in kidney transplant patients¹⁰. Tolerance refers to non-responsiveness to antigens¹¹. Currently tolerance can be divided into three types as, central tolerance mediated by transplanted donor hematopoietic cells which recognizes donor antigens as ‘self’ antigen, peripheral tolerance mediated by pharmacological therapy leading to depletion or suppression of self-reactive T-cells and operational tolerance in patients who left immunosuppression on their own for more than one year and no destructive alloimmune response was observed¹². More than 200 cases of operational tolerance have been reported¹³ but the exact mechanism remains very vague. Therefore, a deep insight into the operational tolerance related molecular mechanism and biomarkers is necessary.

Identifying transplant recipients in whom immunological tolerance is established or is developing would allow an individually tailored approach to their post transplantation management. An important role in graft rejection and transplant tolerance is played by B-lymphocytes through antibody production, presentation of antigen to T-cells and cytokine production¹⁴. B-lymphocytes pass through different stages of maturation before differentiating into mature plasma cells as shown in Fig-5.

Immature B-cells released from bone marrow are called Transitional B-cells^{15,16}. The memory B response is more potent to antigens than the primary B responses and produces responses with greater affinity and immunoglobulin recombination¹⁴. B- Lymphocytes (BL) can also secrete cytokines that are pro- and anti-inflammatory, modulating the T cell response¹⁷. Naive B-Lymphocytes can differentiate into different cytokine-producing effector subpopulations depending on Th1 or Th2 cells¹⁴.

Specific subpopulations of BLs with immunosuppressive capabilities called regulatory B cells (Breg) have been shown to play an important role in tolerance by inhibiting effector responses. Breg cells are a minority population

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that can arise at various stages of BLs maturational development being abundant within the transitional B, memory B, and plasmablast subpopulations¹⁸.

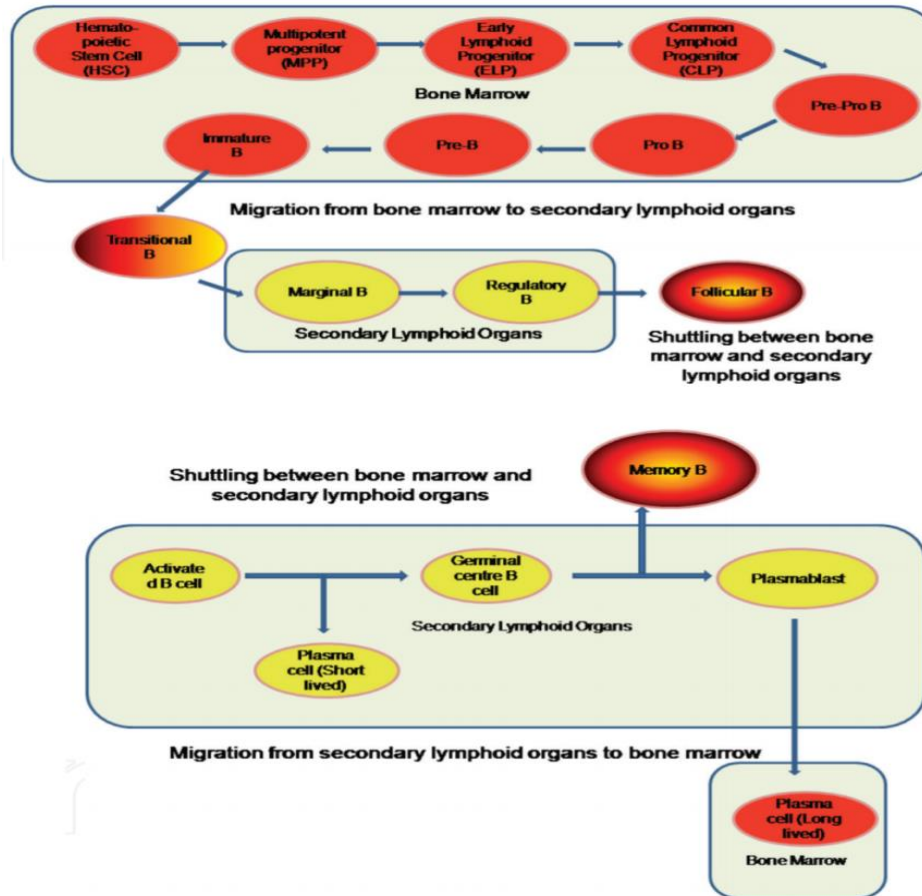


Figure 5: Immune cells production and formation of memory cells in the pathway of immune recognition.

