Chapter 14

HIV, SIV and the Pathogenesis of AIDS
Lentiviruses

- HIV is a member of the lentiviruses.
- In common with all retroviruses, lentiviruses have 3 major genetic loci — *gag, pol, env* — that encode core proteins, reverse transcriptase and integrase, and envelope proteins.
HIV – has distinguishing features from non-lenti retroviruses:

1. HIV possesses six accessory genes that encode non-structural proteins. Two of these genes (\textit{tat} and \textit{rev}) are required for replication in cell cultures. Four (\textit{vpr}, \textit{vpu}, \textit{vif}, \textit{nef}) accessory genes are not absolutely necessary for replication in cell culture systems but at least some (such as \textit{nef}) are required for full virulence in vivo (Figure 14.1).

2. The viral attachment protein (the surface envelope protein, known as SU or gp120) binds to the CD4 molecule that is found on the CD4+ subset of T lymphocytes and monocytoïd cells (macrophages, microglia and dendritic cells) and this determines its cellular host range.
HIV – has distinguishing features from non-lenti retroviruses:

3. HIV can replicate in non-dividing (as well as dividing) cells in contrast to other retroviruses that replicate only in dividing cells. In the case of lentiviruses, the pre-integration complex of reverse transcribed viral DNA and proteins can be imported across the nuclear envelope, permitting infection of non-dividing cells.

4. HIV causes lifelong infections that are associated with a number of chronic diseases, including AIDS, but they do not encode oncogenes.

5. HIV is a strictly exogenous virus and host genomes do not include copies of HIV sequences.
Virus-cell interactions

- Entry of HIV into host cells is a multistep process involving a primary receptor and co-receptor.
- In all cases, the primary receptor is CD4.
- The co-receptor is one of several members of the chemokine receptor family – CCR5 and CXCR4.
- CCR5 is found on CD4+ T cells and macrophages.
- CXCR4 is expressed on CD4+ T cells but at very low levels on macrophages.
Virus-cell interactions:
HIV strains vary in ability to bind to co-receptors

- Viruses isolated from patients may be roughly classified into 3 groups:

1. those that utilize mainly CCR5 (often called R5 viruses)
2. those that utilize mainly CXCR4 (X4 viruses)
3. those that utilize both CCR5 and CXCR4 (R5X4, or ‘dual-tropic’viruses).
HIV entry is a multi-step process

**Figure 14.2** Entry of HIV showing a hypothetical reconstruction of stepwise conformational changes in the SU (gp120) and TM (gp41) proteins. The gp120 protein binds to the CD4 receptor. Binding to CD4 triggers a conformational change (A) that leads to major conformational change (B), this one in the TM (gp41) protein, which unfolds to expose and insert the fusion sequence at its N terminus into the plasma membrane of the cell, producing the pre-hairpin intermediate. In a third conformational change (C), helices in the N and C domains of gp41 associate, producing the hairpin configuration that brings the viral envelope and the plasma membrane into close approximation. Finally, the two membranes fuse, leaving gp41 on the external surface. After Chan DC, Kim PS. HIV entry and its inhibition. *Cell* 1998, 93: 681–684, with permission.
HIV replication & host cell response

• HIV-1 replicates slowly in permissive cells.
• If infected lymphocyte is actively dividing, then viral replication proceeds at a maximal rate.
• If infected T cells is resting, provirus can enter the nucleus & integrate but remains latent until T cell divides & then HIV replication proceeds.
HIV replication & host cell response

- Differentiated cell types – circulating monocytes, tissue macrophages, brain microglia and DCs – are derived from monocyte precursors in bone-marrow.
- Each of these cells plays a role in pathogenesis of HIV infection and some are permissive for HIV.
- Mature DCs are able to bind, sequester & conserve infectious virions at extracellular surface or within endosomes.
- Immature DCs express CCR5 and support low levels of HIV replication.
Cell killing in vitro

• HIV varies in its ability to cause cell killing in permissive cells – depending both on cell type and viral strain.
• Replication in T cell cultures results in varied cytopathology based on replication rate of virus and co-receptor usage.
• HIV replication in macrophage cultures is typically low and cultures can remain viable for weeks.
• Death of T cells is usually due to apoptosis initiated by the tat protein. Alternatively, death can arise through syncytium formation.
Cell killing in vivo

• Immune-mediated destruction of infected cells is known to occur in vivo.
• HIV-infected patients mount a cellular immune response in which CD8 T cells recognize and lyze cells that present viral peptides via class I.
• During clinical latency, only a small % of CD4 T cells are infected at any time suggesting other mechanisms involved in C4 T cell depletion.
Transmission, portal of entry and sequential spread of virus

HIV is transmitted by 3 major routes:
1) Sexual contact (accounting for >90% of infections worldwide)
2) Mother to child
3) Blood/blood products

SIV studies have provided important insight into how HIV may be transmitted via sexual contact.

