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The role role of MHC-specific IgE in antibody-mediated transplant rejection

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Section of Transplantation Immunology, Department of Surgery, Medical University of Vienna, Vienna, Austria

Background

Transplantation is the gold standard for treatment of patients with end-stage organ failure. While the 1-year graft survival rate has increased significantly, long-term allograft survival is still limited due to antibody-mediated rejection (ABMR) caused by either pre-existing or *de novo* donor specific antibodies (DSA) [1]. Especially the occurrence of DSAs of the IgG isotype is strongly correlated with an increased risk of graft loss [2, 3]. To our knowledge our group was the first to describe DSAs of the IgE isotype in mice and humans upon allograft rejection [4].

Methods

Hearts from fully mismatched BALB/c (H-2^d) or Bm12.K^d.IE mice, a recipient model for studies on chronic ABMR, are transplanted heterotopic into C57BL/6 (H-2^b) mice [8]. The parental strains of this donor model have to be revitalized from frozen embryos by our animal facility. As an additional murine model we will employ CCR5KO recipients as a model for acute ABMR established by the group of Fairchild et al. [9]. For measurements of MHC-specific IgE and IgG1 a custom-made ELISA employing MHC monomers, allowing us to distinguish between MHC class I and class II DSA specificities, is used. IgE effector cells during cardiac allograft rejection are analyzed in peripheral blood and in the graft using flow cytometry. Therapeutic interventions targeting IgE, mast cells or eosinophils are performed using α-IgE, cromoglicat or α-IL5, respectively.

Preliminary Data

Our laboratory recently demonstrated the occurrence of IgE specific for MHC antigens in mice and highly sensitized kidney transplant patients. IgE specific for donor MHC class I or class II antigens was detectable in WT C57BL/6 mice as early as 2 weeks after transplantation of a fully mismatched skin or cardiac allograft until at least 12 months post-transplant. Importantly, IgE-DSAs also developed in two murine models of humoral rejection employing either Bm12.Kd.IE cardiac donors as model for chronic ABMR (**Fig. 1**) or CCR5KO recipients as model of acute ABMR (Data not shown). Through a rat basophil leukemia cell degranulation assay and a cutaneous type I hypersensitivity reaction we demonstrated *in vitro* and *in vivo* that MHC-specific IgE that developed in murine recipients undergoing rejection was functional (**Fig. 2**).

Hypothesis & Specific Aims

IgG-DSAs mediate graft injury mainly by targeting endothelial cells causing vascular lesions through complement activation, FcγR-mediated antibody-dependent cellular cytotoxicity or target cell activation through cross-linking. In contrast, IgE-DSAs mainly target mast cells, basophils and eosinophils in the graft, thus the hypothesized effector functions would be distinct from IgG-DSAs. Therefore, we hypothesize, that donor-specific IgE antibodies might play a specific and distinct role in mediating immunologic graft injury.

IgE is associated with Th2-type immune responses, such as type I allergy, and parasite infections [5]. Auto-antibodies of the IgE isotype were observed in Th2-drive autoimmune diseases such as lupus, atopic dermatitis and atherosclerosis [6]. Recently, it was shown that it may also play a protective role in certain cancer types [7].
So far, the occurrence of IgE and its effector cells (e.g. mast cells, basophils, eosinophils) in graft-directed immunity has not been investigated.

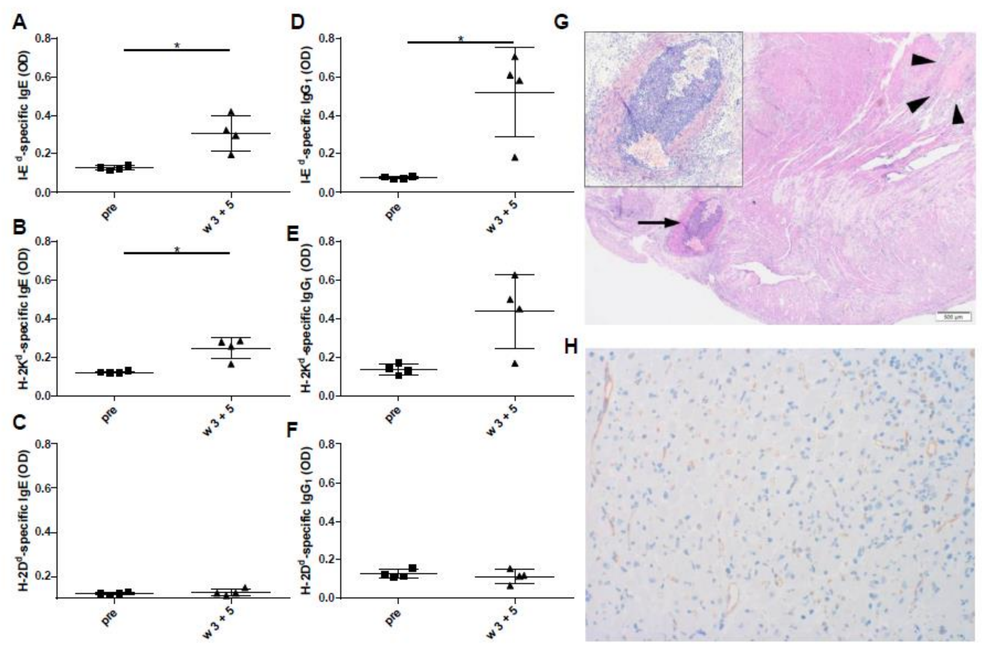

Figure 1: MHC-specific IgE is induced in a murine model of chronic humoral heart allograft rejection.

