Idiotypic Networks Incorporating T-B cell Cooperation. The Conditions for Percolation.

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Previous work was concerned with symmetric immune networks of idiotypic interactions amongst B cell clones. The behaviour of these networks was contrary to expectations. This was caused by an extensive percolation of idiotypic signals. Idiotypic activation was thus expected to affect almost all ($> 10^7$) B cell clones. We here analyse whether the incorporation of helper T cells (Th) into these B cell models could cause a reduction in the percolation. Empirical work on idiotypic interactions between Th and B cells however, would suggest that two different idiotypic Th models should be developed: 1) a Th which recognises native B cell idiotypes, i.e. a non-MHC-restricted "ThId" model, and 2) a "classical" MHC-restricted helper T cell model.

In the ThId model, the Th-B cell interaction is symmetric. A 2-D model of a Th and a B cell clone that interact idiotypically with each other accounts for various equilibria (i.e. one virgin and two immune states). Introduction of antigen does indeed lead to a state switch from the virgin to the immune state; such a system is thus able to "remember" its exposure to antigen. Idiotypic signals do however, percolate in ThId models via these "B-Th-B-Th" pathways: proliferating Th and B cell clones that interact idiotypically, will always activate each other reciprocally.

In the MHC-restricted Th model, Th-B interactions are asymmetric. Because the B cell idiotypes are processed and subsequently presented by MHC molecules, the Th receptor and the native B cell receptor are not expected to be complementary. Thus the Th and the B cells are unable to activate each other reciprocally, and a 2-D Th-B cell model cannot account for idiotypic memory. In contrast to the ThId model, idiotypic activation cannot percolate via "B-Th-B-Th" interactions. Due to the assymmetry idiotypic actiavtion stops at the first Th level. A Th clone cannot activate a subsequent B cell clone: if the B cells recognise the Th cells, they see idiotype but get no help; if the Th cells see the B cells, the B cells are helped but see no idiotype.

The percolation along "B-B-B" pathways in these two models is next analysed. Two B cells clones, each helped by one Th clone, are connected by a symmetric idiotypic interaction. It turns out that in both models the second (i.e. anti-idiotypic) B cells (B_2) never proliferate. The anti-idiotypic B cells are activated whenever the first (idiotypic) B cells (B_1) proliferate, but because the activated B_2 cells are not being helped, they fail to proliferate. Thus, because the third level Th cells (T_3) are not being activated by the idiotypic cascade, signals never percolate along "B-B-B" pathways in both models.

Percolation is subsequently analysed in 200-D networks, comprised of random mixtures of "B-B" and "B-Th" pathways. As soon as the connectivity of the symmetric Th-B interactions exceeds a threshold level of two connections per clone, the idiotypic cascade percolates extensively in the ThId model. Conversely, due to the asymmetry, signals do not percolate at all in the MHC-restricted networks.

Helper T cells thus set the conditions for idiotypic signal percolation. Firstly,

considering "B-B" cell idiotypic interactions, it turns out to be essential to consider the activation of helper T cells (i.e. T_3) for the anti-idiotypic B cells (B_2). Secondly, due to the asymmetry, MHC-restricted Th and ThId cells differ with respect to the constitution of functional networks based on a "B-Th-B-Th" topology. MHC-restricted Th cells may altogether prevent the development of functional idiotypic interactions. Thus, idiotypic networks are not necessarily "unavoidable".

1. Introduction

Idiotypic network theory [Jerne, 1974] is presented as an "unavoidable" [Jerne, 1984] implication of the extremely large ($>10^7$) number of different lymphocyte clones which collectively, can virtually recognise anything. The theory states that, if the repertoire of receptors can recognise any antigen, receptors should also be able to recognise other receptors. The antigenic determinants presented by receptors are called "idiotypes" [Jerne, 1974]; the interaction between receptors therefore "idiotypic" interactions. Via idiotypic interactions, clones should thus be able to interact in a stimulating and/or inhibiting manner. Apart from being "unavoidable", idiotypic interactions are suppossed to be desirable: they are supposed to play an important role in the behaviour of immune systems. Idiotypic networks have often been compared to neural networks, with respect to both structure (i.e. an almost infinite number of connected cells or clones) and to function (i.e. memory and/or cognitive properties) [Jerne, 1974; Kelsoe *et al.*, 1981; Hoffmann, 1986; Farmer *et al.*, 1986, Varela *et al.*, 1988].

In order to analyse whether high-D immune networks do indeed have such network properties, we previously analysed an idiotypic network model that aimed to be "fundamental", i.e. that was based on simple and reasonable assumptions. Focussing on the important immunological properties of antigen specific "regulation" (i.e. proliferation control) and "immunity" (i.e. memory), we concluded that our idiotypic networks fail to account for this. Proliferation control is impossible in symmetric immune networks, because proliferating clones suppress their suppressors [De Boer & Hogeweg, 1989_b]. Immunity, i.e. antigen specific memory, is impossible in high-D immune networks (i.e. networks consisting of many clones) because of (semi) chaotic behaviour and extensive percolation of idiotypic signals [De Boer 1989_a, 1989_b; De Boer & Hogeweg, 1989_c]. We have tested the robustness of these results by the incorporation of "longe-range inhibition" and of "circulating antibodies"; the result was that the network behaviour only got worse.

In previous work we defined the "extensive percolation" problem of high-D networks [De Boer & Hogeweg, 1989_c]; see also [De Boer, 1988; 1989_a; 1989_b; De Boer & Hogeweg, 1989_a]. Extensive percolation essentially means that all clones are eventually expected to become activated and/or affected by the idiotypic signal that is initiated by the first antigen that enters the system. Subsequent antigens are therefore unable to perturb the system. We think that, if the immune system does indeed function by means of an idiotypic network, the network responses should remain localised, i.e. most of clones should remain in the virgin state in order to be able to respond to subsequent different antigens. Empirical data also suggest that idiotypic network responses remain more or less localised [Wikler *et al.* 1979; Bona & Pernis, 1984; Bottomly, 1984]. The extensive percolation is however a very robust property of our models. It hinges upon: 1) the (statistical) connectivity properties of idiotypic networks (i.e. a graph theoretical result), and 2) the (reasonable) assumption that sustaining the proliferation (i.e. the immune state) of a large clone consisting of very many cells, requires a higher idiotypic stimulus than the activation of the few cells of a subsequent small (virgin) clone.

In order to achieve localised network responses, it thus seems necessary to assume that the initiation of an idiotypic interaction with a subsequent (virgin) clone is somehow more difficult that the maintenance of an established idiotypic interaction. Helper T cells (Th) that produce the necessary growth and differentiation factors for B cells can possibly account for this. Initiation of new idiotypic interactions would require both idiotypic activation and the recruitment of sufficient specific helper

cells (due to threshold effects the latter might be difficult [De Boer & Hogeweg, 1986, 1987; Kevrekidis *et al.* 1988]). Ongoing idiotypic interactions have already accumulated these helper cells; their maintenance therefore merely requires idiotypic activation (which itself might even be easier in the presence of helper T cells). Note, however, that this argument is a subtle one: idiotypic network theory requires that the B cell clones that are activated by antigen (i.e. level 1) are able to activate the anti-idiotypic B cell clones (i.e. level 2) they can interact with. These anti-idiotypic clones are expected to be in the virgin state and have not yet recruited helper T cells. The percolation should obviously fade at some higher level and not at level 2.

Nevertheless, we will incorporate helper T cells in our previous idiotypic B cell model. It will become clear that, because of MHC-restriction, we have to discern between two different types of helper T cells. These two models differ essentially with respect to our percolation problem: one cannot solve it because percolation proceeds via the Th cells, the other makes it obsolete because all idiotypic interactions beyond level 2 (i.e. beyond the Th level, see Fig. 1) are non-functional. Furthermore, it will turn out that we have to discern between two different pathways of signal percolation. Firstly, idiotypic activation might generally go via B cell clones (i.e. the "B-B-B" pathway); anti-idiotypic helper T cells help the B cells on the pathway but do not transfer the idiotypic signal (see Fig. 1a). Secondly, the signal might generally switch between B and Th clones (i.e. the "B-Th-B-Th" pathway), see Fig. 1b. Because of the expected randomness of the Th and B cell repertoires, we even have to consider mixtures of both pathways. The structure of this paper is thus rather complex: two models and two percolation pathways are treated.

