AIDS RESEARCH AND HUMAN RETROVIRUSES
Volume 20, Number 1, 2004, pp. 111–126

Mini-Review

The CCR5 and CXCR4 Coreceptors—Central to Understanding the Transmission and Pathogenesis of Human Immunodeficiency Virus Type 1 Infection

JOHN P. MOORE,[1] SCOTT G. KITCHEN,[2] PAVEL PUGACH,[1] and JEROME A. ZACK[2]

ABSTRACT

In this review, we will discuss what is known, what is suspected, and what still remains obscure about the central role played by coreceptor expression and usage in the transmission and pathogenic consequences of human immunodeficiency virus type 1 (HIV-1) infection. An emphasis will be on the HIV-1 phenotypic variants that are defined by their usage of the CCR5 or CXCR4 coreceptors, and how the different cellular tropism of these variants influences how and where HIV-1 replicates *in vivo*. We will also review what might happen when coreceptor antagonists are used clinically to treat HIV-1 infection.

INTRODUCTION

The independent discoveries in late 1995 by Lusso and Gallo that CC-chemokines could inhibit HIV-1 replication, and by Berger in early 1996 that the CXC-chemokine receptor CXCR4 was the long sought-after coreceptor for some strains of HIV-1, opened up an entirely new area of AIDS research.[1,2] Soon it was shown that the counterpart to CXCR4 for the most commonly transmitted strains of HIV-1 was the CC-chemokine receptor CCR5, a receptor for the same chemokines that had been shown to be inhibitors of HIV-1 replication only a few months earlier.[3–7] Moreover, the complete absence of CCR5 from some humans (those homozygous for the defective, Δ32-CCR5 allele) was found to be strongly protective against HIV-1 infection *in vitro* and *in vivo*, while decreased CCR5 expression caused by heterozygosity for the Δ32-CCR5 allele reduced the rate of disease progression in HIV-1-infected people.[8–11] In the 7 years since these early, seminal observations, the central role that coreceptor expression and usage play in HIV-1 pathogenesis has become increasingly obvious.

In this article, we will focus on the role played in pathogenesis and transmission by the HIV-1 phenotypic variants that are defined by their usage of CCR5 or CXCR4. It is not our intent to rigorously review every aspect of the relevant literature that already exists in abundance. For example, there are several, complex ways in which phenotypic variants can affect how the human immune system responds to HIV-1 infection that we will not discuss. The complexities of all the possible interactions between HIV-1 and the many cell types of the immune system are also beyond the scope of this article. Instead, a recent and thorough review by Douek *et al.*[12] should be consulted. HIV-1 infection of the brain will also be ignored, since this organ represents *terra incognita* for the senior authors. Instead, we will highlight some specific topics that we believe merit further discussion.

CORECEPTORS: THE BASICS

The two coreceptors that are the most relevant to HIV-1 replication *in vivo* are CCR5 and CXCR4. More than a dozen other G-protein-coupled receptors can mediate the entry of some HIV-1 strains when they are expressed in transfected cells *in vitro*.[13,14] However, with very rare exceptions,[15–19] these receptors are not used by HIV-1 to enter primary $CD4^+$ cells *in vitro*, and probably not *in vivo*. Thus, HIV-1 replication in primary cells *in vitro* is usually completely blocked by inhibitors specific for CCR5 or CXCR4, and there is no correlation between disease progression and the use of coreceptors other than

[1]Department of Microbiology and Immunology, Weill Medical College of Cornell University, New York, New York 10021.
[2]Departments of Medicine and of Microbiology, Immunology and Molecular Genetics, UCLA AIDS Institute, David Geffen School of Medicine at UCLA, Los Angeles, California 90095.

CCR5 or CXCR4.[16–18,20–26] *In vivo,* the absence of CCR5 is strongly protective against HIV-1 infection, which is powerful evidence for the paramount importance of this coreceptor for viral replication.[8,9,11] Moreover, so far as is known, the rare individuals that do acquire HIV-1 infection despite their genetic lack of CCR5 expression are infected by strains that use CXCR4 (and sometimes CCR5 *in vitro*), but not other potential coreceptors.[27–30] Thus despite the opportunity to diversify their coreceptor usage, the CXCR4-using strains of HIV-1 do not do so under *in vivo* conditions. For these reasons, only CCR5 and CXCR4 are the foci of drug development aimed at inhibiting HIV-1 entry at the coreceptor binding stage.[31–33] This is not to say that other coreceptors are not, or could not be, relevant to HIV-1 pathogenesis—only that there is at present little evidence in favor of their importance.

The simian immunodeficiency virus (SIV) strains that naturally or experimentally infect macaque monkeys almost never use CXCR4. In contrast, they all use CCR5 and one or more of a wide range of other G-protein-coupled receptors to enter receptor-transfected cells *in vitro.*[13,14,34] There is some evidence that some SIV (and HIV-2) strains can use an unknown coreceptor(s) to enter primary cells *in vitro*, but whether this coreceptor(s) is relevant to viral replication in macaques is not yet clear.[24,35] However, the paramount importance of CCR5 and CXCR4 for HIV-2 replication, and of CCR5 for SIV replication, seems apparent.[24,35–38]

Years before the coreceptors were identified, it was recognized that two major phenotypic variants of HIV-1 existed, and that they had different impacts on the rate of disease progression in infected people.[39,40] In those days, the phenotypic variants were differentiated by their ability to replicate, and form syncytia, in the MT-2 cell line. The viruses that were able to form syncytia in MT-2 cells were named "syncytium-inducing" (SI) viruses, while those that could not were, logically enough, designated "non-syncytium-inducing" (NSI) viruses.[39] The isolation of SI viruses from an infected person was a poor prognostic indicator; the overt appearance of these viruses was associated with an accelerated rate of loss of $CD4^+$ T cells, and a relatively rapid progression to AIDS and death.[40–43] Fortunately, it was also apparent that SI viruses were relatively rarely transmitted (or, more accurately, that they were relatively rarely found during primary and early stage infection).[40,44] Many people who died of AIDS did so with only NSI viruses detectable in their blood. However, in perhaps 50% of infected individuals after about 5 years of infection, SI viruses became detectable.[41,45] The appearance of these viruses became known as the "phenotypic switch" and, as noted above, it heralded a poor prognosis for the patient.

With the identification of the coreceptors, the SI/NSI nomenclature became archaic, once it was realized that SI viruses were able to use CXCR4 (and sometimes CCR5 as well) whereas the NSI viruses used CCR5 only.[20,25] The MT-2 cell assay "worked" and was valuable because MT-2 cells, like most permanent $CD4^+$ T cell lines, expressed CXCR4 but not CCR5. Hence the formation of syncytia in MT-2 cells heralded the presence of CXCR4-using viruses. A new nomenclature based on coreceptor usage was soon introduced; CCR5-using viruses were designated R5, CXCR4-using viruses, X4, and viruses able to use both receptors, R5X4.[46] Some clonal viruses have been proven to be able to use both CCR5 and CXCR4 and properly justify the designation R5X4.[47,48] However, most isolates that replicate on both CCR5-positive and CXCR4-positive cells probably do so because they can contain a mixture of R5 viruses and X4 viruses. A better term for such isolates would be R5 + X4, but since it is technically very difficult to distinguish between a true dual-tropic virus and an isolate containing a mixture of phenotypic variants, we will continue to use the now-conventional term, R5X4.

Just as many humans die of AIDS without the overt acquisition of X4 viruses, so do macaques experimentally infected with SIVmac and related viruses. In this model, there is good evidence that R5 SIV strains can evolve to become more virulent, but without broadening their tropism to gain CXCR4 usage.[49] The same phenomenon has also been reported to occur in some humans.[50]

ARE THERE BLOCKS TO X4 VIRUS TRANSMISSION, OR POSTINFECTION REPLICATION BLOCKS?

As noted above, X4 viruses rarely predominate in the early years of HIV-1 infection. Does this mean that there is a block to the transmission of X4 viruses that occurs at, for example, the genital or rectal mucosa, perhaps due to the lack of CXCR4 expression at or near the sites of virus deposition? It has been argued that R5 viruses are preferentially transmitted because of the patterns of expression of coreceptors and their ligands at mucosal sites after virus deposition during sexual intercourse.[51,52]

There are many uncertainties about the mechanism(s) by which HIV-1 is transmitted sexually, and controversies surround the identity of the first infected cells.[50,53] Genital and rectal subepithelia stromal tissues are densely populated with dendritic cells, macrophages, and T cells that express CD4, CCR5, and, to a lesser extent, CXCR4.[54–56] Each of these cell types is, therefore, susceptible to HIV-1 infection. In the macaque model, infection of all three cell types can be detected within 1 hr of the addition of SIV to the macaque vagina, most commonly where the epithelium is abraded to allow the virus better access to the underlying, target-cell-enriched tissues.[54] Immature dendritic cells present in the epithelium express 10-fold more CCR5 than CXCR4,[57] and they selectively replicate R5 strains.[58] The Langerhans cell, a member of the dendritic cell family, has been proposed to play a particularly important role in the early stages of sexual transmission.[51] CXCR4 is usually undetectable on human Langerhans cells, whereas CCR5 is present at low levels on a significant minority of these cells,[59] rendering a low percentage (<5%) susceptible to productive R5 HIV-1 infection *ex vivo.*[60] It has been suggested that the level of CCR5 on Langerhans cells is a major determinant of sexual transmission, and an important factor in the preferential transmission of R5 viruses.[51] The expression of CCR5 but not CXCR4 on intestinal epithelial cells may also be relevant to the preferential transmission of R5 viruses via the rectal route.[52,61] The high levels of the CXCR4 blocking ligand, SDF-1, that are present in the intestinal lumen could also be a factor in suppressing the transmission of X4 viruses.[62]

One model of sexual transmission is, therefore, that CCR5 on target cells within or near the sexual mucosa acts as a local

"gatekeeper." According to this model, the level of CCR5 expression has a major influence on the efficiency of HIV-1 transmission, and the more abundant local expression of CCR5 compared to CXCR4 explains the R5 phenotype of the most commonly transmitted viruses.

There may, however, be a different or additional explanation for the apparent block to the sexual transmission of X4 viruses. The dominance of R5 viruses early after infection is true in both adults and children, so any block to the transmission of X4 viruses must be relevant to not just sexual, but also vertical[63–70] and intravenous[71,72] infection routes. Moreover, epidemiological analyses do not suggest that the route of infection is a major influence on the rate of disease progression.[72,73] This would not be the case if X4 viruses generally dominated the infections of those infected directly via the blood; in that scenario, rapid progression to AIDS and death would be more common in intravenous drug users (IVDU) and hemophilia cohorts than is, in fact, the case.[71–73] Furthermore, X4 SHIVs are perfectly infectious for macaques after atraumatic vaginal deposition,[74] again arguing against the presence of an insuperable physical block to X4 virus transmission.

If X4 viruses can be successfully transmitted, why do they not dominate the overall pool of virus that becomes amplified during primary HIV-1 infection, a stage at which R5 viruses almost always dominate? After all, *in vitro*, X4 viruses have more cellular targets in lymphoid cell cultures and in lymphoid tissue blocks,[75] and there are many more $CXCR4^+$ $CD4^+$ T cells in the blood than there are $CCR5^+$ $CD4^+$ T cells.[12,76] We will discuss this critical issue at more length later in the article. However, in the setting of acute infection, a key point may be that, *in vivo*, R5 viruses have a selective advantage over X4 strains due to their preferential tropism for the dendritic cell in the context of dendritic cell–T cell conjugates.[58,60,77] Members of the dendritic cell family can bind HIV-1 particles efficiently via CCR5- and CXCR4-independent mechanisms, then internalize the virus into intracellular endocytic vacuoles without being productively infected themselves.[79,80] Dendritic cells are natural sentinel cells that sample incoming pathogens or their antigens at, for example, the vaginal epithelium, transport them to regional lymph nodes, and there present them to T and B cells for the initiation of adaptive immune responses.[81] Unfortunately, after internalization, HIV-1 can remain infectious within a dendritic cell for up to 5 days.[79,80] When the dendritic cell interacts with, and activates, $CD4^+$ T cells within T cell-rich regions of the lymph nodes, the virus is in a perfect environment for its rapid and efficient amplification, with R5 viruses dominating because of the expression of CCR5 on activated $CD4^+$ T cells.[80–82] According to this model, then, it is not the expression of CCR5 on the transporting dendritic cell that determines the preferential transmission of R5 viruses, it is the advantage provided to R5 virus replication by dendritic cell–T cell conjugates within lymphoid tissues. The selective amplification of R5 viruses within the lymphoid tissue would presumably operate irrespective of the route of transmission, be it rectal, vaginal, vertical, or intravenous, for it should not matter how the virus reaches the lymph node. The same factors would apply also to HIV-1 infection of tissue macrophages, because most R5 viruses replicate more efficiently than do most X4 strains in these cells.[34,83]

Whether their advantage is gained during the early events of transmission or later during the initial amplification stage within the first infected lymph node, R5 viruses then go on to dominate the generally homogeneous quasispecies pool that is present during primary infection. There are, perhaps, no specific, insurmountable obstacles to the transmission of X4 viruses in any setting; only a limitation to their relative replication capacity once viral dissemination to the most active sites of replication has occurred. Two anecdotal transmission cases, one by intramuscular inoculation, the other by needle-stick, are quite revealing.[84,85] In both examples, X4 viruses were identifiable in the donor, and the first samplings of blood from the recipients suggested that X4 viruses had, in fact, been successfully transmitted. However, as primary infection progressed, R5 viruses came to dominate the pool of viruses in the blood of the recipients, while the X4 viruses became a minor, and eventually almost invisible species.[84,85] Although several explanations of these phenomena are possible, the one that we favor, as outlined below, is that R5 viruses simply outcompete their X4 counterparts in the race to replicate *in vivo*, once they are successfully transmitted. It may even be that X4 viruses evolve to become R5 strains under the conditions of primary infection.

WHERE DOES HIV-1 REPLICATION OCCUR *IN VIVO*, AND WHY? THE ROLE OF GALT

There is an increasing appreciation of the central role that the gut-associated lymphoid tissue (GALT) plays in the replication of HIV-1 *in vivo*, and hence in the pathogenesis of HIV-1 infection overall. This important issue has been reviewed in detail by Veazey *et al.*[86] The GALT contains over half the human body's total of T-lymphocytes, and these cells tend to be more activated than T cells of the peripheral blood—a reflection of the role played by GALT in combating intestinal and food-borne pathogens. Moreover, the T cells of the GALT are not in rapid equilibrium with their counterparts in the peripheral blood, so that events such as viral replication that affect one organ do not necessarily have the same effect on the other.[86] This is nicely revealed by the outcome of infection of macaques with SHIV-162P.[87] That virus caused rapid and extensive depletion of $CD4^+$ T cells from GALT, but there was no significant loss of $CD4^+$ T cells from the blood at the time the GALT was being destroyed; only later did peripheral $CD4^+$ T cell counts decline. Thus an apparently minimally pathogenic viral infection, judged by what is happening in the blood, was actually an infection that was having a severe impact on a major lymphoid organ.[87]

It is notable that SHIV-162P is an R5 virus. In contrast, an X4 strain, SHIV-SF33A.2, depleted circulating CD4 T cells, while sparing the T cells in the GALT.[87] A similar situation occurs in macaques infected with SIV, which also causes a rapid depletion of $CD4^+$ T cells that, initially, is seen exclusively in the GALT.[88,89] The use of CCR5 by SIV is highly relevant in this context, once it is appreciated that the majority of the GALT $CD4^+$ T cells are $CCR5^+$, compared with only 5% in peripheral blood.[90–92] The high levels of expression of the chemokine ligands for CCR5 that are present in the GALT may direct the trafficking of memory $CCR5^+$ T-lymphocytes to this tissue.[93] The $CCR5^+$ subset of $CD4^+$ T cells is known to be preferentially depleted soon after infection with HIV-1 or SIV, in par-

ticular at mucosal sites.[55,92] Furthermore, CXCR4 expression in the GALT is inherently low.[87,90] The localized production of the SDF-1 ligand for CXCR4 may further occlude this coreceptor and prevent its use for HIV-1 entry, both in mucosal tissues, and elsewhere.[62,94] One significant implication of this pattern of coreceptor expression in the GALT will be discussed below in the context of the R5 to X4 phenotypic switch and why it does and does not occur.

Overall, it can be argued that HIV-1 and SIV infection might be considered as predominantly an infection of the GALT, at least in the early years of infection when R5 strains are the most abundant.[86] This is not to say that R5 virus replication does not occur elsewhere in the body, such as the peripheral lymph nodes; instead, we argue (as have others[86]) that virus production in the GALT may initially dwarf production in other lymphoid tissues. Measuring CCR5 expression levels in the blood may allow only an imprecise and indirect understanding of factors that influence the replication of R5 viruses in environments such as the GALT. On the one hand, the reduction in CCR5 expression in the blood caused by CCR5-Δ32 heterozygosity is likely to be predictive of reductions in CCR5 expression elsewhere, because the genetic lesion will have the same effect in all cells. But conversely, the pronounced effects that CCR5 promoter polymorphisms can have on HIV-1 transmission and disease progression rates may be mediated by influences on CCR5 mRNA transcription that are cell-type specific or otherwise affected by the local milieu within a lymphoid tissue.[95–102] In other words, measurements of CCR5 expression on $CD4^+$ T cells derived from the peripheral blood may not reveal every influence that a promoter polymorphism can have on CCR5 protein expression in all tissues relevant to HIV-1 replication *in vivo*.

A similar explanation may account for the apparent paradox that HIV-1-infected infants have very high levels of plasma viremia, usually involving R5 viruses[103–106] yet CCR5 expression is very low on $CD4^+$ T cells derived from the blood of infants (cord blood).[107] Again, this speaks to the limitations of relying on measurements of CCR5 expression that involve only cells from the blood. Perhaps in infants, as in adults, the more relevant levels of CCR5 expression are those present in the GALT. For understandable reasons, measurements of CCR5 expression levels, and the extent of HIV-1 replication, in the GALT of young infants have not been performed. Perhaps such studies could be done in the macaque models.

