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Mechanisms Involved in Non-progressive HIV Disease

Meriet Mikhail, Bin Wang and Nitin K. Saksena

Retroviral Genetics Laboratory, Center for Virus Research, Westmead Millennium Institute, Westmead Hospital, The University of Sydney, Westmead, Sydney NSW 2145, Australia

Abstract

HIV disease is a culmination of a complex interplay between both viral and host factors. As the underlying biological and molecular mechanisms involved in determining disease status are not fully understood, the relationship between the two can be better extrapolated using long-term non-progressing individuals who harbor the virus but clinically show some form of immunologic control over it. Only a fraction of individuals comprising less than 1% of the total HIV-infected population show no clinical sign of infection for an extended period of time. Continued immunologic characterization of such non-progressing individuals will lead to the delineation of anti-HIV mechanisms and development of immuno-therapeutic modulators for controlling HIV. In this article, we present recent progress made in non-progressive HIV disease, and summarize the vast array of literature on factors and mechanisms which determine the effectiveness of viral and antiviral responses in maintaining a non-progressive state of HIV infection.

Key words

HIV-1. Non-progression. Viral factors. AIDS. Cytokines. Immunological control. Gene defects.

Introduction

Both viral and host factors participate in defining HIV-1 disease progression rates. In infected individuals, the latent phase of HIV infection is characterized by three distinct rates of progression towards HIV disease, which vary from one infected individual to another. One clinical group is termed "rapid progressors" who, following primary infection, deteriorate in their T-cell counts within a period of two to three years from initial infection with the virus, and rapidly progress to AIDS[1]. One of the main features of rapid progression is the maintenance of high plasma viral loads, which do not decline after primary infection. Another group of individuals consists of "typical progressors", whose immune systems remain intact during the early part of infection, but show gradual deterioration over a period of 10-15 years in their T-cell counts in concomitance with increasing plasma viral loads[2]. The third category comprises of individuals who have remained healthy, with normal CD4+ and CD8+ T-cell counts and low but detectable plasma viremia, or below detection viremia. These are termed "long term non-progressors", and represent 8-10% of the total HIV-1 infected population. A considerable number of these individuals remain therapy naive and asymptomatic, with persistently low plasma viremia for >15 years[3]. Investigations into correlations between mode of HIV acquisition and long-term non-progressors show no distinction, revealing the presence of these asymp-

Correspondence to:
Nitin Saksena
Retroviral Genetics Laboratory,
Center for Virus Research,
Westmead Millennium Institute,
Westmead Hospital
Darcy Road, Westmead NSW 2145
Sydney, Australia
Phone: (61-2) 9845 9119
Fax: (61-2) 8824 3572
E-mail: nitin_saksena@wmi.usyd.edu.au

tomatic individuals in all infected categories such as heterosexuals, homosexuals, injecting drug users, and transfusion- and perinatally-acquired cases[4].

Since its first discovery 20 years ago, the various HIV-1 disease-progression rates have shown evidence for yet another category of a rare subset of HIV-infected individuals. These individuals are termed "true non-progressors" and represent 0.8% of the total HIV-infected population[5]. These therapy-naive individuals maintain high CD4+ and CD8+ T-cell counts, undetectable plasma viremia, unculturable virus, very low copies of integrated provirus in the genome, and absence of viral evolution throughout the course of infection[6,7]. The median time of HIV infection in these individuals varies from >15 or 20 years. The rarity of such individuals has rekindled considerable interest in initiating studies on this rare subset of HIV-infected patients who remain resistant to the pathological effects of HIV infection. It is thought that knowledge of the type of anti-HIV immune responses and antiviral factors which remain intact in such chronically-infected non-progressive individuals may assist the development of novel natural therapeutic agents, which can lead to the possible cure or even the permanent remission of HIV disease in others. Here we have discussed the background and overall progress made to date in understanding the natural mechanisms that provide long-term protection against HIV disease development.

What should be the definition of long-term non-progressors?