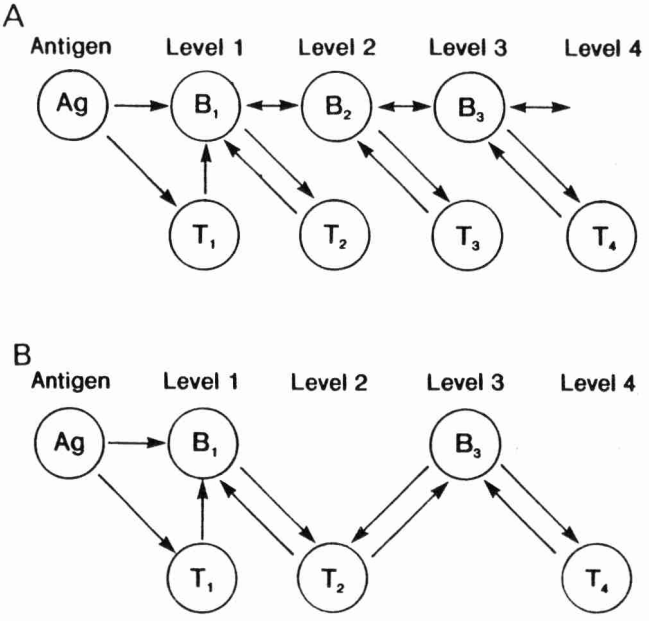

Figure 1. Two schemes of idiotypic Th-B cell interactions. The upper panel (A) displays a "B-B-B" pathway, i.e. a network of idiotypic B cell interactions (these are always symmetric) in which each B cell clone is helped by an anti-idiotypic Th clone (B_i by T_{i+1} etcetera). The Th clone therefore belongs to the next idiotypic level. This Th-B interaction is symmetric in the ThId model and asymmetric if we incorporate MHC-restriction. The bottom panel (B) displays a "B-Th-B-Th" pathway, i.e. a network devoid of idiotypic B cell interactions. Idiotypic activation along the subsequent levels now switches between B and Th cells. In the actual models we ignore the antigen specific helper T cells, i.e. the T_1 population that is shown in both panels.

2. The previous symmetric B cell model

We [De Boer, 1988; De Boer & Hogeweg, 1989_b, 1989_c] considered one class of cells, i.e. clones of B-lymphocytes. We assumed that B-lymphocyte populations are regulated by three processes: 1) influx of newborn cells from the bone marrow, 2) normal turnover (decay) of cells, and 3) proliferation. It was further assumed that idiotypic interactions influence the rate of cell proliferation. (It was implicitly assumed that idiotypic B cell interactions are T cell independent or that T cell help is always sufficient). Because idiotypic recognition is based on complementary matching, followed by receptor crosslinking, idiotypic interactions seem necessarily symmetric. Hoffmann [1979, 1980] first proposed this simple and appealing symmetry theory. Because B cells are most probably activated by the crosslinking of the antigen receptors (surface Ig), it is to be expected that the rate of cell activation increases with the concentration of the crosslinking agent (here antigen or anti-idiotypic antibody). However, whenever these concentrations become too high, the efficacy of cell activation by receptor crosslinking decreases. This argument corresponds to a (log) bell shaped proliferation dose response curve, i.e. to one that can be found in any textbook on immunology.

The results showed that these "reasonable" B cell models can easily account for memory phenomena (i.e. immunity) in networks incorporating very few B cell clones, i.e. low-D networks. Memory is generated by stable state switches from a virgin state to the "correct" immune state [De Boer & Hogeweg, 1989_b]. The general validity of these results was however questioned because, in high-D networks, similar state switches necessarily occur for clones at higher idiotypic levels. Thus idiotypic activation percolates deeply into the network and is expected to affect most of the clones [De Boer, 1989_a; De Boer & Hogeweg, 1989_c]. We considered this behaviour to be unrealistic.

3. Toward a realistic model of idiotypic T-B interactions

Empirical data show that B cells can only proliferate and mature if they are sufficiently "helped" by soluble factors produced by antigen (or idiotype) specific helper T cells (Th) [Melchers & Anderson, 1986]. This is also true for idiotypic interactions: in order to elicit an anti-idiotypic response one usually introduces idiotype plus adjuvant [Kawahara *et al.*, 1986] or dentritic cells [Francotte & Urbain, 1985] which enhance the Th response. The necessity of incorporating T-B interactions in our models arises, firstly, from our inability to account for reasonable network behaviour with our previous models of idiotypic B-cell networks [De Boer, 1988; De Boer & Hogeweg, 1989_c]. Our aim is now to determine whether the regulatory properties of Th cells might solve these earlier problems. Secondly, if we consider a "random" network of Th and B clones, idiotypic interactions between T and B cells in such a network might be different from those amongst B-cell clones. However, such an extended model can be developed along two alternative lines of evidence derived from empirical data.

The most classical approach is the line that has demonstrated that Th cells can only see B cell idiotypes that are presented by the (class II) MHC molecules embedded in the B cell surface. Because antigens are processed (i.e. degraded) before they are presented, the Th cells cannot be expected to recognise the native immunoglobulin (Ig). These aspects are treated in our second model. Our first model considers the other, more simple but more controversial, line of evidence which shows that some Th cells form bonds with complementary immunoglobulins [Bottomly, 1984; Janeway, 1984, 1988_a]. Tite *et al.* [1986] describe a murine Th cell line that 1) activates B cells with complementary receptors (surface Ig), and 2) is itself activated by such complementary antibodies. Such idiotypic interactions are not MHC restricted. These Th cells are called ThId cells [Bottomly, 1984; Janeway, 1984, 1988_a].

The ThId model. If Th and B cell receptor molecules do indeed match complementarily, and if such matching does indeed suffice for cellular activation, these idiotypic Th-B interactions are expected to be symmetric. If a Th cell is activated by anti-idiotypic (i.e. complementary) Ig, the same Th receptors should also be able to activate the corresponding B cell. We believe that this is confirmed by

the empirical data on reciprocal activation [Tite *et al.*, 1986]. Therefore we asume that both the B-B and the Th-B idiotypic interactions in the ThId model are symmetric. Apart from the symmetric idiotypic Th-B interaction, the ThId model incorporates an asymmetric helper interaction, i.e. Th cells provide help for B cells (and not vise versa). These non-idiotypic helper interactions (e.g. aspecific local factor production) are however steered by the idiotypic interactions: B cells receive help from the T cells they interact with idiotypically. We assume that individual Th cells always help themselves sufficiently (Th cells generally produce their own growth factors); i.e. we ignore possible proliferation threshold effects [De Boer & Hogeweg, 1986, 1987].

In accordance with our previous models, both the idiotypic activation of B and T cells by anti-idiotypic Ig, and that of B cells by anti-idiotypic Th receptors, follows the general (log) bell-shaped dose response curve [De Boer & Hogeweg, 1989_b]. Thus extremely high concentrations of anti-idiotype are assumed to be suppressive for both Th and B cells. We again assume that a B cell that is activated by external antigen is always sufficiently helped by antigen specific Th cells (i.e. T_1 cells). This seems a quite realistic assumption; such Th cells are activated by antigen presenting cells. In our model antigen directly provides "help" for antigen specific B cells; this enables us to simplify the model by omitting antigen specific Th cells (T_1).