A final point is that the abundance of activated, $CD4^+$ $CCR5^+$ T cells in GALT may be an important factor when considering why receptive anal intercourse is such a high-risk sexual practice for HIV-1 transmission,[61] particularly when the high levels of the HIV-1 attachment factor, DC-SIGN, on the rectal mucosa are also taken into account.[108]

WHERE DOES HIV-1 REPLICATION OCCUR *IN VIVO*, AND WHY? THE ROLE OF THE THYMUS

It has been apparent ever since AIDS was first identified as a clinical syndrome in the early 1980s that the loss of $CD4^+$ T cells was its central feature.[109,110] Furthermore, HIV-1 is a cytopathic virus *in vitro*, causing massive destruction of $CD4^+$ T cells. This is particularly true of the X4 viruses that were the first HIV-1 isolates to be extensively studied. Of course, these viruses have the X4 phenotype, because early isolations tended to be from symptomatic AIDS patients, and because permanent T cell lines were used for the isolations.[12,111,112] It is, therefore, not remotely controversial to state that HIV-1 kills T cells. However, while T cell killing may be necessary for AIDS to occur, it is highly unlikely to be sufficient.[113,114] The immune system has abundant regenerative capacity that it can use to repair some of the damage caused by T cell destruction, or at least prolong the period before the damage becomes fatal. Extremely high levels of replication of SIV variants can occur in naturally infected monkeys (e.g., SIVagm infection of African green monkeys or SIVsm infection of sooty mangabeys) without the onset of immunodeficiency; lentivirus production per se need not be lethal to the host.[114–117] For humans to develop AIDS, it is becoming ever-more accepted that an important contributory factor must be viral impairment of the regenerative capacity of the immune system.[12,118–124] This capacity is finite and it is age dependent, in that older people develop the symptoms of AIDS much sooner than younger people do.[121,125,126] It is hard to imagine that T cell destruction is a function of patient age, but both T cell replenishment capacity[126,127] and immune function[124] certainly are. The impressive $CD4^+$ T cell recovery in children treated with HAART, compared to the lesser response in adults, speaks for itself.[128]

The major organ of naive T cell production is the thymus, long thought to be dormant in adults but now known not to be. The central role of the thymus in HIV-1 pathogenesis has been reviewed by McCune, and will not be discussed at length here.[118] However, a highly relevant issue is the role that HIV-1 phenotypic variants play in impairing thymic function, and in particular the local damage that can be caused to the thymus by X4 viruses that target the majority of developing T cells in the thymus.[129–134] R5 viruses also replicate in the thymus, but they do not appear to cause the same level of destruction of developing T cells as do X4 strains, as discussed further below. Regardless of the strain, destruction of immature thymocytes either by direct infection or by alteration of thymic stromal function will lead to decreased production of naive T cells. If very immature cells are affected, this will result in decreased production of both naive $CD4^+$ and $CD8^+$ lineages, and the body's fight against HIV-1 or other infection may be diminished.

The pattern of CXCR4 and CCR5 expression in the human thymus is an important influence on the destruction of immature T cells, and overall, on the pathogenesis of HIV-1 infection. CXCR4 and CCR5 are independently regulated during T cell development. The modulation of each coreceptor renders $CD4^+$ cells at different stages of development differentially susceptible to HIV-1 infection. A summary of CD4 and coreceptor expression patterns during lymphopoiesis is provided in Figure 1. A low level of CXCR4 expression can be observed on a subset of the earliest T cell progenitor, the $CD34^+$ bone marrow hematopoietic progenitor cell, prior to its entry into the thymus.[129,135] Furthermore, some $CD34^+$ cells express CD4, which renders them infectable, albeit poorly, by HIV-1, at least *in vitro*.[136–138] However, it is unclear as to whether $CD34^+$ cells are infected *in vivo* and whether any such infection might have direct immunological consequences. Although low levels of virus have been reported in the $CD34^+$ progenitor cell popula-

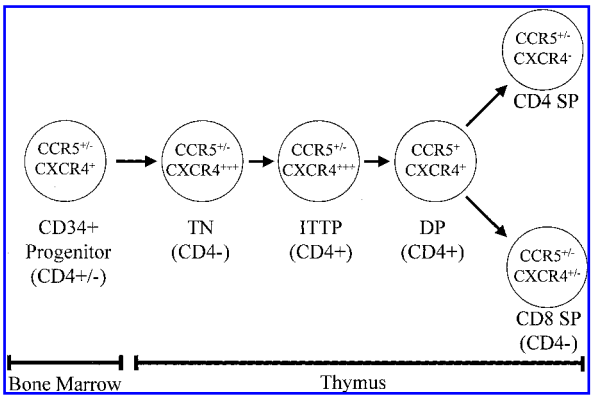

FIG. 1. CXCR4 and CCR5 expression during various stages of lymphoid development. The presence of CD4 on these cells is indicated below each subset. Maturation proceeds from left to right.

tion in a minority of studies, more often these cells have been found to not be infected[139–141] (reviewed in ref. 142). In the fetal lymphoid system, CXCR4 expression is at its greatest in the thymus.[143] The receptor is expressed throughout thymopoiesis and appears to have a functional, SDF-1 activated signaling role in early T cell development.[135] This function of CXCR4 may not be absolutely required: T cells can develop normally in mice engineered to lack CXCR4 expression, although the overall CXCR4 null genotype was developmentally lethal.[144]

CXCR4 is expressed on approximately 60–70% of primary human thymocytes.[145,146] In the thymus, the highest amounts CXCR4 are found on early thymocytes, particularly the $CD3^-CD4^-CD8^-$ [triple negative (TN)], the $CD3^-CD4^{lo}CD8^-$ intrathymic T cell progenitor (ITTP), and the $CD3^+CD4^+CD8^+$ [CD4CD8 double positive (DP)] populations.[129,146,147] CXCR4 is down-regulated during the DP stage of development, more specifically during the transition from the $CD3^{lo}CD4^{lo}CD8^{lo}$ stage to $CD3^{hi}CD4^{hi}CD8^{hi}$ stage.[146,147] Further down-regulation occurs at the cells mature into the $CD3^+CD4^+CD8^-$ and $CD3^+CD4^-CD8^+$ [CD4 or CD8 single positive (SP)] populations.

CCR5 expression is relatively low during T cell development, the converse of what is seen with CXCR4.[129,145,147,148] There is some evidence that CCR5 is expressed on the bone marrow-derived and thymic-resident $CD34^+$ T cell progenitor populations.[129,147,149] In the thymus, CCR5 is generally found on $< 5\%$ of total thymocytes.[129,143,147,148,150] In general, CCR5 appears to be detectable at low levels on a few cells from every thymocyte subset. However, there is little consensus as to which particular subset(s) express(es) the highest levels of CCR5. In contrast to CXCR4, CCR5 is expressed poorly on $CD34^+$ progenitors and ITTPs while it is present on a larger percentage of cells from the more mature populations. The use of different monoclonal antibodies that detect low levels of CCR5 with different efficiencies may explain some of the discrepant results from multiple laboratories. The anti-CCR5 monoclonal antibodies (mAbs) 3A9 and 5C7 appear to cross-react with CCR8, which is also expressed on thymocytes and where it can function as an HIV-1 coreceptor.[15] The 2D7 mAb, however, is specific for CCR5.[150] Cross-reactivity could therefore help explain discrepant results found in different studies. Some of the highest reported CCR5 levels (although still typically $< 5\%$) are present on DP cells.[129,143,147,148] CCR5 is also consistently expressed on a low percentage of thymic CD4 and CD8 SP cells. Within the thymus, while its expression is relatively low compared to CXCR4, CCR5 is present at approximately equal distributions on both the $CXCR4^+$ and $CXCR4^-$ populations.[143] Further, similar to CXCR4 expression, CCR5 expression in the thymus is clearly not obligatory, as humans homozygous for the CCR5-Δ32 mutation produce normal T cells.[151] Furthermore, T cells develop normally in CCR5-deficient mice, although they appear to have some functional differences compared to T cells from wild-type mice.[152] Thus, the differential distribution and expression of CXCR4 and CCR5 during T cell development suggest that different HIV-1 strains may target different cells at different stages of development; in some cases the virus may target the same subset of developing cells (i.e., $CCR5^+/CXCR4^+/CD4^+$ cells). This pattern of tropism could dramatically influence viral pathogenesis within this primary lymphoid compartment.

The expression of both CXCR4 and CD4 at one of the earliest stages of T cell development in the thymus (i.e., ITTPs) renders these cells susceptible to infection and destruction by X4 HIV-1 strains. The different cellular transcriptional and replicative activity of the various thymocyte subsets also influences their susceptibility to productive infection. For example, the highly transcriptionally active DP^{Bright} subset is very vulnerable to infection with X4 strains whereas the less transcriptionally active CD4 SP population is less likely to be productively infected.[153,154] Thus, multiple factors render thymocytes more or less permissive to HIV-1 infection. When thymocytes are infected with X4 strains either *in vitro* or in the SCID-hu mouse, the immature CD4 subset is depleted first, then the mature subsets.[146,155–160] *In vitro* and in the SCID-hu mouse model, X4 HIV-1 isolates primarily deplete the immature $CD4^+$ population that expresses the highest level of CXCR4.[146,161] In the SCID-hu mouse, these $CXCR4^+$ cells are regenerated following antiretroviral therapy.[161,162] Hence X4 viruses do not permanently alter the ability of progenitor cells to express CXCR4 during development, even if the cells had previously been exposed to the virus.

R5 viruses are less cytopathic than X4 strains in cultured thymocytes, and they replicate to lower titers *in vivo* in the SCID-hu mouse.[130,132,148,155,158,163,164] Children from whom X4 viruses can be isolated have a greater impairment of thymic function than those infected with R5 viruses; their ability to generate new $CD4^+$ T cells is much diminished.[133] The relatively high levels of CXCR4 but low levels of CCR5 in the thymus condemn this organ to be particularly vulnerable to X4 strains. However, viruses with *env* gene sequences suggestive of the R5 phenotype have been identified in thymuses from HIV-1-infected individuals at autopsy.[165] Furthermore, R5 viruses can cause significant cellular alterations in infected thymocytes, and the outright destruction of these cells *in vivo* and *in vitro*.[130,131,158,164,165]

Infection and involution of the thymus occurs as macaques infected with SIV (an R5 virus) progress to disease.[166–168] The majority of SIV-infected cells in the thymus are found in the medullary region,[167,168] where the mature thymocytes reside and where CCR5-expressing cells are present in the greatest

amount.[169] That CCR5 is expressed at lower levels than CXCR4 in the thymus may explain the decreased pathogenesis of R5 strains in thymocytes *in vivo*, as the extent of infection correlates with CCR5 expression level.[148] Although the mechanism for viral spread into new target cells is unclear, CCR5 may be transiently expressed by thymocytes during T cell development, which could then render these cells susceptible to R5 viruses. Indeed, it was shown recently that HIV-1 expands its tropism within the thymus by up-regulating CCR5.[170] This seems to occur through induction of interferon-alpha (IFN-α) by intrathymic predendritic cells that have responded to HIV-1 infection.[170] This induction of CCR5 was restricted to the TN and ITTP populations, which rendered these progenitors susceptible to HIV-1 infection and depletion. Hence this mechanism could explain the pathogenic effects of R5 strains in the human thymus, despite the low levels of CCR5 expressed by uninfected thymocytes. Recent studies have shown that the interleukin (IL)-7 cytokine alone[171] or the combination of IL-4 plus IL-7[172] increases the expression of CXCR4 on mature thymocytes cultured *in vitro*, so the cytokine microenvironment may influence coreceptor expression and therefore HIV replication in the thymus. Of further note is that two other components of the T cell arm of the immune response—immunoregulatory $CD161^+$ natural killer (NK) T cells and T cell receptor (TCR)$\gamma\delta^+$ T cells—are also present in the thymus. A significant fraction of the TCR$\gamma\delta^+$ cells expresses CD4 in the thymus, but CD4 expression is lost in the periphery or gut. $CD161^+$ T cells express CD4 both in the thymus and periphery.[173] In the thymus, these cell types both express CXCR4 and CCR5 and are susceptible to infection by multiple HIV-1 strains. Thus, HIV-1 can target diverse subsets of cells within the thymus. By doing so, the virus has a dramatic effect on the development of immature components of the immune system, which may contribute substantially to the immune impairment that is a hallmark of systemic HIV-1 infection.

THE R5 TO X4 PHENOTYPIC SWITCH: WHY DOES IT OCCUR (OR NOT)?

Experimentally, an R5 virus can be converted to an X4 virus by engineering a few (two or three) amino acid changes into the V3 loop of gp120.[174–176] Given the extent of HIV-1 replication *in vivo*[177] and the associated error rate,[178] the conversion of an R5 virus into an X4 strain should occur rapidly, in every infected person. Moreover, *in vitro*, both in cell culture and in lymphoid tissue blocks, X4 viruses usually replicate more rapidly than R5 strains, and have more cellular targets available to them.[12,75,76] Target cell availability is also not a driving force for the phenotypic switch *in vivo*, at least so far as can be determined from measurements of coreceptor expression on T cells of the peripheral blood.[179] Thus, measured after a year of infection, the percentage of $CXCR4^+$ $CD4^+$ T cells in the blood was *inversely* correlated with the later development of X4 viruses, whereas there was no relationship between the percentage of $CCR5^+$ $CD4^+$ T cells and the phenotypic switch.[179] X4 viruses are also more pathogenic *in vitro*, by a variety of mechanisms including direct cell killing and Env-induced apoptosis,[180–183] yet the reduced levels of $CXCR4^+$ $CD4^+$ T cells found in cohort members with advanced disease and X4 variants is not simply due to virus-mediated killing of this T cell subset.[179] HIV-1 infection, of course, causes immune activation that leads to the up-regulation of CCR5 expression on $CD4^+$ T cells.[184,185] Overall, CCR5 and CXCR4 expression levels in the blood do not influence the rate of evolution of X4 variants; the availability of blood $CXCR4^+$ $CD4^+$ T cells does not cause X4 viruses to evolve more rapidly; and the ability to use CXCR4 is not an escape mechanism used by HIV-1 to overcome a limiting number of $CCR5^+$ $CD4^+$ target cells.[179] As noted earlier, however, different forces might influence coreceptor expression levels in other lymphoid organs that cannot readily be sampled in large cohorts.

So why do X4 viruses not dominate HIV-1 pathogenesis in every infected individual, causing AIDS to be a disease of rapid progression? The answer must presumably be that a selection pressure acts against the evolution and dominance of X4 viruses under *in vivo* conditions. It is still unclear whether this pressure has a virological or an immunological basis. Yet it is important to know which of these two possibilities applies, or indeed whether both do. The answer could help resolve a question that has been asked since SI (X4) viruses were first described[39,40]: Is the emergence of X4 viruses the cause or consequence of severe immune system impairment?

As outlined earlier, arguments can be made that R5 viruses have a selective replication advantage under *in vivo* conditions. Hence, the early bias toward R5 viruses *in vivo* might be a fundamental property of the underlying virology of X4 and R5 virus replication. Moreover, the burst size (virions released per infected cell) has been reported to be about an order of magnitude greater for R5 viruses than for X4 viruses, as measured in lymphoid tissue blocks *in vitro*.[186] The $CD4^+$ T cells infected by and releasing R5 viruses were activated memory cells and those producing X4 viruses were resting, naive cells. If the relative burst sizes are similar *in vivo*, the capacity of R5 viruses to outcompete their X4 counterparts over multiple replication cycles is obvious, particularly if much replication occurs in a tissue such as GALT in which CCR5 is abundant, CXCR4 sparse (see above).

There is also evidence that the selection pressure against X4 strains might have an immunological basis, which would imply that the emergence of X4 strains late in infection reflects the cumulative erosion of the suppressive capacity of the immune system. Several mechanisms have been proposed, mostly based on *in vitro* studies.[187–190] Indirect evidence for an immunological component to the X4 suppressive mechanism is the observation that the development of X4 viruses is associated with low $CD4^+$ T cell numbers, and hence with immunological impairment.[179] More direct, but still inferential, support is provided by a recent study in the SHIV-infected rhesus macaque model: an R5 virus (SHIV-162P3) replicates to dominance over a dual-infecting X4 strain (SHIV-33A).[190a] The mechanism appears to involve preferential suppression of the X4 virus by the immune system. It is unlikely that there is any differential effect of neutralizing antibodies on the two SHIVs,[190a] and earlier *in vitro* studies also argue against the involvement of this humoral response.[191,192] Instead, there is evidence based on the use of a depleting anti-CD8 mAb that $CD8^+$ cytotoxic T-lymphocyte (CTL) may be responsible for the suppression of X4 viruses.[190a] One possibility is that the R5 SHIV has a greater propensity than its X4 counterpart for replication

in macrophages, and that these cells constitute a privileged site that is relatively resistant to CTL lysis. There is some independent evidence to support the latter concept.[188] The X4 SHIV, on the other hand, is argued as replicating predominantly in $CD4^+$ T cells, which are efficiently suppressed by CTL activity, at least while the immune system remains intact, so fewer X4 viruses are produced. Determining how general these observations are and how broadly they relate to HIV-1 infection of humans will require more studies, perhaps with additional SHIV or SIV strains. Different primary X4 HIV-1 strains and X4 SHIVs vary greatly in their abilities to replicate efficiently in macrophages, compared to $CD4^+$ T cells.[83,193,194] Hence, even focusing on the above immunological model of preferential X4 virus suppression by $CD8^+$ CTL, the likelihood of the R5 to X4 phenotypic switch might have a strong virological influence. And of course, any host genetic factors that affect the potency of the CTL response[195] would also have a significant impact on what happens, and when.

Adding to the complexity, environmental influences might also be important. Viruses with the X4 phenotype are infrequently found in individuals with infections with subtype C viruses, strains that are common in sub-Saharan Africa.[196,197] The apparent preference for the R5 phenotype among subtype C strains might conceivably account for the relatively rapid heterosexual spread of HIV-1 in southern Africa.[197] One possibility is that the envelope glycoproteins of subtype C viruses are in some fundamental way different from those of the subtype B strains that are the most common in North America and Europe, disfavoring the R5 to X4 phenotypic switch on protein structure grounds. However, we doubt this is the explanation; subtype C X4 viruses certainly exist.[196,198,199] Studies on cohorts that contain individuals infected with several different HIV-1 subtype yield little or no evidence that the subtype of the infecting virus is an important variable in disease progression rates.[197,200–202] *In vitro*, subtype C R5 viruses are actually *less* fit than B R5 strains.[202a] Instead, we favor the argument that the local environment is a more important influence on the spread of subtype C strains, and that this is influenced by coreceptor-expression patterns, not by any fundamental virological property of HIV-1 subtype C strains.

Supporting this hypothesis is a study that measured CCR5 protein levels on blood lymphocytes of Italians and Ugandans resident in Italy and Uganda.[203,204] CCR5 expression varied significantly, but showed no correlation with the racial grouping of the blood donor; instead, the highest CCR5 levels were found in Ugandan residents, be they of Italian or Ugandan extraction; conversely, Italian residents had relatively low CCR5 expression.[203] These observations were attributed to the relatively high levels of immune system activation found in Ugandan residents, particularly activation caused by parasitic infections.[203,204] Immune activation is known to increase CCR5 expression on T cells of the peripheral blood.[76,184,185] Sustained, high-level CCR5 expression, particularly if it also occurred in solid lymphoid tissues, could be an important selection pressure in favor of the retention of the R5 phenotype during HIV-1 infections in the African environment.

Intuitively, one might suppose that chronically elevated CCR5 levels might accelerate disease progression since a reduction in expression caused by Δ32-CCR5 heterozygosity is associated with a decreased rate of progression.[9] However, if there is any selection against X4 strains caused by high CCR5 expression, this would work in the opposite direction, i.e., to slow the rate of progression. The complexities are nicely illustrated by the cohort-based studies of van Rij *et al.* summarized above.[179] Overall, the net effect on disease progression of elevating CCR5 expression is hard to predict without taking into account other variables, although higher CCR5 expression, local or systemic, is likely to act to increase the probability of a successful transmission and amplification of HIV-1 in the first place. It should also be noted that the elevated CCR5 expression in the blood of African residents may actually be a marker of what is happening elsewhere in the body. For example, intestinal parasitic infections may induce the up-regulation of CCR5 in the GALT, and this may be a more important influence on virus replication and disease progression than anything that is measured in the blood (see above). We are not aware of any comparative studies of CCR5 expression in the genital or rectal mucosa, or the GALT, of African and European/North American residents, but perhaps these experiments should be done to gain further insight into why HIV-1 spreads so rapidly in sub-Saharan Africa.