Since the discovery of HIV-1 twenty years ago, tremendous variability in disease progression rates has been observed in HIV-infected individuals. The large majority of HIV-infected individuals eventually progress to HIV disease; very few HIV-infected, therapy-naive individuals (<1%) do not show any signs of HIV disease for >15 years. Thus according to this definition, only those disease-free, therapy-naive HIV-infected individuals (with a median infection time >15 years), who maintain below-detection plasma viremia (<50 copies), and stable/high CD4+ and CD8+ T-cell counts (>500/mm^3), should be classified as "true non-progressors". This is the subset of HIV-infected individuals who may harbor clues to future HIV vaccines and natural therapeutics that can be used in other HIV patients[5,6].

Epidemiology of HIV-infected long-term non-progressors

Many potential cofactors to disease progression have been investigated, including geographical factors, age, gender, genetics, route of HIV infection, smoking, nutrition, psychology and other infectious diseases. These analyses remained confined to well-defined cohorts of Caucasian men in developed countries, with little information available on differences based on the potential cofactors listed above. Significant between-cohort differences were found in the proportion of long-term non-progressors (LTNPs) in Amsterdam, Vancouver, Sydney, and San Francisco cohorts of homosexual men comprising the intercontinental seroconverter study[8]. However, the variability may have arisen from differences in measurement of CD4+T cell counts[9]. There have been few other studies with direct comparisons of the prevalence of LTNPs by geography. To date no association has been made between ethnicity and disease progression. What remains worth investigating is the epidemiology of LTNPs in developing countries where antiretroviral therapy has only recently been introduced. Such untreated individuals and their future clinical follow-up may yield important information on natural durability of the immune responses in controlling HIV.

Correlation of route of HIV transmission with HIV disease progression

It is clearly evident that new HIV infections are increasing across the globe. However, whether a certain transmission route determines the progression rate or not has not yet been conclusively elucidated. This is especially the case when conducting a comparison of mortality among injecting drug users (IDUs) and transfusion-acquired HIV recipients. Another study carried out by Pérez-Hoyos in 1999 applied the Kaplan and Meier extension in addition to other models for estimation of survival time in different sexes, ages, and year of seroconversion in a cohort of IDU seroconverters from Spain. Results showed HIV median-incubation period of 11 years as previously reported by many scientists[10].

The Italian seroconverter study found a nominally lower proportion of LTNPs among homosexual men than IDUs; however the difference found was not sufficient to achieve statistical significance. Similarly, in a Madrid study comprising of IDUs, women, and homosexual men, LTNPs were more likely to be male with previous injecting drug use history[11]. However, a study conducted by Prins, et al. revealed no significant difference between the mode of acquisition within IDUs and homosexual men obtained from 12 cohorts[12]. The difficulty in establishing mortality rates among IDUs falls predominantly in the substantial pre-AIDS causes of death. This complicates studies comparing the prevalence of non-progression in this risk group since rapidly-progressing IDUs predominantly die due to non-HIV-related causes, leaving the proportion defined as LTNPs among the surviving IDUs spuriously increased. A recent study, which examined HIV prevalence in Thailand in a mixed group of predominantly homosexual, IDU and male clients, revealed that 13.5% percent of 235 HIV-infected Thai patients showed the presence of SDF-1.3A polymorphism. This statistically-significant result suggests that this group of patients may be expected to carry long-term non-progressing disease status after HIV-1 infection[13]. Additionally, there has been no evidence to suggest any effect on disease progression by recreational drug use in homosexual men[14]. Using universal definitions of

non-progression as described previously, only 6 to 8% of hemophiliacs have been classified to be within the range of LTNPs[15,16].

Effect of age at the time of infection on disease progression

The relevance of age at HIV infection in determining the rate to AIDS disease is not yet fully investigated. However, preliminary evidence suggests no significant association to limited ranges of age of the seroconverters. Most of the studies conducted revealed small sample sizes in association to limited ranges of age of the seroconverters. The estimated effect of age on risk of AIDS is calculated to occur at 1.5% per every 10 years of age, with discrepancies occurring for hemophiliacs with a value of 1.6 and a value of 1.4 for homosexual men. The extrapolated median time to AIDS from the Webull model for individuals infected at an age of 20 was 10.2 years, which encompasses the period required for long-term non-progressive status, and a drastic decrease in this prediction for individuals infected at 40 years of age, with a median expected time of 7.4 years till clinical AIDS manifestation[17]. In a study conducted by Pezzotti, et al., no evidence of differences in rate of development of AIDS by exposure category was detected. However, there was a strong tendency for more rapid development in older subjects for the various rates of disease progression. This supports the view that external cofactors do not play a major role in AIDS pathogenesis, but that age is of fundamental importance and warrants further investigation[18].

