The immune system and HIV

How HIV damages the immune system

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<th>Source: Aidsmap: <a href="http://www.aidsmap.com">http://www.aidsmap.com</a></th>
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| A patient oriented educational website
| Adapted by Luc Kestens for SCART 2005 students
| Recommended reading with the lecture
| 23/8/05: Virologic and immunologic basis of HIV infection + HIV pathogenesis |

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Summary: How HIV damages the immune system

Primary infection

- HIV can enter the body through the sexual organs, the bloodstream and the mouth. Many people experience a flu-like illness a few weeks after being exposed to HIV.

- The `seroconversion illness' may be accompanied by a rash, ulcers in the mouth, severe headaches, diarrhoea, fevers, aches and pains and swollen glands.

- This illness is followed by the development of antibodies to HIV, and the immune system brings HIV under control.

- Not everyone experiences such an illness, but within a few months after becoming infected everyone begins to produce antibodies to HIV.

- A small minority of people seems to be at least partially resistant to infection with HIV because of their genes.

How AIDS develops

- The immune system is gradually disrupted. HIV kills cells in the lymph nodes (clusters of immune cells that trap foreign organisms), and this throws the immune system out of balance.

- Virus levels in the blood and the lymph nodes increase because HIV can escape the antibodies and other immune cells which normally control infections.

- Each generation of viruses is slightly different, and this constant evolution helps HIV keep one step ahead of the immune system. The immune system cells are always looking for viruses that resemble the previous generation of HIV, so gradually the virus can `escape' and the viral load increases.

- The CD4 lymphocytes, or CD4 T-cells, gradually decline in number because they are killed by HIV, and are killed more rapidly as HIV levels increase.

- HIV destroys the immune system's memory. CD4 cells which have been programmed to recognise infections become depleted. This is why opportunistic infections like PCP and candida can develop when the CD4 count falls low.
The pathway to disease

The process by which a micro-organism causes disease is known as pathogenesis. Despite extensive research into AIDS pathogenesis since 1981, large gaps remain in our knowledge of the steps between HIV infection and the development of AIDS. A simple concept of pathogenesis is with one factor causing a change in another, which in turn changes another, and so on. For example, A affects B, B affects C, and so on, until Y affects Z (the Z is the end result, such as a disease).

A simple orthodox view of pathogenesis for AIDS is: HIV infection leads to loss of CD4 cells; this loss of cell-mediated immunity results in increased susceptibility to AIDS-related conditions.

In reality there are many more steps in the pathogenesis of AIDS. There are also human and environmental factors influencing almost every step, and changes in one factor may influence other functions of the body along other pathways.

From the very beginning of AIDS research there has been disagreement between virologists and immunologists about the relative importance of the virus and the immune system in the pathogenesis of AIDS.

The virus and the immune system

From the perspective of virologists, the important factor driving the pathway is the success of the virus in replicating (measured as viral load). They are interested in why different strains of HIV have different preferences for either CD4 cells or macrophages as host cells and for different places in the body, such as the gut or brain. These variations in function are called phenotypes and they are related to single or multiple changes in the viral genome (genes). There are even different phenotypes in terms of sensitivity (or resistance) to drugs. An important area of interest is how the virus evades the immune system (or drugs) by mutating and varying the structure of proteins. Virologists are interested in how the virus itself could kill CD4 cells directly (such as during the process when HIV buds away from cells). A priority is the development of anti-HIV drugs. The majority of basic science publications on HIV are virological.

Immunologists are interested in how CD4 cells could be killed by immune responses and the abnormalities which develop in the immune system during HIV infection. They are interested in measuring the functions of particular immune cells and the effects of cytokines. Ultimately, they would like to find treatments that restore the immune abnormalities in HIV infection. They would also like to develop a vaccine that prevents HIV infection (in a person without HIV infection) or even a therapeutic vaccine (in people with HIV infection) by inducing immune responses that eliminate HIV.
The points of view of virologists and immunologists are not necessarily in conflict as each would recognise that both the virus (HIV) and the immune system (the human host) are inseparable players in the process leading to AIDS. There are differences of opinion as to whether the better treatments will be those directed against the virus or those to help the immune system; in practice, a combination of the two is likely to be needed for people whose immunity has already been substantially impaired by HIV. Of course, a consequence of AIDS research has been a huge advance in basic knowledge about viruses and the immune system which will be helpful in the fight against AIDS as well as other diseases.

**Exposure and primary infection**

HIV is not able to enter through most of the surface linings of the body. However, it can enter through some wet mucous membranes such as in the rectum, the female genital tract, the glans penis (particularly in uncircumcised men), and the urethra (the tube that carries urine). Blood and genital secretions in men and women are the infectious fluids for people with HIV infection.

It is not clear whether HIV is infectious as free virus outside cells or as HIV in CD4 T-cells passing to another person. It is also not clear which cells are first infected by HIV, but these may be lymphocytes and macrophages in the linings of the rectum and sexual organs. There is good evidence that other sexually transmitted diseases can increase the risk of HIV transmission, by increasing the number of cells in genital secretions or causing ulceration.