We propose the following model. Consider a set of B and Th clones, i.e. we have a total number (say N) of B cell clones B_i and Th clones T_j (i.e. $1 \le i,j \le N$). Let αId_{Bi} be the total amount of anti-idiotype for clone B_i; this is divided over anti-idiotypic Th (αIdT_{Bi}) and anti-idiotypic B (αIdB_{Bi}) cells. Our Th clones cannot see the other Th clones: the total amount of anti-idiotype for clone T_j is αId_{Tj} which exclusively consists of αIdB_{Tj} (i.e. of anti-idiotypic B cells). The B cells only respond to antigen (Ag_i): antigen specific Th cells were omitted (see above). Hence, the growth functions "G" for B respectively Th cells become:

$$G_B(B_i, Ag_i, \alpha Id_{Bi}) = \frac{Ag_i + \alpha Id_{Bi}}{G_{1B} + F \circ B_i + Ag_i + \alpha Id_{Bi}} \circ \frac{G_{2B}}{G_{2B} + \alpha Id_{Bi}} \qquad (1)$$

$$G_T(T_j, \alpha Id_{Tj}) = \frac{\alpha Id_{Tj}}{G_{1T} + F \circ T_j + \alpha Id_{Tj}} \circ \frac{G_{2T}}{G_{2T} + \alpha Id_{Tj}} \qquad (2)$$

The B_i clone receives "help" from its anti-idiotypic Th cells (αIdT_{Bi}) according to a saturation function "H":

$$H(B_i, Ag_i, \alpha IdT_{Bi}) = \frac{Ag_i + \alpha IdT_{Bi}}{H_1 + F \circ B_i + Ag_i + \alpha IdT_{Bi}} \qquad (3)$$

Thus B and Th cells grow at a rate:

$$B_i' = S_{Bi} - D \circ B_i + P \circ B_i \circ G_B(B_i, Ag_i, \alpha Id_{Bi}) \circ H(B_i, Ag_i, \alpha IdT_{Bi}) \qquad (4)$$

$$T_j' = S_{Tj} - D \circ T_j + P \circ T_j \circ G_T(T_j, \alpha Id_{Tj}) \qquad (5)$$

Where S_{Bi} and S_{Tj} specify the influx of cells per clone "i" or "j" from the bone marrow and thymus respectively, D is the rate of cell turnover, and P is the maximum rate of proliferation. For reasons of simplicity we assume that each antigen is recognised by only one B clone. Antigen (Ag_i) cannot grow, has a low turnover rate R, and is removed by the (one and only) B cell clone (B_i) that recognises this particular antigen:

$$Ag_i' = - R \circ Ag_i - \frac{K \circ Ag_i \circ B_i}{K_1 + B_i} \qquad (6)$$

All saturation functions are buffered with a F parameter (F<<1). Buffering ensures that large idiotypic populations cannot be stimulated by small antigen and/or anti-idiotype concentrations [De Boer & Hogeweg, 1989_b]. The general parameter setting is the same as the one used before: $S_B \approx S_T \approx 10$ cells d^{-1}, D=1 d^{-1}, P=1.5 d^{-1}, $G_{1B}=G_{1T}=10^3$, $G_{2B}=G_{2T}=10^6$, F=0.01, R=0.1 d^{-1}, K=1, $K_1=10^5$. B cells require about H_1=100 Th cells for half maximal help; this resembles our previous estimate for the effect of help (i.e. IL2) from helper cells [De Boer & Hogeweg, 1986, 1987]. The virgin population density equals S/D ≈10 cells. The influx is slightly different for each clone (to prevent settlement in unstable equilibria): S has a mean of 10 cells per day with a 10% standard deviation. Virgin populations are too small to evoke proliferation ($S_B/D \approx S_T/D << P_1$): idiotypic interactions are negligable in the virgin state. Recent empirical data confirm this assumption: virgin B cell populations are indeed unable to activate resting T cells [Lassila *et al.*, 1988]. Moreover, in our models virgin Th populations are insufficient for helping B cells ($H_1 > S_T/D$): only by proliferation can Th populations become effective helpers. All cells are short-lived, i.e. they live about one day. Maximum proliferation proceeds at a rate P-D=0.5 cells per cell per day (this corresponds to a doubling time of about 16 hours). See [De Boer & Hogeweg, 1989_b] for a more detailed discussion of these parameters.

The ThMHCId model. A general dogma in immunology is that Ig bonds with native antigen, whereas Th receptors bond with processed antigen fragments presented by class II MHC molecules. Via the surface Ig receptors B cells should thus be able to recognise anti-idiotypic (i.e. complementary) T cell receptors. Th cells, by contrast, should not bond with complementary Ig, but should only be able to interact with Ig that is internalized, fragmented, and represented by the B cell MHC. Because B cells 1) do express class II MHC, 2) do process antigens, and 3) do internalize their Ig, such MHC-restricted Th-B idiotypic interactions are indeed expected to take place. MHC-restricted idiotypic interactions between Th and B cells are indeed described in the literature [Celada, 1988; Kawahara *et al.*, 1986]; this is a class of anti-idiotypic Th cells separate from the ThId cells described above [Janeway, 1988_a].

Importantly, however, this MHC-restricted interaction structure is no longer expected to be symmetric. A (3-D shaped) B cell receptor molecule (Ig) that matches complementary to a (3-D shaped) T cell receptor molecule, is not expected to be complementary to the same TcR if the Ig is processed and presented as a (linear) fragment in combination with MHC. Thus the Th cell that activates a particular anti-idiotypic B cell is itself expected to become activated by different anti-idiotypic B cell clones. Moreover, note that "anti-idiotypic" or "complementary" now has two meanings: 1) complementary native molecules (i.e. Ig vs. Ig or Ig vs. Th receptor), and 2) Ig fragments that in combination with MHC are somehow "complementary" to Th receptors. In this context we simply conclude that, within the framework of MHC-restriction, idiotypic Th-B interactions are a viable possibility if they are asymmetric.

MHC-restriction has even more implications. Th cells are activated by B cells by cell to cell contacts [Kupfer *et al.* 1986], and not by picking up free antibodies from the circulation. It has even been shown that Th cells orient the release of lymphokines toward the B cell that presents the antigen [Kupfer *et al.*, 1987; Poo *et al.*,1988]. Older empirical reports do indeed describe that Th and B cells are "monogamous" [Waldmann *et al.*, 1976; Phillips & Waldmann, 1977; Sullman & Feinstein, 1977], i.e. one Th cell interacts with only one B cell. Because each Th cell apparently sees only one B cell, this means that a high concentration of anti-idiotypic B cells is not necessarily suppressive to Th cells. We therefore assume that B cells activate Th cells up to a certain maximum; the state of Th activation cannot decrease if B cells further increase in number. In order to incorporate the

"monogamous" activation of Th by B cells, the corresponding term (eq. 2') in the model saturates to both Th and B cells. An increase in Th cells thus requires an equivalent increase in B cells. We consider networks in which 1) B cell clones recognise each other symmetrically by complementary matching of Ig, 2) B cells recognise Th receptors, and 3) processed B cell idiotypes are recognised by Th cells. The idiotypic interactions of B cells again follow the bell-shaped dose response curve. The other assumptions are the same as above.

Let again αId_{Bi} be the total amount of anti-idiotype for clone B_i; this again consists of anti-idiotypic B (αIdB_{Bi}) and Th (αIdT_{Bi}) cells. Th cells again only see B cells (and not each other): the total amount of anti-idiotype for clone T_j is αId_{Tj} which exclusively consists of αIdB_{Tj}. Let $T\alpha Id_{Bi}$ be the total amount of Th that recognise the B_i idiotype: this is the (weighed) number of Th cells that provide help for B_i growth and maturation. Thus the growth function G_B of the B cells (eq. 1) can remain the same but its helper function H now incorporates $T\alpha Id_{Bi}$ (eq. 3') instead of αIdT_{Bi} (eq. 3). The growth function G_T of the Th cells (eq. 2') no longer incorporates the suppressive term and saturizes to both T and B cells because of the monogamous interactions:

$$G_T(T_j, \alpha Id_{Tj}) = \frac{\alpha Id_{Tj}}{G_{1T} + T_j + \alpha Id_{Tj}} \qquad (2')$$

$$H(B_i, Ag_i, T\alpha Id_{Bi}) = \frac{Ag_i + T\alpha Id_{Bi}}{H_1 + F \circ B_i + Ag_i + T\alpha Id_{Bi}} \qquad (3')$$

Thus B cells grow at a rate:

$$B_i' = S_{Bi} - D \circ B_i + P \circ B_i \circ G_B(B_i, Ag_i, \alpha Id_{Bi}) \circ H(B_i, Ag_i, T\alpha Id_{Bi}) \qquad (4')$$

The growth of Th cells (eq. 5) and the removal of antigen (eq. 6) remain equal. In order to compare the two models (ThId and ThMHCId), we also keep the parameters the same.

