

Chapter 15. Self-nonsel self discrimination and autoimmunity

This above all:
to thine own self be true
-Shakespeare,
Hamlet (1601)

The immune system can make antibodies to just about anything that is *foreign* (not part of the body) that happens to come along. At the same time it cannot normally make antibodies to its own proteins, carbohydrates, red cells, or any of the other components of *self*. The immune system is constantly being exposed to a great variety of molecules, and it needs to have a basis for deciding which ones are foreign and which ones are part of self. For example, bovine serum albumin ("BSA"), a protein that is present at high concentration in the blood of cattle, induces the production of antibodies when injected into mice, but not when it is injected into cattle, while mouse serum albumin ("MSA") induces the production of antibodies when injected into cattle, but not in mice. There are no obvious differences between the physical or chemical properties of BSA and MSA that would constitute a basis for these differences. They are proteins of approximately the same size and very similar amino acid sequence. The question of how the immune systems of mice, cows and people learn to differentiate between self and nonself components is a central problem of cellular immunology.

<u>Antigen</u>	<u>Injected in species</u>	<u>Response</u> ²²⁵
mouse serum albumin	mouse	no
mouse serum albumin	cow	yes
bovine serum albumin	mouse	yes
bovine serum albumin	cow	no

The clonal selection theory provided some tentative explanations for self-nonsel self discrimination. One was that before lymphocytes are fully mature, they pass through a stage in which encounters with the antigen kill them rather than stimulates them. As the immune system develops, it would then be purged of all self-reactive clones. The system would then be unresponsive (tolerant) to self antigens. This idea has been called the clonal deletion or clonal abortion theory of self tolerance.

²²⁵ An immune response occurs to a foreign protein but not a self protein. This is an example of criss-cross specificity, as described in chapter 5. In this case matching between columns one and two precludes a response.

A number of experimental results suggest that the clonal deletion theory of self-tolerance is wrong. For example, if we inject an animal with a foreign antigen that is similar to but not identical with a self antigen, it can cause antibodies to be produced which react with both the foreign antigen and the self antigen. That is, we can break self-tolerance using a “cross-reacting antigen”. For example, as described in chapter 6, when rat red blood cells are used as an antigen in mice they induce an immune response that includes immunity to the mouse's own red blood cells, causing hemolytic anemia (reference 54). This shows that cells reactive to self antigens are present, but are somehow normally prevented from secreting antibodies, in spite of the presence of the self antigen. By default, such findings lead to an explanation for self tolerance that involves an active role for suppressor T cells.

Self tolerance must be learned

If a mouse of strain A is immunized with lymphocytes from a mouse B, it makes anti-B antibodies, and similarly, if a mouse of strain B is immunized with cells of strain A, it makes anti-A antibodies. An F1 hybrid mouse of the AxB type has both the antibody genes of the strain A mouse, and the antigens of the strain B mouse. In spite of the fact that it has both the antigens and the appropriate genes for making antibodies to them, it normally makes neither anti-A antibodies in response to immunization with A strain cells, nor anti-B antibodies in response to immunization with B strain cells. Self-tolerance must therefore be learned, and is not simply due to a lack of the appropriate antibody genes.

Self tolerance in the context of the symmetrical network theory

A good reason for including network interactions in a theory of self-nonsel self discrimination is that the picture that emerges is simpler than the picture we obtain when we do not include those interactions. Each lymphocyte has a specific receptor that distinguishes it from most other lymphocytes. A given lymphocyte has a V region that may recognize self molecules or non-self molecules or both. So we begin with the fundamental fact that the V regions of lymphocytes recognize anything, including the V regions of other lymphocytes. We then assume that self antigens, like foreign antigens, are stimulatory (already postulated for self MHC molecules) and each self antigens can induce the specifically suppressed state for itself during development.

How many self antigens are there, and is it possible for the system to really be in the suppressed state for all of them? We can address this question by ordering self antigens according to their level of immunogenicity, that is, the extent to which they can facilitate the stimulation of T cells, since the

suppressed state is T cell mediated. We envisage building a repertoire that is self antigen based, one self antigen at a time, such that it is suppressed with regard to all the self antigens.