HIV is probably trapped by dendritic and macrophage cells and carried to the lymph nodes where it is presented to and recognised by a few T- and B-cells. Under T-cell control the B-cells produce clones of plasma cells which can release antibodies that specifically attack HIV in the bloodstream. The appearance of these antibodies against HIV antigens in the blood is called seroconversion. HIV viruses outside cells can become coated with antibodies and may then be attacked and destroyed by cytotoxic cells. By a process of evolution, HIV repeatedly varies the antigens on its coat (called envelope antigens), enabling it to escape specific antibodies.

**Early detection of HIV**

Several different tests can be used to establish whether a person is in the early stages of HIV infection.

The most sensitive test which can be used is HIV polymerase chain reaction (PCR) testing, which is the same principle as the viral load test. This test may be able to detect HIV within 48 hours of infection in some cases. However, caution should be exercised in interpreting the results of such tests when the viral load measurement appears very low. Brown University School of Medicine in the United States has reported on three false-positive diagnoses of HIV infection by PCR testing: initial PCR testing yielded measurements of 1254, 1574 and 1300 copies/ml respectively, but subsequent PCR tests and antibody tests failed to confirm infection (Rich 1999).
Some laboratories may offer a proviral DNA test which is a highly sensitive and specific test for early HIV infection. An Australian clinic reported that 14 cases of HIV seroconversion were correctly identified by both the proviral DNA test and HIV RNA test. Viral load was above 15,000 copies/ml in all these cases (Medland 2003).

p24 antigen testing may also be used to determine infection, although this method has been superceded by PCR testing.

Antibody testing is unlikely to detect HIV antibodies until at least six weeks after infection in the majority of cases, and a small proportion of people (less than 1%) take up to six months to develop antibodies. The HIV antibody test is known as an enzyme-linked immunosorbent assay (ELISA) test.

A comparative study of diagnostic methods in people with seroconversion illness found that the p24 antigen and ELISA methods together detected only 79% cases of seroconversion. In contrast, the PCR viral load test detected HIV in every infected person. The third generation ELISA antibody test, designed to detect antibodies earlier than standard tests, was only as accurate as the p24 antigen test.

However, in this study the viral load test wrongly diagnosed the presence of HIV infection in a small number of individuals. The authors noted that in each case, viral load was reported to be below 3000 copies/ml, leading them to suggest that any viral load result below 5000 copies/ml in a patient with suspected primary HIV infection should be regarded as a false positive result and the patient should be re-tested.

The authors recommended that patients with suspected primary infection should be tested with a third generation ELISA test and a PCR test, with repeat PCR testing if the ELISA test is negative. If viral load remains higher than 5000 copies/ml while the ELISA result is negative, HIV infection should be presumed. However, ELISA testing should be continued on a regular basis until the presence of antibodies is confirmed (Hecht 2002).

Other experts have advocated use of the PCR test in conjunction with the proviral DNA test and routine antibody testing as the most reliable and accurate way of establishing the presence of HIV infection shortly after exposure or when seroconversion illness is suspected (Medland 2003).

**Seroconversion illness**

Seroconversion typically occurs two to twelve weeks after infection and may be associated with a flu-like illness. The most commonly reported symptoms include:

- Fever
- Rash.
- Aches and pains.
- Oral ulcers.
- Sore throat.
- Weight loss of more than 2.6 kg.
- Fatigue.
- Nausea.
Fever and rash together are the most strongly predictive symptoms of HIV seroconversion illness, according to a study carried in San Francisco (Hecht 2002). It is unclear if the presence of a larger number of these symptoms is a stronger predictor of HIV seroconversion. The duration of the illness and the severity of the symptoms are associated with a higher viral load during and just after seroconversion (Lavreys 2002).

It is thought that somewhere between 50 and 80% of newly infected people will experience most or all of these symptoms. A smaller proportion may develop diarrhoea, ulceration of the mouth, throat or genitals, anorexia or swollen lymph nodes (Clark 1991). Weight loss, abdominal pain, loss of appetite, oral candidiasis, as well as neurological abnormalities, have been reported as symptoms of seroconversion, but only loss of weight and appetite were associated with HIV seroconversion in another study which sought out people with apparent symptoms of seroconversion in order to test them for HIV infection (Vanhems 1999).

A study of seroconverters in Zambia has suggested a slightly different profile for seroconversion illness. Influenza, rash and sore throat were not linked to HIV seroconversion. Instead, malaria, diarrhoea, swollen glands, inflammation, night sweats and weakness were associated with HIV seroconversion (Fideli 2003). A study of women in Uganda and Zambia has also shown the women with acute or early HIV infection are more likely to have fever, severe headaches, abdominal pain, pelvis tenderness, fatigue, abnormal vaginal discharge and itching and abnormalities of the vagina and cervix than HIV-negative women, along with an increased incidence of vaginal infections (Morrison 2004). These study may point to regional or racial differences in the nature of seroconversion illness.

There is some evidence that people who experience symptoms during primary infection tend to have a more rapid rate of disease progression than people who have few or no symptoms upon initial infection with HIV.

