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An Autoimmune Mechanism for AIDS' T4 Lymphopenia

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Abstract.
We put forward a new model for the T4 lymphopenia occurring in AIDS by suggesting a mechanism whose net effect is blocking the generation of T4 cells during HIV infection. Supporting evidence for this mechanism is derived from experiments in the recent literature.

The etiological agent of AIDS has been identified in the HIV virus. HIV is a retrovirus with very long latency, whose action is directed against a subpopulation of T cells. T cells, a fundamental component of our immune system, can be divided in two subpopulations: T4 cells and T8 cells. The first is characterized by the expression of glycoprotein CD4 as a surface receptor, the second by glycoprotein CD8; both subpopulations are essential for properly mounting an immune response. HIV does not affect T8 cells, but infects and ultimately causes the death of T4 cells by specifically binding to their CD4 receptor. Though the immune system mounts a vigorous humoral response to HIV, within a few years AIDS patients progressively develop a characteristic T4 lymphopenia. Once T4 cells become sufficiently few, the immune system no longer works properly, AIDS patients fall prey to all kinds of opportunistic infections, and eventually die.

There still is, however, a mysterious aspect to this illness.

A mystery. HIV infects and ultimately kills T4 cells by binding to CD4. It is thus quite natural to attribute the slowly developing T4 lymphopenia of AIDS patients to the selective depleting action of HIV. But things are not that simple: the number of T4 cells infected by HIV is *negligible* with respect to the greater number of "missing" T4 cells.

Indeed, many AIDS researchers (including Gallo and Montagnier [1]) do not think that HIV's direct killing of T4 cells is sufficient for explaining the depletion seen in AIDS, and ask which indirect mechanisms may also be at work. We wish to suggest such a mechanism.

An Indirect Mechanism

Receptors CD4 and CD8 play, respectively, a major role in the activation of T4 and T8 cells (review by Bierer, Sleckman, Ratnofsky, Burakoff [2]). Indeed, T4 cells recognize antigen in the context of MHC class II proteins (ligands for CD4), and T8 cells in the context of MHC class I proteins (ligands for CD8). An equally important role is played by CD4 and CD8 during thymic development. As shown by Ramsdell and Fawlkes [3], their engagement is *required* for the maturation of, respectively, T4 and T8 cells. Indeed, it is in the thymus that maturing T cells are selected on the basis of their capability of recognizing antigen. We now ask the following question:

What will happen if the thymus is injected with soluble CD4?

Our answer is that *the maturation of T4 cells will be inhibited.* In the experiments of Ramsdell and Fawlkes, the role of CD4 in T4-cell maturation was established by injecting anti-CD4 antibodies that do not deplete peripheral T4 cells nor double positive thymocytes. Simply *blocking* --with a proper mAb-- the CD4 receptors of maturing T4 cells also blocked their development. We believe that the main effect of this blocking consisted of preventing the interaction of CD4 with thymic class II MHC molecules, and that in turn this caused the blocking of development. However, this interaction may be prevented in a different, but symmetric manner, namely, by blocking thymic class II MHC proteins, something that may be accomplished by soluble CD4.* At this point, a second question naturally arises. Namely,

Is soluble CD4 ever injected in the thymus?

We now argue that this event may indeed be an indirect result of HIV infection:

> As for all foreign organisms, our body must react to HIV by producing antibodies to all of its antigenic determinants. One of these determinants is known to be a binding site for CD4. Thus we hypothesize that one of the (a group of) antibodies raised against HIV must have a binding affinity *very close* to the one of CD4. Let's call *CD4like* such an antibody, or group of antibodies. Like the copy of a key, *CD4like* may be vastly different from CD4, but will essentially have its functional value. Thus, it is capable of binding to MHC-II molecules of thymic cells, preventing the development of maturing T4 cells.

Let us further elucidate this mechanism. The release into the blood of *CD4like* caused by HIV will not be a one-time affair. HIV has a very long latency, during which it seems to be "invisible" to the immune system. In these conditions *any* virus would remain present for a long time. Assume, in fact, that some virus *V* manages to infect many cells of the organisms before any defense against it can be mounted. At this point, each infected cell has a separate, individual destiny. Thus, within the infected cells, the proper conditions for *V* to become active will occur at *random* and *independent* times.

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* An indirect confirmation that blocking class I (II) MHC molecules may block the development of maturing T4 (T8) lymphocytes, may be inferred from the results of Zijlstra, Bix, Simister, Loring, Raulet, and Jaenish [6]. They show that genetically engineered mice not expressing class I MHC proteins (not in the thymus nor anywhere else) lack $CD4^{-}8^{+}$ cytolytic cells.