Might methodological considerations also be relevant? Although X4 subtype C infections appear to be rare, X4 viruses are much more common when subtype B viruses are the infecting strains, as is the case in North America and Europe.[39–41] But how common are X4 viruses? Do most infected people have a low-level X4 HIV-1 infection or are some people completely "X4 free"? One issue is techniques used to detect and quantify X4 and R5 viruses. In the early years of AIDS research, HIV-1 isolations were performed using transformed $CD4^+$ T cell lines, which are usually CCR5 negative but CXCR4 positive. Not surprisingly, the early isolates were X4 or, in some cases, R5X4 viruses, because of the obvious bias caused by the cell substrate. When peripheral blood mononuclear cells (PBMC) became the cell of choice for virus isolations, it was (and still is) standard procedure to activate them with phytohemagglutinin (PHA) plus IL-2, which up-regulates CCR5 expression.[76] Consequently, there is probably a bias in today's standard isolation procedures toward finding R5 viruses. Other phenotypic methods using cell lines expressing CCR5 or CXCR4 tend to detect the presence of R5 or X4 viruses in plasma, without truly quantifying their relative abundance[205]; the same concern applies to the MT-2 cell assay for detecting X4 viruses. At best, all these cell-based assays are semiquantitative when it comes to estimating the relative amounts of R5 and X4 viruses in the plasma. Moreover it is entirely possible that X4 viruses will sometimes not be detected if they are present only at low levels relative to R5 viruses, or are present only in other anatomical sites, such as the thymus (see above).

An alternative method to quantify R5 and X4 viruses is a genetic procedure based on the detection of "signature" amino acids in the V3 loop of gp120 that are associated with the R5 or X4 phenotype.[174,206] Although the genetic methods can be quite laborious and the correlation between the presence of signature residues and the X4 phenotype is probably not 100%, they may be more reliable for quantification purposes than their cell-based counterparts. Using the genetic method, Shankarappa *et al.* identified the presence of X4 viruses (or their genetic

signatures) in all HIV-1-infected individuals involved in the study.[207] Of note is that a variable frequency of X4 viruses was observed throughout the course of infection, so an inability to detect X4 viruses in a patient at a single time point could be a false-negative finding, particularly if sampling were done following a period of viremia decline.

Chimpanzees experimentally infected with an X4 virus do have viral loads that are about 1–2 logs lower than those in animals challenged with an R5 strain.[208] However, the small number of animals involved and the multiple differences between the challenge viruses other than coreceptor usage limit the robustness of any conclusions that could be drawn. Additional, although again still limited, evidence that X4 viruses are rarely produced in large quantities during HIV-1 infection of humans comes from the studies of the few individuals who are homozygous for the defective Δ32-CCR5 allele, yet who become infected with HIV-1. Viral load data from most of the cases are missing, but where it is available, it does suggest that X4-specific viremia is not particularly high.[27,28,209] Despite the moderate-to-low viral loads in these people, the rates of $CD4^+$ T cell loss are high, again emphasizing the particularly destructive effect of the X4 virus on the immune system. CCR5 wild-type individuals infected with high levels of both R5 and X4 viruses, or genuine R5X4 viruses, most likely suffer the worst of both worlds—high levels of virus production and T cell loss caused by the R5 viruses, combined with a relatively modest level of additional viremia and the associated much greater immune system, particularly thymic, impairment caused by the X4 virus component. Hence their rapid progressor status.[40,43,71]

WHAT WILL HAPPEN WHEN CORECEPTOR-SPECIFIC ENTRY INHIBITORS ARE USED IN THE CLINIC?

Specific inhibitors of HIV-1 entry via CCR5 or CXCR4 are now in human clinical trials.[33,210,211,211a] Studies in the SCID mouse models suggest that they will have a beneficial effect on viral load,[211,212] and emerging evidence from human studies is also encouraging.[211a] Of course, a CCR5-specific inhibitor will block only the entry of R5 viruses, and conversely for a CXCR4-specific inhibitor. But will blocking one coreceptor drive the virus toward using the other? More specifically, will inhibiting HIV-1 entry via CCR5 drive the virus to use CXCR4, thereby increasing the prevalence of X4 viruses and exacerbating disease? Perhaps but not necessarily. In most, but not all,[213] studies a reduction in CCR5 expression caused by Δ32-CCR5 heterozygosity does not lead to an increased rate of emergence of X4 variants *in vivo*.[45,189,214,215] Moreover, a low number of $CCR5^+$ $CD4^+$ target cells in the blood does not drive the evolution of X4 variants from R5 viruses.[179] Again, however, the situation in solid lymphoid tissues may not be the same as in the blood.

To a large extent, the above questions can be resolved only by clinical trials under the most carefully monitored conditions, but some *in vitro* studies are relevant. Most studies of the escape pathways used by HIV-1 to avoid coreceptor antagonists have not been set up to permit coreceptor switching to occur (cell lines were used that expressed either CCR5 or CXCR4, not both). The outcome was predictable; the escape mutant still used the same coreceptor, but in a way that ignored the presence of the inhibitor.[216–220] In other words, the virus either evolved a different binding site for the coreceptor that was not affected by the inhibitor, and/or it developed a higher affinity for the coreceptor, so outcompeted the inhibitor. Three studies, however, were designed to permit coreceptor switching to occur. In two of these, CXCR4 antagonists were added to PBMC cultures containing either mixtures of R5 and X4 viruses, or an X4 virus alone.[221,222] In both cases, the replication of X4 isolates was effectively inhibited, so that only R5 viruses replicated in the cultures. If the input inoculum contained a mixture of R5 and X4 viruses, only the former replicated; if the input was an X4 virus, it mutated to an R5 strain, which grew to dominance in the culture. Thus the intuitively predictable events occurred.[221,222] However, a different outcome was observed when an R5 virus was cultured in the presence of a CCR5-specific antagonist, AD101.[223] Although the input virus was derived from a patient in whom X4 viruses later grew to readily detectable levels, in the *in vitro* experiment no coreceptor switching to CXCR4 usage was seen; instead, the escape mutant still used CCR5 but in an inhibitor-insensitive manner. Similar results were observed with a second CCR5 antagonist, SCH-C, and a different R5 input virus.[223] Thus, for whatever reason, CCR5 was the preferred coreceptor under the *in vitro* conditions that apply to a PBMC culture, at least in those experiments. Whether this is always the case will await the outcome of additional studies with other inhibitors and/or different HIV-1 strains.

What might happen *in vivo*? We can imagine two scenarios that depend very much on whether R5 and X4 viruses replicate in two independent pools of cells, or whether they fight for the same pool with the X4 viruses normally being outcompeted by their R5 cousin compound. In the former scenario, inhibiting the replication of R5 viruses by the use of a CCR5-specific compound would not in any way affect the X4 viruses, which would continue to replicate to the same extent in their own population of target cells (Fig. 2). For example, if R5 viruses are replicating predominantly in the GALT, inhibiting their replication would not necessarily cause X4 viruses to now replicate in GALT, because very little free CXCR4 is expressed in that tissue.[90] However, in the second scenario, the selective inhibition of R5 viruses would make available an evolutionary niche into which the X4 viruses could expand (Fig. 2). Time and experience will tell which of these scenarios is correct, or whether there will be an intermediate or alternative course of events.

OVERVIEW

We argue that HIV-1 infection of humans might almost be viewed as two separate diseases caused by two different lentiviruses that have different cellular tropisms: R5 HIV-1 strains and their close cousins, the X4 viruses. The most commonly transmitted viruses, the R5 strains, replicate preferentially in activated $CD4^+CCR5^+$ lymphocytes and macrophages, cells found in abundance in the GALT. In that tissue, which is rarely sampled for analysis, localized virus production and T cell destruction occur for a prolonged period of time. Of course, R5 virus replication occurs also in other lymphoid organs, particularly the peripheral lymph nodes. In such solid tis-

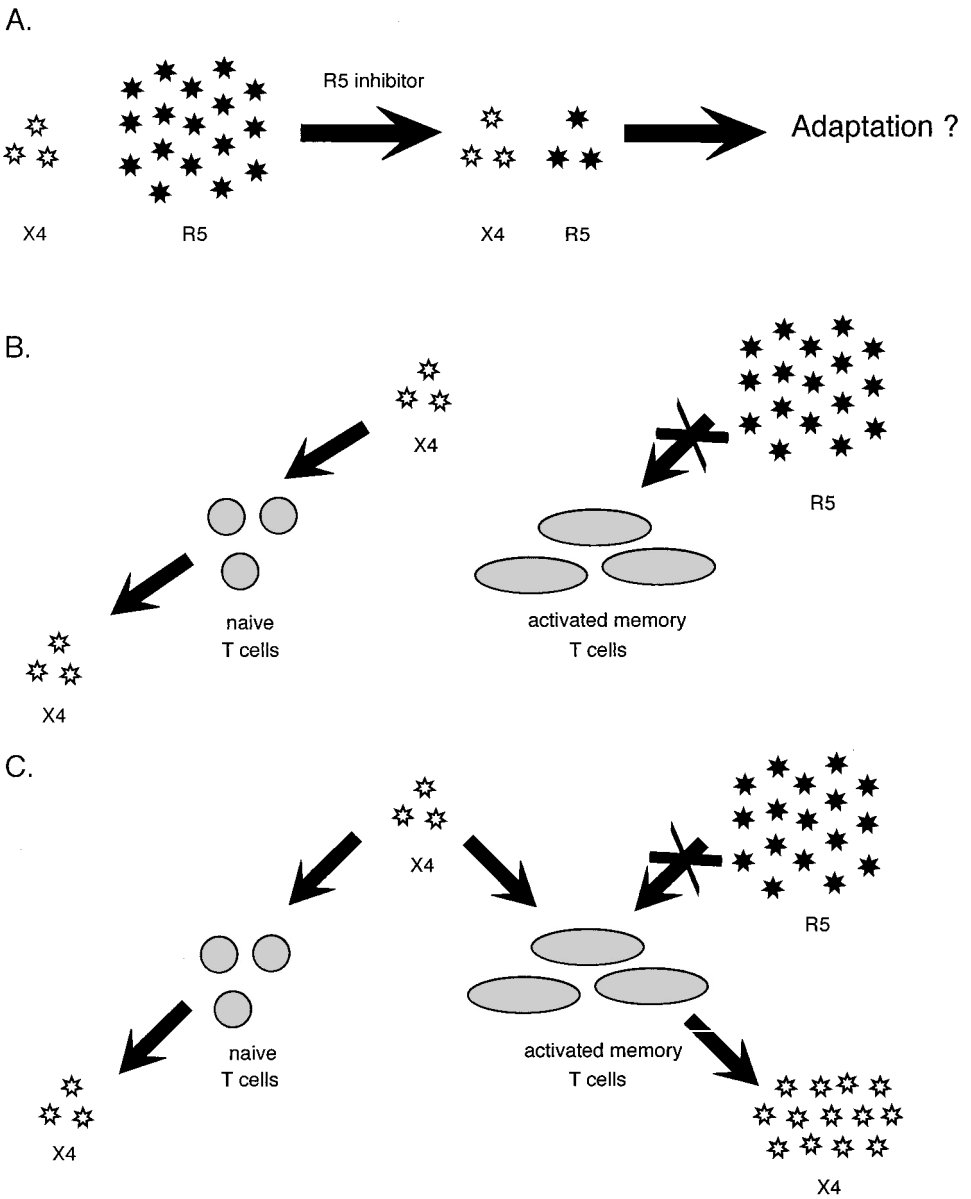

FIG. 2. Possible outcomes of suppressing R5 HIV-1 replication in an individual infected with R5 and X4 viruses. (**A**) Application of a CCR5 inhibitor initially suppresses the R5 population, followed by outgrowth of escape variants. In principle, the escape viruses could still use CCR5, or they could switch to use CXCR4 or even another coreceptor (not shown). (**B**) If X4 and R5 viruses normally replicate in independent pools of cells, suppression of the R5 population by a CCR5 inhibitor does not affect the X4 population. (**C**) If R5 viruses normally outcompete the X4 viruses for the pool of activated memory $CD4^+$ T cells, then suppression of the R5 population allows a niche into which the X4 population can expand, contributing to a greater amount of X4 virus.

sue environments, influences on the CCR5 promoter may be quite different from what applies in the blood where CCR5 expression is normally measured. Within the GALT, and perhaps within lymph nodes, there is little selection pressure for HIV-1 to evolve to CXCR4 usage, particularly if CXCR4 is occluded by SDF-1 or otherwise down-regulated. Moreover, if the relative burst sizes for R5 and X4 virus production are the same *in vivo* as they might be in lymphoid tissue blocks *in vitro*, and we suspect they might be, R5 viruses will simply outcompete their X4 counterparts. There has been a recent paradigm shift in HIV-1 pathogenesis, with the rejection of the simplistic and incorrect "tap-and-drain" model of T cell turnover[224] in favor of one based on chronic immune activation as the proximal cause of T cell loss.[225] Immune activation will continue to generate $CCR5^+$ $CD4^+$ T cell targets for R5 viruses, because of the up-regulation of CCR5 on these activated T cells, so immune activation and HIV-1 replication go hand in hand.[123,204,225] Eventually, however, the reservoir of $CCR5^+$ $CD4^+$ target cells in the GALT and elsewhere becomes exhausted, and other cells elsewhere in the body may become the

engine room of virus production. In macaques, there is good evidence that the macrophage can produce abundant quantities of virions in the later stages of an X4 SHIV infection, and presumably this applies earlier in infection as well.[193] Macrophages can be productively infected by both R5 and X4 viruses, the archaic "macrophage-tropic and T cell tropic" nomenclature notwithstanding:[30,83,193,194,226] X4 viruses may be present in most people at low-to-intermediate levels for most of the time, and they may be responsible for a disproportionate amount of immune system damage, particularly via their capacity to infect and destroy naive $CD4^+$ T cells, and especially in the thymus. The rare individuals in whom X4 or R5X4 viruses predominate early after infection can lose their immune systems with stunning rapidity,[40,43,71] just as do macaques infected with acutely pathogenic, X4 SHIVs.[193,227,228] In rapid progressors, the destruction of $CD4^+$ T cell is often so swift and comprehensive that T cell help for B cells is insufficient even to permit seroconversion to the viral antigens.[227,229,230] This could be considered as an entirely separate disease manifestation to that caused by the still lethal, but less rapidly destructive, R5 strains.[228] And, as outlined above, the transition from the dominance of R5 viruses early in infection to the emergence of X4 strains in the later years is likely to be influenced by not just virological factors but also immunological and conceivably even environmental ones as well. Such an eclectic gallimaufry of interdependent variables defies simplistic analysis.

Indeed, when considering HIV-1 pathogenesis, it is important to fight the natural tendency for oversimplification by homogenization. HIV-1 can evolve complex phenotypes, humans are an outbred species (as too are macaques), and the efficiency with which the immune system responds to a viral infection varies between individuals.[12] These are complex, interlocking variables. To fully understand HIV-1 pathogenesis on a population basis is, therefore, no easy task; each individual's overall response to the infecting strain is usually subtly, and sometimes profoundly, distinctive. Hence different answers to questions of HIV-1 pathogenesis can be obtained that depend upon who is studied, when during the course of his or her infection, and what properties are possessed by the infecting virus, a parameter that can itself vary markedly over time. Until recently, there has been a tendency for immunologists to overlook the complexities caused by the HIV-1 phenotypic variants, for virologists to consider all $CD4^+$ T cells as being much the same as one other, and for mathematical modelers to ignore most or all of the above variables as being beyond the scope of their equations. Perhaps the ever increasing knowledge of coreceptor expression and usage will enable HIV-1 infection to eventually be better understood than it is now.

ACKNOWLEDGMENTS

We thank Cecilia Cheng-Mayer, Mike McCune, and Cheryl Stoddart for the communication of unpublished results. We appreciate helpful comments from Shawn Kuhmann, Andre Maroszan, Ron Veazey, and Barbara Shacklett. This work was funded by RO1 AI41420 (JPM) and by AI036554 and R37 AI036059 (JAZ). J.P.M. is a Stavros S. Niarchos Scholar. The Department of Microbiology and Immunology at the Weill Medical College gratefully acknowledges the support of the William Randolph Hearst Foundation.

REFERENCES

1. Cocchi F, De Vico AL, Garzino-Demo A, Arya SK, Gallo RC, and Lusso P: Identification of RANTES, MIP-1 alpha, and MIP-1 beta as the major HIV-suppressive factors produced by CD8+ T cells. Science 1995;270:1811–1815.
2. Feng Y, Broder CC, Kennedy PE, and Berger EA: HIV-1 entry cofactor: Functional cDNA cloning of a seven-transmembrane, G protein-coupled receptor. Science 1996;272:872–877.
3. Alkhatib G, Combadiere C, Broder CC, *et al.*: CC CKR5: A RANTES, MIP-1alpha, MIP-1beta receptor as a fusion cofactor for macrophage-tropic HIV-1. Science 1996;272:1955–1958.
4. Choe H, Farzan M, Sun Y, *et al.*: The beta-chemokine receptors CCR3 and CCR5 facilitate infection by primary HIV-1 isolates. Cell 1996;85:1135–1148.
5. Deng H, Liu R, Ellmeier W, *et al.*: Identification of a major co-receptor for primary isolates of HIV-1. Nature 1996;381:661–666.
6. Doranz BJ, Rucker J, Yi Y, *et al.*: A dual-tropic primary HIV-1 isolate that uses fusin and the beta-chemokine receptors CKR-5, CKR-3, and CKR-2b as fusion cofactors. Cell 1996;85: 1149–1158.
7. Dragic T, Litwin V, Allaway GP, *et al.*: HIV-1 entry into CD4+ cells is mediated by the chemokine receptor CC-CKR-5. Nature 1996;381:667–673.
8. Liu R, Paxton WA, Choe S, *et al.*: Homozygous defect in HIV-1 coreceptor accounts for resistance of some multiply-exposed individuals to HIV-1 infection. Cell 1996;86:367–377.
9. Dean M, Carrington M, Winkler C, *et al.*: Genetic restriction of HIV-1 infection and progression to AIDS by a deletion allele of the CKR5 structural gene. Hemophilia Growth and Development Study, Multicenter AIDS Cohort Study, Multicenter Hemophilia Cohort Study, San Francisco City Cohort, ALIVE Study. Science 1996;273:1856–1862.
10. Paxton WA, Martin SR, Tse D, *et al.*: Relative resistance to HIV-1 infection of CD4 lymphocytes from persons who remain uninfected despite multiple high-risk sexual exposure. Nat Med 1996;2:412–417.
11. Samson M, Libert F, Doranz BJ, *et al.*: Resistance to HIV-1 infection in caucasian individuals bearing mutant alleles of the CCR-5 chemokine reeptor gene. Nature 1996;382:722–725.
12. Douek DC, Picker LJ, and Koup RA: T cell dynamics in HIV-1 infection. Annu Rev Immunol 2003;21:265–304.
13. Doms RW: Chemokine receptors and HIV entry. AIDS 2001;15(Suppl. 1):S34–35.
14. Berger EA, Murphy PM, and Farber JM: Chemokine receptors as HIV-1 coreceptors: Roles in viral entry, tropism, and disease. Annu Rev Immunol 1999;17:657–700.
15. Lee S, Tiffany HL, King L, Murphy PM, Golding H, and Zaitseva MB: CCR8 on human thymocytes functions as a human immunodeficiency virus type 1 coreceptor. J Virol 2000;74:6946-6952.
16. Sharron M, Pohlmann S, Price K, *et al.*: Expression and coreceptor activity of STRL33/Bonzo on primary peripheral blood lymphocytes. Blood 2000;96:41-49.
17. Zhang YJ, Zhang L, Ketas T, Korber BT, and Moore JP: HIV type 1 molecular clones able to use the Bonzo/STRL-33 coreceptor for virus entry. AIDS Res Hum Retroviruses 2001;17: 217–227.
18. Zhang YJ, Dragic T, Cao Y, *et al.*: Use of coreceptors other than CCR5 by non-syncytium-inducing adult and pediatric isolates of human immunodeficiency virus type 1 is rare in vitro. J Virol 1998;72:9337–9344.
19. Willey SJ, Reeves JD, Hudson R, *et al.*: Identification of a subset of human immunodeficiency virus type 1 (HIV-1), HIV-2, and simian immunodeficiency virus strains able to exploit an alternative coreceptor on untransformed human brain and lymphoid cells. J Virol 2003;77:6138–6152.