We first analyse these models in simple low-D situations: a B_1 clone interacts with one T_2 clone and/or with one B_2 clone. In order to analyse signal percolation along "B-Th-B-Th" and "B-B-B" pathways (see Fig.1) two situations are respectively distinguised: "Th-B interactions" and "B-B interactions". For reasons of simplicity, we first omit the affinity of the interactions (i.e. we assume maximum affinity). Then the equations simplify because in the ThId model $\alpha IdB_{B1} = B_2$, $\alpha IdT_{B1} = T_2$, and $\alpha Id_{T2} = B_1$. In the MHC-restricted model $\alpha Id_{B1} = B_2 + T_2$, $\alpha Id_{T2} = B_1$ and $T\alpha Id_{B1} = T_2$. See Fig. 1 for explanation. Thus we analyse 2-D to 4-D models consisting of one or two B_i clones each helped by a Th clone. These models are analysed numerically by GRIND [De Boer, 1983]; GRIND uses the ROW4A integrator [Gottwald & Wanner, 1981]. We secondly specify affinity matrices and analyse 200-D networks using a variable step size Runge-Kutta-Merson integrator implemented in NAG [1984].

4. Results

Th-B interactions. Consider an idiotypic interaction between the B cell clone B_1 and the Th clone T_2. We thus have a 2-D network comparable to those analysed before [Hoffmann, 1979; Gunther & Hoffmann, 1982; De Boer, 1988; De Boer & Hogeweg, 1989$_b$], except for the fact that here we study an explicit Th-B interaction. Here we analyse two such Th-B networks: the ThId model (Fig. 2-3) and the MHC-restricted model (Fig. 4).

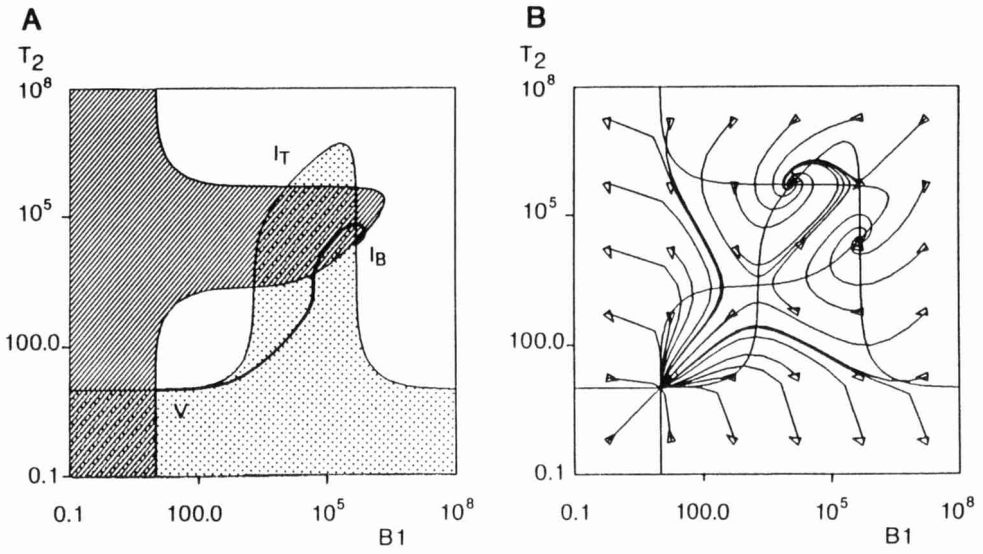

Figure 2. The Th-B interaction in the symmetric ThId model. Fig. 2a shows the $B_1'=0$ and the $T_2'=0$ isoclines. The isoclines intersect in 3 stable equilibria: the virgin state (V), and two immune states for T_2 (I_T) and for B_1 (I_B). The region of Th proliferation (i.e. $T_2'>0$) is dotted, that of B cell proliferation is striped. The fat line marks the trajectory initiated by the introduction of antigen ($Ag_1=10^5$). The phase portrait (Fig. 2b) shows the basins of attraction of these three equilibria.The ThId model.

The 0-isoclines of the ThId model (Fig. 2a) are very similar to those of our previous B-B models. Again we find three stable equilibria: one virgin state (V in the Figure) and two immune states (for T_2 (I_T) and B_1 (I_B) respectively). The dynamic analysis, in the absence of antigen (Fig. 2b), shows the basins of attraction of the three stable equilibria. Most trajectories return to the virgin state. The I_T state attracts somewhat more trajectories than the B_T state because B cells require Th cells for both idiotypic stimulation and helper factors, whereas Th cells require B cells only for idiotypic stimulation. However, following the introduction of antigen in this system (the thick line in Fig. 2a), this Th-B network switches to the immune state for the B cells (I_B). The B cells recognise antigen and hence gain a proliferative advantage over the Th clone. In the immune state both the B_1 and the T_2 clone remain enlarged due to reciprocal stimulation. Because both clones are enlarged, reintroduction of this antigen in the I_B state leads to rapid antigen rejection, i.e. to immunity. Thus this Th-B network accounts for antigen specific immunological memory. Experimental data have indeed suggested a role for regulatory T cells in "idiotypic memory" [Kelsoe *et al.*, 1981].

Because the Th-B interaction in the ThId model is so similar to the B-B interaction of our previous models, we also expect the (nasty) extensive percolation of the previous model to occur in the present ThId model. Indeed, if we consider a third clone B_3 that interacts with T_2 in the I_B state, it is expected to proliferate. If T_2 is able to sustain the proliferation of the large (immune) clone B_1, it should also be able to induce proliferation for the few (~10) cells of clone B_3. The Th clone T_2 provides 1) a strong antigenic (i.e. idiotypic) stimulus for B_3, and 2) sufficient help for B_3 proliferation. The proliferating B_3 can in turn activate an anti-idiotypic T_4 clone. Therefore we conclude that, if we consider "B-Th-B-Th" paths (see Fig. 1b) in the ThId network, we again find

extensive percolation of idiotypic signals. Note that this already occurs if such a network has two connections per clone (see Fig. 1b, and below).

In Fig. 3 we test the sensitivity of these results for the Th parameters, 1) H_1: the number of helper cells required for providing sufficient help, and 2) the onset and offset of Th proliferation (i.e. G_{1T} and G_{2T}). Fig. 3a shows that if H_1 becomes too large (i.e. $H_1 >> 10^3$) B cells fail to proliferate. In such situations a Th population of about $G_{1T}=10^3$ cells suffices for providing (half maximal) idiotypic B cell stimulation, but is insufficient for providing "help" for B cell proliferation. If $H_1 \leq 10^3$, the $B_1'=0$-isocline is independent of H_1. $H_1=100$ thus seems a very reasonable choice. The relation between the onset and offset of Th proliferation (i.e. G_{1T} and G_{2T}) is compared with that of B cell proliferation (G_{1B} and G_{2B}) in Fig. 3b. We analyse the sensitivity of G_{1T} and G_{2T} with the Q parameter (Fig. 3b): let G_{1T} be $Q \circ G_{1B}$ and G_{2T} be $Q \circ G_{2B}$ (i.e. Q=1 generates our general model). The Figure shows that both immune states exist if Th and B cell proliferation are sufficiently similar (i.e. Q≈1); Q may vary considerably however. For simplicity reasons we choose for Q=1, i.e. $G_{1T}=G_{1B}=10^3$ and $G_{2T}=G_{2B}=10^6$.