During early infection HIV remains concentrated in the lymph nodes, where it replicates in huge numbers and infects more CD4 T-cells. Swollen lymph nodes are often the only clinical feature seen in a person with HIV infection for the first months or years after infection.

**Immune response during primary infection**

The presence of high levels of HIV in the body stimulates the immune system, and huge numbers of HIV-specific CD4 T-cells are produced in an attempt to contain the virus. However, these activated CD4 T-cells are also prime targets for the virus. Therefore, rather than controlling the infection, they are rapidly infected and destroyed, leaving the body with only weak anti-HIV immune responses. Long-term non-progressors, on the other hand, tend to have strong immune responses such as high levels of HIV-specific CD8 T-cells, which effectively control HIV replication and keep viral load low.

CD8 T-cell production during primary infection is the only immune parameter which has a significant association with the level of viral load, suggesting that these cells play a key role in controlling viral replication (Routy 2000).
For further information on the immune response to anti-HIV treatment during primary infection, see Treatment during primary infection in Anti-HIV therapy: When to start treatment.

**Viral dynamics during primary infection**

During the first few days of infection, HIV replicates without being checked by the immune system. Consequently, very high levels of HIV are present in the blood and individuals are likely to be highly infectious. For example, one study reported that individuals had an average viral load above one million copies/ml 13 days after infection (Kaufmann 2000).

During this initial period of infection, HIV establishes ‘viral reservoirs’ by infecting a range of different cells. Some of these cells become inactive ‘memory’ cells which harbour HIV's genetic material within the cells' DNA, thus holding the potential to produce more virus in the future. These reservoirs are seemingly unaffected by anti-HIV drugs.

**Follicular dendritic cells in the lymphoid tissue are known to be a major site of HIV trapping.** It seems that HIV rapidly accumulates in lymphatic tissue following infection (Schacker 2000).

When the body's anti-HIV immune response begins, symptoms of seroconversion may develop and viral load falls. One study found a relationship between viral load levels during primary infection, the severity of symptoms during primary infection and the level at which viral load subsequently stabilised, or the 'set-point'. A higher set-point has been found to predict the speed of progression to AIDS, as has the speed with which a patient reaches the set point: patients who reach their set point more quickly have a slower progression to AIDS (Pedersen 1997; Blattner 2004).

**Key research**

Blattner (2004) identified 22 patients with primary HIV infection from the Trinidad Seroconverter Cohort and followed them for 7 years. All those with viral load set points above the median of 25,000 copies/ml progressed to AIDS within 4.5 years, but half of those with set points below this value had not progressed to AIDS after 7 years. The 11 patients with a viral load decline after acute infection slower than the median of 0.63 log_{10} per month had progressed to AIDS within 5 years, but a quarter of those with faster declines had progressed to AIDS within 7 years. Fast initial clearance of HIV and maintaining a set point for over 20 months was associated with a 10-fold lower chance of progressing to AIDS, and having a set-point over 25,000 copies/ml was associated with a 6.5-fold increased chance.

Hecht reported a survey of 258 people screened for potential primary HIV infection (newly exposed individuals). The study was carried out by Positive Health Program at San Francisco General Hospital, which advertised in the community for people with suspected high risk exposures such as unprotected sex or needle sharing with an HIV-positive partner, and/or suspected symptoms of HIV seroconversion. Individuals were tested for HIV using a variety of methods, and the researchers assessed which symptoms occurred more frequently in people who were subsequently found to have HIV infection. The significant predictors were: Fever and rash together (OR 8.4); fever (OR 5.2); rash (OR 4.8); oral ulcers (OR 3.1); joint pain (OR 2.6); sore throat (
OR 2.6); loss of appetite (OR 2.5); weight loss of greater than 5lbs (2.5kg) (OR 2.8); muscle pain (OR 2.1); fatigue (OR 2.2); nausea (OR 1.9). Other previously reported symptoms, such as headaches, night sweats, diarrhoea, ulcers on the genitals and vomiting were just as likely to occur in people who did not have infection. The study was not able to identify whether the presence of a greater number of these symptoms had stronger predictive power, due to the relatively small number of people who were diagnosed with primary HIV infection (40 out of 258). There was no difference in the length of time that key symptoms lasted between people with HIV infection and people without. On average, symptoms lasted for ten days or less. The only exception was genital ulcers, which lasted significantly longer in patients with HIV infection (27 days vs 9.5 days).

Lavreys identified 74 women who seroconverted and for whom viral load measurements were available. Women were tested for HIV on a regular basis after being recruited to the cohort study, which enrolled a total of 1295 women. Fever, vomiting, headache, fatigue, arthralgia, myalgia, sore throat, skin rash and being too sick to work were associated with a significantly higher viral load. Asymptomatic women had a median viral load of 216 copies/ml (a level that would be regarded as a potential false positive result in at-risk individuals seen in everyday clinical practice). In women with one symptom, the median viral load was 32,111 copies/ml, compared with approximately 4 million copies/ml in women with five or more symptoms.