20. Connor RI, Sheridan KE, Ceradini D, Choe S, and Landau NR: Change in coreceptor use correlates with disease progression in HIV-1-infected individuals. J Exp Med 1997;185:621–628.
21. de Roda Husman AM, van Rij RP, Blaak H, Broersen S, and Schuitemaker H: Adaptation to promiscuous usage of chemokine receptors is not a prerequisite for human immunodeficiency virus type 1 disease progression. J Infect Dis 1999;180:1106–1115.
22. Edinger AL, Hoffman TL, Sharron M, Lee B, O'Dowd B, and Doms RW: Use of GPR1, GPR15, and STRL33 as coreceptors by diverse human immunodeficiency virus type 1 and simian immunodeficiency virus envelope proteins. Virology 1998;249: 367–378.
23. Kreisberg JF, Kwa D, Schramm B, *et al.*: Cytopathicity of human immunodeficiency virus type 1 primary isolates depends on coreceptor usage and not patient disease status. J Virol 2001;75: 8842–8847.
24. Pohlmann S, Lee B, Meister S, *et al.*: Simian immunodeficiency virus utilizes human and sooty mangabey but not rhesus macaque STRL33 for efficient entry. J Virol 2000;74:5075–5082.
25. Simmons G, Wilkinson D, Reeves JD, *et al.*: Primary, syncytium-inducing human immunodeficiency virus type 1 isolates are dual-tropic and most can use either Lestr or CCR5 as coreceptors for virus entry. J Virol 1996;70:8355–8360.
26. Zhang YJ and Moore JP: Will multiple coreceptors need to be targeted by inhibitors of human immunodeficiency virus type 1 entry? J Virol 1999;73:3443–3448.
27. Sheppard HW, Celum C, Michael NL, *et al.*: HIV-1 infection in individuals with the CCR5-delta32/delta32 genotype: Acquisition of syncytium-inducing virus at seroconversion. J Acquir Immune Defic Syndr 2002;29:307–313.
28. Michael NL, Nelson JA, KewalRamani VN, *et al.*: Exclusive and persistent use of the entry coreceptor CXCR4 by human immunodeficiency virus type 1 from a subject homozygous for CCR5 delta32. J Virol 1998;72:6040–6047.
29. Gorry PR, Zhang C, Wu S, *et al.*: Persistence of dual-tropic HIV-1 in an individual homozygous for the CCR5 delta32 allele. Lancet 2002;359:1832–1834.
30. Naif HM, Cunningham AL, Alali M, *et al.*: A human immunodeficiency virus type 1 isolate from an infected person homozygous for CCR5delta32 exhibits dual tropism by infecting macrophages and MT2 cells via CXCR4. J Virol 2002;76:3114–3124.
31. Moore JP and Stevenson M: New targets for inhibitors of HIV-1 replication. Nat Rev Mol Cell Biol 2000;1:40–49.
32. De Clercq E: Strategies in the design of antiviral drugs. Nat Rev Drug Discov 2002;1:13–25.
33. Moore JP and Doms RW: The entry of entry inhibitors: A fusion of science and medicine. Proc Natl Acad Sci USA 2003;100: 10598–10602.
34. Clapham PR and McKnight A: Cell surfce receptors, virus entry and tropism of primate lentiviruses. J Gen Virol 2002;83: 1809–1829.
35. Zhang Y, Lou B, Lal RB, Gettie A, Marx PA, and Moore JP: Use of inhibitors to evaluate coreceptor usage by simian and simian/human immunodeficiency viruses and human immunodeficiency virus type 2 in primary cells. J Virol 2000;74:6893–6910.
36. Schramm B, Penn ML, Palacios EH, Grant RM, Kirchhoff F, and Goldsmith MA: Cytopathicity of human immunodeficiency virus type 2 (HIV-2) in human lymphoid tissue is coreceptor dependent and comparable to that of HIV-1. J Virol 2000;74:9594–9600.
37. Morner A, Bjorndal A, Albert J, *et al.*: Primary human immunodeficiency virus type 2 (HIV-2) isolates, like HIV-1 isolates, frequently use CCR5 but show promiscuity in coreceptor usage. J Virol 1999;73:2343–2349.
38. Rey-Cuille MA and Hu SL: Conserved CXCR4 usage and enhanced replicative capacity of HIV-2/287, an isolate highly pathogenic in Macaca nemestrina. AIDS 2001;15:2349–2357.
39. Schuitemaker H, Kootstra NA, de Goede RE, de Wolf F, Miedema F, and Tersmette M: Monocytotropic human immunodeficiency virus type 1 (HIV-1) variants detectable in all stages of HIV-1 infection lack T-cell line tropism and syncytium-inducing ability in primary T-cell culture. J Virol 1991;65:356–363.
40. Schuitemaker H, Koot M, Kootstra NA, *et al.*: Biological phenotype of human immunodeficiency virus type 1 clones at different stages of infection: Progression of disease is associated with a shift from monocytotropic to T-cell-tropic virus population. J Virol 1992;66:1354–1360.
41. Richman DD and Bozzette SA: The impact of the syncytium-inducing phenotype of human immunodeficiency virus on disease progression. J Infect Dis 1994;169:968–974.
42. Maas JJ, Gange SJ, Schuitemaker H, Coutinho RA, van Leeuwen R, and Margolick JB: Strong association between failure of T cell homeostasis and the syncytium-inducing phenotype among HIV-1-infected men in the Amsterdam Cohort Study. AIDS 2000;14:1155–1161.
43. Karlsson A, Parsmyr K, Sandstrom E, Fenyo EM, and Albert J: MT-2 cell tropism as prognostic marker for disease progression in human immunodeficiency virus type 1 infection. J Clin Microbiol 1994;32:364–370.
44. Fenyo EM, Morfeldt-Manson L, Chiodi F, *et al.*: Distinct replicative and cytopathic characteristics of human immunodeficiency virus isolates. J Virol 1988;62:4414–4419.
45. de Roda Husman AM, Koot M, Cornelissen M, *et al.*: Association between CCR5 genotype and the clinical course of HIV-1 infection. Ann Intern Med 1997;127:882–890.
46. Berger EA, Doms RW, Fenyo EM, *et al.*: A new classification of HIV-1. Nature 1998;391:240.
47. Singh A and Collman RG: Heterogeneous spectrum of coreceptor usage among variants within a dualtropic human immunodeficiency virus type 1 primary-isolate quasispecies. J Virol 2000; 74:10229–10235.
48. Cho MW, Lee MK, Carney MC, Berson JF, Doms RW, and Martin MA: Identification of determinants on a dualtropic human immunodeficiency virus type 1 envelope glycoprotein that confer usage of CXCR4. J Virol 1998;72:2509–2515.
49. Kimata JT, Kuller L, Anderson DB, Dailey P, and Overbaugh J: Emerging cytopathic and antigenic simian immunodeficiency virus variants influence AIDS progression. Nat Med 1999;5: 535–541.
50. Kwa D, Vingerhoed J, Boeser B, and Schuitemaker H: Increased in vitro cytopathicity of CC chemokine receptor 5-restricted human immunodeficiency virus type 1 primary isolates correlates with a progressive clinical course of infection. J Infect Dis 2003;187:1397–1403.
51. Kawamura T, Gulden FO, Sugaya M, *et al.*: R5 HIV productively infects Langerhans cells, and infection levels are regulated by compound CCR5 polymorphisms. Proc Natl Acad Sci USA 2003;100:8401–8406.
52. Bomsel M and David V: Mucosal gatekeepers: Selecting HIV viruses for early infection. Nat Med 2002;8:114–116.
53. Pope M and Haase AT: Transmission, acute HIV-1 infection and the quest for strategies to prevent infection. Nat Med 2003;9: 847–852.
54. Miller CJ and Shattock RJ: Target cells in vaginal HIV transmission. Microbes Infect 2003;5:59–67.
55. Veazey RS, Marx PA, and Lackner AA: Vaginal CD4+ T cells express high levels of CCR5 and are rapidly depleted in simian immunodeficiency virus infection. J Infect Dis 2003;187: 769–776.
56. Patterson BK, Landay A, Andersson J, *et al.*: Repertoire of chemokine receptor expression in the female genital tract: Implications for human immunodeficiency virus transmission. Am J Pathol 1998;153:481-490.

57. Lee B, Sharron M, Montaner LJ, Weissman D, and Doms RW: Quantification of CD4, CCR5, and CXCR4 levels on lymphocyte subsets, dendritic cells, and differentially conditioned monocyte-derived macrophages. Proc Natl Acad Sci USA 1999;96: 5215–5220.
58. Granelli-Piperno A, Delgado E, Finkel V, Paxton W, and Steinman RM: Immature dendritic cells selectively replicate macrophagetropic (M-tropic) human immunodeficiency virus type 1, while mature cells efficiently transmit both M- and T-tropic virus to T cells. J Virol 1998;72:2733–2737.
59. Zaitseva M, Blauvelt A, Lee S, *et al.*: Expression and function of CCR5 and CXCR4 on human Langerhans cells and macrophages: Implications for HIV primary infection. Nat Med 1997;3: 1369–1375.
60. Kawamura T, Cohen SS, Borris DL, *et al.*: Candidate microbicides block HIV-1 infection of human immature Langerhans cells within epithelial tissue explants. J Exp Med 2000;192:1491–1500.
61. Meng G, Sellers MT, Mosteller-Barnum M, Rogers TS, Shaw GM, and Smith PD: Lamina propria lymphocytes, not macrophages, express CCR5 and CXCR4 and are the likely target cell for human immunodeficiency virus type 1 in the intestinal mucosa. J Infect Dis 2000;182:785–791.
62. Agace WW, Amara A, Roberts AI, *et al.*: Constitutive expression of stromal derived factor-1 by mucosal epithelia and its role in HIV transmission and propagation. Curr Biol 2000;10:325–328.
63. Lathey JL, Tsou J, Brinker K, Hsia K, Meyer WA 3rd, and Spector SA: Lack of autologous neutralizing antibody to human immunodeficiency virus type 1 (HIV-1) and macrophage tropism are associated with mother-to-infant transmission. J Infect Dis 1999;180:344–350.
64. Fiore JR, Bjorndal A, Peipke KA, *et al.*: The biological phenotype of HIV-1 is usually retained during and after sexual transmission. Virology 1994;204:297–303.
65. Scarlatti G, Hodara V, Rossi P, *et al.*: Transmission of human immunodeficiency virus type 1 (HIV-1) from mother to child correlates with viral phenotype. Virology 1993;197:624–629.
66. Balotta C, Vigano A, Riva C, *et al.*: HIV type 1 phenotype correlates with the stage of infection in vertically infected children. AIDS Res Hum Retroviruses 1996;12:1247–1253.
67. Salvatori F and Scarlatti G: HIV type 1 chemokine receptor usage in mother-to-child transmission. AIDS Res Hum Retroviruses 2001;17:925-935.
68. van't Wout AB, Kootstra NA, Mulder-Kampinga GA, *et al.*: Macrophage-tropic variants initiate human immunodeficiency virus type 1 infection after sexual, parenteral, and vertical transmission. J Clin Invest 1994;94:2060–2067.
69. Long EM, Rainwater SM, Lavreys L, Mandaliya K, and Overbaugh J: HIV type 1 variants transmitted to women in Kenya require the CCR5 coreceptor for entry, regardless of the genetic complexity of the infecting virus. AIDS Res Hum Retroviruses 2002;18:567–576.
70. Casper CH, Clevestig P, Carlenor E, *et al.*: Link between the X4 phenotype in human immunodeficiency virus type 1-infected mothers and their children, despite the early presence of R5 in the child. J Infect Dis 2002;186:914–921.
71. Yu XF, Wang Z, Vlahov D, Markham RB, Farzadegan H, and Margolick JB: Infection with dual-tropic human immunodeficiency virus type 1 variants associated with rapid total T cell decline and disease progression in injection drug users. J Infect Dis 1998;178:388–396.
72. Spijkerman IJ, Koot M, Prins M, *et al.*: Lower prevalence and incidence of HIV-1 syncytium-inducing phenotype among injecting drug users compared with homosexual men. AIDS 1995;9: 1085–1092.
73. Lyles CM, Graham NM, Astemborski J, *et al.*: Cell-associated infectious HIV-1 viral load as a predictor of clinical progression and survival among HIV-1 infected injection drug users and homosexual men. Eur J Epidemiol 1999;15:99–108.
74. Miller CJ and Hu J: T cell-tropic simian immunodeficiency virus (SIV) and simian-human immunodeficiency viruses are readily transmitted by vaginal inoculation of rhesus macaques, and Langerhans' cells of the female genital tract are infected with SIV. J Infect Dis 1999;179(Suppl. 3):S413–417.
75. Grivel JC, Penn ML, Eckstein DA, *et al.*: Human immunodeficiency virus type 1 coreceptor preferences determine target T-cell depletion and cellular tropism in human lymphoid tissue. J Virol 2000;74:5347–5351.
76. Bleul CC, Wu L, Hoxie JA, Springer TA, and Mackay CR: The HIV coreceptors CXCR4 and CCR5 are differentially expressed and regulated on human T lymphocytes. Proc Natl Acad Sci USA 1997;94:1925–1930.
77. Frank I and Pope M: The enigma of dendritic cell-immunodeficiency virus interplay. Curr Mol Med 2002;2:229–248.
78. Kawamura T, Cohen SS, Borris DL, *et al.*: Candidate microbicides block HIV-1 infection of human immature Langerhans cells within epithelial tissue explants. J Exp Med 2000;192:1491–1500.
79. Pohlmann S, Baribaud F, and Doms RW: DC-SIGN and DC-SIGNR: Helping hands for HIV. Trends Immunol 2001;22: 643–646.
80. Kwon DS, Gregorio G, Bitton N, Hendrickson WA, and Littman DR: DC-SIGN-mediated internalization of HIV is required for trans-enhancement of T cell infection. Immunity 2002;16: 135–144.
81. Pope M: Mucosal dendritic cells and immunodeficiency viruses. J Infect Dis 1999;179(Suppl. 3):S427–430.
82. Geijtenbeek TB, Kwon DS, Torensma R, *et al.*: DC-SIGN, a dendritic cell-specific HIV-1-binding protein that enhances trans-infection of T cells. Cell 2000;100:587–597.
83. Simmons G, Reeves JD, McKnight A, *et al.*: CXCR4 as a functional coreceptor for human immunodeficiency virus type 1 infection of primary macrophages. J Virol 1998;72:8453–8457.
84. Cornelissen M, Mulder-Kampinga G, Veenstra J, *et al.*: Syncytium-inducing (SI) phenotype suppression at seroconversion after intramuscular inoculation of a nonsyncytium-inducing/SI phenotypically mixed human immunodeficiency virus population. J Virol 1995;69:1810–1818.
85. Pratt RD, Shapiro JF, McKinney N, Kwok S, and Spector SA: Virologic characterization of primary human immunodeficiency virus type 1 infection in a health care worker following needlestick injury. J Infect Dis 1995;172:851–854.
86. Veazey RS, Marx PA, and Lackner AA: The mucosal immune system: Primary target for HIV infection and AIDS. Trends Immunol 2001;22:626–633.
87. Harouse JM, Gettie A, Tan RC, Blanchard J, and Cheng-Mayer C: Distinct pathogenic sequela in rhesus macaques infected with CCR5 or CXCR4 utilizing SHIVs. Science 1999;284:816–819.
88. Smit-McBride Z, Mattapallil JJ, McChesney M, Ferrick D, and Dandekar S: Gastrointestinal T lymphocytes retain high potential for cytokine responses but have severe CD4(+) T-cell depletion at all stages of simian immunodeficiency virus infection compared to peripheral lymphocytes. J Virol 1998;72:6646–6656.
89. Veazey RS, DeMaria M, Chalifoux LV, *et al.*: Gastrointestinal tract as a major site of CD4+ T cell depletion and viral replication in SIV infection. Science 1998;280:427–431.
90. Veazey RS, Mansfield KG, Tham IC, *et al.*: Dynamics of CCR5 expression by CD4(+) T cells in lymphoid tissues during simian immunodeficiency virus infection. J Virol 2000;74:11001–11007.
91. Poles MA, Elliott J, Taing P, Anton PA, and Chen IS: A preponderance of CCR5(+) CXCR4(+) mononuclear cells enhances gastrointestinal mucosal susceptibility to human immunodeficiency virus type 1 infection. J Virol 2001;75:8390–8399.
92. Shacklett BL, Cox CA, Sandberg JK, Stollman NH, Jacobson MA,