We begin with MHC class II. Tissue distribution is important for determining which self antigens are the most immunogenic. The presence of MHC class II on accessory cells, that provide a second signal for proliferation for T cells, has the result that MHC class II selects helper T cells that in turn select T cells that have an image of MHC class II, as discussed in the chapter on I-J. Tolerance to MHC class II is envisaged as involving many helper T cells, and in principle could involve just one or a few MHC class II image (I-J) clones. This is a nonlinear co-selection process; the particular helper cells that are selected depend on which I-J clones are selected and vice versa. In chapter 13 I described an *in vitro* experiment in which it was shown that self tolerance to MHC class II can be broken by stimulation with I-J disparate cells (reference 198). I gave an interpretation of this finding in the context of the model in which helper T cells are selected to recognize MHC class II and there is idiotypic co-selection of the helpers and a population of suppressor T cells.

The next antigens to consider are MHC class I, that also have a profound impact on the repertoire, especially on the CD8 expressing T cells, that include suppressors and cytotoxic T cells. MHC class I and MHC class II are important boundary conditions for the overall T cell repertoire, and the diverse population of T cells that are selected to recognize MHC class I may be selected also by positive selection to recognize the diverse idiotypes of MHC class II specific helper T cells. Then we would have the additional picture of Figure 15-1:

Figure 15-1. Another co-selection model. CD4 bearing T lymphocytes are selected to recognise MHC class II and CD8 lymphocyte idiotypes, while CD8 bearing T lymphocytes are selected to recognise MHC class I and CD4 lymphocyte idiotypes. This is not inconsistent with the focusing model of **Figure 13-3**, since I-J is expressed primarily on suppressor T cells, that are primarily CD8 cells.

	anti-MHC class II	anti-MHC class I	
MHC class II -----	CD4 lymphocyte -----	CD8 lymphocyte -----	MHC class I
	idiotypes	idiotypes	

The validity of Figure 15-1 can be tested by determining whether T cell receptors and antibodies that are images of MHC class II of a given strain are antiidiotypic to T cell receptors and antibodies that are images of MHC class I of that strain. Referring to second symmetry, this can potentially be determined using some monoclonal antibodies derived from an A anti-B immune animal that are anti-MHC class I^B, and others that are anti-MHC class II^B. In the converse, B anti-A immune animal we would expect to see MHC class I image

(anti-anti-MHC class I^B) and MHC class II image (anti-anti-MHC class II^B) antibodies. These may be separable using the respective monoclonal antibodies from the A anti-B immunization. If so, the prediction is that the anti-anti-MHC class I^B and anti-anti-MHC class II^B antibodies can then be shown to be antiidiotypic to each other. Alternatively, a population of anti-anti-self MHC idiotypes may be produced, that is both anti-anti-MHC class I and anti-anti-MHC class II²²⁶.

Impinging on the repertoire of idiotypes, in addition to the self antigens MHC class I and MHC class II, is the repertoire of other self antigens. We consider the gedankenexperiment (thought experiment) of adding the influence of the self antigens one at a time. There is a diversity of these self shapes, and for each one of them we can ask, does it have more complementarity to the idiotypes of CD4 bearing T lymphocytes or more complementarity to the idiotypes of CD8 bearing T lymphocytes? Some self antigens will stimulate one population more, some the other. If the next most immunogenic self antigen has more complementarity to CD4 T cell idiotypes than CD8 T cell idiotypes, it will induce an increase in CD4 T cell clones that are specific for itself, which will in turn cause an increase in corresponding CD8 clones, and the basic structure of Figure 15-1 may remain the same. Clones with idiotypes that recognise both MHC (class I or class II or both) and this self antigen will be selected in preference to clones that recognise only one or the other. Each self antigen in the series (with each one being weaker than the preceding one) will cause a smaller perturbation of the system, and again clones that recognise multiple self antigens will be favoured. As we descend the list of stimulatory self antigens, additional clones with cross-reaction with self MHC are added, and eventually a suppressed state for all of the self antigens may be reached. So ultimately the repertoire of CD4 and CD8 idiotypes that are selected is determined not only by MHC class II and MHC class I, but by the complete repertoire of self antigens. Idiotypes for a given self antigen will also be antiidiotypes for other self antigens. Mathematically modelling this repertoire generation process may yield additional insights, but at this stage it seems at least highly plausible that the resulting structure is self stabilizing, just as the suppressed state is in our two-variable model (chapter 10), because the resulting picture is similar to that picture. The resulting centre-pole of the repertoire is then determined to some extent by all self antigens, and is instrumental in maintaining self tolerance to all self antigens.

