Sequence Space Localization in the Immune System Response to Vaccination and Disease

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We introduce a model of protein evolution to explain limitations in the immune system response to vaccination and disease. The phenomenon of original antigenic sin, wherein vaccination creates memory sequences that can increase susceptibility to future exposures to the same disease, is explained as stemming from localization of the immune system response in antibody sequence space. This localization is a result of the roughness in sequence space of the evolved antibody affinity constant for antigen and is observed for diseases with high year-to-year mutation rates, such as influenza.

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INTRODUCTION

Our immune system protects us against death by infection. A major component of the immune system is generation of antibodies, protein molecules that bind specific antigens. To recognize invading pathogens, the immune system performs a search of the amino acid sequence space of possible antibodies. To find useful antibodies in the effectively infinite protein sequence space, the immune system has evolved a hierarchical strategy. The first step involves creating the DNA sequences for B cells that code for moderately effective antibodies through rearrangement of immune-system-specific gene fragments from the genome. This process is called VDJ recombination. This combinatorial process can produce on the order of $10^{14}$ different antibodies through recombination of pieces of antibodies. The second step, which occurs when a specific antigen invades our body, is somatic hypermutation. Somatic hypermutation is the process of mutation that occurs when the B cells that produce the antibodies divide and multiply. Only those B cells that produce antibodies that bind the antigen with higher affinity are propagated by this mutation and selection process, and another name for this process is affinity maturation. Somatic hypermutation is essentially a search of the amino acid sequence space at the level of individual point mutations.

The consequence of an immune system response to antigen is the establishment of a state of memory. Immunological memory is the ability of the immune system to respond more rapidly and effectively to antigens that have been encountered previously. Specific memory is maintained in the DNA of long-lived memory B cells that can persist without residual antigen.

Although our immune system is highly effective, some limitations have been reported. The phenomenon known as “original antigenic sin” is the tendency for antibodies produced in response to exposure to influenza virus antigens to suppress the creation of new, different antibodies in response to exposure to different versions of the flu. Roughly speaking, the immune system responds only to the antigen fragments, or epitopes, that are in common with the original flu virus. As a result, individuals vaccinated against the flu may become more susceptible to infection by mutated strains of the flu than would individuals receiving no vaccination. The details of how original antigenic sin works, even at a qualitative level, are unknown.

In this Letter, we offer an explanation for the reported limitations in the immune system response using a model of protein evolution. We describe the dynamics of affinity maturation by a search for increased binding constants between antibody and antigen in antibody sequence space. It is shown that an immune system response to an antigen generates localized memory B cell sequences. This set of localized sequences reduces the ability of the immune system to respond to subsequent exposures to different but related antigens. It is this competitive process between memory sequences and the VDJ recombinations of secondary exposure that is responsible for the reported limitations in the immune system.

We use a random energy model to represent the interaction between the antibodies and the influenza proteins. This model captures the essence of the correlated ruggedness of the interaction energy in the variable space, the variables being the antibody amino acid sequences and the identity of the disease proteins, and the correlations being mainly due to the physical structure of the antibodies. The random energy model allows study of the sequence-level dynamics of the immune/antigen system, which would otherwise be an intractable problem at the atomic scale, with $10^4$ atoms per antibody, $10^8$ antibodies per individual, $6 \times 10^9$ individuals, and many possible influenza strains. Use of random energy theory to treat correlations in otherwise intractable physical systems goes back at least to Bohr’s random matrix theory for nuclear cross sections and has been used for quantum chaos, disordered mesoscopic systems, QCD, and quantum gravity. Close to the present application is the study of spin glasses by random energy models, protein folding by coarse-grained models, and
evolutionary systems by NK-type models \cite{12, 13, 14, 15}.