In a multicenter study across Europe enrolling patients showing operational tolerance were compared to patients on different immunosuppression regimens, patients showing chronic rejection, and healthy controls. In this study, in-vitro assays showed an expansion of peripheral blood B and NK lymphocytes, fewer activated CD4+ T cells, a lack of donor-specific antibodies, donor-specific hyporesponsiveness of CD4+ T cells, and among tolerant group a high ratio of forkhead box P3 to α -1,2-mannosidase gene expression.¹⁹ In the maintenance of immunologic tolerance, increased levels of regulatory T cells (T-Regs) play an important role by suppressing overt immune responses.²⁰

In a study of kidney transplantation where B cell depletion with Rituximab was obtained prior to transplantation resulted in higher acute cellular rejections, similar results were observed in islet cell transplantation.²¹ Patients with operational tolerance were also characterized by a higher number of B cells in the blood compared with patients with stable graft function under immunosuppression and patients with antibody-mediated chronic rejection.²² These and other studies have described B cells to differentiate into B regulatory cells (B-Regs) that have important role in immune homeostasis, nicely reviewed Peng B et.al.²³ Studies with Basiliximab induction resulting in an increase of regulatory T cells and various biomarkers that are also observed in patients with operational tolerance.²⁴

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In an earlier study Newel et.al have shown that B cell genetic signature in OT patients was similar to the healthy controls suggesting a B cell dominant state.²⁵ In the same study patients who were using immunosuppression had less B cell genetic expression profiles. This points towards a significant role of B-Regs in the development of immune tolerance and possible use of cell therapies to achieve this goal. This also indicated towards opportunities to develop assays to identify patients with potential of tolerance and gradual withdrawal of immunosuppressive medications. In a recent editorial by Schwarz and Wekerle in Transplantation highlights the problems and the slow but achievable journey of immunetolreance.^{26,27}

Another recently published study in which monitoring of B-Lymphocytes subpopulations was performed by flow cytometry before transplantation and three and six months after transplantation showed an increase in transitional B-Lymphocytes and plasma blasts in conjunction with a better kidney function and lower acute rejection incidence. Transplant recipients with a decreased transitional B-Lymphocytes and plasma blasts were associated with lower kidney function and higher acute rejection in post-transplant period in comparison to those who had an increase in transitional B-lymphocytes during Post transplant period and a better clinical outcome. The increase in transitory B-Lymphocytes during post-transplant period was also associated with an increase in Tregs²⁷.

Our patient had developed OT and recovered from an episode of ATN following acute gastroenteritis. Even in the presence of a kidney biopsy that did not reveal rejection or significant IFTA, and self-discontinuation of AZA, we did not have the courage to stop prednisolone. We therefore continued with small dose of prednisolone, despite the lack of evidence whether its presence is beneficial or might disturb the immune tolerant profiles of T-Regs and B-Regs. We however did not find any study among OT reporting the effects of restarting immunosuppression. Identification of underlying mechanism and targeted cell therapy is probably the holy grail of Immune tolerance and a reality not far away.

Advancements have been made in the recent years in the field of transplant tolerance in an effort to improve graft survival and decreasing the need of long-term immunosuppression (IS) and ultimately saving the patient from their side effects improving mortality and morbidity. Although, research work done in recent years have confirmed the feasibility for inducing transplant tolerance; however, there remains a gap in translating these findings to the clinic in a fear of losing a functioning allograft. To prevent graft loss, immunosuppression regimens are selected based on the likelihood of graft rejection for specific organs with skin and intestine carrying the highest risk of rejection and while heart, kidney and liver carrying a lower risk. Although similar transplant strategies apply to all these organ transplants, the diversity in immunogenicity across tissues poses a challenge in predicting the outcome. Since immunosuppression always comes with a list of side effects, every effort has been made to cut down IS at any time during the life span of patients without compromising graft function. The recent breakthrough is pairing allogeneic Hematopoietic stem cell transplant (HSCT) with solid organ transplant(SOT).²⁹ Besides dramatically increasing the chance of finding a suitable donor for the organ transplant, combining allogeneic HSCT with SOT can positively impact allograft survival and overall clinical outcomes including a decreased or no IS need. However the work is still under progress in this game changing innovation and may lead us to IS free SOTs.

In conclusion Operational tolerance is like the Holy Grail of transplantation and gradual progress is ultimately lead us to achieve a immunosuppression transplantation.

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