References
Lavreys L et al. Virus load during primary human immunodeficiency virus (HIV) type 1 infection is related to the severity of acute HIV illness in Kenyan women. Clinical Infectious Diseases 35(1): 77-81, 2002.
Strains of HIV

HIV undergoes a continual process of evolution after primary infection, and it seems that the balance between evolutionary pressures on HIV and the success of the virus population in adapting to these pressures are the major determinants of the rate of disease progression. In the body as a whole, this ongoing struggle is reflected in the level of viral load and the CD4 cell count, but within the virus population this struggle is evident in changes in the genetic characteristics of viruses.

During primary infection, viruses which are adapted to gaining entry to cells displaying the CCR5 chemokine receptor are likely to predominate. However, viruses which can also gain access to cells by using the additional route of the CXCR4 chemokine receptor (fusin) come to predominate over the CCR5 variants, and are associated with a much more rapid loss of CD4 cells. This is because HIV which is adapted to infecting cells which display the CXCR4 chemokine receptor will have a preference for infecting what are known as TH-2 type CD4 cells. HIV replication in TH-2 CD4 cells is far greater than in TH-1 type cells, which are preferentially infected by CCR5-tropic virus.

This switch towards a virus population which can infect more hospitable host cells is accompanied by other events, most notably the change from non-syncytium inducing (NSI) to syncytium-inducing (SI) virus. This means that viruses can begin to induce cell killing in uninfected CD4 cells.

Mechanisms of CD4 T-cell destruction

A number of theories have been developed to explain how HIV infection results in the destruction or loss of CD4 T-cells. Although many of these studies took place relatively early in the epidemic, the observations are still relevant in explaining the range of mechanisms by which CD4 T-cell death takes place in the body.

Syncytium formation

In a laboratory setting, a few HIV-infected CD4 T-cells can form a clump (syncytium) of fused cells. Strains of HIV found in late infection are more likely to have the ability to cause syncytium formation: the cytokine interleukin-15 (IL-15) may play a role in the transition from non-syncytium inducing (NSI) to syncytium inducing (SI) viral strains.

It is not known whether the clumping of CD4 T-cells is actually important inside the body. It is known, however, that SI strains are able to replicate faster than NSI strains and the actual turnover of virus may be the mechanism killing CD4 T-cells rather than syncytium formation itself.
Excessive gp120
During the time when HIV is replicating in CD4 T-cells there is excessive production of HIV proteins which can be released into the bloodstream and act as antigens. Free proteins called gp120 could attach to CD4 molecules on uninfected CD4 cells leading to an attack on the cell by antibodies and cytotoxic cells. Possibly, the attachment of gp120 to CD4 also inhibits the function of the CD4 T-cell.

Apoptosis
Normally, all cells can be programmed to die if they need replacing or eliminating. This is called apoptosis. An abnormally large proportion of infected and uninfected CD4 T-cells seem to be programmed for apoptosis in people with HIV infection. The reason is unclear but may involve the attachment of free HIV proteins to CD4 cells or due to cell over-activation.

One possibility is that HIV antigens act as 'superantigens' that can activate a larger range of CD4 T-cells than normal and which can cause increased replication of HIV. Changes in cytokine levels, such as increased tumour necrosis factor (TNF) and decreased interleukin-1 (IL-1), may also induce apoptosis. Interleukin-2 (IL-2) may rescue some cells from apoptosis. Interestingly, CD8 T-cells, natural killer cells and B-cells are also prone to apoptosis during HIV infection. However, while apoptosis of T-cells is common, there seems to be no apoptosis of HIV-infected macrophages or monocytes (Goldberg 1999).

Major histocompatibility complex and gp120 homology
It has been found that parts of the HIV envelope protein gp120 are similar to the major histocompatibility complex (MHC) molecules, the antigens present on human cell surfaces that signal to the immune system that the cell belongs to the host. The normal T-cell receptor learns to recognise foreign antigens when they are presented in association with MHC 'self' molecules. However, free gp120 can also attach to T-cell receptors by mimicking MHC proteins. This causes the T cell to be activated, possibly explaining the excessive activation of T cells during HIV infection.

Auto-immunity
One of the most controversial theories for CD4 T-cell death is based on the idea that HIV induces auto-immunity. This theory posits that host CD4 T-cells are no longer recognised as self but as foreign cells, which are destroyed by the immune system. Possible causes of this include the similarity or 'homology' between HIV antigens and non-self MHC molecules, with HIV antigens on the surface of CD4 T-cells causing them to be recognised as foreign cells.

Excessively activated B cells may also produce anti-lymphocyte antibodies. One effect of recognising host cells as foreign could be the generalised activation of the immune system which is seen during HIV infection. Conditions characteristic of auto-immunity are known to appear during HIV infection, including reduced platelet cell counts (thrombocytopenia), dermatitis, psoriasis, vasculitis (inflammation of blood vessels) and arthritis. It seems likely that genetic factors influence the risk of auto-immunity.
These characteristics are similar to those seen when foreign material is grafted into the body, such as during an infection or after a transplant (graft versus host disease). The use of immune suppressant therapies, such as corticosteroids, may therefore be of benefit in reducing the over-stimulation of the immune system, and CD4 T-cell death seen during chronic HIV-infection.

References


Immune responses to HIV

Most people infected with HIV will mount an effective immune response to the virus during the first few months of infection. However, over time this response will prove ineffective.