In detail, the generalized NK model we use considers three different kinds of interactions within an antibody: interaction within a subdomain ($U_{\alpha}$), interactions between subdomains ($U_{\text{sd}}$), and direct binding interaction between antibody and antigen ($U_{c}$). In the context of protein evolution, parameters of the model have been calibrated \cite{12, 16, 17}. The energy function of a protein is given by

$$U = \sum_{i=1}^{M} U_{\alpha} + \sum_{i>j=1}^{M} U_{ij} + \sum_{i=1}^{P} U_{c},$$

where $M$ is the number of antibody secondary structural subdomains, and $P$ is the number of antibody amino acids contributing directly to the binding. The subdomain energy $U_{\alpha}$ is

$$U_{\alpha} = \frac{1}{\sqrt{M}} \sum_{j=1}^{N-K+1} \sigma_{\alpha}(a_{j}, a_{j+1}, \cdots, a_{j+K-1}),$$

where $N$ is the number of amino acids in a subdomain, and $K$ is the range of local interaction within a subdomain. All subdomains belong to one of $L = 5$ different types (e.g., helices, strands, loops, turns, and others). The quenched Gaussian random number $\sigma_{\alpha}$ is different for each value of its argument for a given subdomain type, $\alpha$. All of the Gaussian $\sigma$ values have zero mean and unit variance. The energy of interaction between secondary structures is

$$U_{\text{sd}} = \sqrt{\frac{2}{DM(M-1)}} \sum_{k=1}^{D} \sigma_{ij}^{k}(a_{j1}^{i}, \cdots, a_{jk/2}^{i}, a_{jK/2+1}^{i}, \cdots, a_{jk}^{i}).$$

We set the number of interactions between secondary structures at $D = 6$. Here $\sigma_{ij}^{k}$ and the interacting amino acids, $j_{1}, \cdots, j_{K}$, are selected at random for each interaction $(k, i, j)$. The chemical binding energy of each antibody amino acid to the antigen is given by $U_{c} = \sigma_{i}(a_{i})/\sqrt{P}$. The contributing amino acid, $i$, and the unit-normal weight of the binding, $\sigma_{i}$, are chosen at random. Using experimental results, we take $P = 5$ amino acids to contribute directly to the binding event. Here we consider only five chemically distinct amino acid classes (e.g., negative, positive, polar, hydrophobic, and other) since each different type of amino acid behaves as a completely different chemical entity within the random energy model.

The generalized NK model, while a simplified description of real proteins, captures much of the thermodynamics of protein folding and ligand binding. In the model, a specific B cell repertoire is represented by a specific set of amino acid sequences. Moreover, a specific instance of the random parameters within the model represents a specific antigen. An immune response that finds a B cell that produces an antibody with high affinity constant to a specific antigen corresponds in the model to finding a sequence having a low energy for a specific parameter set.

The random character of the generalized NK model makes the energy rugged in antibody sequence space. The energy is, moreover, correlated by the local antibody structure ($K = 4$), the secondary antibody structure ($U_{\text{sd}}$), and the interaction with the influenza proteins ($U_{c}$). As the immune system explores the space of possible antibodies, localization is possible if the correlated ruggedness of the interaction energy is sufficiently great.

Since the variable region in each light and heavy chain of an antibody is about 100 amino acids long, and most of the binding occurs in the heavy chain, we choose a sequence length of 100. We choose $M = 10$ since there are roughly 10 secondary structures in a typical antibody and thus choose $N = 10$. The immune system contains of the order of $10^{8}$ B cells divided into different specificities, and the frequency of a specific B cell participating in the initial immune response is roughly 1 in $10^{6}$ \cite{12}. Hence, we use $10^{3}$ sequences during an immune response.

The hierarchical strategy of the immune system is used to search the antibody sequence space for high affinity antibodies. Initial combination of optimized subdomains is followed by a point mutation and selection procedure \cite{12}. To mimic combinatorial joining of gene segments during B cell development, we produce a naive B cell repertoire by choosing each subdomain sequence from pools that have $N_{\text{pool}}$ amino acid segments obtained by minimizing the appropriate $U_{\alpha}$. To fit the theoretical heavy-chain diversity of $3 \times 10^{11}$ \cite{12}, we choose $N_{\text{pool}} = 3$ sequences among the top 300 sequences for each subdomain type.