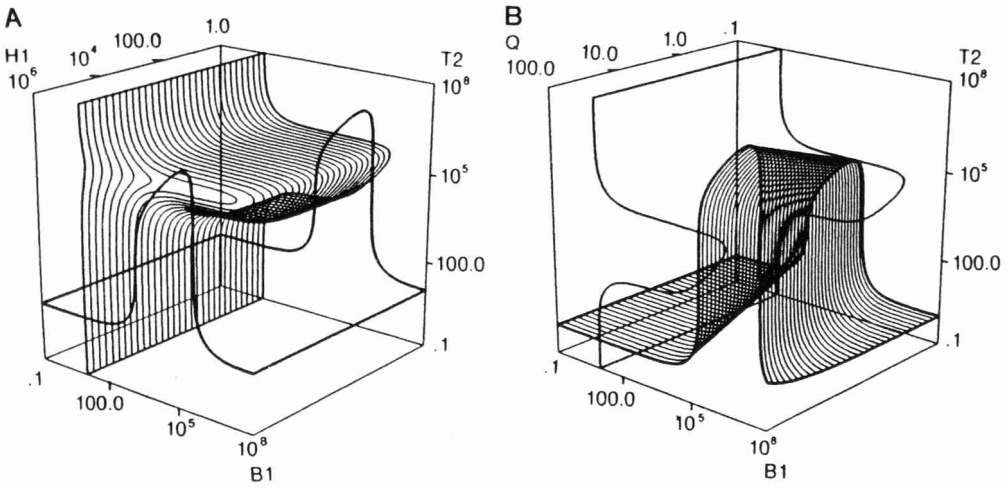

Figure 3. Parameter sensitivity analysis of the ThId model. Fig. 3a shows the $B_1'=0$ and $T_2'=0$ isoclines as a function of the H_1 parameter, i.e the number of helpers required for half maximal help. The isoclines are insensitive to H_1 for $H_1 \leq 10^3$; an increase of H_1 reduces B cell proliferation whatever the helper density. We choose for $H_1=100$, i.e. B cells easily proliferate. Fig. 3b shows the effect of changes in the Th bell-shaped dose response curve: G_{1T} is defined as $Q \circ G_{1B}$ and G_{2T} as $Q \circ G_{2B}$ (i.e. Q=1 generates our general model). Here the $T_2'=0$ plane is shaded. Both immune states exist if Th and B cell proliferation are sufficiently similar (i.e. Q≈1); Q may vary considerably however.

The ThMHCId model. Because the MHC-restricted idiotypic Th-B interaction is asymmetric, we now have to consider two different situations in two dimensions. In the first (Fig. 4a) the Th clone T_2 recognises clone B_1; in the second (Fig. 4b) B_1 recognises T_2. Because Ig fragments presented in MHC context are expected to be very different from native Ig receptors, B_1 and T_2 are not expected to see each other. If the Th clone recognises a B clone, it is perfectly capable of proliferation: Fig. 4a displays a large region of Th proliferation. However, although the B cell receives help from T_2, it cannot proliferate because it does not receive any antigenic (here idiotypic) stimulus. Thus, the

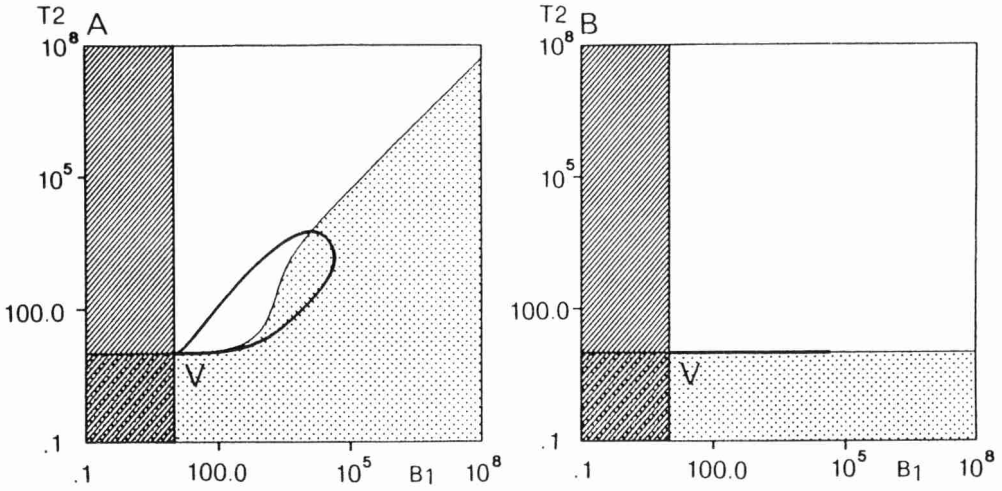

Figure 4. The Th-B interaction in the asymmetric MHC-restricted model. Fig. 4a shows the $B_1'=0$ and the $T_2'=0$ isoclines of a model in which T_2 sees B_1; Fig. 4b those of a model in which B_1 sees T_2. The shading of the respective Th and B cell proliferation regions is the same as that in Fig. 1. Both MHC-restricted models only have a virgin state. The fat lines are trajectories initiated by antigen (i.e. $Ag_1=10^5$).

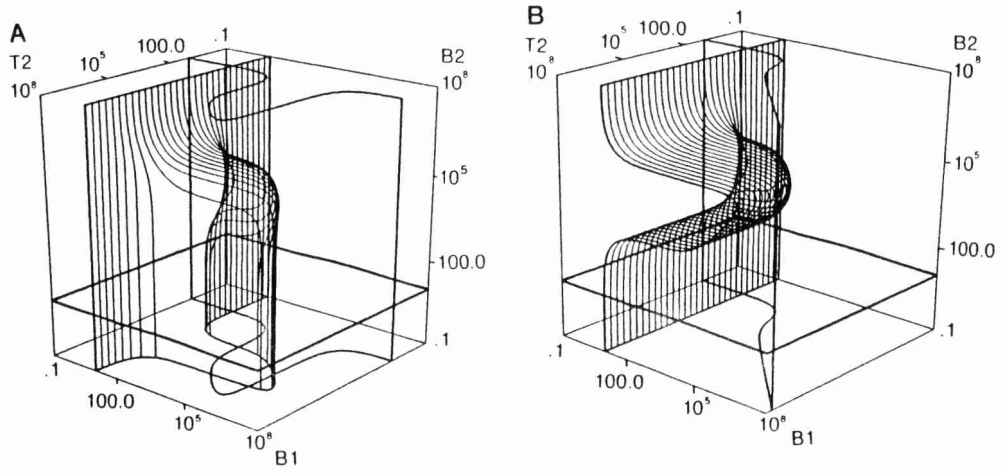

Figure 5. Static analysis of B-B interactions in 4-D models: B_1 and B_2 recognise each other. B_1 is helped by T_2 and B_2 is helped by T_3. The Figure shows the $B_1'=0$, the $B_2'=0$, and the $T_2'=0$ isoclines as a function of B_1, B_2 and T_2; T_3 is kept at the virgin state in the Figure (i.e. $T_3\approx10$). The $B_1'=0$ isocline plane is shaded, and the $B_2'=0$ isocline is flat. Fig. 5a displays the 0-isoclines of the ThId model (in which the Th-B interactions is symmetric). Fig. 5b those of the MHC-restricted model: the Th clones recognise (and help) the B cell clones but not vice-versa.

isoclines intersect in only one equilibrium: the virgin state. If, on the other hand (Fig. 4b), the B cells recognise the Th clone, we find hardly any interaction. The Ig receptor of the B cells can be activated by the Th cells (if Th > G_{1B}), but the B cells do not receive help because the T_2 cells are not activated; hence the B cells cannot proliferate.