The response comes in two forms: cellular and humoral. The cellular response refers to the activity of the CD4 and CD8 T-cells, the latter known as cytotoxic lymphocytes (cell-killing immune cells). The humoral response refers to antibody production and activity.

Cellular immune responses
In the first few weeks after infection, the number of CD8 T-cells increases to up to 20-fold above the normal range, whilst CD4 T-cell numbers fall sharply.

There is a decline in the immune functions which are governed by CD4 T-cells, sometimes leading to the appearance of infections such as Candida, herpes and Pneumocystis pneumonia (PCP) during seroconversion illness. Six months after infection, CD4 T-cell function improves except in relation to HIV-specific antigen (Musey 1999). The few people who maintain strong HIV-specific CD4 T-cell responses have lower viral loads than people with poor responses. For more detail on CD4 T-cell decline following HIV infection see page 9: Mechanisms of CD4 T-cell destruction.

CD8 T-cells in HIV infection
CD8 T-cells play a crucial role in controlling HIV replication during the early phase of infection. HIV-specific CD8 T-cells are targeted at the dominant viral variant and their emergence is associated with a rapid fall in viral load, before the development of an antibody response. Most of the CD8 T-cells generated during primary infection die within a few weeks, leaving a reservoir of HIV-specific CD8 memory cells which will persist regardless of the presence of an antigen or CD4 helper cells.

CD8 T-cells appear to act against HIV in two ways during primary infection: by killing HIV-infected cells and by secreting chemokines. HIV-specific CD8 T-cells recognise
a specific genetic sequence of HIV and are primed to copy themselves if this sequence is encountered again in the future.

It is not fully understood why some people continue to exhibit strong HIV-specific CD8 T-cell responses which control viral load.

Several theories have attempted to account for the gradual failure of CD8 T-cells to control HIV replication. One theory (known as 'viral escape') is that the cells begin to lose the ability to recognise HIV's genetic sequences due to the high level of viral turnover and mutation. A second theory is that HIV may actually kill off some of the CD8 T-cell repertoire.

In support of the 'viral escape' theory, one study found that CD8 T-cells lose their ability to recognise and kill viral variants, even though they may be responsive to wild-type viruses. The researchers compared killer T-cells from HIV infected asymptomatic individuals with those from symptomatic AIDS patients. They examined the cells' ability to eliminate target cells infected with laboratory strains of HIV and with virus isolated from the patient. They found that killer T-cells from asymptomatic individuals can recognise and kill both types of target cells. In contrast, the killer T-cells from symptomatic patients, while still able to recognise and eliminate the laboratory strain targets, no longer killed target cells that were infected with their own virus.

While there may still be high numbers of killer T-cells late in HIV disease, the researchers found that they are likely to be programmed only to kill wild-type HIV. As HIV mutates in the body due to several factors, including pressure from antiretroviral medications, these killer T cells become increasingly irrelevant. Without helper T-cells, which slowly disappear during HIV disease, the killer T-cells are unable to programme themselves to recognise the increasingly diverse population of HIV inside the body.

These findings are significant because they show that conventional measurements of killer T-cell responses in people with HIV, which focus on responses to laboratory strains, overestimate the response to the person's own virus.

Another theory is that rapid progression and death may be linked to a defect in the activation and proliferation of HIV-specific CD8 T-cells. Studies by Walker and colleagues have found that HIV-specific CD8 T-cells do recognise viral variants, calling into question the 'viral escape' theory outlined above. Further evidence to support Walker's theory has come from German researchers who reported CD8 T-cells which specifically target 3TC (lamivudine/Epivir)-resistant virus in individuals with 3TC resistance, indicating the body's ability to adapt to viral variation even during advanced HIV disease (Schmitt 2000). A poor immune response to HIV may not therefore be due to a lack of HIV-specific CD8 T-cells, but to a lack of activity by these cells.

A subset of CD8 T-cells called CD8 / CD28 T-cells seem to be the most important cytotoxic cells, but their number decreases during HIV infection. Very rarely, an efficient CD8 T-cell response can occur before HIV has started to replicate in CD4 T-
cells or macrophages. This can prevent HIV infection and the production of HIV antibodies. This may occur more frequently in new-born babies than in adults.

A study of seven long-term non-progressors has found that they had relatively low levels of HIV-1 specific CD8 T-cells targeting Gag and Env HIV proteins, but high levels of CD8 precursor cells. Three of six long term non-progressors had high HIV-1 p24-specific CD8 T-cell responses (Greenough 2000). This study provides evidence that HIV-specific CD8 T-cell responses are crucial in control of disease progression.

More recently, research with a group of long-term non-progressors has identified that the CD8 T-cells of non-progressors could divide and proliferate more readily when called upon to fight infection. In addition, they also produced higher levels of a molecule called perforin which assists in the destruction of HIV-infected cells (Migueles 2002).

Another group has identified another set of proteins, called α-defensins-1, 2 and 3, which are secreted by CD8 T-cells. However, when α-defensins were isolated from the CD8 T-cells of long-term non-progressors, investigators found that the HIV-inhibiting properties of these proteins were minimal (Zhang 2002).