Somatic hypermutation occurs at the rate of roughly one mutation per variable regions of light and heavy chains per cell division, which occurs every 6 to 8 hours during intense cell proliferation \cite{14}. Hence, in our simulation, we do 0.5 point mutations per sequence, keep the best (highest affinity) $x = 20\%$ sequences, and then amplify these back up to a total of $10^{3}$ copies in one round, which corresponds to 1/3 day. That is, the probability of picking one of the, possibly mutated, sequences for the next round is $p_{\text{select}} = 1/200$ for $U < U_{200}$ and $p_{\text{select}} = 0$ for $U > U_{200}$, where $U_{200}$ is the 200th best energy of the $10^{3}$ sequences after the mutation events, and this equation is employed $10^{3}$ times to select randomly the $10^{3}$ sequences for the next round. Given a specific antigen, i.e. a specific set of interaction parameters, we do 30 rounds (10 days) of point mutation and selection in one immune response. In this way, memory B cells for the antigen are generated.

The affinity constant is given as a function of energy,

$$K_{\text{eq}} = \exp(a - bU),$$

(4)
where $a$ and $b$ are determined by the dynamics of the mutation and selection process. Affinity constants resulting from VDJ recombination are roughly $10^4$, affinity constants after the first response of affinity maturation are roughly $10^6$, and affinity constants after a second response of affinity maturation are roughly $10^7$ [18] (values that fix the selection strength, $x$). By comparison to the dynamics of the model, we obtain $a = -18.56$, $b = 1.67$.

The memory B cells, key to immunological memory, give rapid and effective response to the same antigen due to their increased affinity for previously-encountered antigens. We focus on the role of the memory cells in the immune response to different antigens. The distance between a first antigen and the second antigen is given by the probability, $p$, of changing parameters of interaction within subdomain ($U^{sd}$), subdomain-subdomain ($U^{sd-sd}$), and chemical binding ($U^c$) terms. Within $U^{sd}$, we change only the subdomain type, $\alpha_i$, not the parameters $\sigma_\alpha$, which are probably fixed by structural biology and should be independent of the antigen.

We estimate the number of memory and naive B cells that participate in the immune response to the second antigen by the ratio of the respective affinity constants. From the definition of the affinity constant, $K^{eq} = \frac{\text{[Antigen : Antibody]}}{\{\text{[Antigen] [Antibody]}\}}$, the binding probability is proportional to the affinity constant and to the concentration of antigen-specific antibody, which is $10^2$ times greater for the memory sequences [13]. We measure the average affinity for the second antigen of the $10^4$ memory cells, $K^{eq}_m$, and that of the $10^5$ B cells from the naive repertoire of optimized subdomain sequences, $K^{eq}_n$. The ratio $10^2K^{eq}_m/K^{eq}_n$ gives the ratio of memory cells to naive cells. For exposure to the second antigen, we perform 30 rounds (10 days) of point mutation and selection, starting with $10^3K^{eq}_m/(10^2K^{eq}_m + K^{eq}_n)$ memory cells and $10^3K^{eq}_n/(10^2K^{eq}_m + K^{eq}_n)$ naive cells, since both memory and naive sequences participate in the secondary response [20].

Figure 1 shows the evolved affinity constant to a second antigen if the exposure to a first antigen exists (solid line) or not (dashed line) as a function of the difference between the first and second antigen, $p$, or "antigenic distance" [21]. When the difference is small, the exposure to a first antigen leads to higher affinity constant than without exposure, which is why immune system memory and vaccination is effective. For a large difference, the antigen encountered in the first exposure is uncorrelated with that in the second exposure, and so immune system memory does not play a role. Interestingly, the immunological memory from the first exposure actually gives worse protection, i.e. a lower affinity constant, for intermediate differences—which is original antigenic sin.