Self antigens that are present even at very low levels must play a role in the T cell repertoire selection process. We know this because of the phenomenon of low dose tolerance, and especially ultra-low dose tolerance, in which minute amounts of foreign antigen have a detectable impact on the T cell repertoire, as manifest in the ability of an animal to respond to the antigen (references 20, 21,

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44,126). It is then only reasonable that self antigens that are continuously present should also have an impact.

Elimination of clones with high affinity for MHC

The positive selection paradigm does not preclude the possibility that cells whose receptors have high affinity for self MHC would be not only suppressed (inhibited by antiidiotypic clones) but actually eliminated. If a cell's specific receptors have a high affinity for MHC on a macrophage's surface, such that (in the case of a T cell) it cannot free itself by secreting specific T cell factors, it would likely be phagocytosed by the macrophage and thus be eliminated. This could be the mechanism of the "veto cell" discovered by Miller and his colleagues.²²⁷ The positive selection process would then apply for cells with lower affinity for self MHC to give the MHC restricted repertoire.

Autoimmune diseases and MHC class II

A failure of self-nonsel self discrimination constitutes immunity against self, or autoimmunity. Many diseases involve the immune system producing antibodies or T cell immunity specific for self tissue. Autoimmune diseases include systemic lupus erythematosus, diabetes, rheumatoid arthritis, myasthenia gravis, hemolytic anemia and many more. Self-nonsel self discrimination is not perfect also in a healthy animal; some anti-self antibodies can often be detected transiently during immune responses to foreign antigens.

An early hypothesis to explain autoimmunity was that self-reactive V regions are somehow eliminated during development to give self-tolerance, and that later in life mutations produce new V regions, some of which are self-reactive. The frequency of autoimmune diseases increases with age, so this hypothesis predicts that the frequency of mutations increases with age. Mutations in the V regions of old mice following antigenic challenge are however rare compared with the rate in young mice²²⁸, which speaks against the mutation theory.

Many diseases are MHC linked. MHC linkage means that people with a particular MHC allele (gene) are more prone (or in the case of negative linkage, less prone) to contract a particular disease than others that lack that allele. Over 40 autoimmune diseases fall into this category, including rheumatoid arthritis,

²²⁷ R. G. Miller and H. Derry (1979) A cell population in nu/nu spleen can prevent generation of cytotoxic lymphocytes by normal spleen cells against self antigens of the nu/nu spleen. *J. Immunol.* 122, 1502-1509.

²²⁸ C. Miller and G. Kelsoe (1995) IgV_H hypermutation is absent in the germinal centres of aged mice. *J. Immunol.* 155, 3377-3384.

insulin-dependent diabetes mellitus (IDDM) and multiple sclerosis. For autoimmune diseases the linkage is primarily to MHC class II. In the case of IDDM some alleles are positively associated with the disease, with relative risks of up to 20-fold higher than other alleles, while another allele is negatively associated with the disease (relative risk of 0.25)^{229,230}. MHC class II therefore appears to be a boundary condition of the network that is important in determining how the system eventually becomes unstable.