CD8 T-cells and activation markers
Researchers from the University of California have published work which confirms that CD8 cells are crucial in determining the speed of HIV disease progression. Comparing ten long-term non-progressors and eleven progressors who were matched for age, race, sex and time of seroconversion, the researchers found that long-term non-progressors had a low viral load, as well as significantly lower levels of the CD38-positive subset of CD8 T-cells. These differences were unrelated to viral type, variations in the CCR5 receptor or antibodies (Barker 1998; Mackewicz 1998).

Similarly, a study of men with advanced HIV disease found that high levels of the CD38 activation marker on CD4 and CD8 T-cells was associated with shorter survival time. In contrast, people with high levels of the HLA-DR activation marker on their CD8 T-cells had a longer survival time. Other factors such as high viral load, naive cell numbers and co-receptor usage did not impact on survival time (Giorgi 1999). Another group reported that the activation marker C1.7 Ag exhibited on activated CD8 T-cells was associated with disease progression (Peritt 1999).

Chemokine responses
CD8 cytotoxic T-cells appear to produce a substance (provisionally called CD8 antiviral factor or CAF) that inhibits HIV replication, and may or may not directly kill CD4 T-cells. While some researchers believe that CAF is composed of the RANTES, MIP-1α and MIP-1β chemokines that are thought to block the CCR5 receptor, when these cytokines are blocked with monoclonal antibodies, another HIV-suppressive factor still appears to be at work.

There is conflicting evidence about the impact of chemokine activity on HIV disease progression. Some research has suggested that chemokines suppress HIV replication, and that high levels of certain chemokines are associated with delayed HIV disease progression (Garzino-Demo 1999). However, one group of researchers has found either no consistent relationship between beta-chemokine production and
HIV replication or an unexpected correlation between high β-chemokine levels and high-level virus replication (Greco 1998).

Chemokine activity appears to decline during the course of HIV infection. The only test of an artificial chemokine as a therapy so far found no anti-viral efficacy, and researchers are now looking at whether they need to give higher doses or to administer chemokines in a different way.

Chemokine receptor genotype also affects the response to antiretroviral treatment. An analysis of 272 individuals in the ACTG 343 study of indinavir (Crixivan)-based triple therapy found that those with the genotype CCR5+/+, CCR2+/+ and CCR5-59029 A/A were 2.5-fold less likely to sustain undetectable viral load than patients with other genotypes. Furthermore, mean viral load reduction was 2.12 log10 for this group compared with 2.64 log10 for all other groups combined (O’Brien 2000).

**Antibody responses**

Antibody responses begin to develop four to eight weeks after infection, and tend to follow the CD8 T-cell response. Antibodies are chiefly targeted against free floating virions, although some antibodies may destroy HIV-infected cells. Antibodies cannot prevent the transmission of HIV from one cell to another. The antibodies are primed to recognise particular genetic sequences of HIV, and are also evaded by the gradual mutation of the virus. It is unclear how much evolutionary pressure antibodies place upon the virus.

A very small number of people infected with HIV do not develop antibodies to HIV. A study of six HIV-infected people found that the absence of antibodies was due to individual immune dysfunction (Ellenberger 1999).

**Intracellular responses**

A vital step between the entry of HIV into a human cell and the conversion of viral RNA into DNA is the removal, or uncoating, of the protective shell surrounding the virus’s genetic material. This coat, called the capsid, must be removed before HIV can insert its genetic material into a human cell and make copies of itself.

In early 2004, scientists announced in the journal *Nature* the discovery of an intracellular immune protein which blocks this process in monkeys, and to a lesser extent in humans (Stremlau 2004; Goff 2004). This molecule, called TRIM5-α in humans, specifically targets HIV’s capsid. In humans the protein has some ability to block HIV, and investigators are speculating that it could help to explain why some people infected with HIV do not experience disease progression.

Investigators believe that TRIM5-α chops up HIV’s capsid, preventing the orderly uncoating the virus must undergo before it replicates. TRIM5-α is one of a number of intracellular proteins with anti-HIV effects that may be expressed more or less strongly according to genetic inheritance.

**References**

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Immune disruption

The net loss of CD4 cells that develops during HIV infection has many direct and indirect consequences during HIV infection. There is less response by CD4 cells to antigens and less production of some cytokines. There is less response by CD8 cells to virus- and bacteria-infected cells. CD4 memory cells are lost so that the more efficient responses of acquired immunity decrease against familiar micro-organisms.

Memory B cells continue to function and produce excessively a wide range of antibodies against previously encountered antigens but there is an impaired response to new antigens. Natural killer cells have less activity against virus-infected cells because of cytokine abnormalities.

CD8 cells

CD8+ lymphocytes play an important role in controlling viral infections, and CD8+ cytotoxic lymphocytes are thought to control HIV through direct killing of cells infected by HIV, and by secretion of chemokines and other soluble factors which can suppress HIV replication.