and Nixon DF: Trafficking of human immunodeficiency virus type 1-specific CD8(+) T cells to gut-associated lymphoid tissue during chronic infection. J Virol 2003;77:5621–5631.
93. Olsson J, Poles M, Spetz AL, *et al.*: Human immunodeficiency virus type 1 infection is associated with significant mucosal inflammation characterized by increased expression of CCR5, CXCR4, and beta-chemokines. J Infect Dis 2000;182:1625–1635.
94. Coulomb-L'Hermine A, Emilie D, Durand-Gasselin I, Galanaud P, and Chaouat G: SDF-1 production by placental cells: A potential mechanism of inhibition of mother-to-fetus HIV transmission. AIDS Res Hum Retroviruses 2000;16:1097–1098.
95. Carrington M, Dean M, Martin MP, and O'Brien SJ: Genetics of HIV-1 infection: Chemokine receptor CCR5 polymorphism and its consequences. Hum Mol Genet 1999;8:1939–1945.
96. An P, Martin MP, Nelson GW, *et al.*: Influence of CCR5 promoter haplotypes on AIDS progression in African-Americans. AIDS 2000;14:2117–2122.
97. Gonzalez E, Dhanda R, Bamshad M, *et al.*: Global survey of genetic variation in CCR5, RANTES, and MIP-1alpha: Impact on the epidemiology of the HIV-1 pandemic. Proc Natl Acad Sci USA 2001;98:5199–5204.
98. Kostrikis LG, Huang Y, Moore JP, *et al.*: A chemokine receptor CCR2 allele delays HIV-1 disease progression and is associated with a CCR5 promoter mutation. Nat Med 1998;4:350–353.
99. Mangano A, Gonzalez E, Dhanda R, *et al.*: Concordance between the CC chemokine receptor 5 genetic determinants that alter risks of transmission and disease progression in children exposed perinatally to human immunodeficiency virus. J Infect Dis 2001; 183:1574–1585.
100. Martin MP, Dean M, Smith MW, *et al.*: Genetic acceleration of AIDS progression by a promoter variant of CCR5. Science 1998;282:1907–1911.
101. Mummidi S, Ahuja SS, Gonzalez E, *et al.*: Genealogy of the CCR5 locus and chemokine system gene variants associated with altered rates of HIV-1 disease progression. Nat Med 1998;4:786–793.
102. O'Brien TR, McDermott DH, Ioannidis JP, *et al.*: Effect of chemokine receptor gene polymorphisms on the response to potent antiretroviral therapy. AIDS 2000;14:821–826.
103. Cao Y, Krogstad P, Korber BT, *et al.*: Maternal HIV-1 viral load and vertical transmission of infection: The Ariel Project for the prevention of HIV transmission from mother to infant. Nat Med 1997;3:549–552.
104. Sleasman JW, Aleixo LF, Morton A, Skoda-Smith S, and Goodenow MM: CD4+ memory T cells are the predominant population of HIV-1-infected lymphocytes in neonates and children. AIDS 1996;10:1477–1484.
105. Luzuriaga K and Sullivan JL: Pediatric HIV-1 infection: Advances and remaining challenges. AIDS Rev 2002;4:21–26.
106. Richardson BA, Mbori-Ngacha D, Lavreys L, *et al.*: Comparison of human immunodeficiency virus type 1 viral loads in Kenyan women, men, and infants during primary and early infection. J Viral 2003;77:7120–7123.
107. Mo H, Monard S, Pollack H, *et al.*: Expression patterns of the HIV type 1 coreceptors CCR5 and CXCR4 on CD4+ T cells and monocytes from cord and adult blood. AIDS Res Hum Retroviruses 1998;14:607–617.
108. Jameson B, Baribaud F, Pohlmann S, *et al.*: Expression of DC-SIGN by dendritic cells of intestinal and genital mucosae in humans and rhesus macaques. J Virol 2002;76:1866–1875.
109. Dalgleish AG, Beverley PC, Clapham PR, Crawford DH, Greaves MF, and Weiss RA: The CD4 (T4) antigen is an essential component of the receptor for the AIDS retrovirus. Nature 1984;312:763–767.
110. Klatzmann D, Champagne E, Chamaret S, *et al.*: T-lymphocyte T4 molecule behaves as the receptor for human retrovirus LAV. Nature 1984;312:767–768.
111. Barre-Sinoussi F, Chermann JC, Rey F, *et al.*: Isolation of a T-lymphotropic retrovirus from a patient at risk for acquired immune deficiency syndrome (AIDS). Science 1983;220:868–871.
112. Gallo RC, Salahuddin SZ, Popovic M, *et al.*: Frequent detection and isolation of cytopathic retroviruses (HTLV-III) from patients with AIDS and at risk for AIDS. Science 1984;224:500–503.
113. Sousa AE, Carneiro J, Meier-Schellersheim M, Grossman Z, and Victorino RM: CD4 T cell depletion is linked directly to immune activation in the pathogenesis of HIV-1 and HIV-2 but only indirectly to the viral load. J Immunol 2002;169:3400–3406.
114. Silvestri G, Sodora DL, Koup RA, *et al.*: Nonpathogenic SIV infection of sooty mangabeys is characterized by limited bystander immunopathology despite chronic high-level viremia. Immunity 2003;18:441-452.
115. Broussard SR, Staprans SI, White R, Whitehead EM, Feinberg MB, and Allan JS: Simian immunodeficiency virus replicates to high levels in naturally infected African green monkeys without inducing immunologic or neurologic disease. J Virol 2001;75: 2262–2275.
116. Rey-Cuille MA, Berthier JL, Bomsel-Demontoy MC, *et al.*: Simian immunodeficiency virus replicates to high levels in sooty mangabeys without inducing disease. J Virol 1998;72:3872–3886.
117. Chakrabarti LA, Lewin SR, Zhang L, *et al.*: Normal T-cell turnover in sooty mangabeys harboring active simian immunodeficiency virus infection. J Virol 2000;74:1209–1223.
118. McCune JM: The dynamics of CD4+ T-cell depletion in HIV disease. Nature 2001;410:974–979.
119. Hazenberg MD, Hamann D, Schuitemaker H, and Miedema F: T cell depletion in HIV-1 infection: How CD4+ T cells go out of stock. Nat Immunol 2000;1:285–289.
120. Rowland-Jones S: HIV infection: Where have all the T cells gone? Lancet 1999;354:5–7.
121. Douek DC, Betts MR, Hill BJ, *et al.*: Evidence for increased T cell turnover and decreased thymic output in HIV infection. J Immunol 2001;167:6663–6668.
122. Haynes BF, Markert ML, Sempowski GD, Patel DD, and Hale LP: The role of the thymus in immune reconstitution in aging, bone marrow transplantation, and HIV-1 infection. Annu Rev Immunol 2000;18:529–560.
123. Grossman Z, Meier-Schellersheim M, Sousa AE, Victorino RM, and Paul WE: CD4+ T-cell depletion in HIV infection: Are we closer to understanding the cause? Nat Med 2002;8:319–323.
124. Kalayjian RC, Landay A, Pollard RB, *et al.*: Age-related immune dysfunction in health and in human immunodeficiency virus (HIV) disease: Association of age and HIV infection with naive CD8+ cell depletion, reduced expression of CD28 on CD8+ cells, and reduced thymic volumes. J Infect Dis 2003;187: 1924–1933.
125. Collaborative: Time from HIV-1 seroconversion to AIDS and death before widespread use of highly-active antiretroviral therapy: A collaborative re-analysis. Lancet 2000;355:1131–1137.
126. Bestilny LJ, Gill MJ, Mody CH, and Riabowol KT: Accelerated replicative senescence of the peripheral immune system induced by HIV infection. AIDS 2000;14:771–780.
127. Fagnoni FF, Vescovini R, Passeri G, *et al.*: Shortage of circulating naive CD8(+) T cells provides new insights on immunodeficiency in aging. Blood 2000;95:2860–2868.
128. Essajee SM, Kim M, Gonzalez C, *et al.*: Immunologic and virologic responses to HAART in severely immunocompromised HIV-1-infected children. AIDS 1999;13:2523–2532.
129. Berkowitz RD, Beckerman KP, Schall TJ, and McCune JM: CXCR4 and CCR5 expression delineates targets for HIV-1 disruption of T cell differentiation. J Immunol 1998;161:3702–3710.
130. Berkowitz RD, van't Wout AB, Kootstra NA, *et al.*: R5 strains of human immunodeficiency virus type 1 from rapid progressors

lacking X4 strains do not possess X4-type pathogenicity in human thymus. J Virol 1999;73:7817–7822.
131. Berkowitz RD, Alexander S, and McCune JM: Causal relationships between HIV-1 coreceptor utilization, tropism, and pathogenesis in human thymus. AIDS Res Hum Retroviruses 2000;16:1039–1045.
132. Camerini D, Su HP, Gamez-Torre G, Johnson ML, Zack JA, and Chen IS: Human immunodeficiency virus type 1 pathogenesis in SCID-hu mice correlates with syncytium-inducing phenotype and viral replication. J Virol 2000;74:3196–3204.
133. Correa R and Munoz-Fernandez MA: Viral phenotype affects the thymic production of new T cells in HIV-1-infected children. AIDS 2001;15:1959–1963.
134. Blaak H, van't Wout AB, Brouwer M, Hooibrink B, Hovenkamp E, and Schuitemaker H: In vivo HIV-1 infection of CD45RA(+)CD4(+) T cells is established primarily by syncytium-inducing variants and correlates with the rate of CD4(+) T cell decline. Proc Natl Acad Sci USA 2000;97:1269–1274.
135. Hernandez-Lopez C, Varas A, Sacedon R, *et al.*: Stromal cell-derived factor 1/CXCR4 signaling is critical for early human T-cell development. Blood 2002;99:546–554.
136. Muench MO, Roncarolo MG, and Namikawa R: Phenotypic and functional evidence for the expression of CD4 by hematopoietic stem cells isolated from human fetal liver. Blood 1997;89: 1364–1375.
137. Chelucci C, Casella I, Federico M, *et al.*: Lineage-specific expression of human immunodeficiency virus (HIV) receptor/coreceptors in differentiating hematopoietic precursors: Correlation with susceptibility to T- and M-tropic HIV and chemokine-mediated HIV resistance. Blood 1999;94:1590–1600.
138. Ruiz ME, Cicala C, Arthos J, *et al.*: Peripheral blood-derived CD34+ progenitor cells: CXC chemokine receptor 4 and CC chemokine receptor 5 expression and infection by HIV. J Immunol 1998;161:4169–4176.
139. Neal TF, Holland HK, Baum CM, *et al.*: CD34+ progenitor cells from asymptomatic patients are not a major reservoir for human immunodeficiency virus-1. Blood 1995;86:1749–1756.
140. Koka PS, Jamieson BD, Brook DG, and Zack JA: Human immunodeficiency virus type 1-induced hematopoietic inhibition is independent of productive infection of progenitor cells in vivo. J Virol 1999;73:9089–9097.
141. Stanley SK, Kessler SW, Justement JS, *et al.*: CD34+ bone marrow cells are infected with HIV in a subset of seropositive individuals. J Immunol 1992;149:689–697.
142. Moses A, Nelson J, and Bagby GC: The influence of human immunodeficiency virus-1 on hematopoiesis. BLood 1998;91: 1479–1495.
143. Kitchen SG and Zack JA: Distribution of the human immunodeficiency virus coreceptors CXCR4 and CCR5 in fetal lymphoid organs: Implications for pathogenesis in utero. AIDS Res Hum Retroviruses 1999;15:143–148.
144. Zou YR, Kottmann AH, Kuroda M, Taniuchi I, and Littman DR: Function of the chemokine receptor CXCR4 in haematopoiesis and in cerebellar development. Nature 1998;393:595–599.
145. Zamarchi R, Allavena P, Borsetti A, *et al.*: Expression and functional activity of CXCR-4 and CCR-5 chemokine receptors in human thymocytes. Clin Exp Immunol 2002;127:321–330.
146. Kitchen SG and Zack JA: CXCR4 expression during lymphopoiesis: Implications for human immunodeficiency virus type 1 infection of the thymus. J Virol 1997;71:6928–6934.
147. Zaitseva MB, Lee S, Rabin RL, *et al.*: CXCR4 and CCR5 on human thymocytes: Biological function and role in HIV-1 infection. J Immunol 1998;161:3103–3113.
148. Pedroza-Martins L, Gurney KB, Torbett BE, and Uittenbogaart CH: Differential tropism and replication kinetics of human immunodeficiency virus type 1 isolates in thymocytes: Coreceptor expression allows viral entry, but productive infection of distinct subsets is determined at the postentry level. J Virol 1998;72:9441–9452.
149. Majka M, Rozmyslowicz T, Ratajczak J, *et al.*: The limited infectability by R5 HIV of CD34(+) cells from thymus, cord, and peripheral blood and bone marrow is explained by their ability to produce beta-chemokines. Exp Hematol 2000;28:1334–1342.
150. Taylor JR Jr, Kimbrell KC, Scoggins R, Delaney M, Wu L, and Camerini D: Expression and function of chemokine receptors on human thymocytes: Implications for infection by human immunodeficiency virus type 1. J Virol 2001;75:8752–8760.
151. Huang Y, Paxton WA, Wolinsky SM, *et al.*: The role of a mutant CCR5 allele in HIV-1 transmission and disease progression. Nat Med 1996;2:1240–1243.
152. Zhou Y, Kurihara T, Ryseck RP, *et al.*: Impaired macrophage function and enhanced T cell-dependent immune response in mice lacking CCR5, the mouse homologue of the major HIV-1 coreceptor. J Immunol 1998;160:4018–4025.
153. Brooks DG, Kitchen SG, Kitchen CM, Scripture-Adams DD, and Zack JA: Generation of HIV latency during thymopoiesis. Nat Med 2001;7:459–464.
154. Jamieson BD and Zack JA: In vivo pathogenesis of a human immunodeficiency virus type 1 reporter virus. J Virol 1998;72: 6520–6526.
155. Uittenbogaart CH, Anisman DJ, Jamieson BD, *et al.*: Differential tropism of HIV-1 isolates for distinct thymocyte subsets in vitro. AIDS 1996;10:F9–16.
156. Aldrovandi GM, Feuer G, Gao L, *et al.*: The SCID-hu mouse as a model for HIV-1 infection. Nature 1993;363:732–736.
157. Bonyhadi ML, Rabin L, Salimi S, *et al.*: HIV induces thymus depletion in vivo. Nature 1993;363:728–732.
158. Jamieson BD, Pang S, Aldrovandi GM, Zha J, and Zack JA: In vivo pathogenic properties of two clonal human immunodeficiency virus type 1 isolates. J Virol 1995;69:6259–6264.
159. Kitchen SG, Uittenbogaart CH, and Zack JA: Mechanism of human immunodeficiency virus type 1 localization in CD4-negative thymocytes: Differentiation from a CD4-positive precursor allows productive infection. J Virol 1997;71:5713–5722.
160. Rosenzweig M, Clark DP, and Gaulton GN: Selective thymocyte depletion in neonatal HIV-1 thymic infection. AIDS 1993;7: 1601–1605.
161. Kitchen SG, Killian S, Giorgi JV, and Zack JA: Functional reconstitution of thymopoiesis following HIV infection. J Virol 2000;74:2943–2948.
162. Withers-Ward ES, Amado RG, Koka PS, *et al.*: Transient renewal of thymopoiesis in HIV-infected human thymic implants following antiviral therapy. Nat Med 1997;3:1102–1109.
163. Kaneshima H, Su L, Bonyhadi ML, Connor RI, Ho DD, and McCune JM: Rapid-high, syncytium-inducing isolates of human immunodeficiency virus type 1 induce cytopathicity in the human thymus of the SCID-hu mouse. J Virol 1994;68:8188–8192.
164. Scoggins RM, Taylor JR Jr, Patrie J, van't Wout AB, Schuitemaker H, and Camerini D: Pathogenesis of primary R5 human immunodeficiency virus type 1 clones in SCID-hu mice. J Virol 2000;74:3205–3216.
165. Alves K, Canzian M, and Delwart EL: HIV type 1 envelope quasispecies in the thymus and lymph nodes of AIDS patients. AIDS Res Hum Retroviruses 2002;18:161–165.
166. Baskin GB, Murphey-Corb M, Martin LN, Davison-Fairburn B, Hu FS, and Kuebler D: Thymus in simian immunodeficiency virus-infected rhesus monkeys. Lab Invest 1991;65:400–407.
167. Wykrzykowska JJ, Rosenzweig M, Veazey RS, *et al.*: Early regeneration of thymic progenitors in rhesus macaques infected with simian immunodeficiency virus. J Exp Med 1998;187:1767–1778.
168. Lackner AA, Vogel P, Ramos RA, Kluge JD, and Marthas M: Early events in tissues during infection with pathogenic (SIV-