Correlation of lifestyle cofactors on disease progression

In relation to lifestyle cofactors, studies comparing LTNPs to relatively rapid progressors have found only minor between-group differences. One study of HIV-infected individuals from Madrid reported higher levels of alcohol use in LTNPs compared with rapid progressors[11]; however reduced alcohol consumption as a result of disease progression in the rapid progressor group may contribute to this association. A recent paper published by Balbin, et al. investigated the effect of emotional expression and depth processing of past trauma in delaying the onset of clinical disease[19]. Results obtained revealed an existent correlation between long-term survival in association with medication adherence, perceived stress and social support[20]. However, despite these recent findings, greater sample sizes need to be incorporated in such studies to confirm any of the above mentioned cofactors and their medical relevance in delaying the rate of HIV disease progression.

The overall evidence for epidemiologically-linked cofactors, including geographical location, route of transmission, age, social status, or lifestyle cofactors, has not yet shown strong correlations to determining delayed AIDS progression. Possible correlations between long-term survivors and nutrition, gender, age at the time of infection, and other possible cofactors, warrant the need for further investigations in this field of research.

Viral factors associated with non-progression

The hallmark of HIV is its ability to undergo genetic change due to the lack of proof-reading ability of its reverse transcriptase (RT) protein[21,22]. The genetic variants, which emerge as a consequence of the error-prone RT enzyme, are selected according to conformational constraints imposed by viral structure, function, and immune pressure[23].

A number of viral factors such as plasma viral load[24], enhancement of viral transcription and replication[25-27], emergence of more cytopathic strains[28] and infection with attenuated strains[29-31], have been recognized to play a vital role in progressive and/or non-progressive HIV disease.

Several studies have confirmed that the host-induced control over HIV-1 replication in LTNPs was apparently due to viral loads in plasma and peripheral blood mononuclear cells (PBMC) of orders of magnitude lower than those typically found in subjects with progressive disease[32]. This was in association with low numbers of infected cells that appeared to be infected with a less cytopathic virus strain. Often such strains proved difficult in isolating infectious virus *in vitro*, which correlates with low copies of HIV provirus in cells of aviremic non-progressors with high CD4+ and CD8+ T-cells counts[33]. This obvious control over HIV can be attributed to the degree of virus trapping in the follicular-dendritic cell (FDC) network in the lymph nodes of subjects with long-term non-progressive HIV infection, which paralleled the extent of lymph node-germinal center formation. It is hypothesized that the lower quantity of circulating virus may be reflected in the lower degree of virus trapping in association with the decreased rate of tissue activation observed in LTNPs with low viral loads[34]. LTNPs have also been reported to have lower levels of multiply-spliced RNA[15]. These findings are consistent with those of a large number of published reports, suggesting the rate of disease progression is driven by an increasing viral burden[32-34]. The multicenter hemophilia cohort study revealed that HIV-1 RNA levels during early chronic HIV-1 infection is a strong age-independent predictor of clinical outcome, and low levels define persons with a high probability of long-term AIDS-free survival[35]. While measurement of plasma viremia offers important prognostic information, the mechanism for prolonged control of viral replication in LTNPs remains as yet unclear.

Viral diversity and its correlation with rates of disease progression

Evidence suggests that HIV disease progression results from the attainment of a threshold level of viral antigenic diversity above which the host immune system is non-responsive[36]. Loss of immune

containment and triggering of disease progression have also been attributed to clonal dominance or deceptive imprinting of the immune response toward the virus established in infection. This is partially aided by possible saturating levels of circulating viral antigen[37].