CD8+ cells have been shown to express CD4+ markers on their surface after activation through the T-cell receptor. Expression of the CD4+ molecule permits infection by HIV. Robert Gallo and colleagues have demonstrated that CD8+ cells can become infected in the laboratory, and have proposed that this is the mechanism...
by which CD8 cells become depleted in HIV infection, leading eventually to the exhaustion of HIV-specific CD8 cells and the loss of immunological control over HIV. Most CD8+ cells do no co-express CD4 and this mechanism may only account for marginal CD8 depletion/dysfunction.

However, other researchers have proposed an alternative mechanism for CD8+ depletion, in which HIV-specific T-helper cells play the key role. CD4+ T-helper cells specifically targeted at HIV emerge during primary infection, but these cells are probably the first to be infected and killed by HIV precisely because of their tendency to proliferate in the presence of HIV antigen, thus offering new targets for infection.

CD4+ T-helper cells activate CD8+ cells, and strong CD4+ T-helper cell and CD8+ responses have been shown to correlate with long-term non-progression in several cohorts, and it has been suggested that restoring the HIV-specific CD4+ T-helper response by the use of a DNA vaccine would contribute to suppression of HIV and maintenance of a strong CD8+ response to HIV.

**Dendritic cells**

Follicular dendritic cells in the lymph tissue are a major source and reservoir for HIV. Dendritic cells in lymph nodes are destroyed during HIV infection. This means that T- and B-cells will have antigen presented to them by less efficient macrophage cells. Cytotoxic CD8 cells are highly activated in early HIV-infection and may continue to be activated in people who do not progress rapidly to HIV disease. They may secrete several newly discovered cytokines which inhibit the replication of HIV. CD8 T-cells decrease in number and function as HIV-infection progresses to AIDS even though they are not directly infected by HIV.

**Cytokines**

There are many long-standing changes to chemical messengers known as cytokines during HIV infection. IL-6 (interleukin-6) levels are increased and this activates most B-cells to release more of their antibodies – even though the antibodies may not be needed to fight infections all of the time. Elevated levels of a wide range of antibodies in the blood (hypergammaglobulinaemia) are a feature of HIV infection. B-cell activation may be one factor involved in the formation of B-cell non-Hodgkin lymphomas – a type of cancer which is more frequent in HIV infection.

Other cytokine changes also affect immune function.

- **Interleukin-2 (IL-2)** normally stimulates division of T-and B-cells and the killing of virus-infected cells by Natural Killer (NK) cells. IL-2 levels decrease in HIV-infection.

- Interferon gamma is also reduced. Normally, interferon gamma inhibits viruses replicating inside cells and stimulates cytotoxic cells.

- **tumour necrosis factor (TNF)** is increased and activates T-cells and HIV replication, as well as causing fever, weight loss and malaise (tiredness). Drugs that may reduce levels of TNF are expentifylline and thalidomide.
One theory to explain changes during HIV-infection concerns an apparent shift in the roles of T helper cells known as CD4 Th1 and Th2 cells. Early on, Th1 cells predominate and they are associated with production of IL-2 and interferon gamma and high activity by cytotoxic cells. This may help clear lots of HIV by cell-mediated immunity. Later, Th2 cells are dominant with production of IL-4, IL-6 and IL-10 which activates B-cells and antibody production – probably this humoral immunity is a less efficient way of eliminating HIV. It may be possible to use synthetic cytokines or drugs to correct these abnormalities and several drug trials are under way to test these as treatments.

Other factors such as surgery may enhance immune suppression in people with HIV although this has not been proven. It is known, however, that major surgery leads to suppression of Th1 cells and the cytokines it produces - IL-2 and interferon gamma.

**Antibody production**
HIV will evade the specific antibodies made against it by varying the structure of its proteins (antigens) – especially gp120 and gp41 found in the envelope of the virus. Even compared to other viruses, HIV’s ability to mutate is one of the fastest rates known. Early in HIV infection the B cells may keep up with the new antigens produced but this response is slow or absent in late infection. Possibly the B cell response gets exhausted. Direct HIV infection of B cells may also undermine their functioning. Potential HIV vaccines have been made which mimic parts of gp120 that are the least likely to vary over time.

Lastly, HIV may infect the stem cells in the bone marrow from which immune cells originate, which could interfere with their replacement.

**Why do CD4 T-cells disappear in HIV infection?**

Early in 1995 several scientists, including Dr David Ho, published new work which changed previous ideas about the turnover of HIV replication and the replacement of CD4 cells by the body. The people with HIV who were studied in the experiments were not necessarily typical of all people with HIV-infection as they had already had HIV-infection for many years and had relatively high quantities of HIV (viral load) in their blood. The measurements were possible because of the availability of new drugs which temporarily blocked most HIV replication and allowed CD4 cells to increase much more substantially than with previous treatments. Without these drugs the CD4 cell count and HIV viral load do not change much day by day.