mac239) and nonpathogenic (SIVmac1A11) molecular clones of simian immunodeficiency virus. Am J Pathol 1994;145:428–439.
169. Zhang L, He T, Talal A, Wang G, Frankel SS, and Ho DD: In vivo distribution of the human immunodeficiency virus/simian immunodeficiency virus coreceptors: CXCR4, CCR3, and CCR5. J Virol 1998;72:5035–5045.
170. Stoddart C, Keir ME, and McCune JM: Personal communication. Gladstone Institute of Virology and Immunology, San Francisco, CA.
171. Schmitt N, Chene L, Boutolleau D, *et al.*: Positive regulation of CXCR4 expression and signaling by interleukin-7 in CD4(+) mature thymocytes correlates with their capacity to favor human immunodeficiency X4 virus replication. J Virol 2003;77:5784–5793.
172. Pedroza-Martins L, Boscardin WJ, Anisman-Posner DJ, Schols D, Bryson YJ, and Uittenbogaart CH: Impact of cytokines on replication in the thymus of primary human immunodeficiency virus type 1 isolates from infants. J VIrol 2002;76:6929–6943.
173. Gurney KB, Yang OO, Wilson SB, and Uittenbogaart CH: TCR gamma delta+ and CD161+ thymocytes express HIV-1 in the SCID-hu mouse, potentially contributing to immune dysfunction in HIV infection. J Immunol 2002;169:5338–5346.
174. Fouchier RA, Groenink M, Kootstra NA, *et al.*: Phenotype-associated sequence variation in the third variable domain of the human immunodeficiency virus type 1 gp120 molecule. J Virol 1992;66:3183–3187.
175. Shioda T, Levy JA, and Cheng-Mayer C: Small amino acid changes in the V3 hypervariable region of gp120 can affect the T-cell-line and macrophage tropism of human immunodeficiency virus type 1. Proc Natl Acad Sci USA 1992;89:9434–9438.
176. De Jong JJ, De Ronde A, Keulen W, Tersmette M, and Goudsmit J: Minimal requirements for the human immunodeficiency virus type 1 V3 domain to support the syncytium-inducing phenotype: Analysis by single amino acid subtitution. J Virol 1992;66: 6777–6780.
177. Wei X, Ghosh SK, Taylor ME, *et al.*: Viral dynamics in human immunodeficiency virus type 1 infection. Nature 1995;373: 117–122.
178. Coffin JM: HIV population dynamics in vivo: Implications for genetic variation, pathogenesis, and therapy. Science 1995;267: 483–489.
179. van Rij RP, Hazenberg MD, van Benthem BHB, *et al.*: Early viral load and CD4+ T cell count, but not percentage of CCR5+ or CXCR4+ CD4+ T cells, are associated with R5-to-X4 HIV type 1 virus evolution. AIDS Res Hum Retroviruses 2003;19: 389–398.
180. Penn ML, Grivel JC, Schramm B, Goldsmith MA, and Margolis L: CXCR4 utilization is sufficient to trigger CD4+ T cell depletion in HIV-1-infected human lymphoid tissue. Proc Natl Acad Sci USA 1999;96:663–668.
181. Arthos J, Cicala C, Selig SM, *et al.*: The role of the CD4 receptor versus HIV coreceptors in envelope-mediated apoptosis in peripheral blood mononuclear cells. Virology 2002;292:98–106.
182. Kwa D, Vingerhoed J, Boeser-Nunnink B, Broersen S, and Schuitemaker H: Cytopathic effects of non-syncytium-inducing and syncytium-inducing human immunodeficiency virus type 1 variants on different CD4(+)-T-cell subsets are determined only by coreceptor expression. J Virol 2001;75:10455–10459.
183. Jekle A, Keppler OT, De Clercq E, Schols D, Weinstein M, and Goldsmith MA: In vivo evolution of human immunodeficiency virus type 1 toward increased pathogenicity through CXCR4-mediated killing of uninfected CD4 T cells. J Virol 2003;77: 5846–5854.
184. Ostrowski MA, Justement SJ, Catanzaro A, *et al.*: Expression of chemokine receptors CXCR4 and CCR5 in HIV-1-infected and uninfected individuals. J Immunol 1998;161:3195–3201.
185. Hazenberg MD, Stuart JW, Otto SA, *et al.*: T-cell division in human immunodeficiency virus (HIV)-1 infection is mainly due to immune activation: A longitudinal analysis in patients before and during highly active antiretroviral therapy (HAART). Blood 2000;95:249–255.
186. Eckstein DA, Penn ML, Korin YD, *et al.*: HIV-1 actively replicates in naive CD4(+) T cells residing within human lymphoid tissues. Immunity 2001;15:671–682.
187. Lee S, Lapham CK, Chen H, *et al.*: Coreceptor competition for association with CD4 may change the susceptibility of human cells to infection with T-tropic and macrophagetropic isolates of human immunodeficiency virus type 1. J Virol 2000;74: 5016–5023.
188. Schutten M, van Baalen CA, Guillon C, *et al.*: Macrophage tropism of human immunodeficiency virus type 1 facilitates in vivo escape from cytotoxic T-lymphocyte pressure. J Virol 2001;75: 2706–2709.
189. Kwa D, Vin Rij RP, Boeser-Nunnink B, Vingerhoed J, and Schuitemaker H: Association between an interleukin-4 promoter polymorphism and the acquisition of CXCR4 using HIV-1 variants. AIDS 2003;17:981-985.
190. Llano A, Barretina J, Gutierrez A, *et al.*: Interleukin-7 in plasma correlates with CD4 T-cell depletion and may be associated with emergence of syncytium-inducing variants in human immunodeficiency virus type 1-positive individuals. J Virol 2001;75: 10319–10325.
190a. Harouse JM, Buckner C, Gettie A, *et al.*: $CD8^+$ T cell-mediated CRC chemokine receptor 4—simian/human immunodeficiency virus suppression in dually infected rhesus macaques. Proc Natl Acad Sci USA 2003;100:10977–10982.
191. Trkola A, Ketas T, Kewalramani VN, *et al.*: Neutralization sensitivity of human immunodeficiency virus type 1 primary isolates to antibodies and CD4-based reagents is independent of coreceptor usage. J Virol 1998;72:1876–1885.
192. Montefiori DC, Collman RG, Fouts TR, *et al.*: Evidence that antibody-mediated neutralization of human immunodeficiency virus type 1 by sera from infected individuals is independent of coreceptor usage. J VIrol 1998;72:1886–1893.
193. Igarashi T, Brown CR, Byrum RA, *et al.*: Rapid and irreversible CD4+ T-cell depletion induced by the highly pathogenic simian/human immunodeficiency virus SHIV(DH12R) is systemic and synchronous. J Virol 2002;76:379–391.
194. Tokunaga K, Greenberg ML, Morse MA, Cumming RI, Lyerly HK, and Cullen BR: Molecular basis for cell tropism of CXCR4-dependent human immunodeficiency virus type 1 isolates. J Virol 2001;75:6776–6785.
195. Carrington M and O'Brien SJ: The influence of hla genotype on AIDS. Annu Rev Med 2003;54:535–551.
196. Cilliers T, Nhlapo J, Coetzer M, *et al.*: The CCR5 and CXCR4 coreceptors are both used by human immunodeficiency virus type 1 primary isolates from subtype C. J Virol 2003;77:4449-4456.
197. Tatt ID, Barlow KL, Nicoll A, and Clewley JP: The public health significance of HIV-1 subtypes. AIDS 2001;15(Suppl.5):S59–71.
198. Batra M, Tien PC, Shafer RW, Contag CH, and Katzenstein DA: HIV type 1 enevlope subtype C sequences from recent seroconverters in Zimbabwe. AIDS Res Hum Retroviruses 2000;16:973-979.
199. Johnston ER, Zijenah LS, Mutetwa S, Kantor R, Kittinunvorakoon C, and Katzenstein DA: High frequency of syncytium-inducing and CXCR4-tropic viruses among human immunodeficiency virus type 1 subtype C-infected patients receiving antiretroviral treatment. J Virol 2003;77:7682–7688.
200. Alaeus A, Lidman K, Bjorkman A, Giesecke J, and Albert J: Similar rate of disease progression among individuals infected with HIV-1 genetic subtypes A-D. AIDS 1999;13:901–907.
201. Amornkul PN, Tansuphasawadikul S, Limpakarnjanarat K, *et al.*: Clinical disease associated with HIV-1 subtype B′ and E infection among 2104 patients in Thailand. AIDS 1999;13:1963–1969.

202. Weisman Z, Kalinkovich A, Borkow G, Stein M, Greenberg Z, and Bentwich Z: Infection by different HIV-1 subtypes (B and C) results in a similar immune activation profile despite distinct immune backgrounds. J Acquir Immune Defic Syndr 1999;21: 157–163.
202a. Ball SC, Abraha A, Collins KR, *et al.*: Comparing the *ex vivo* fitness of CCR5-tropic human immunodeficiency virus type 1 isolates of subtypes B and C. J Virol 2003;77:1021–1038.
203. Clerici M, Butto S, Lukwiya M, *et al.*: Immune activation in Africa is environmentally-driven and is associated with upregulation of CCR5. Italian-Ugandan AIDS Project. AIDS 2000;14: 2083–2092.
204. Bentwich Z, Maartens G, Torten D, Lal AA, and Lal RB: Concurrent infections and HIV pathogenesis. AIDS 2000;14: 2071–2081.
205. Trouplin V, Salvatori F, Cappello F, *et al.*: Determination of coreceptor usage of human immunodeficiency virus type 1 from patient plasma samples by using a recombinant phenotype assay. J Virol 2001;75:251–259.
206. Resch W, Hoffman N, and Swanstrom R: Improved success of phenotype prediction of the human immunodeficiency virus type 1 from envelope variable loop 3 sequence using neural networks. Virology 2001;288:51–62.
207. Shankarappa R, Margolick JB, Gange SJ, *et al.*: Consistent viral evolutionary changes associated with the progression of human immunodeficiency virus type 1 infection. J Virol 1999;73: 10489–10502.
208. ten Haaft P, Murthy K, Salas M, *et al.*: Differences in early virus loads with different phenotypic variants of HIV-1 and SIV(cpz) in chimpanzees. AIDS 2001;15:2085–2092.
209. Michael NL and Moore JP: HIV-1 entry inhibitors: Evading the issue. Nat Med 1999;5:740–742.
210. Hendrix CW, Flexner C, MacFarland RT, *et al.*: Pharmacokinetics and safety of AMD-3100, a novel antagonist of the CXCR-4 chemokine receptor, in human volunteers. Antimicrob Agents Chemother 2000;44:1667–1673.
211. Strizki JM, Xu S, Wagner NE, *et al.*: SCH-C (SCH 351125), an orally bioavailable, small molecule antagonist of the chemokine receptor CCR5, is a potent inhibitor of HIV-1 infection in vitro and in vivo. Proc Natl Acad Sci USA 2001;98:12718–12723.
211a. Meanwell NA, Kadow JF. Inhibitors of the entry of HIV into host cells. Curr Opin Drug Discov Develop 2003;6:451–461.
212. Datema R, Rabin L, Hincenbergs M, *et al.*: Antiviral efficacy in vivo of the anti-human immunodeficiency virus bicyclam SDZ SID 791 (JM 3100), an inhibitor of infectious cell entry. Antimicrob Agents Chemother 1996;40:750–754.
213. D'Aquila RT, Sutton L, Savara A, Hughes MD, and Johnson VA: CCR5/delta(ccr5) heterozygosity: A selective pressure for the syncytium-inducing human immunodeficiency virus type 1 phenotype. NIAID AIDS Clinical Trials Group Protocol 241 Virology Team. J Infect Dis 1998;177:1549–1553.
214. de Roda Husman AM, Blaak H, Brouwer M, and Schuitemaker H: CC chemokine receptor 5 cell-surface expression in relation to CC chemokine receptor 5 genotype and the clinical course of HIV-1 infection. J Immunol 1999;163:4597–4603.
215. Michael NL, Chang G, Louie LG, *et al.*: The role of viral phenotype and CCR-5 gene defects in HIV-1 transmission and disease progression. Nat Med 1997;3:338–340.
216. Aarons EJ, Beddows S, Willingham T, Wu L, and Koup RA: Adaptation to blockade of human immunodeficiency virus type 1 entry imposed by the anti-CCR5 monoclonal antibody 2D7. Virology 2001;287:382–390.
217. de Vreese K, Kofler-Mongold V, Leutgeb C, *et al.*: The molecular target of bicyclams, potent inhibitors of human immunodeficiency virus replication. J Virol 1996;70:689–696.
218. Kambara K, Sato S, Tanuma J, *et al.*: Biological and genetic characterization of a human immunodeficiency virus strain resistant to CXCR4 antagonist T134. AIDS Res Hum Retroviruses 2001;17:615–622.
219. Maeda Y, Foda M, Matsushita S, and Harada S: Involvement of both the V2 and V3 regions of the CCR5-tropic human immunodeficiency virus type 1 envelope in reduced sensitivity to macrophage inflammatory protein 1alpha. J Virol 2000;74:1787–1793.
220. Schols D, Este JA, Cabrera C, and De Clercq E: T-cell-line-tropic human immunodeficiency virus type 1 that is made resistant to stromal cell-derived factor 1alpha contains mutations in the envelope gp120 but does not show a switch in coreceptor use. J Virol 1998;72:4032–4037.
221. Este JA, Cabrera C, Blanco J, *et al.*: Shift of clinical human immunodeficiency virus type 1 isolates from X4 to R5 and prevention of emergence of the syncytium-inducing phenotype by blockade of CXCR4. J Virol 1999;73:5577–5585.
222. Gotoh K, Yoshimori M, Kanbara K, *et al.*: Increase of R5 HIV-1 infection and CCR5 expression in T cells treated with high concentrations of CXCR4 antagonists and SDF-1. J Infect Chemother 2001;7:28–36.
223. Trkola A, Kuhmann SE, Strizki JM, *et al.*: HIV-1 escape from a small molecule, CCR5-specific entry inhibitor does not involve CXCR4 use. Proc Natl Acad Sci USA 2002;99:395–400.
224. Ho DD, Neumann AU, Perelson AS, Chen W, Leonard JM, and Markowitz M: Rapid turnover of plasma virions and CD4 lymphocytes in HIV-1 infection. Nature 1995;373:123–126.
225. Grossman Z and Paul WE: The impact of HIV on naive T-cell homeostasis. Nat Med 2000;6:976–977.
226. Verani A, Pesenti E, Polo S, *et al.*: CXCR4 is a functional coreceptor for infection of human macrophages by CXCR4-dependent primary HIV-1 isolates. J Immunol 1998;161:2084–2088.
227. Parker RA, Regan MM, and Reimann KA: Variability of viral load in plasma of rhesus monkeys inoculated with simian immunodeficiency virus or simian-human immunodeficiency virus: Implications for using nonhuman primate AIDS models to test vaccines and therapeutics. J Virol 2001;75:11234–11238.
228. Feinberg MB and Moore JP: AIDS vaccine models: Challenging challenge viruses. Nat Med 2002;8:207–210.
229. Michael NL, Brown AE, Voigt RF, *et al.*: Rapid disease progression without seroconversion following primary human immunodeficiency virus type 1 infection—evidence for highly susceptible human hosts. J Infect Dis 1997;175:1352–1359.
230. Montagnier L, Brenner C, Chamaret S, *et al.*: Human immunodeficiency virus infection and AIDS in a person with negative serology. J Infect Dis 1997;175:955–959.

Address reprint requests to:
John P. Moore
Weill Medical College of Cornell University
Department of Microbiology and Immunology
1300 York Avenue
W-805
New York, New York 10021

E-mail: jpm2003@med.cornell.edu

This article has been cited by:

1. Matthew D. Marsden, Jerome A. Zack. 2013. HIV/AIDS eradication. *Bioorganic & Medicinal Chemistry Letters* . [CrossRef]
2. Sabrina Wai-Chi To, Jonathan Hon-Kwan Chen, Wing-Cheong Yam. 2013. Current assays for HIV-1 diagnostics and antiretroviral therapy monitoring: challenges and possibilities. *Future Virology* **8**:4, 405-419. [CrossRef]
3. Tiffany N. Walker, Lisa M. Cimakasky, Ebony M. Coleman, M. Nia Madison, James E.K. Hildreth. 2013. Antibody Against Integrin Lymphocyte Function-Associated Antigen 1 Inhibits HIV Type 1 Infection in Primary Cells Through Caspase-8-Mediated Apoptosis. *AIDS Research and Human Retroviruses* **29**:2, 371-383. [Abstract] [Full Text HTML] [Full Text PDF] [Full Text PDF with Links]
4. Katarzyna Zwolińska, Brygida Knysz, Katarzyna Rybka, Monika Pazgan-Simon, Jacek Gąsiorowski, Maciej Sobczyński, Andrzej Gładysz, Egbert Piasecki. 2013. Protective Effect of CCR5-Δ32 Against HIV Infection by the Heterosexual Mode of Transmission in a Polish Population. *AIDS Research and Human Retroviruses* **29**:1, 54-60. [Abstract] [Full Text HTML] [Full Text PDF] [Full Text PDF with Links]
5. Annalisa Saracino, Laura Monno, Luigia Scudeller, Giuseppe Bruno, Nicoletta Ladisa, Grazia Punzi, Anna Volpe, Antonella Lagioia, Gioacchino Angarano. 2013. X4 viruses are frequently archived in patients with long-term HIV infection but do not seem to influence the "inflamm-aging" process. *BMC Infectious Diseases* **13**:1, 220. [CrossRef]
6. Shinsuke Nakagawa, Victor Castro, Michal Toborek. 2012. Infection of human pericytes by HIV-1 disrupts the integrity of the blood-brain barrier. *Journal of Cellular and Molecular Medicine* **16**:12, 2950-2957. [CrossRef]
7. Laurence Morand-Joubert, Jade Ghosn, Constance Delaugerre, Boniface Giffo, Caroline Solas, Assia Samri, Alexandrina Pinta, Aurore Triglia, François Raffi. 2012. Lack of benefit of 3-month intensification with enfuvirtide plus optimized background regimen (OBR) versus OBR alone in patients with multiple therapeutic failures: The INNOVE study. *Journal of Medical Virology* **84**:11, 1710-1718. [CrossRef]
8. Verónica Briz, Dolores García, Gema Méndez-Lagares, Ezequiel Ruiz-Mateos, Miguel de Mulder, David Moreno-Pérez, M. Luisa Navarro, Juan A. León-Leal, M. Isabel de José, José T. Ramos, M. José Mellado, M. Isabel González-Tomé, Manuel Leal, M. Ángeles Muñoz-Fernández. 2012. High Prevalence of X4/DM-Tropic Variants in Children and Adolescents Infected With HIV-1 by Vertical Transmission. *The Pediatric Infectious Disease Journal* **31**:10, 1048-1052. [CrossRef]
9. Cathia Soulie, Vincent Calvez, Anne-Geneviève Marcelin. 2012. Coreceptor usage in different reservoirs. *Current Opinion in HIV and AIDS* **7**:5, 450-455. [CrossRef]
10. Martin Obermeier, Jori Symons, Annemarie M.J. Wensing. 2012. HIV population genotypic tropism testing and its clinical significance. *Current Opinion in HIV and AIDS* **7**:5, 470-477. [CrossRef]
11. Georgios Pollakis, William A. Paxton. 2012. Use of (alternative) coreceptors for HIV entry. *Current Opinion in HIV and AIDS* **7**:5, 440-449. [CrossRef]
12. Patrick Dorr, Blanda Stammen, Elna van der RystDiscovery and Development of Maraviroc, a CCR5 Antagonist for the Treatment of HIV Infection 196-226. [CrossRef]
13. Christof Geldmacher, Richard A. Koup. 2012. Pathogen-specific T cell depletion and reactivation of opportunistic pathogens in HIV infection. *Trends in Immunology* **33**:5, 207-214. [CrossRef]
14. Mary-Anne Trabaud, Vinca Icard, Caroline Scholtes, Thomas Perpoint, Joseph Koffi, Laurent Cotte, Djamila Makhloufi, Jean Claude Tardy, Patrice André. 2012. Discordance in HIV-1 Co-receptor use prediction by different genotypic algorithms and phenotype assay: Intermediate profile in relation to concordant predictions. *Journal of Medical Virology* **84**:3, 402-413. [CrossRef]
15. Nirjal Bhattarai, Jack T. Stapleton. 2012. GB virus C: the good boy virus?. *Trends in Microbiology* . [CrossRef]

16. Saleta Sierra, Hauke Walter. 2012. Targets for Inhibition of HIV Replication: Entry, Enzyme Action, Release and Maturation. *Intervirology* **55**:2, 84-97. [CrossRef]

17. Quazi Ataher, Simon Portsmouth, Laura A Napolitano, Sybil Eng, Anna Greenacre, Andrew Kambugu, Robin Wood, Sharlaa Badal-Faesen, Randy Tressler. 2012. The epidemiology and clinical correlates of HIV-1 co-receptor tropism in non-subtype B infections from India, Uganda and South Africa. *Journal of the International AIDS Society* **15**:1, 2. [CrossRef]

18. Ivona Pandrea, Alan L. LandayImplications for Therapy 81-132. [CrossRef]

19. Alan J. CannReplication 103-131. [CrossRef]

20. Lavina Gharu, Rajesh Ringe, Jayanta Bhattacharya. 2011. Evidence of extended alternate coreceptor usage by HIV-1 clade C envelope obtained from an Indian patient. *Virus Research* . [CrossRef]

21. C. F. Kelley, R. E. Haaland, P. Patel, T. Evans-Strickfaden, C. Farshy, D. Hanson, K. Mayer, J. L. Lennox, J. T. Brooks, C. E. Hart. 2011. HIV-1 RNA Rectal Shedding Is Reduced in Men With Low Plasma HIV-1 RNA Viral Loads and Is Not Enhanced by Sexually Transmitted Bacterial Infections of the Rectum. *Journal of Infectious Diseases* **204**:5, 761-767. [CrossRef]

22. Jorge Parra, Joaquín Portilla, Federico Pulido, Rainel Sánchez-de la Rosa, Carlos Alonso-Villaverde, Juan Berenguer, José L. Blanco, Pere Domingo, Fernando Dronda, Carlos Galera, Félix Gutiérrez, José M. Kindelán, Hernando Knobel, Manuel Leal, Jose López-Aldeguer, Ana Mariño, Celia Miralles, José Moltó, Enrique Ortega, José A. Oteo. 2011. Clinical Utility of Maraviroc. *Clinical Drug Investigation* **31**:8, 527-542. [CrossRef]

23. H. A. Pilch-Cooper, S. F. Sieg, T. J. Hope, A. Koons, J.-M. Escola, R. Offord, R. S. Veazey, D. E. Mosier, B. Clagett, K. Medvik, J. K. Jadlowsky, M. R. Chance, J. G. Kiselar, J. A. Hoxie, R. G. Collman, N. E. Riddick, V. Mercanti, O. Hartley, M. M. Lederman. 2011. Circulating human CD4 and CD8 T cells do not have large intracellular pools of CCR5. *Blood* **118**:4, 1015-1019. [CrossRef]