If the expected life time of the infected cells and the expected latency of virus *V* both are --say-- several years, the result of the initial infection is that *V* (or its proteins) will reappear at random times for several years. If the cells initially infected are sufficiently many, these "random, reappearance times" will be frequent enough for *V* (or its products) to be essentially continually present and detectable in the organism for a long time. (This would hold even if the infected cells were not T4 ones and the immune system had mastered the *instant* killing of virus *V* whenever it becomes visible.) Thus HIV "continual reappearance," continually elicits powerful secondary responses of the immune system. Consequently with our hypothesis, (different) plasma cells will continually produce (different) antibodies against HIV, *including CD4like* ones. In fact, despite its constant mutability, HIV maintains its capability of binding CD4; thus, once a *CD4like* antibody has been successfully "manufactured," its continual production will be guaranteed by the *memory* of the immune system, by the *continual reappearance* of HIV, and by the presence on the virus' surface of an *identical* binding site for CD4. (It is important to notice that our hypothesis is distinct from what at first glance may resemble it. Namely, antigenic mimicry and anti-idiotype networks involving an internal image of the antigen.)

An important, novel feature of our hypothesized mechanism is that it provides a better model for the T4-cell depletion that is the hallmark of AIDS; a model, that is, that explains the mentioned mystery away. In fact, HIV *does not need* to directly kill lots of T4 cells to cause AIDS' impressive T4 lymphopenia. (In principle, it might not need to directly kill a single T4 cell!) It would be sufficient for it to be "visible" for a long time to the immune system, so as to elicit for a long time the production of *CD4like*, and thus mislead the organism into producing fewer T4 cells. These cells have a finite life time and must be replaced; tampering with their replacement may be HIV's most insidious action. In a few years time, it may easily cause the typical T4 lymphopenia of AIDS patients, even without any direct killing. (If HIV only caused a modest, selective depletion of *easily replaceable* T4 cells, AIDS patients might perhaps adjust to living with it.)

The emerging etiology for AIDS' T4 lymphopenia is thus that of an autoimmune mechanism. This is in agreement with Giorgi's and Dentels' [4] remark that T4 depletion occurs only after antibody formation against HIV, though the presence of antigen can be documented prior to seroconversion. The emerging picture is also easily reconcilable with the fact that the body produces a vigorous response to HIV, as it is exactly this powerful response that causes T4 cell loss.

Testing The Mechanism

Ideally, one would like to develop an essay for *CD4like* antibodies and then determine whether there is a correlation between the quantity of *CD4like* present and the decline of T4 cells. However, more indirect tests may also be useful.

Such an indirect test may consist of monitoring T-cell reconstitution of irradiated animals, both in the presence and in the absence of soluble CD4. Radiation, causing a sudden drop in T-cell level, may allow one to conduct a shorter experiment. Working with soluble CD4 avoids isolating *CD4like* among many candidate antibodies, and a new successful method for producing soluble

rat CD4 has recently been obtained by Davis, Ward, Puklavec, Willis, Williams, and Barkley [5].**

Thymus cultures and bone-marrow cells may be used to verify that the thymic tissue of AIDS patients indeed produces fewer T4 cells than normal. Of course, an effective method must be used so to trace only newly generated T cells.

An elegant test has been suggested by Herman Eisen [7] for determining whether the non-polymorphic regions of thymic MHC-II molecules of AIDS patients have some proteins bound to it, thus providing an indirect evidence for the existence of *CD4like*. The test consists of showing that some (fluorescent) antibodies for non-polymorphic epitopes of MHC-II molecules fail to bind the thymic cells of AIDS patients.*** The same experiment can be conducted using B cells rather than thymic cells, since they too express MHC-II molecules.

In Sum

Above we have described the most plausible way, in the light of established biological mechanisms, for *CD4like* to influence the level of T4 cells. So little, however, is known about T-cell regulation, that many other possibilities exist for *CD4like* to affect the level of T4 cells, even in mature adults whose thymus may be atrophic. (For instance, an anti-antibody may be raised against *CD4like* capable of binding CD4 and depleting T4 cells. For all we know, it may even be that the level of T4 cells is controlled by the *total* amount of CD4 --whether or not on cell surface-- thus allowing *CD4like* to mislead the organism into believing that there are many more T4 cells than actually present.) For this reason let us summarize our autoimmune model for AIDS' T4 lymphopenia in a more open-ended manner. Namely,

CD4like causes loss of T4 cells.

Models have a fundamental role in organizing our thoughts and pointing out new possibilities for deeper understanding, but, of course, the last word always belongs to the Experiment.

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** It is, however, important to notice that, due to the difference in molecular weight and structure between *CD4like* and soluble CD4, the risk exists that only the former protein may successfully block thymic MHC-II cells. Moreover, soluble CD4 would last in circulation much less than an antibody, and a viable method must be found to keep high level of it in the blood.

*** A more refined version of the test consists of using both antibodies (labelled "green") for the polymorphic regions of MHC-II proteins, and (labelled "red") for the non-polymorphic regions, and then studying the green/red binding ratio for B cells of AIDS patients and healthy individuals.

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