Research published in 1995 and 1996 indicated that the half-life (the time it takes for 50% of the virus in the body to enter a CD4 T-cell, replicate and spread to a new T-CD4 cell) was about 1.2 days, of which about 0.9 days was spent inside a T-CD4 cell and 0.3 days outside T-CD4 cells. However, more recent research from David Ho’s team in 1999 estimated that HIV has a half-life of between 28 and 110 minutes. This means that HIV is cleared from the blood ten times faster than previously estimated. That is, about half of the HIV particles in the blood die and are replaced by newly produced virions every hour.
Based on data from four individuals, Ho’s team estimated that between 1,995-15,849 million new virions are produced and released into the blood every day. More than 99% of all HIV is produced from newly infected CD4 cells; the rest is from macrophages. Consequently, billions of CD4 cells are killed and replaced every day. If CD4 cells were not replaced they would all be killed within several weeks. Usually 2% of CD4 cells are present in the blood and the rest in tissues such as lymph nodes.

These measurements can be interpreted along with evidence that large amounts of HIV replication and CD4 killing take place in lymph nodes (outside the bloodstream) to mean that an extraordinary battle between HIV and the immune system takes place with an amazing capacity for lost CD4 cells to be replaced. Eventually, however, the immune system becomes exhausted, and is unable to match the rate at which new virus is produced.

Further investigation of the capacity for CD4 cell renewal has suggested that HIV causes an increased rate of CD4 cell proliferation and death. When antiretroviral therapy is administered, the rate of CD4 cell death is reduced, leading to an increase in the number of CD4 cells (Kovacs; Mohri)

Counter-arguments
However this model has been questioned by Dutch and American researchers who argue that the gradual decrease in CD4 cells is in part a consequence of the increased trapping of CD4 lymphocytes in the lymph nodes as viral infection in these sites worsens over time. This trend has been observed in macaques infected with simian immunodeficiency virus (Schenkel). The rapid rise in CD4 cell counts in the weeks after beginning potent anti-retroviral therapy is a consequence of the shut-down of virus replication in the lymph nodes, and the redistribution of CD4 cells back into circulation. CD4 cell renewal could also be affected by damage to the bone marrow (either by destruction of precursor cell production capacity by HIV, or by a direct effect of anti-retroviral drugs); by interference with CD4 cell production in the thymus; or by interference with cell cycling and signaling.

In early 1999 a team of researchers from the University of California at San Francisco published a paper in the journal Nature Medicine which opposed David Ho's work on HIV and T-cell dynamics. Using their own technique for labeling immune cells, they studied the half-life and production rates of CD4 and CD8 cells in nine uninfected people, seven untreated HIV-positive people, and five HIV-positive people who had undergone twelve weeks of highly active antiretroviral therapy (HAART). In the untreated HIV-positive group, the half-life of each T-cell sub-population was found to be less than a third of that of the seronegative controls. Contrary to Ho's proposed model, the Californian researchers found that this reduced cell lifespan was not compensated for by increased cell production. Following viral suppression after twelve weeks of HAART, they found a considerable increase in circulating CD4 and CD8 cells, though for CD4 cells this was due to greater production rather than a longer half-life. The authors concluded that the loss of CD4 cells which typically occurs in the course of HIV infection is not due (as Ho suggests) to the exhaustion which follows a sustained over-production of CD4 cells, but instead to a shortened survival time and to a failure to increase the production of CD4 cells (Hellerstein).
This group updated their findings in June 1999 with data comparing CD4 survival time after 12 weeks of HAART and after 18 months of HAART. They found that after 18 months of successful viral suppression, CD4 survival times had increased until they were almost equal to those seen in a healthy uninfected control group, from 14 to 78 days. Production was elevated above normal levels during the first months of therapy but returned to normal or below average after 18 months (Hellerstein 1999b).

Complementing these findings, Professor Frank Miedema of Amsterdam University has proposed that the key mechanism leading to loss of CD4 cells is interference with the production of T-cell progenitors. These are produced in the bone marrow and migrate to the thymus where they are transformed into mature T-cells of all types. In long-term non-progressors, Miedema has found that progenitor production is unchanged after six years of infection, whereas production declines in those with disease progression. HAART improves T-cell progenitor production and the improvement in naive CD4 cell reconstitution seen on HAART is correlated to the degree of improvement in progenitor production (Miedema; Nielsen).

Another indicator that progenitor cell production is impaired during untreated HIV-infection is the observation that lymphocyte counts, white blood cell counts, neutrophil and platelet counts all increase at the same time, and at a similar rate to CD4 counts after commencing HAART (McCune).

Another theory, proposed by Haynes Sheppard, is that CD4 depletion in HIV disease is a consequence of a confused immune system. According to Sheppard's model, the immune system mistakes HIV virions for CD4 cells because HIV is able to mimic the CD4 receptor, and so decreases the rate of CD4 cell replacement in order to maintain homeostasis (a stable level of CD4 cells). However, this model assumes a substantial increase in viral load over time, a trend questioned by the findings of the Royal Free Hospital Haemophiliac Cohort. This study found that most people followed had viral load changes of less than 1 log10 copies/ml over 10 years (Sabin).

Other researchers have also found that there is little difference in CD4 turnover between HIV-negative and HIV-infected but asymptomatic individuals. However, CD8+ cells do turn over faster in HIV-infected people. There is also evidence from a number of different studies that CD8 cells migrate from the lymphoid tissue in response to anti-retroviral therapy, and that anti-retroviral therapy stimulates proliferation of CD4 and CD8 cells even in the absence of HIV-infection.

References


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