Viral genetic diversity has been shown to have a profound influence on HIV disease development and on rates of disease progression[38-40]. The accumulation of viral sequence diversity is slower in individuals who progress rapidly to AIDS, or in other terms, homogeneity in viral quasispecies is required at the progressive phase of HIV disease[39-41]. In contrast, there is an accumulation of higher genetic diversity over time in infected individuals who either slowly progress or do not progress at all. Although both non-synonymous and synonymous site substitutions increase over time, notable differences are seen between patients with diverse rates of disease progression[38,40]. HIV-infected non-progressing and slowly-progressing individuals show a higher accumulation of non-synonymous base substitutions (that typify positive selection), suggesting that higher genetic diversity is the determinant of slow progression and/or non-progression in HIV disease. Recently, a study by Ross and Rodrigo has further demonstrated, by analyzing eight patients longitudinally, that the broad genetic diversity of HIV-1 in an infected individual is a consequence of site-specific positive selection for diversity, a likely consequence of immune recognition. According to this study, positive selection appears to be a good indicator and predictor of disease duration. This positive selection in long-term progressors persisted over time and appears to be associated with helper T-cell epitopes. In contrast, sites under positive selection shift from one time point to another in normal progressors. Thus, a broad and persistent immunologic response is associated with a slower rate of disease progression or non-progressive HIV disease[5,42]. In contrast, individuals who mount a limited and shifting immunologic response to HIV have fewer and less persistent positively selected sites, and progress more rapidly to AIDS.

Non-coding LTR, and structural genes (*gag*, *pol*, and *env*)

Some LTNPs appear to be infected with attenuated viral strains[30,31,43-45,] resulting in possible impaired replication kinetics, low viral plasma levels and a consequent delay in disease progression.

Analysis of HIV-1 LTR region sequences from LTNPs has shown tight conservation of functional motifs such as binding sites for NF-kβ, SP1 and the viral trans-activator *Tat* and an absence of any gross deletions and insertions[46]. The only exception is the SBBC cohort in which all six individuals were infected with a *Nef*-LTR-defective virus through blood transfusion[44].

One report outlining several unusual polymorphisms in the HIV genome derived from longitudinal sequences obtained from eight LTNPs implicated a single deletion in the *env* gp41 gene[47]. Also in this study, five of eight individuals showed no defects in the *gag* gene, while three individuals harbored a defective *nef* gene, and one showed a 4 amino acid insertion in the *vpu* region[47]. No reports have documented *pol* gene polymorphisms in relation to long-term non-progression or slow progression of HIV disease. However, our recent study provides a compelling documentation of the presence of several stop codons appearing in the p17 and p24 region of the *gag* gene, possibly contributing to the lack of viral evolution in one unique "true non-progressor"[6]. Despite the overwhelming literature available on all the diverse occurrences of HIV-1 mutations, listing all of the reported polymorphisms would require an exhaustive amount of effort, which may not necessarily elude to significant correlating markers of disease progression or non-progression.

Of marked interest is the highly variable envelope gene. Although previously higher peptide variability (3-21%) has been seen in HIV strains derived from LTNPs and slow progressors, as opposed to normal progressors[40], there has been little evidence of *env* gene polymorphisms related to rates of HIV disease. Wang, et al. reported a unique V2 region extension that was identified only in slow and non-progressors[40]. The functional role of this V2 region extension in the same set of patients was recently confirmed to be in the maintenance of CCR5 usage over time in both slow and long-term non-progressing HIV disease[48]. These data imply that certain gene changes, similar to that seen in the *env* V2 region, may play a pivotal role in favoring the selection of less pathogenic viral variants leading to slow and/or non-progressive HIV disease.

Roman, et al. identified uncommon amino acid substitutions in the V3 loop regions of HIV-1 strains obtained from infected patients from Rwanda. The frequency of these variations was greater in LTNPs compared with late-stage patients ($P = 0.006$), particularly in a sequence region that has crucial interactions with the cell surface, and is highly relevant for the host's immune response[49]. These variants might reflect a viral response to a strong immune pressure, or represent attenuated HIV-1 strains infecting LTNPs[49]. Additional reports have also included amino acid substitutions located in the variable loop V3 regions in addition to the conserved regions of the C2 and V4 in sequences obtained from PBMC in slow and non-progressors. These amino acid substitutions were found to be maintained through long-term passage in cell culture and are hence less likely to be reversible[47]. This was also shown to be true in an animal model as reported by Cherpelis, et al., whereby DNA-immunization with a V2-deleted HIV genome was found to elicit protective antibodies in macaques[50].