In both MHC-restricted systems, stimulation of the B_1 clone with antigen (Ag_1) always leads to B_1 proliferation (because of our implicit Ag-specific helper "cells"). Both systems however fail to switch to "immune" states because the two clones never stimulate each other reciprocally (i.e. there are no immune states, Fig. 4). We conclude that 2-D MHC-restricted Th-B interactions cannot account for immunological memory. The second idiotypic level, here the T_2 clone (Fig. 1), can only be activated if the Th clone sees the B clone (Fig. 4a). And, even if this is the case, Th proliferation is temporary: the systems return to the virgin state. We conclude that signal percolation along "B-Th-B-Th" paths remains absent due to the asymmetry: any B_3 clone that would recognises such a proliferating T_2 clone cannot respond due to a lack of help (cf. Fig. 4b), and any B_3 clone that would be recognised by this T_2 clone cannot proliferate due to a lack of idiotype (cf. Fig. 4a).

We can do the same parameter sensitivity analysis as we did in Fig. 3. Whatever the magnitude of the H_1 or G_{1T} and G_{2T} parameters, we never find any immune state. Furthermore, note that if we were to abandon our assumption that B cells cannot suppress Th cells, i.e. if we were to use eq. 2 instead of eq. 2', we would only obtain an instable intersect between the $T_2'=0$ and the $B_1'=0$ isoclines. Thus, if the Th isocline were to fold back due to a suppressive excess of B cells, this would not generate an immune state.

We conclude for the two Th-B interaction systems that 1) in the ThId model Th-B idiotypic interactions are rather similar to the B-B interactions of our previous models, and hence that idiotypic signals percolate extensively, and 2) that MHC-restricted, i.e. asymmetric, helper interactions fail to give rise to stable immunity phenomena by mutual stimulation. As a consequence signals do not percolate at all.

B-B interactions. In order to analyse percolation via "B-B-B" paths (Fig. 1a) we now consider situations in which clone B_1 interacts with clone B_2 (symmetrically for both models), and in which each B clone is recognised by a Th clone (symmetrically or asymmetrically): B_1 by T_2 and B_2 by T_3 (see Fig. 1a). We thus analyse 4-D systems with three idiotypic interactions (i.e. one B-B and two Th-B interactions). These 4-D systems are statically analysed in 3-D state spaces, i.e. the idiotypic B_1-B_2 interaction is analysed as a function of T_2 (i.e. help for B_1). The dynamic analysis (by numerical integration) of these systems is performed in the complete 4-D network.

Static analysis. In the symmetric ThId model the B_1 clone has two idiotypic interaction partners: its helper clone T_2 and the clone B_2 (Fig. 5a); in the asymmetric MHC-restricted model B_1 is recognised by the same two clones (i.e. T_2 and B_2), but recognises only B_2 (Fig. 5b). In the Figure we have shaded the $B_1'=0$ isocline plane. B_1 can only proliferate in response to B_2 at sufficiently high T_2 levels. In the ThId system, large T_2 populations are suppressive for B_1 proliferation (see the section on Th-B interactions); in Fig. 5b this is not the case (because B_1 doesn't recognise T_2). The respective $T_2'=0$ isoclines are identical to those described above: T_2 is never influenced by B_2. The most important result from this static analysis is the $B_2'=0$ isocline plane: in both models this isocline is straight, i.e. is independent of B_1 and T_2. Because B_2 is not being helped by its helper T_3 (T_3 is virgin, i.e. $T_3=S_{T3}/D\approx 10$ in the Figure), B_2 cannot respond anyhow. In the dynamic analysis this will turn out to be the normal situation.

Dynamic analysis. In Figure 6 we stimulate these systems with antigen. In both systems this evokes proliferation of the B_1 population (B_1 is activated and "helped" by antigen). The expansion of B_1 in turn evokes T_2 proliferation. However, in both systems (i.e. Fig. 6a and 6b) B_2 fails to respond to this expansion of B_1; B_2 is activated (by B_1) but is not being helped (by T_3). T_3 is never activated by these idiotypic cascades (because B_2 fails to respond) and thus remains at the virgin level of S_{T3}/D. We conclude that due to a lack of help of the second level B cells (i.e. B_2) the idiotypic network reaction remains confined to 1) the B cell clones that respond to antigen, and 2) the Th clones

that recognise these particular B clones. Thus, whether or not Th-B interactions are MHC-restricted, B cells of the second idiotypic level fail to proliferate. Considering "B-B" idiotypic interactions, it thus turns out to be necessary to consider the concomitant activation of helper T cells. Previous theoretical idiotypic network models have, however, neglected explicit T-B coorperation.

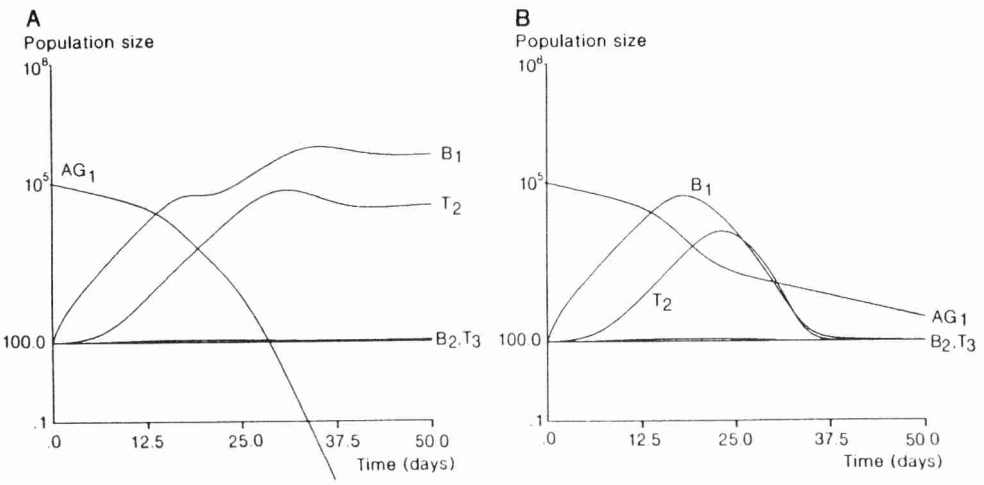

Figure 6. Dynamic analysis of B-B interactions in 4-D models, i.e. the immune reaction of these models to antigen (Ag_1). In both models (Fig. 6a and 6b) the B_1 clone proliferates (being helped by the implicit antigen specific Th) which induces the proliferation of T_2. However, B_2 and T_3 always remain virgin. In the ThId model (Fig. 6a) the system remains in the immune state described in Fig. 2. The MHC-restricted system (Fig. 6b) cannot account for immunity (see Fig. 4).

In conclusion, the two models differ in the generation of immunity: the symmetric ThId system remains immune (by means of mutual T_2-B_1 stimulation, Fig. 6a), the MHC-restricted systems returns to the virgin state (Fig. 6b). It is important to note that both systems do have attractors in which the B_1 and the B_2 are both enlarged (i.e. immune or suppressed). These states are attained if we start with high T_2 and T_3 densities. Thus, the fact that B_2 and T_3 are not being activated by the idiotypic cascade of Fig. 6 is a dynamic feature of the models. The clones of the second and third level are perfectly capable of interacting with each other, and thus of switching to different states, but due to the initial absence of "help" for B_2 such interactions fail to develop.