The dynamics of the immune response (Fig. 1) depend upon the constants of Nature, i.e. the parameters of the model. For example, in an organism with a smaller immune system, such as the mouse, there are fewer starting sequences and less favorable binding constants are measured in the same number of rounds: a factor of 0.5 reduction in the number of starting sequences leads to a 0.64 reduction in the evolved binding constant, but a similar degree of original antigenic sin as in Fig. 1. On the other hand, if more rounds are performed, better binding constants are found in the primary and secondary responses, but the secondary response is not as improved over the primary response as when using 30 rounds, because the evolved sequences are becoming more localized in ever deeper wells. The degree of original antigenic sin is, however, similar in the range of 30 to 60 rounds per response. Similarly, if the roughness of the energy upon disease mutation is increased, for example by assuming that mutation of the influenza actually changes the $\sigma_\alpha$, the degree of original antigenic sin increases substantially, by a factor of 2, because the barriers between the regions of localization in sequence space are increased. On the other hand, if the concentration of the memory cells is decreased, the contribution of the memory cells to the dynamics is reduced, and the original antigenic sin phenomenon decreases, almost disappearing when the memory and naive antibody concentrations are equal. The average number of mutations leading to the best antibody, a measure of the localization length when original antigenic sin occurs, is 15 for the first response and rises from 5 to 15 for the second response in the range $0 \leq p < 0.30$. Interestingly, the average number of mutations rises slightly above 15 in the range $0.30 \leq p < 0.70$, indicating that in the original antigenic sin region more
mutations are necessary for the compressed ensemble of memory sequences from the primary response to evolve to a suitable new state in the secondary response. Larger selection strengths \( (x < 20\%) \) cause more localization and original antigenic sin, in shallower wells for small \( x \), and smaller strengths \( (x > 20\%) \) lead to less evolution.

For small values of \( p, \ p < 0.19 \), the memory B cells produce antibodies with higher affinities, \( K_{eq}^m > 10^4 \), for the new antigen than do naive B cells. The binding constant of the memory antibodies steadily decreases with \( p \), reaching the non-specific value of \( K_{eq}^m = 10^2 \) at \( p = 0.36 \) (see inset in Fig. 1), which, interestingly, is less than the range to which original antigenic sin extends, \( 0.23 < p < 0.60 \). These model predictions are in good agreement with experimental data on cross-reactivity, which ceases to occur when the amino acid sequences are more than 33–42% different.

The ineffectiveness of immune system memory over a window of \( p \) values can be understood from the localization of memory B cell sequences. Figure 2 displays distributions of memory and naive affinity constants for the second antigen. Notice that the memory sequences are highly homogeneous and lack diversity compared to the naive sequences. Indeed, original antigenic sin arises mainly because the memory sequences from the primary response suppress use of naive sequences in the tail of the distribution for the secondary response. Although those naive sequences initially look unpromising, they may actually evolve to sequences with superior binding constants. The inset to Fig. 2 illustrates this phenomenon. Interestingly, when half of the distribution is removed, the reduction in the binding constant is just about that which occurs in original antigenic sin, Fig. 1. In summary, the generalized NK model is shown to successfully model immune system dynamics. A localization mechanism for the original antigenic sin phenomenon observed in the flu is explained. Localization of antibodies in the amino acid sequence space around memory B cell sequences is shown to lead to a decreased ability of the immune system to respond to diseases with year-to-year mutation rates within a critical window. This localization occurs because of the ruggedness of the evolved affinity constant in amino acid sequence space. From the model dynamics, we find that memory sequences can both outcompete the non-vaccinated immune response and become trapped in local minima. Memory sequences with affinity constants initially superior to those from naive sequences can be selected by the dynamics, and these memory sequences can lead to poorer evolved affinity constants, to the detriment of the immune system for intermediate disease mutation rates. These results suggest several implications for vaccination strategy: the difference between vaccinations administered on a repeated basis should be as great as practicable, and suppression of the memory B cell response may be helpful during vaccination against highly variable antigens.

![FIG. 2: The affinity distribution of antibodies from naive B cells (hatched) or memory cells (open) for the antigen of second exposure. In the inset is shown the binding constant that evolves in the primary response when an initial selection step retains only the top fraction of the naive cells.](image)

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