Relevance of mutations in accessory genes (*vif, vpr*, and *vpu*)

Increasing evidence suggests that HIV-1 *vpr* along with the other accessory genes play a pivotal role in viral pathogenesis, as their functions are closely linked to viral activation, suppression of

immune functions along with depletion of host cells and viral maturation[4]. A comparison of proviral accessory genes conducted between HIV non-progressors and progressors revealed the presence of attenuated viral quasispecies derived especially from LTNPs. This quasispecies derived from LTNPs was characterized by the accumulation of high genetic diversity in HIV accessory genes. The presence of mutated accessory genes in viral populations correlated with their reduced replication, in addition to inducing host immune responses[51].

A growing body of literature suggests that the HIV accessory proteins *Vpr* and *Nef* could be involved in the depletion of $CD4^+$ and non-$CD4^+$ cells, tissue atrophy, and in delaying the death of HIV-infected cells[52,53]. In a recent paper, Somasundaran, et al. reported a Q3R polymorphism in *Vpr* as an independent cytopathogenic determinant, with effects that are uncoupled from viral replication *in vitro* and *in vivo*[52]. It is also evident from a longitudinal study of an HIV-infected non-progressing mother-child pair that polymorphisms contained within the N-terminal α-helix turn-α-helix of *Vpr* may be both unique and a possible contributor to the lack of disease progression of its patients[30,54]. This study showed further evidence of the impact of natural mutations in the *vpr* gene on the functional activities of *Vpr*, such as cell cycle G2 arrest, induction of cell death and nuclear localization. These activities were found to be highly conserved in fast-progressing AIDS patients, but critical substitutions or single amino acid changes seen in HIV strains from non-progressors were shown to impair these functions[54].

The role of the viral infectivity factor (*Vif*) protein in HIV maturation and pathogenesis revolves around cell-to-cell transmission of HIV and HIV replication in primary lymphocytes and macrophages and is largely maintained intact *in vivo*[55,56]. One interesting signature change found located in 16 members of a cohort of 42 LTNPs was at position 132 of the *vif* gene, correlating with the low plasma viral load ranges found existent between the non-progressing members of the study[57]. Though the HIV-1 *nef* gene has been termed as an accessory gene in function, due to the immense literature available on its genetic polymorphism and its correlation to disease progression, we have treated it as a separate entity in this review. This is in order to provide an unbiased picture of *nef* and its correlation with various stages of HIV disease.

Accessory *Nef* gene polymorphisms

Early accounts report no correlation between defective n*ef* genes from patients with divergent rates of disease progression and their corresponding rates of disease progression[45]. This was further confirmed through functional studies conducted by Huang in the same year[58]. Defective *nef* sequences, possibly attributing to AIDS-free status in humans, were first described by Deacon, et al. (1995) in the long-term surviving members of the epidemiologically-linked Sydney Blood Bank Cohort (SBBC). A study by Rhodes and workers showed the presence of a 29 base pair (bp) *nef* deletion, which was related to a reduced pathogenic potential of *nef*-defective viruses[59]. Moreover, recent studies on the SBBC members have shown continuous viral evolution, with larger deletions in the *nef/LTR* region in members having higher plasma viral load[60]. Thus, it appears that larger *nef* deletions in members who harbor these deleted *nef* genes contribute to HIV disease progression, which may be in association with highly activated CD8 T-cells, and strong HIV-specific CTL responses. These results describing *nef* gene alterations over time, and their correlation with disease progression, overlap with a study conducted on Rhesus macaques infected with SIVmac239[61]. Thus, these two findings in human and non-human primate models suggest strong selective pressure to restore expression of truncated *nef* protein.

Another recent account of the suppressive activity of *nef* on viral replication is that of Tobiume, et al., which investigated *nef* sequences in a cohort of 14 Japanese hemophiliacs, showing inefficient enhancement of viral infectivity and CD4 down-regulation by the patient-derived *nef* alleles. They attributed these changes to the non-progressive disease status of the cohort members[62].