There are many different diseases in which a dysfunctional immune system has been implicated, and these diseases occur at varying ages. Each disease may correspond to a particular mode of failure of the idiotypic network. This concept is supported by findings of skewing of V region repertoires in many diseases, including cancer,²³¹ autoimmune diseases^{232,232,233,234} and graft versus host disease²³⁵.

Why do most people live from 60 to 90 years?

²²⁹ J. A. Todd, J. I. Bell, H. O. McDevitt (1987) HLA-DQ β gene contributes to susceptibility and resistance to insulin-dependent diabetes mellitus. *Nature* 329, 599-604.

²³⁰ A. Svejgaard, P. Platz and L. P. Ryder (1983) HLA and disease 1982 – a survey. *Immunol. Rev.* 70, 193-218.

²³¹ H. Pilch, H. Hohn, C. Neukirch, K. Freitag, P. G. Knapstein, B. Tanner and M. J. Maeurer (2002) Antigen-driven T-cell selection in patients with cervical cancer as evidenced by T-cell receptor analysis and recognition of autologous tumor. *Clin. Diagn. Lab. Immunol.* 9, 267-278.

²³² K. W. Wucherpfennig, J. Newcombe, H. Li, C. Keddy, M. L. Cuzner and D. A. Hafler (1992) T cell receptor V α -V β repertoire and cytokine gene expression in active multiple sclerosis lesions. *J. Exp. Med.*, 175, 993-1002.

²³³ L. Imberti, A. Sottini, A. Bettinardi, M. Puoti, D. Primi, (1991) Selective depletion in HIV infection of T cells that bear specific T cell receptor V β sequences. *Science*, 254, 860-862.

²³⁴ N. Rebai, G. Pantaleo, J. F. Demarest, C. Ciurli, H. Soudeyans, J. W. Adelsberger, M. Vaccarezza, R. E. Walker, R. F. Sekaly and A. S. Fauci (1994) Analysis of the T-cell receptor β -chain variable-region (V β) repertoire in monozygotic twins discordant for human immunodeficiency virus: evidence for perturbations of specific V β segments in CD4 $^{+}$ T cells of the virus-positive twins. *Proc. Natl. Acad. Sci. (USA)* 91, 1529-1533.

²³⁵ F. S. Smith, S. D. Rencher, H. E. Heslop and J. L. Hurwitz (1995) T cell receptor repertoire of CD4 $^{+}$ and CD8 $^{+}$ T cell subsets in the allogeneic bone marrow transplant recipient. *Cancer Immunol. Immunotherapy.* 41, 104-110.

There has been a lot of speculation that the immune system is implicated in the aging process. We know that people (and animals) contract a variety of diseases, that together conspire to kill within a fairly narrow time span. In the case of people this is mostly between the ages of 60 and 90 years. This narrow range allows us to legislate a retirement age of 65. If death were governed by a single event that has a fixed probability per unit time of occurring, regardless of age, we would have an exponential death rate. There would then be a wider distribution of ages, with (for the same life expectancy) many more people dying before 60 and many more living much longer than 90. Theories of aging therefore have to account for the organism wearing out relatively abruptly. A finite life span for the immune system is a plausible possibility, since there are many autoimmune diseases, and other important diseases including cancer have an immune system related aspect, such as skewing of the repertoire of V regions. The typically narrow range of human ages at death may be due to an inherent instability in the idiotypic network, leading to the skewing of V region repertoires followed by the collapse of the system. A proteomic method of analysis of immune system V regions has been formulated, that I anticipate will enable us to accurately characterize the V region repertoire skewing in various diseases, and furthermore to counteract such skewing.²³⁶

Serum IgG and self tolerance

In chapter 17 I describe a model that includes three suppressor T cell populations, two helper T cell populations and serum IgG. I make the case that the V regions of serum IgG antibodies are images of self antigens, and the IgG plays a role in self tolerance through interactions with T cell idiotypes and by eliminating anti-self clones.

²³⁶ G. W. Hoffmann (2003) Proteomic analyser with applications to diagnostics and vaccines. *J. theoret. Biol.* 228, 459-465.