Experimental vaginal infection of SIV reveals first detection of the virus in submucosal/lymphoid tissue primarily in resting CD4+ T cells.

Within a few days, local lymphoid tissue is heavily infected and virus begins to spread to other lymph nodes.
Transmission, portal of entry and sequential spread of virus

FIGURE 14.4 Virus replication and depletion of CD4+ lymphocytes in the GALT of monkeys during acute SIV infection. Rhesus macaques were infected with an R5 SIV strain (mac251) and infection was followed in the colonic lamina propria (submucosal layer). There was a drastic loss of CD4+ lymphocytes associated with an acute round of SIV replication in CD4+ T cells, most of which were ‘resting’ since they were negative for surface markers of proliferation or activation (such as Ki67). After Li Q, Duan L, Estes JD et al. Peak SIV replication in resting memory CD4+ T cells depletes gut lamina propria CD4+ T cells. Nature 2005, 434: 1148–1152, with permission.

Greatest loss of CD4+ T cells is in GALT (gastrointestinal-associated lymphoid tissue), particularly following infection with R5 virus. Naïve T cells in GALT are CCR5- whereas memory T cells are CCR5+ rendering them susceptible to viral infection.
Viremia, cell counts and incubation period

HIV produces a viremia that persists throughout lifetime and can be used to monitor the course of infection. Within the blood, HIV is present both with infected cells and as free infectious virus in plasma. Another useful surrogate for disease course is the concentration of CD4+ T cells in blood which is inversely related to virus titer and a harbinger for functional loss of immune responses during clinical AIDS.
Viremia, cell counts and incubation period

- In absences of antiretroviral therapy, there is an acute phase of infection with high titer viremia followed by a subclinical phase of modest levels of viremia – can last 1 -> 20 years before death, followed by a phase of clinical AIDS that lasts 1-4 years before death.
- During acute infection, a mononucleosis-like syndrome occurs accompanied by a peak in viremia and an acute drop in CD4+ T cells in blood.
- This is followed by induction of an immune response which dampens the infection and is associated with a dramatic drop in blood virus.
- However, infection is never completely cleared and viremia usually stabilizes 4-6 months after infection at a level often called the virus “setpoint”.

Viremia, cell counts and incubation period

Outcome of infection is related to virus setpoint. In cohort of infected patients, 90% of quartile with highest setpoints progress to AIDS in 5 years while <10% of quartile with lowest setpoint has developed AIDS in that time. Patients with slowest progression are often dubbed long-term non-progressors, generally defined as subjects who are AIDS-free 10 years after infection.
Opportunistic infections and neoplasms

- Drop in CD4+ T cells counts below a critical threshold (200-300 cells/μl blood) is often accompanied by rise in virus set-point signaling the advent of AIDS-defining illnesses.
- Constitutional symptoms include fever, fatigue, malaise, lymphadenopathy, GI symptoms, and oral candidiasis.
- Infections are caused by wide spectrum of parasites including protozoa, fungi, bacteria, and viruses.
- In addition, increased risk of neoplasms also occur including Burkitt’s lymphoma, cervical carcinoma, and Kaposi’s sarcoma.
Immune response to HIV

Most patients develop detectable Abs against HIV-1 within 2 months of infection, with highest reactivity against gag and env. Early neutralizing responses are often narrow but will broaden gradually over several years. Weak neutralizing responses are likely due to structure of the env protein in which the binding site to CD4 are heavily glycosylated making ab binding difficult. Only a few broadly neutralizing Abs have been identified:

1) CD4 binding site on gp120
2) Glycosylation sites on gp120
3) CD4-induced site on gp120
4) Base of gp41
Immune response to HIV

FIGURE 14.8 Average immune response of rhesus macaques following infection with SIV, where virus strain, dose, route and time of infection can be controlled. In this example, SIV infection progresses relatively slowly and the pattern of responses appears to be similar to HIV infection of humans. Virus-specific CD8+ effector T cells were measured in an ELISPOT assay and are shown as a per cent of total T cells (CD3+ cells). Anti-SIV IgG antibody was measured in an ELISA but the titer of neutralizing antibody (not shown) would be minimal. After Gould PJ, Watkins DI. HIV and SIV CTL escape: implication for vaccine design. *Nature Reviews Immunology* 2004, 4: 630–640, with permission.
CD8+ T cell responses are critical in protection against SIV infection.