Regulatory genes (*Tat* and *Rev*) and their relevance in HIV disease progression

HIV also encodes two essential regulatory proteins, *Tat* and *Rev*, from partial overlapping reading frames, which function as potent post-transcriptional modulators and promoters of nuclear export of viral mRNA, respectively. The work conducted by Shugurova (2002) presented supportive evidence of cell proliferation enhancement under the effect of the *tat* gene, and that this form of differentiation varies in different types of cells[63]. In addition, recent studies have incorporated the use of HIV gene epitopes as initiators for the immune system. One such report examined serum anti-*Tat* and anti-*Vpr* IgG by ELISA in a cohort of HIV-1 seropositive slow/non-progressors and fast-progressors, and in seronegative controls. Findings revealed that higher levels of serum anti-*Tat* IgG, but not anti-*Vpr* IgG, were associated with maintenance of non-progression status in HIV-1 infection[64]. This provided evidence that vaccination with the *Tat* toxoid induces humoral immune responses to *Tat* similar to those observed in stable non-progressors, which is encouraging for future vaccine strategies. In addition to genetic polymorphisms and their possible contribution to future vaccine developments, Shugurova, et al. additionally revealed the significance of regulatory genes in the induction of cell-specific changes in growth and morphology conducted in different rat cell types. Hence, the possibility that the regulatory effects of its HIV-1 accessory proteins are not only host-specific, but are also cell-specific, is significant in wholly understanding the mechanisms involved in the journey towards clinical progression to AIDS[63].

Accounts on the effect of attenuated *Rev* proteins in asymptomatic individuals first correlated a mutation at codon 78 causing the amino acid change from Leu

to Ile, which was found to correlate to the non-disease status of LTNP members[65]. Thus, in the light of the large body of data discussed above, it is evident that there are no consensus changes in non-coding LTR, structural, accessory and regulatory genes which can be attributed to non-progression of HIV disease. Under the assumption that HIV causes immune dysfunction by directly destroying cells, it is plausible to think that the cytopathology of HIV variants is one of the major contributors to disease progression. And, at late stages of disease progression highly cytopathic variants are seen. As cytopathic variants arise at late stages of HIV infection, several lines of evidence suggest that this could be a consequence of immune deterioration induced by the non-cytopathic forms (a preferential selection of rapidly growing HIV strains in the face of diminishing immune selection pressure). Furthermore, in one-third to half of the AIDS cases reported, HIV never attains cytopathic form. Another alternative explanation is that it is not the ability of the virulence factor to kill CD4+ T-cells, but rather the relative ability to disrupt regulation of the immune system through a myriad of physiologic signals *in vivo*. Thus, it appears that strain variability, coupled with host-induced divergence over time, may act as a vital determinant of virulence. Therefore, the molecular factors which contribute to rates of HIV disease, should be interpreted in conjunction with host factors. Overall, the identification of viral gene polymorphisms and their effects on inhibition or attenuation of viral replication in the host cell marks them as potential targets for therapeutic intervention and possible incorporation into the design of antiretroviral strategies with greater efficacy and less clinical hazards.

Rare molecular changes in true non-progressive HIV disease

All the studies done to date have shown that viral gene changes between infected individuals are highly variable. Strain-specific variation and defects in various genes are commonly seen in a given host. Thus different HIV strains, with different gene defects appearing in different individuals, is a likely possibility that may or may not be incumbent upon the similar outcome and/or the rates of HIV disease progression. Recently, a comprehensive analysis of a rare true non-progressor, which emphasizes the relevance of viral attenuation in non-progression, has shown a significant difference between this rare individual and other LTNPs previously identified[5,6]. In this study, a complete lack of viral evolution, and the presence of only a single integrant, was detected for the last 18 years. The only notable molecular changes seen in the viral sequences obtained from this patient were the presence of stop codons in the structural genes p24, RT and p17 and G-A hypermutations. It is thus clear that this is the first individual with a replication-incompetent strain. Despite the rarity of such cases in nature, such findings lead us to believe that permanent remission from HIV disease may be possible in this rare subset of non-progressing HIV infected individuals. These data lend further credence to the role of viral diversity in conjunction with host factors as determinants of HIV disease.