24. Xue-qing Chen, Chang Liu, Xiao-hong Kong. 2011. The role of HIV replicative fitness in perinatal transmission of HIV. *Virologica Sinica* **26**:3, 147-155. [CrossRef]

25. K. C. Brown, K. B. Patterson, S. A. Malone, N. J. Shaheen, H. M. Asher Prince, J. B. Dumond, M. B. Spacek, P. E. Heidt, M. S. Cohen, A. D. M. Kashuba. 2011. Single and Multiple Dose Pharmacokinetics of Maraviroc in Saliva, Semen, and Rectal Tissue of Healthy HIV-Negative Men. *Journal of Infectious Diseases* **203**:10, 1484-1490. [CrossRef]

26. LPR Vandekerckhove, AMJ Wensing, R Kaiser, F Brun-Vézinet, B Clotet, A De Luca, S Dressler, F Garcia, AM Geretti, T Klimkait, K Korn, B Masquelier, CF Perno, JM Schapiro, V Soriano, A Sönnerborg, A-M Vandamme, C Verhofstede, H Walter, M Zazzi, CAB Boucher. 2011. European guidelines on the clinical management of HIV-1 tropism testing. *The Lancet Infectious Diseases* **11**:5, 394-407. [CrossRef]

27. Ken Y. C. Chow, Françoise BachelerieCXCR4 as a Therapeutic Target 239-278. [CrossRef]

28. Kerina Duri, White Soko, Felicity Gumbo, Knut Kristiansen, Munyaradzi Mapingure, Babill Stray-Pedersen, Fredrik Muller, and the BHAMC Group. 2011. Genotypic Analysis of Human Immunodeficiency Virus Type 1 env V3 Loop Sequences: Bioinformatics Prediction of Coreceptor Usage Among 28 Infected Mother–Infant Pairs in a Drug-Naive Population. *AIDS Research and Human Retroviruses* **27**:4, 411-419. [Abstract] [Full Text HTML] [Full Text PDF] [Full Text PDF with Links]

29. Alan Perelson, Ariel Weinberger. 2011. Persistence and emergence of X4 virus in HIV infection. *Mathematical Biosciences and Engineering* **8**:2, 605-626. [CrossRef]

30. Valence M. K. Ndesendo, Viness Pillay, Yahya E. Choonara, Lisa C. Du Toit, Leith C. R. Meyer, Eckhart Buchmann, Pradeep Kumar, Riaz A. Khan. 2011. In vivo evaluation of the release of zidovudine and polystyrene sulfonate from a dual intravaginal bioadhesive polymeric device in the pig model. *Journal of Pharmaceutical Sciences* **100**:4, 1416-1435. [CrossRef]

31. K. Allers, G. Hutter, J. Hofmann, C. Loddenkemper, K. Rieger, E. Thiel, T. Schneider. 2011. Evidence for the cure of HIV infection by CCR5 32/ 32 stem cell transplantation. *Blood* **117**:10, 2791-2799. [CrossRef]

32. Ines Frank, Melissa Robbiani. 2011. Attachment and Fusion Inhibitors Potently Prevent Dendritic Cell-Driven HIV Infection. *JAIDS Journal of Acquired Immune Deficiency Syndromes* **56**:3, 204-212. [CrossRef]
33. N. Brieu, P. Portales, M.-J. Carles, P. Corbeau. 2011. Interleukin-7 induces HIV type 1 R5-to-X4 switch. *Blood* **117**:6, 2073-2074. [CrossRef]
34. Steven J. Reynolds, Pascal O. Bessong, Thomas C. QuinnHuman Retroviral Infections in the Tropics 541-558. [CrossRef]
35. S. ALIZON, B. BOLDIN. 2010. Within-host viral evolution in a heterogeneous environment: insights into the HIV co-receptor switch. *Journal of Evolutionary Biology* **23**:12, 2625-2635. [CrossRef]
36. Anne-Laure Fiser, Thierry Vincent, Natalie Brieu, Yea-Lih Lin, Pierre Portalès, Clément Mettling, Jacques Reynes, Pierre Corbeau. 2010. High CD4+ T-Cell Surface CXCR4 Density as a Risk Factor for R5 to X4 Switch in the Course of HIV-1 Infection. *JAIDS Journal of Acquired Immune Deficiency Syndromes* **55**:5, 529-535. [CrossRef]
37. Beatriz Perez-Sweeney, Rob DeSalle, John L. Ho. 2010. An introduction to a novel population genetic approach for HIV characterization. *Infection, Genetics and Evolution* **10**:8, 1155-1164. [CrossRef]
38. Conor J. Meehan, Jessica A. Hedge, David L. Robertson, Grace P. McCormack, Simon A.A. Travers. 2010. Emergence, dominance, and possible decline of CXCR4 chemokine receptor usage during the course of HIV infection. *Journal of Medical Virology* **82**:12, 2004-2012. [CrossRef]
39. M. Calado, P. Matoso, Q. Santos-Costa, M. Espirito-Santo, J. Machado, L. Rosado, F. Antunes, K. Mansinho, M.M. Lopes, F. Maltez, M.O. Santos-Ferreira, J.M. Azevedo-Pereira. 2010. Coreceptor usage by HIV-1 and HIV-2 primary isolates: The relevance of CCR8 chemokine receptor as an alternative coreceptor. *Virology* **408**:2, 174-182. [CrossRef]
40. Weijing He, John Castiblanco, Elizabeth A Walter, Jason F Okulicz, Sunil K Ahuja. 2010. Mendelian randomization: potential use of genetics to enable causal inferences regarding HIV-associated biomarkers and outcomes. *Current Opinion in HIV and AIDS* **5**:6, 545-559. [CrossRef]
41. Bo Yang, Sangya Singh, Rafael Bressani, Georgette D. Kanmogne. 2010. Cross-talk between STAT1 and PI3K/AKT signaling in HIV-1-induced blood-brain barrier dysfunction: Role of CCR5 and implications for viral neuropathogenesis. *Journal of Neuroscience Research* **88**:14, 3090-3101. [CrossRef]
42. ChiYu Zhang, Na Ding, KePing Chen, RongGe Yang. 2010. Complex positive selection pressures drive the evolution of HIV-1 with different co-receptor tropisms. *Science China Life Sciences* **53**:10, 1204-1214. [CrossRef]
43. Lewis Kaufman, Michael J. RossBiomarkers of HIV 381-400. [CrossRef]
44. Benedikt Simon, Katharina Grabmeier-Pfistershammer, Armin Rieger, Mario Sarcletti, Brigitte Schmied, Elisabeth Puchhammer-Stöckl. 2010. HIV coreceptor tropism in antiretroviral treatment-naive patients newly diagnosed at a late stage of HIV infection. *AIDS* **24**:13, 2051-2058. [CrossRef]
45. Steven G Deeks, Joseph M McCune. 2010. Can HIV be cured with stem cell therapy?. *Nature Biotechnology* **28**:8, 807-810. [CrossRef]
46. Jonathan Pitcher, Saori Shimizu, Silvia Burbassi, Olimpia Meucci. 2010. Disruption of neuronal CXCR4 function by opioids: Preliminary evidence of Ferritin Heavy Chain as a potential etiological agent in neuroAIDS. *Journal of Neuroimmunology* **224**:1-2, 66-71. [CrossRef]
47. Paul A Volberding, Steven G Deeks. 2010. Antiretroviral therapy and management of HIV infection. *The Lancet* **376**:9734, 49-62. [CrossRef]
48. Panel de expertos de Gesida, Plan Nacional sobre el Sida. 2010. Documento de consenso del Grupo de Estudio de Sida/Plan Nacional sobre el Sida respecto al tratamiento antirretroviral en adultos infectados por el virus de la inmunodeficiencia humana (actualización enero 2010). *Enfermedades Infecciosas y Microbiología Clínica* **28**:6, 362.e1-362.e91. [CrossRef]
49. Luke C Swenson, Richard Boehme, Alexander Thielen, Rachel A McGovern, P Richard Harrigan. 2010. Genotypic determination of HIV-1 tropism in the clinical setting. *HIV Therapy* **4**:3, 293-303. [CrossRef]

50. Vanessa Pirrone, Shendra Passic, Brian Wigdahl, Robert F. Rando, Mohamed Labib, Fred C. Krebs. 2010. A Styrene-alt-Maleic Acid Copolymer Is an Effective Inhibitor of R5 and X4 Human Immunodeficiency Virus Type 1 Infection. *Journal of Biomedicine and Biotechnology* **2010**, 1-11. [CrossRef]
51. Annalisa Saracino, Laura Monno, Donatella C. Cibelli, Grazia Punzi, Gaetano Brindicci, Nicoletta Ladisa, Alessandra Tartaglia, Antonella Lagioia, Gioacchino Angarano. 2009. Co-receptor switch during HAART is independent of virological success. *Journal of Medical Virology* **81**:12, 2036-2044. [CrossRef]
52. Chuanyi Nie, Kei Sato, Naoko Misawa, Hiroko Kitayama, Hisanori Fujino, Hidefumi Hiramatsu, Toshio Heike, Tatsutoshi Nakahata, Yuetsu Tanaka, Mamoru Ito. 2009. Selective infection of CD4+ effector memory T lymphocytes leads to preferential depletion of memory T lymphocytes in R5 HIV-1-infected humanized NOD/SCID/IL-2Rγnull mice. *Virology* **394**:1, 64-72. [CrossRef]
53. Wei Huang, Jonathan Toma, Eric Stawiski, Signe Fransen, Terri Wrin, Neil Parkin, Jeannette M. Whitcomb, Eoin Coakley, Frederick M. Hecht, Steven G. Deeks, Rajesh T. Gandhi, Susan H. Eshleman, Christos J. Petropoulos. 2009. Characterization of Human Immunodeficiency Virus Type 1 Populations Containing CXCR4-Using Variants from Recently Infected Individuals. *AIDS Research and Human Retroviruses* **25**:8, 795-802. [Abstract] [Full Text HTML] [Full Text PDF] [Full Text PDF with Links]
54. P. Frange, J. Galimand, C. Goujard, C. Deveau, J. Ghosn, C. Rouzioux, L. Meyer, M.-L. Chaix. 2009. High frequency of X4/DM-tropic viruses in PBMC samples from patients with primary HIV-1 subtype-B infection in 1996-2007: the French ANRS CO06 PRIMO Cohort Study. *Journal of Antimicrobial Chemotherapy* **64**:1, 135-141. [CrossRef]
55. Kaustuv Banerjee, Sofija Andjelic, Per Johan Klasse, Yun Kang, Rogier W. Sanders, Elizabeth Michael, Robert J. Durso, Thomas J. Ketas, William C. Olson, John P. Moore. 2009. Enzymatic removal of mannose moieties can increase the immune response to HIV-1 gp120 in vivo. *Virology* **389**:1-2, 108-121. [CrossRef]
56. Martin J. Stone, Sara Chuang, Xu Hou, Menachem Shoham, John Z. Zhu. 2009. Tyrosine sulfation: an increasingly recognised post-translational modification of secreted proteins. *New Biotechnology* **25**:5, 299-317. [CrossRef]
57. Cathia Soulié, Roland Tubiana, Anne Simon, Sidonie Lambert-Niclot, Isabelle Malet, Ana Canestri, Christel Brunet, Robert Murphy, Christine Katlama, Vincent Calvez, Anne-Geneviève Marcelin. 2009. Presence of HIV-1 R5 Viruses in Cerebrospinal Fluid Even in Patients Harboring R5X4/X4 Viruses in Plasma. *JAIDS Journal of Acquired Immune Deficiency Syndromes* **51**:1, 60-64. [CrossRef]
58. Andrew A. Lackner, Mahesh Mohan, Ronald S. Veazey. 2009. The Gastrointestinal Tract and AIDS Pathogenesis. *Gastroenterology* **136**:6, 1966-1978. [CrossRef]
59. C. G. Anastassopoulou, T. J. Ketas, P. J. Klasse, J. P. Moore. 2009. Resistance to CCR5 inhibitors caused by sequence changes in the fusion peptide of HIV-1 gp41. *Proceedings of the National Academy of Sciences* **106**:13, 5318-5323. [CrossRef]
60. Mattia C.F. Prosperi, Iuri Fanti, Giovanni Ulivi, Alessandro Micarelli, Andrea De Luca, Maurizio Zazzi. 2009. Robust Supervised and Unsupervised Statistical Learning for HIV Type 1 Coreceptor Usage Analysis. *AIDS Research and Human Retroviruses* **25**:3, 305-314. [Abstract] [Full Text PDF] [Full Text PDF with Links]
61. Jean-Michel Garcia, Anhui Gao, Pei-Lan He, Joyce Choi, Wei Tang, Roberto Bruzzone, Olivier Schwartz, Hugo Naya, Fa-Jun Nan, Jia Li, Ralf Altmeyer, Jian-Ping Zuo. 2009. High-throughput screening using pseudotyped lentiviral particles: A strategy for the identification of HIV-1 inhibitors in a cell-based assay. *Antiviral Research* **81**:3, 239-247. [CrossRef]
62. Ghalib Alkhatib. 2009. The biology of CCR5 and CXCR4. *Current Opinion in HIV and AIDS* **4**:2, 96-103. [CrossRef]
63. Amelia Hughes, Mark Nelson. 2009. HIV entry: new insights and implications for patient management. *Current Opinion in Infectious Diseases* **22**:1, 35-42. [CrossRef]
64. Gilles Peytavin, Vincent Calvez, Christine Katlama. 2009. Les antagonistes du récepteur CCR5 : une nouvelle classe d'antirétroviraux. *Thérapie* **64**:1, 9-16. [CrossRef]

65. Jay A Levy. 2009. HIV pathogenesis: 25 years of progress and persistent challenges. *AIDS* **23**:2, 147-160. [CrossRef]

66. Thorsten Demberg, Marjorie Robert-Guroff. 2009. Mucosal Immunity and Protection Against HIV/SIV Infection: Strategies and Challenges for Vaccine Design. *International Reviews of Immunology* **28**:1-2, 20-48. [CrossRef]

67. Bernhard J. Fromme, Marla Coetsee, Pauline Van Der Watt, Mei-Chi Chan, Karin M. Sperling, Arieh A. Katz, Colleen A. Flanagan. 2008. High-Affinity Binding of Southern African HIV Type 1 Subtype C Envelope Protein, gp120, to the CCR5 Coreceptor. *AIDS Research and Human Retroviruses* **24**:12, 1527-1536. [Abstract] [Full Text PDF] [Full Text PDF with Links]

68. José Alcamí. 2008. Conclusiones y perspectivas. *Enfermedades Infecciosas y Microbiología Clínica* **26**, 49-54. [CrossRef]

69. Fernando Arenzana-Seisdedos. 2008. La entrada viral como diana terapéutica. Situación actual de los inhibidores de la entrada. *Enfermedades Infecciosas y Microbiología Clínica* **26**, 5-11. [CrossRef]

70. Farid Ahmed, Lino Tessarollo, Carol Thiele, Italo Mocchetti. 2008. Brain-derived neurotrophic factor modulates expression of chemokine receptors in the brain. *Brain Research* **1227**, 1-11. [CrossRef]

71. R. S. Veazey, T. A. Ketas, P. J. Klasse, D. K. Davison, M. Singletary, L. C. Green, M. L. Greenberg, J. P. Moore. 2008. Tropism-independent protection of macaques against vaginal transmission of three SHIVs by the HIV-1 fusion inhibitor T-1249. *Proceedings of the National Academy of Sciences* **105**:30, 10531-10536. [CrossRef]

72. Myron S. Cohen, Nick Hellmann, Jay A. Levy, Kevin DeCock, Joep Lange. 2008. The spread, treatment, and prevention of HIV-1: evolution of a global pandemic. *Journal of Clinical Investigation* **118**:4, 1244-1254. [CrossRef]

73. Mariana Manrique, Ewa Micewicz, Pamela A. Kozlowski, Shainn-Wei Wang, Deepti Aurora, Robert L. Wilson, Musie Ghebremichael, Gail Mazzara, David Montefiori, Angela Carville, Keith G. Mansfield, Anna Aldovini. 2008. DNA-MVA Vaccine Protection after X4 SHIV Challenge in Macaques Correlates with Day-of-Challenge Antiviral CD4+ Cell-Mediated Immunity Levels and Postchallenge Preservation of CD4+ T Cell Memory. *AIDS Research and Human Retroviruses* **24**:3, 505-519. [Abstract] [Full Text PDF] [Full Text PDF with Links]

74. Shawn E. Kuhmann, Oliver Hartley. 2008. Targeting Chemokine Receptors in HIV: A Status Report. *Annual Review of Pharmacology and Toxicology* **48**:1, 425-461. [CrossRef]

75. Per Johan Klasse, Robin Shattock, John P. Moore. 2008. Antiretroviral Drug–Based Microbicides to Prevent HIV-1 Sexual Transmission. *Annual Review of Medicine* **59**:1, 455-471. [CrossRef]

76. C Soulié, A Derache, C Aimé, A-G Marcelin, G Carcelain, A Simon, C Katlama, V Calvez. 2008. Comparison of two genotypic algorithms to determine HIV-1 tropism*. *HIV Medicine* **9**:1, 1-5. [CrossRef]

77. Jason D. Barbour, Steven G. DeeksClinical Implications of HIV Fitness and Virulence 161-169. [CrossRef]

78. Gemma Moncunill, Mercedes Armand-Ugón, Eduardo Pauls, Bonaventura Clotet, José A Esté. 2008. HIV-1 escape to CCR5 coreceptor antagonism through selection of CXCR4-using variants in vitro. *AIDS* **22**:1, 23-31. [CrossRef]

79. Mohsin M. Sidat, Anne M. Mijch, Sharon R. Lewin, Jennifer F. Hoy, Jane Hocking, Christopher K. Fairley. 2008. Incidence of putative HIV superinfection and sexual practices among HIV-infected men who have sex with men. *Sexual Health* **5**:1, 61. [CrossRef]

80. Alireza Minagar, Deborah Commins, J. Steven Alexander, Romy Hoque, Francesco Chiappelli, Elyse J. Singer, Behrooz Nikbin, Paul Shapshak. 2008. NeuroAIDS. *Molecular Diagnosis & Therapy* **12**:1, 25-43. [CrossRef]

81. Matthew J Dolan, Hemant Kulkarni, Jose F Camargo, Weijing He, Alison Smith, Juan-Manuel Anaya, Toshiyuki Miura, Frederick M Hecht, Manju Mamtani, Florencia Pereyra, Vincent Marconi, Andrea Mangano, Luisa Sen, Rosa Bologna, Robert A Clark, Stephanie A Anderson, Judith Delmar, Robert J O'Connell, Andrew Lloyd, Jeffrey Martin, Seema S Ahuja, Brian K Agan, Bruce D Walker, Steven G

Deeks, Sunil K Ahuja. 2007. CCL3L1 and CCR5 influence cell-mediated immunity and affect HIV-AIDS pathogenesis via viral entry-independent mechanisms. *Nature Immunology* **8**:12, 1324-1336. [CrossRef]