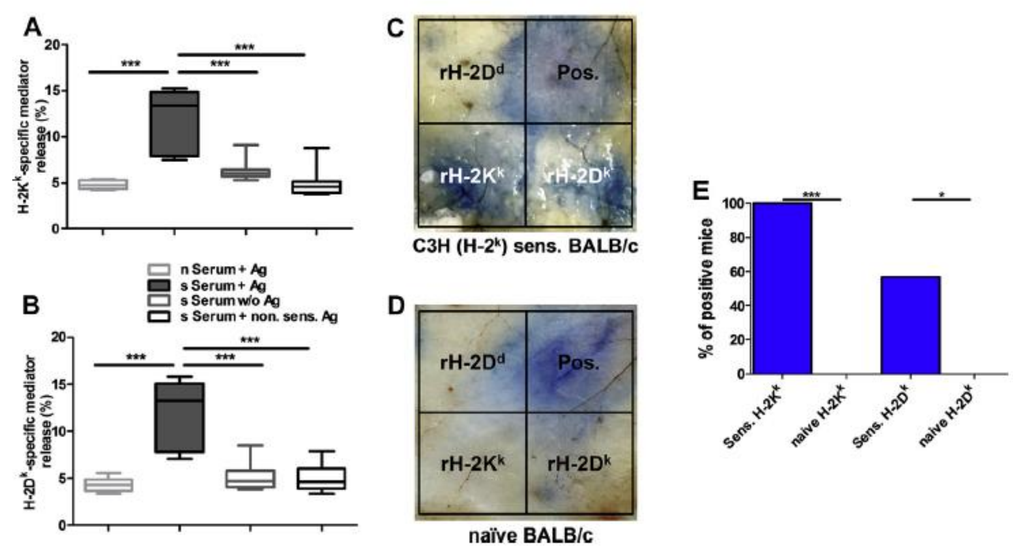

Figure 2: Functional activity of donor-specific IgE developing after allogeneic murine allo-transplantation.

The overall goal of this project is to determine the role of IgE specific for donor MHC antigens in the pathology of ABMR.

Specific Aim 1

To investigate the effect of eliminating IgE on immune-mediated graft injury in a murine heart transplant model of chronic ABMR.

Specific Aim 2

To determine the role of mast cells in an IgE-DSA positive murine heart transplant model of chronic ABMR.

Specific Aim 3

To investigate whether therapeutic interventions targeting IgE, mast cells or eosinophils improve outcome in a murine heart transplant model of chronic ABMR.

References

1. Sellares J, de Freitas DG, Mengel M, Reeve J, Einecke G, Sis B, et al. Understanding the causes of kidney transplant failure: the dominant role of antibody-mediated rejection and nonadherence. Am J Transplant. 2012;12(2):388-99.
2. Valenzuela, N.M. and E.F. Reed, *Antibody-mediated rejection across solid organtransplants: manifestations, mechanisms, and therapies.* J Clin Invest, 2017. **127**(7): p. 2492-2504.
3. Loupy, A. and C. Lefaucheur, *Antibody-Mediated Rejection of Solid-Organ Allografts.* N Engl J Med, 2018. **379**(12): p. 1150-1160.
4. Farkas AM, Baranyi U, Böhmig GA, Unger L, Hopf S, Wahrmann M, et al. Allograft rejection is associated with development of functional IgE specific for donor MHC antigens. J Allergy Clin Immunol. 2019;143(1):335-45 e12.
5. Gould, H.J. and B.J. Sutton, *IgE in allergy and asthma today.* Nat Rev Immunol, 2008. **8**(3): p. 205-17.
6. Valenta, R., et al., *Linking allergy to autoimmune disease.* Trends Immunol, 2009. **30**(3): p. 109-16.
7. Crawford, G., et al., *Epithelial damage and tissue gammadelta T cells promote a unique tumor-protective IgE response.* Nat Immunol, 2018. **19**(8): p. 859-870.
8. Ali, J.M., et al., *Diversity of the CD4 T Cell Alloresponse: The Short and the Long of It.* Cell Rep, 2016. **14**(5): p. 1232-45.
9. Nozaki, T., et al., Antibody-mediated rejection of cardiac allografts in CCR5-deficient recipients. J Immunol, 2007. 179(8): p. 5238-45