Percolation. The profound idiotypic networks that we have to consider are however combinations of "B-B" and "Th-B" interactions. Due to the expected randomness of the shapes of idiotypes one would expect a network topology that is comprised of mixtures of "B-Th-B" and "B-B-B" pathways. We have concluded above that the ThId and the MHC-restricted model differ only with respect to the percolation along "B-Th-B" pathways. We will here further analyse this difference in large random networks of "B-B" and "Th-B" interactions. The general model equations (i.e. eqs. 1-5, and 2'-4') remain the same, we just have to specify the multiple interactions between the respective clones. Consider a symmetric matrix "BB" of affinities of the respective B-B interactions. Symmetry means that $BB_{ij}=BB_{ji}$; B cells clones never recognise themselves (i.e. all $BB_{ii}=0$). Affinity values are drawn randomly from a uniform distribution between zero and one (i.e. $0 \leq BB_{ij} \leq 1$); see De Boer [1988] and De Boer & Hogeweg [1989_c] for further explanation. In the ThId model, the Th-B interactions are also symmetric: we additionally need a symmetric matrix "TB" that specifies the affinities of the respective Th-B interactions ($TB_{ij}=TB_{ji}$). Note that, clone T_i and clone B_i can interact, i.e. the TB_{ii} values need

not be zero. In the asymmetric MHC-restricted model we, however, need two matrices for the Th-B interactions: the matrix "TB" specifies the affinities with which Th cells see B cells, the matrix "BT" specifies those with which B cells see T cells. In the ThId model we thus obtain the following expressions:

$$\alpha IdB_{Bi} = \sum_{j=1}^{N} BB_{ij} \circ B_j, \qquad \alpha IdT_{Bi} = \sum_{j=1}^{N} TB_{ji} \circ T_j, \qquad \alpha Id_{Ti} = \sum_{j=1}^{N} TB_{ij} \circ B_j.$$

And for the MHC-restricted model we need the following terms:

$$\alpha IdB_{Bi} = \sum_{j=1}^{N} BB_{ij} \circ B_j, \qquad \alpha IdT_{Bi} = \sum_{j=1}^{N} BT_{ij} \circ T_j,$$

$$\alpha Id_{Ti} = \sum_{j=1}^{N} TB_{ij} \circ B_j, \qquad T\alpha Id_{Bi} = \sum_{j=1}^{N} TB_{ji} \circ T_j.$$

Varying the connectivity of the various affinity matrices, we study networks consisting of a 100 Th and a 100 B cell clones (i.e. N=100). We thus analyse networks with an average of one to four B-B and Th-B interactions per clone (Fig. 7). For reasons of simplicity we keep the connectivity of the TB and the BT matrix the same (if we would not do this, results would depend on the connectivity of the matrix that has the least connections). In the virgin state of these 200-D networks we introduce a random antigen for a period of 25 days (i.e. $Ag_i=10^5$). This triggers the idiotypic cascade. The system is simulated (by numerical integration) until it settled into an equilibrium. In this equilbrium we score for each clone whether or not is affected by the percolated idiotypic signal (i.e. we score the number of virgin clones). This is depicted in Figure 7. See De Boer [1989_a] or De Boer & Hogeweg [1989_c] for further explanation of these methods.

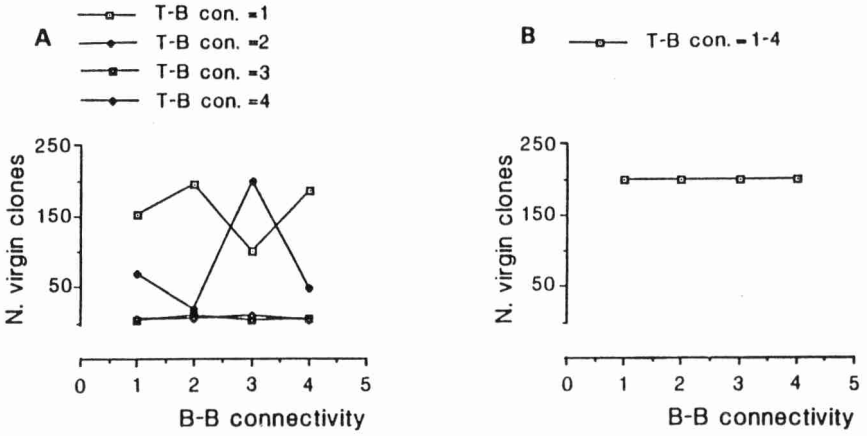

Figure 7. Percolation in 200-D networks. As a function of the number of B-B and Th-B interactions we plot the number of clones that manage to remain virgin (and hence responsive) following antigenic perturbation of the network. In ThId systems (Fig. 7a) we find extensive percolation whenever the Th-B connectivity exceeds a certain threshold. In the MHC-restricted systems (Fig. 7b) we never find any percolation.

The Figure clearly shows the absence of percolation in the MHC-restricted model (Fig. 7b: all 200 clones remain virgin whatever the connectivity). In the ThId model, conversely, signals do percolate extensively via "B-Th-B" pathways: percolation is largely independent of the BB connectivity but

depends strongly on the TB connectivity. This is in full accordance with the results described above: due to the absence of helper activation "B-B" topologies cannot become functional. Signals can only proceed along "B-Th-B" pathways (i.e. in the ThId model). This "B-Th-B" percolation however affects most of the clones, and is (again) unrealistic [De Boer & Hogeweg, 1989_c]. Additionally, it is worth mentioning that affected Th clones tend to become immune, whereas affected B cell clones tend to end up suppressed. This was already explained by the basins of attraction displayed in Fig. 2b.

Signals do percolate extensively in the ThId model whenever the TB connectivity exceeds a threshold level of two connections per clone (see Fig. 1b, Fig. 7a). At the threshold (i.e. the T-B con.=2 line) it is a matter of change whether the antigen triggers the largest connected structure (which explains why this line is so erratic). In our previous B-B networks this connectivity threshold was located at an average of one connection per clone [De Boer, 1989_a, 1989_b; De Boer & Hogeweg, 1989_c]. The threshold is doubled here because connecting two B cell clones (via a Th clone) now requires two idiotypic connections (see Fig. 1b) instead of one direct B-B connection. A connectivity of two idiotypic interactions per clone is very low [De Boer & Hogeweg, 1989_c]: due to random effects many clones will not be connected to the network at all (and hence require other, i.e. non-idiotypic, regulatory mechanisms). We thus conclude that profound ThId networks cannot account for idiotypic memory: most of the clones are affected by the first antigenic triggering of the network (see Fig. 7a).

Help parameters. Extensive percolation can also be obtained in the MHC-restricted system if we facilitate B and Th proliferation by assuming that a B cell population can be helped by few (e.g. one: H_1=1) Th cells, and that a Th population needs only few (e.g. G_{1T}=1) B cells to proliferate. For such parameters the B and Th clones always proliferate concomitantly: the B cells are helped ($H_1 < S_{Ti}/D \approx 10$) and the Th cells are stimulated idiotypically ($G_{1T} < S_{Bi}/D \approx 10$). In fact, such Th-B models become comparable to our previous B cell models in which we assumed that B cells were always helped sufficiently. If B cells are always helped, they will activate each other reciprocally, and we find extensive percolation (via "B-B-B" pathways). Hence we conclude that MHC-restricted Th cells prohibit idiotypic activation if: 1) the Th population needs to proliferate in order to have a significant helper effect, and 2) that Th populations do not proliferate in response to virgin (i.e. non proliferated) anti-idiotypic B cell clones. Both seem reasonable assumptions (we have always considered proliferative immune reactions); moreover, the latter (i.e. 2) has recently been confirmed [Lassila *et al.*, 1988].

Discussion.

General conclusion. The role that T-B cooperation plays in the percolation of idiotypic signals has turned out to depend strongly on the nature of 1) the network topology (i.e. the percolation pathway), and 2) the helper T cells (i.e. ThId or MHC-restricted). Considering "B-B" topologies, we found that both types of Th cells prohibit the proliferation of the anti-idiotypic B cells. Such a lack of Th activation (i.e. of T_3 cells) explains why, in experiments, the production of anti-idiotypic antibodies in response to idiotypic stimulation is generally low. In theoretical idiotypic network models, Th cells are usually neglected. Because Th activation turns out to be crucially important, results obtained with those previous models can only be valid for helper-independent "B-B" idiotypic interactions (provided these exist). Considering "B-Th-B-Th" pathways, we found a crucial difference between MHC-restricted Th and ThId cells. Due to the asymmetry of MHC-restricted idiotypic interactions, these "classical" Th cells altogether prohibit percolation of idiotypic signals. Thus, if "B-B" idiotypic interactions were to depend on MHC-restricted Th activation, we would conclude that idiotypic networks cannot become functional. And, consequently, that idiotypic network theory should no longer be viewed as a straightforward implication of the extensive (or even complete) receptor repertoires [Jerne 1974, 1984]. If, on the other hand, ThId cells suffice for T-B cooperation, the

"B-Th-B" topology becomes very important since idiotypic signals percolate extensively along such pathways.