82. A SARACINO, L MONNO, G PUNZI, D CIBELLI, A TARTAGLIA, L SCUDELLER, G BRINDICCI, A LAGIOIA, G SCOTTO, G ANGARANO. 2007. HIV-1 biological phenotype and predicted coreceptor usage based on V3 loop sequence in paired PBMC and plasma samples. *Virus Research* **130**:1-2, 34-42. [CrossRef]
83. Kurt Van Baelen, Ina Vandenbroucke, Evelien Rondelez, Veerle Van Eygen, Hans Vermeiren, Lieven J. Stuyver. 2007. HIV-1 coreceptor usage determination in clinical isolates using clonal and population-based genotypic and phenotypic assays. *Journal of Virological Methods* **146**:1-2, 61-73. [CrossRef]
84. Thomas Lengauer, Oliver Sander, Saleta Sierra, Alexander Thielen, Rolf Kaiser. 2007. Bioinformatics prediction of HIV coreceptor usage. *Nature Biotechnology* **25**:12, 1407-1410. [CrossRef]
85. Priscilla Biswas, Andrea Galli, Laura Galli, Chiara Tassan Din, Andrea Vecchi, Mauro Malnati, Adriano Lazzarin, Giuseppe Tambussi. 2007. Does cyclosporin A affect CCR5 and CXCR4 expression in primary HIV-1-infected patients?. *Cytometry Part B: Clinical Cytometry* **72B**:6, 433-441. [CrossRef]
86. Cathia Soulié, Anne-Geneviève Marcelin, Jade Ghosn, Bahia Amellal, Lambert Assoumou, Sidonie Lambert, Claudine Duvivier, Dominique Costagliola, Christine Katlama, Vincent Calvez. 2007. HIV-1 X4/R5 co-receptor in viral reservoir during suppressive HAART. *AIDS* **21**:16, 2243-2245. [CrossRef]
87. Anita De Rossi. 2007. Virus???host interactions in paediatric HIV-1 infection. *Current Opinion in HIV and AIDS* **2**:5, 399-404. [CrossRef]
88. Katherine E. Gantlett, Jonathan N. Weber, Quentin J. Sattentau. 2007. Synergistic inhibition of HIV-1 infection by combinations of soluble polyanions with other potential microbicides. *Antiviral Research* **75**:3, 188-197. [CrossRef]
89. Andrew J Low, Winnie Dong, Dennison Chan, Tobias Sing, Ronald Swanstrom, Mark Jensen, Satish Pillai, Benjamin Good, P Richard Harrigan. 2007. Current V3 genotyping algorithms are inadequate for predicting X4 co-receptor usage in clinical isolates. *AIDS* **21**:14, F17-F24. [CrossRef]
90. R REEVES. 2007. Disparate effects of acute and chronic infection with SIVmac239 or SHIV-89.6P on macaque plasmacytoid dendritic cells. *Virology* **365**:2, 356-368. [CrossRef]
91. Thomas J. Ketas, Shawn E. Kuhmann, Ashley Palmer, Juan Zurita, Weijing He, Sunil K. Ahuja, Per Johan Klasse, John P. Moore. 2007. Cell surface expression of CCR5 and other host factors influence the inhibition of HIV-1 infection of human lymphocytes by CCR5 ligands. *Virology* **364**:2, 281-290. [CrossRef]
92. Thomas J. Ketas, Susan M. Schader, Juan Zurita, Esther Teo, Victoria Polonis, Min Lu, Per Johan Klasse, John P. Moore. 2007. Entry inhibitor-based microbicides are active in vitro against HIV-1 isolates from multiple genetic subtypes. *Virology* **364**:2, 431-440. [CrossRef]
93. Eva Poveda, Verónica Briz, Carmen de Mendoza, José Miguel Benito, Angélica Corral, Natalia Zahonero, Sara Lozano, Juan González-Lahoz, Vincent Soriano. 2007. Prevalence of X4 tropic HIV-1 variants in patients with differences in disease stage and exposure to antiretroviral therapy. *Journal of Medical Virology* **79**:8, 1040-1046. [CrossRef]
94. Rajeev Gautam, Anders Chase Carter, Nathalia Katz, Isolde F. Butler, Mary Barnes, Atsuhiko Hasegawa, Marion Ratterree, Guido Silvestri, Preston A. Marx, Vanessa M. Hirsch, Ivona Pandrea, Cristian Apetrei. 2007. In vitro characterization of primary SIVsmm isolates belonging to different lineages. In vitro growth on rhesus macaque cells is not predictive for in vivo replication in rhesus macaques. *Virology* **362**:2, 257-270. [CrossRef]
95. Lily Tsai, Nataliya Trunova, Agegnehu Gettie, Hiroshi Mohri, Rudolf Bohm, Mohammed Saifuddin, Cecilia Cheng-Mayer. 2007. Efficient repeated low-dose intravaginal infection with X4 and R5 SHIVs in rhesus macaque: Implications for HIV-1 transmission in humans. *Virology* **362**:1, 207-216. [CrossRef]
96. Paul R. Gorry, Rebecca L. Dunfee, Megan E. Mefford, Kevin Kunstman, Tom Morgan, John P. Moore, John R. Mascola, Kristin Agopian, Geoffrey H. Holm, Andrew Mehle, Joann Taylor, Michael Farzan, Hui Wang, Philip Ellery, Samantha J. Willey, Paul R. Clapham, Steven M. Wolinsky, Suzanne M. Crowe, Dana

Gabuzda. 2007. Changes in the V3 region of gp120 contribute to unusually broad coreceptor usage of an HIV-1 isolate from a CCR5 Δ32 heterozygote. *Virology* **362**:1, 163-178. [CrossRef]

97. Christopher N. Scanlan, John Offer, Nicole Zitzmann, Raymond A. Dwek. 2007. Exploiting the defensive sugars of HIV-1 for drug and vaccine design. *Nature* **446**:7139, 1038-1045. [CrossRef]
98. BENHUR LEE. 2007. Envelope-Receptor Interactions in Nipah Virus Pathobiology. *Annals of the New York Academy of Sciences* **1102**:1, 51-65. [CrossRef]
99. Mike Westby. 2007. Resistance to CCR5 antagonists. *Current Opinion in HIV and AIDS* **2**:2, 137-144. [CrossRef]
100. Maarja Adojaan, Tarmo Mölder, Andres Männik, Toomas Kivisild, Richard Villems, Tõnu Krispin, Mart Ustav. 2007. High Prevalence of The CCR5Δ32 HIV-Resistance Mutation among Estonian HIV Type 1-Infected Individuals. *AIDS Research and Human Retroviruses* **23**:2, 193-197. [Abstract] [Full Text PDF] [Full Text PDF with Links]
101. Marjorie Pion, Jean-Francois Arrighi, Jiyang Jiang, Christopher A Lundquist, Oliver Hartley, Christopher Aiken, Vincent Piguet. 2007. Analysis of HIV-1-X4 Fusion with Immature Dendritic Cells Identifies a Specific Restriction that Is Independent of CXCR4 Levels. *Journal of Investigative Dermatology* **127**:2, 319-323. [CrossRef]
102. Andrew A. Lackner, Ronald S. Veazey. 2007. Current Concepts in AIDS Pathogenesis: Insights from the SIV/Macaque Model. *Annual Review of Medicine* **58**:1, 461-476. [CrossRef]
103. Athe M.N. Tsibris, Daniel R. Kuritzkes. 2007. Chemokine Antagonists as Therapeutics: Focus on HIV-1. *Annual Review of Medicine* **58**:1, 445-459. [CrossRef]
104. T. Mugwagwa, G. Witten. 2007. Coreceptor Switching in HIV-1 Subtype B and Subtype C. *Bulletin of Mathematical Biology* **69**:1, 55. [CrossRef]
105. Lucía Pérez-Alvarez, Mercedes Muñoz, Elena Delgado, Celia Miralles, Antonio Ocampo, Valentina García, Michael Thomson, Gerardo Contreras, Rafael Nájera. 2006. Isolation and biological characterization of HIV-1 BG intersubtype recombinants and other genetic forms circulating in Galicia, Spain. *Journal of Medical Virology* **78**:12, 1520-1528. [CrossRef]
106. Fernando Arenzana-Seisdedos, Marc Parmentier. 2006. Genetics of resistance to HIV infection: Role of co-receptors and co-receptor ligands. *Seminars in Immunology* **18**:6, 387-403. [CrossRef]
107. Caitlin Reed, Eric S. Daar. 2006. Novel antiretroviral agents in HIV therapy. *Current Infectious Disease Reports* **8**:6, 489-496. [CrossRef]
108. Eszter Csoma, Tamás Deli, József Kónya, László Csernoch, Zoltán Beck, Lajos Gergely. 2006. Human herpesvirus 6A decreases the susceptibility of macrophages to R5 variants of human immunodeficiency virus 1: Possible role of RANTES and IL-8. *Virus Research* **121**:2, 161-168. [CrossRef]
109. Louis J Picker. 2006. Immunopathogenesis of acute AIDS virus infection. *Current Opinion in Immunology* **18**:4, 399-405. [CrossRef]
110. Marlén M.I. Aasa-Chapman, Keith Aubin, Ian Williams, Áine McKnight. 2006. Primary CCR5 only using HIV-1 isolates does not accurately represent the in vivo replicating quasi-species. *Virology* **351**:2, 489-496. [CrossRef]
111. Eva Poveda, Ver??nica Briz, Miguel Qui??ones-Mateu, Vincent Soriano. 2006. HIV tropism: diagnostic tools and implications for disease progression and treatment with entry inhibitors. *AIDS* **20**:10, 1359??? 1367. [CrossRef]
112. Michael M. Lederman, Robin E. Offord, Oliver Hartley. 2006. Microbicides and other topical strategies to prevent vaginal transmission of HIV. *Nature Reviews Immunology* **6**:5, 371-382. [CrossRef]
113. Leonid Margolis, Robin Shattock. 2006. Selective transmission of CCR5-utilizing HIV-1: the 'gatekeeper' problem resolved?. *Nature Reviews Microbiology* **4**:4, 312-317. [CrossRef]
114. Bhawna Poonia, Steve Nelson, Greg J. Bagby, Ronald S. Veazey. 2006. Intestinal Lymphocyte Subsets and Turnover Are Affected by Chronic Alcohol Consumption. *JAIDS Journal of Acquired Immune Deficiency Syndromes* **41**:5, 537-547. [CrossRef]

115. Maitree Pakarasang, Chantapong Wasi, Surapol Suwanagool, Amphan Chalermchockcharoenkit, Prasert Auewarakul. 2006. Increased HIV-DNA load in CCR5-negative lymphocytes without viral phenotypic change. *Virology* **347**:2, 372-378. [CrossRef]

116. Kevin K. Ariën, Youssef Gali, Abdelkarim El-Abdellati, Leo Heyndrickx, Wouter Janssens, Guido Vanham. 2006. Replicative fitness of CCR5-using and CXCR4-using human immunodeficiency virus type 1 biological clones. *Virology* **347**:1, 65-74. [CrossRef]

117. Steven G Deeks. 2006. Challenges of developing R5 inhibitors in antiretroviral naive HIV-infected patients. *The Lancet* **367**:9512, 711-713. [CrossRef]

118. Zvi Grossman, Martin Meier-Schellersheim, William E Paul, Louis J Picker. 2006. Pathogenesis of HIV infection: what the virus spares is as important as what it destroys. *Nature Medicine* **12**:3, 289-295. [CrossRef]

119. Kelly M. Cheney, Raman Kumar, Adrian Purins, Linda Mundy, Wendy Ferguson, David Shaw, Christopher J. Burrell, Peng LI. 2006. HIV Type 1 Persistence in $CD4^-/CD8^-$ Double Negative T Cells from Patients on Antiretroviral Therapy. *AIDS Research and Human Retroviruses* **22**:1, 66-75. [Abstract] [Full Text PDF] [Full Text PDF with Links]

120. Kalpana Gupta, Per Johan Klasse. 2006. How Do Viral and Host Factors Modulate the Sexual Transmission of HIV? Can Transmission Be Blocked?. *PLoS Medicine* **3**:2, e79. [CrossRef]

121. Jean-Yves Springael, Eneko Urizar, Marc Parmentier. 2005. Dimerization of chemokine receptors and its functional consequences. *Cytokine & Growth Factor Reviews* **16**:6, 611-623. [CrossRef]

122. Francesc Vidal, Joaquim Peraire, Pere Domingo, Montserrat Broch, Hernando Knobel, Enric Pedrol, David Dalmau, Consuelo Vilad??s, Ma Ant??nia Sambeat, Cristina Guti??rrez, Crist??bal Richart. 2005. Lack of Association of SDF-1 3???A Variant Allele With Long-Term Nonprogressive HIV-1 Infection Is Extended Beyond 16 Years. *JAIDS Journal of Acquired Immune Deficiency Syndromes* **40**:3, 276-279. [CrossRef]

123. Ronald S. Veazey, Per Johan Klasse, Susan M. Schader, Qinxue Hu, Thomas J. Ketas, Min Lu, Preston A. Marx, Jason Dufour, Richard J. Colonno, Robin J. Shattock, Martin S. Springer, John P. Moore. 2005. Protection of macaques from vaginal SHIV challenge by vaginally delivered inhibitors of virus–cell fusion. *Nature* **438**:7064, 99-102. [CrossRef]

124. Tina Boadi, Eric Schneider, Stephen Chung, Lily Tsai, Agegnehu Gettie, Marion Ratterree, James Blanchard, A Robert Neurath, Cecilia Cheng-Mayer. 2005. Cellulose acetate 1,2-benzenedicarboxylate protects against challenge with pathogenic X4 and R5 simian/human immunodeficiency virus. *AIDS* **19**:15, 1587???1594. [CrossRef]

125. Jacob Lalezari, Melanie Thompson, Priny Kumar, Peter Piliero, Richard Davey, Kristine Patterson, Anne Shachoy-Clark, Kimberly Adkison, James Demarest, Yu Lou, Michelle Berrey, Stephen Piscitelli. 2005. Antiviral activity and safety of 873140, a novel CCR5 antagonist, during short-term monotherapy in HIV-infected adults. *AIDS* **19**:14, 1443???1448. [CrossRef]

126. Tonie Cilliers, Samantha Willey, W. Mathew Sullivan, Trudy Patience, Pavel Pugach, Mia Coetzer, Maria Papathanasopoulos, John P. Moore, Alexandra Trkola, Paul Clapham, Lynn Morris. 2005. Use of alternate coreceptors on primary cells by two HIV-1 isolates. *Virology* **339**:1, 136-144. [CrossRef]

127. Matthias J. Kleinz, Anthony P. Davenport. 2005. Emerging roles of apelin in biology and medicine. *Pharmacology & Therapeutics* **107**:2, 198-211. [CrossRef]

128. S KRAMERHAMMERLE, I ROTHENAIGNER, H WOLFF, J BELL, R BRACKWERNER. 2005. Cells of the central nervous system as targets and reservoirs of the human immunodeficiency virus. *Virus Research* **111**:2, 194-213. [CrossRef]

129. Melaku Adal, Workenesh Ayele, Dawit Wolday, Kifle Dagne, Tsehaynesh Messele, Tesfaye Tilahun, Ben Berkhout, Shlomo Mayaan, Georgios Pollakis, Wendelien Dorigo-Zetsma. 2005. Evidence of Genetic Variability of Human Immunodeficiency Virus Type 1 in Plasma and Cervicovaginal Lavage in Ethiopian Women Seeking Care for Sexually Transmitted Infections. *AIDS Research and Human Retroviruses* **21**:7, 649-653. [Abstract] [Full Text PDF] [Full Text PDF with Links]

130. Andre J. Marozsan, Shawn E. Kuhmann, Thomas Morgan, Carolina Herrera, Enid Rivera-Troche, Serena Xu, Bahige M. Baroudy, Julie Strizki, John P. Moore. 2005. Generation and properties of a human immunodeficiency virus type 1 isolate resistant to the small molecule CCR5 inhibitor, SCH-417690 (SCH-D). *Virology* **338**:1, 182-199. [CrossRef]

131. Sofia Ribeiro, Richard Horuk. 2005. The clinical potential of chemokine receptor antagonists. *Pharmacology & Therapeutics* **107**:1, 44-58. [CrossRef]

132. Richard Jefferys. 2005. Multidrug-resistant, dual-tropic HIV-1 and rapid progression. *The Lancet* **365**:9475, 1923. [CrossRef]

133. Oliver Hartley, Per Johan Klasse, Quentin J. Sattentau, John P. Moore. 2005. V3: HIV's Switch-Hitter. *AIDS Research and Human Retroviruses* **21**:2, 171-189. [Abstract] [Full Text PDF] [Full Text PDF with Links]

134. Eoin Coakley, Christos J Petropoulos, Jeannette M Whitcomb. 2005. Assessing chemokine co-receptor usage in HIV. *Current Opinion in Infectious Diseases* **18**:1, 9-15. [CrossRef]

135. Polly F Harrison, Trisha L LamphearMicrobicides 190-235. [CrossRef]

136. Patrizia Bagnarelli, Manuela Vecchi, Nicoletta Burighel, Domenico Bellanova, Stefano Menzo, Massimo Clementi, Anita De Rossi. 2004. Genotypic and Phenotypic Correlates of the HIV Type 1 env Gene Evolution in Infected Children with Discordant Response to Antiretroviral Therapy. *AIDS Research and Human Retroviruses* **20**:12, 1306-1313. [Abstract] [Full Text PDF] [Full Text PDF with Links]

137. Steven M. Wolinsky, Ronald S. Veazey, Kevin J. Kunstman, Per Johan Klasse, Jason Dufour, Andre J. Marozsan, Martin S. Springer, John P. Moore. 2004. Effect of a CCR5 inhibitor on viral loads in macaques dual-infected with R5 and X4 primate immunodeficiency viruses. *Virology* **328**:1, 19-29. [CrossRef]

138. Andrea Cara, Maria Teresa Maggiorella, Roberta Bona, Leonardo Sernicola, Silvia Baroncelli, Donatella R.M Negri, Pasqualina Leone, Zahra Fagrouch, Jonathan Heeney, Fausto Titti, Aurelio Cafaro, Barbara Ensoli. 2004. Circular viral DNA detection and junction sequence analysis from PBMC of SHIV-infected cynomolgus monkeys with undetectable virus plasma RNA. *Virology* **324**:2, 531-539. [CrossRef]

139. Joseph E. McDade. 2004. Segue: Brief summaries of articles on pertinent emerging issues published elsewhere. *Emerging Infectious Diseases* **10**:5, 971-972. [CrossRef]

140. Pavel Pugach, Shawn E Kuhmann, Joann Taylor, Andre J Marozsan, Amy Snyder, Thomas Ketas, Steven M Wolinsky, Bette T Korber, John P Moore. 2004. The prolonged culture of human immunodeficiency virus type 1 in primary lymphocytes increases its sensitivity to neutralization by soluble CD4. *Virology* **321**:1, 8-22. [CrossRef]

141. S Staprans, Mark B Feinberg. 2004. The roles of nonhuman primates in the preclinical evaluation of candidate AIDS vaccines. *Expert Review of Vaccines* **3**:4 suppl 1, S5. [CrossRef]