**FIGURE 14.9** The cellular immune response plays an important role in the control of SIV infection. Monkeys infected with dual-tropic virulent SIVmac for >9 months had established stable virus setpoints. They were then treated with a potent antibody against CD8, which reduced the level of CD8 T lymphocytes in the blood by >99%. Data on two animals demonstrate the rise in viremia level during the period of immunosuppression and the reconstitution of immune control when immunosuppressive treatment was terminated. The effect of treatment is more pronounced in monkey A with initially lower viremia. After Schnitz JE, Kuroda MJ, Santra S et al. Control of viremia in simian immunodeficiency virus infection by CD8+ lymphocytes. *Science* 1999, 283: 857–860, with permission.
Virus turnover – HAART
(highly active antiretroviral therapy)

Following commencing HAART, there is a dramatic drop in plasma viremia that can be divided into 3 phases: rapid drop of ~100 fold over first 10 days due to interruption of most cell-to-cell spread of virus and die off of infected activated T cells; slower decrease of ~10-fold over 2 months reflecting death of cells with a longer life (probably macrophages); and a plateau that may be below the level of detection but reflects indefinite persistence of residual latent virus.
Viral phenotypes

Even years of HAART, HIV-1 persists. Evidence for the persistence of a low level of active replication is:

1. the dramatic re-appearance of viremia if HAART is terminated
2. so-called ‘blips’ (transitory appearance of low levels of replicating virus in the blood) in patients who are under long-term HAART
3. mutations in virus sampled at intervals during prolonged HAART, which are seen in some patients
4. the presence of episomal cDNA intermediates that are labile products indicative of active viral replication.
Viral phenotypes

• Majority of viruses obtained early in HIV-1 infection are R5 (macrophage tropic).
• This is probably due to expression of R5 on resting T cells while CXCR4 is minimal.
• Another factor is differential ability of epithelial or dendritic cells to passively capture more R5 viruses than X4 viruses.
Viral phenotypes

**FIGURE 14.12** The virulence of SIV can increase during the course of long-term infection. In this example, macaques (*M. nemestrina*) were infected with a cloned macrophage-tropic strain of SIV (Mne clone 8) and virus was isolated at intervals during infection. This figure compares the inoculated virus and a representative late isolate that were used to infect a new group of macaques (one animal per group is shown) and indicates that the late isolate was more virulent than the original clone. After Kimata JT, Kuller L, Anderson DB, Dailey P, Overbaugh J. Emerging cytopathic and antigenic simian immunodeficiency virus variants influence AIDS progression. *Nature Medicine* 1999, 5: 535–541, with permission.
Pathogenesis of macrophage-tropic and T cell tropic viruses

**FIGURE 14.13** Macrophage-tropic and T lymphocyte-tropic viruses differ in their pathogenesis. This figure compares X4 (T-tropic) and R5 (M-tropic) strains of SHIV (simian human immunodeficiency virus). Upper panel: both viruses produced similar levels of viremia. Lower panel: T-tropic SHIV reduced circulating CD4+ T lymphocytes, while M-tropic SHIV did not reduce. The T-tropic virus infected mainly lymph nodes while the M-tropic virus depleted GALT (gastrointestinal-associated lymphoid tissue). After Harouse JM, Gettle A, Tan RCH, Blanchard J, Cheng-Mayer C. Distinct pathogenic sequelae in rhesus macaques infected with CCR5 or CXCR4-tropic strains of SHIV. *Science*, 1999, 284: 816–881, with permission.
Genetic determinants of susceptibility

<table>
<thead>
<tr>
<th>Genetic locus</th>
<th>Genetic context</th>
<th>Biological effect</th>
<th>Influence on progression to AIDS</th>
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<tbody>
<tr>
<td>CCR5</td>
<td>Homozygous</td>
<td>Δ32 mutation in CCR5 abrogates or reduces CCR5 expression</td>
<td>Protects virus infection</td>
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<td></td>
<td>Heterozygous</td>
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<td>Retards progression</td>
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<tr>
<td>CCR5</td>
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<td>P1 mutation in promoter for CCR5</td>
<td>Accelerates progression</td>
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<tr>
<td>CCR2</td>
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<tr>
<td>CX3CR1</td>
<td>1249 mutation Homozygous</td>
<td>Mutation reduces chemokine binding</td>
<td>Accelerates progression</td>
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<tr>
<td>CCL3L1</td>
<td>Low number of gene duplications</td>
<td>Chemokine ligand for CCR5</td>
<td>Accelerates progression</td>
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<td>Presents peptides with broad HIV representation</td>
<td>Retards progression</td>
</tr>
<tr>
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<td>Homozygous</td>
<td>Presents peptide which is a conserved immunodominant epitope that is under structural constraint</td>
<td>Retards progression</td>
</tr>
<tr>
<td>HLA-A, -B, -C</td>
<td>Homozygous</td>
<td>Reduces number of peptides presented (HLA-B mainly)</td>
<td>Accelerates progression</td>
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**TABLE 14.4** Mutations that influence susceptibility to HIV infection or the rate of progression to AIDS