B-B interactions. In both models idiotypic interactions among B cells fail to become functional. This leads us to speculate that idiotypic B-B interactions are essentially absent from the many (>10^7 different clones) "classical" B cells that do indeed depend on helper T cells. We define "classical" B cells as the B cells that 1) depend on helper T cells, and 2) show a secondary response with 2a) antigen specific memory, 2b) somatic hypermutation, and 2c) a switch to IgG production. For the other class of "helper independent" B cells it would, by contrast, be possible to have functional idiotypic B-B interactions (although these are expected to percolate extensively). Such cells exist; they only produce IgM and do not account for memory. Because the $Ly1^+$ or $CD5^+$ B cells (that predominate early in ontogeny) are largely confined to IgM type [Hayakawa *et al.*, 1984; Möller, 1986], and because they are suppossed to emerge before helper T cells emerge [Martinez-A *et al.*, 1988], they seem reasonable candidates for this helper independent group. Empirical data [Fougereau & Schiff, 1988] seem to confirm this. Indeed, pronounced idiotypic networks have been described for this distinct group of B cells [Vakil & Kearny, 1986; Kearny & Vakil, 1986].

Interestingly, these $CD5^+$ B cells are multispecific [Holmberg *et al.*, 1984, 1986], i.e. they recognise a wide variety of antigens and idiotypes. We demonstrated previously [De Boer, 1988; De Boer & Hogeweg, 1989_c] that networks of clones with many idiotypic connections behave totally different from lowly connected networks. Highly connected systems behave autonomously, i.e. proliferate in the entire absence of antigens. Secondly, such networks are unresponsive to external antigens because the clones in such networks are either immune or suppressed: the perturbation that arises by the antigenic stimulation is usually negligable compared to the strength of the autonomous idiotypic interactions. Both the autonomous activation [Pereira *et al.*, 1986] and the poor responsiveness to antigens [Hayakawa *et al.* 1984] have been described experimentally. With regard to our present results, these previous results would remain correct, i.e. would correctly describe the abberant behaviour of such "neonatal" networks, if "early" $CD5^+$ B cells are indeed helper-independent. Additionally, the present results can now explain how the "classical" B cells escape from the, seemingly inescapable, influence of this highly connected (neonatal) network. The classical B cells will not respond to the abundant $CD5^+$ interactions as long as their helpers are not being activated.

Th-B interaction. In our MHC-restricted model we assume that B cells internalise, process, and present their surface Ig to helper T cells. In our models we have however ignored the empirical data that suggest that antigen presentation is only carried out by activated B cells [Krieger *et al.*, 1985]. Since we made an additional and more stringent assumption, i.e. Th cells can only be activated if the B cells have proliferated, this is not expected to affect the results. It can however easily be incorporated in the model: the fraction of activated B cells is in fact given by the growth function G_B. Hence the amount of anti idiotypic B_i cells for T_j would become $\alpha Id_{Tj} = B_i \circ G_{Bi}$ instead of simply B_i. If it is incorporated, the percolation results remain the same (not shown).

Another complication is the "peptidic self" model proposed by Kourilsky *et al.* [1987]. These authors also assume that idiotypes are processed into peptides that are subsequently presented to T cells. Moreover, they even go one step further: it is assumed that anti-idiotypic B cells that are crosslinked by idiotypic antibodies internalise these complexes, process them, and hence present both idiotypic and anti-idiotypic peptides to the T cells. If this were true, it would have important implications because it would mean that idiotypic and anti-idiotypic B cells can be helped by the same helper T cells. Thus, with respect to our results, B cells of the second level receive help from the Th cells that were activated by the first level B cells. Hence the second level B cells are expected to proliferate. As a consequence, B and Th cells of the third level respond, which, in combination, activate B cells of the fourth level. We conclude that the "peptidic self" model enhances percolation.

ThId cells. If ThId cells do indeed play an important role in immune systems, this role could be comparable to that of the $CD5^+$ B cells. Both cells types generate idiotypic networks whereas the

"classical" B cells and "classical", i.e. MHC-restricted, Th cells do not. It might even be speculated that the lineage of T cells that uses the γδ receptor (instead of the "classical" αβ TcR) accounts for ThId activity [Janeway, 1988_b]. Importantly, it would mean that interactions among "classical" Th and B cells are always non-idiotypic, and that "specialised" cell types account for "separate" idiotypic networks. (See Martinez-A *et al.* [1988] for related speculations). It is possible however that we have overestimated the role of the ThId cells because in some of the empirical reports is is claimed that ThId cells require the concomitant action of conventional MHC-restricted Th cells [Becker Dunn *et al.*, 1986; Janeway, 1988_a]. For our ThId model this would mean that it approximates the MHC-restricted model because the ThId cells are no longer expected to provide help to anti-idiotypic B cells.

T-T interactions. In our analysis we have only considered idiotypic B-B and Th-B interactions; for reasons of clarity we have ignored idiotypic T-T interactions. Idiotypic interactions amongst helper T cells (that we have considered here) are controversial: helper T cells generally fail to express MHC class II antigens which they might need for the presentation of their idiotypes to each other. However, recent empirical data [Lider *et al.* 1988; Ellerman *et al.* 1988; Sun *et al.* 1988] suggest that suppressive idiotypic T-T interaction do play an important immuno- regulatory role. Such suppressive T-T interactions may also be important for our percolation problem. Additionally, networks of idiotypic T cell interactions may (again) be different from the Th-B cell networks that we are critisizing here. Thus this deserves further study: empirical data are needed to substantiate the occurence of idiotypic T-T interactions (e.g. for the different T cell subsets), theoretical analysis is needed to pinpoint the possible significance of such interactions for idiotypic immunoregulation.

Our analysis has given a negative answer to our question on the generation of localised network responses (i.e. of limited percolation) by Th interactions. ThId cells impair the generation of localised responses because such cells help and activate third party B cell clones. Conversely, MHC-restricted Th cells reduce the percolation too rigorously: idiotypic interactions altogether fail to become functional. Idiotypic network theory should therefore no longer be viewed as an "unavoidable idea" [Jerne, 1984]. We conclude that, if idiotypic interactions are to be functional, the present helper T cell models fail to solve our problem of "extensive percolation".

The fact that the results enable us to escape from the "unavoidable" network theory might be important for two reasons. Firstly, no one has yet been able to develop a reasonable high-D idiotypic network model that actually accounts for the most significant immunological phenomena such as regulation, immunity, and self non-self discrimination. This might be a matter of time of course; people have only recently begun to analyse high-D networks [Farmer *et al.* 1986; Segel & Perelson, 1988; Perelson, 1988; Hoffmann *et al.*, 1988; De Boer, 1988]. Alternatively, however, it might be intrinsic to the idiotypic network idea: we have previously described a number of problems that arise in simple, but reasonable, idiotypic network models [De Boer & Hogeweg, 1989_c]. Secondly, immune systems seem perfectly capable of generating these phenomena by interactions at the clonal level. Many models have described important regulatory and/or memory processes that are either intrinsic to [Grossman, 1984; De Boer & Hogeweg, 1986, 1987; Kevrekidis, 1988] or can be added to (in the form of suppression) [Irvine & Savageau, 1985_a, 1985_b; Kaufman & Thomas, 1987] the clonally selected expansion of helper (or effector) arms of the immune system.

In conclusion, helper T cells play an essential role in determining whether a topological idiotypic network can become functional. However, since percolation is either extensive or absent, and cannot remain limited or localised, we conclude that helper T cell activation is not the putative missing element that solves the problems that we have with idiotypic network theory [De Boer & Hogeweg, 1989_c].

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