Immunological and host-genetic factors associated with non-progression

Although viral factors in concomitance with host-genetic factors may contribute to some cases of non-progressing or slowly progressing HIV disease, they do not account for all cases of prolonged disease-free survival. The existence of such patients, who display some form of immunological control over the HIV pathogen, provides the rationale for concerted efforts to better understand the host immune response in combating HIV infection[66,67]. Although a variety of host factors have been examined, a significant correlation has been observed among human lymphocyte antigen (HLA) genes. Several studies suggest that specific alleles of the HLA loci are associated with different rates of progression[68,69] and varying susceptibility to HIV infection[70]. Heterozygosity at all HLA class I loci appears to be protective against HIV infection, while class I alleles B35 and Cω4 have been consistently associated with accelerated progression to AIDS disease[69,71]. In a cohort of an HIV-infected Caucasian population, long-term non-progression in 28 to 40% of non-progressors was ascribed to heterozygosity at all HLA class I loci, the absence of alleles B35 and Cω4, or both[72]. The alleles B57 and B27 most consistently have been associated with long-term non-progression. Homozygosity for HLA-Bω4 was also found to correlate with long-term non-progression of HIV disease[73]. In addition, Migueles, et al. have also demonstrated a dramatic association between the HLA B*5701 class I allele (an allele over represented in HIV-infected non-progressors) and non-progressive infection. Eighty-five percent (11 of 13) of long-term non-progressors (normal CD4 counts, <50 copies/ml of plasma) contained this allele, as opposed to only 11% observed in progressors[74,75]. These findings indicate that, within this phenotypically and genotypically distinct cohort, a host immune factor is highly associated with restriction of virus replication and non-progressive disease. Further characterization of qualitative differences in virus-specific responses that distinguish HLA B*57 LTNPs from progressors may ultimately define mechanisms for effective immune-mediated restriction of virus replication. Furthermore, though the studies by Migueles, et al. have shown the biggest association occurring between B27 and B57 and long-term non-progression, more robust associations with long-term non-progression were observed with B14 and C8 alleles[76].

HIV specific cytotoxic T-cell response

Cellular immune responses play a vital role in HIV pathogenesis. Despite seemingly potent cytotoxic T-cell lymphocyte (CTL) responses, HIV-1 is never

completely eliminated and persists for life in certain individuals at the virologic set point[24]. It appears likely that release of HIV virions from infected cells, which are relatively resistant to CTL-mediated killing, or other immune responses, do contribute to low levels of residual viral replication. The ability of HIV to mutate and escape CTL recognition, and generate broad cellular tropism, is considered to confer viral persistence in infected individuals in the face of vigorous HIV-1-specific CTL responses (Fig. 1).

Investigations carried out on long-term non-progressors, in addition to exposed yet uninfected individuals, have suggested that certain aspects of the host immunity may contribute to protection against HIV infection and lead to slow disease progression. Cellular immune response further incorporates the activity of the cytotoxic T-cell response in addition to the T-helper response. With aid from HIV-specific CD4+T lymphocytes (T-helper cells), HIV-specific CD8+ T-cell lymphocytes, the effector arm of the cellular immune system, kill HIV-infected cells, which present viral peptides associated with HLA class I molecules. CTLs are specialized CD8+ T-cell lymphocytes that kill target cells expressing foreign proteins; hosts use this effector arm of the immune system to control intracellular pathogens including HIV. Several studies have revealed that long-term non-progressors elicit vigorous HIV-specific cytotoxic T-cell activity[5,77] throughout the period of their HIV infection. These data demonstrate not just a highly activated immune system, but in combination a broadly directed HIV-specific CTL response in the setting of very low viral loads, and in association with the emergence of viral escape mutants[66]. This was in direct contrast to studies conducted on progressors, which showed expression of static and narrowly directed CTL responses[78]. Thus, it can be hypothesized that the T-cells in these non-progressors are somewhat less susceptible to HIV infection. However, in a study conducted by Cao, et al., the replication efficiency of the virus in CD8+T cells was relatively similar in the two groups. There was a quantitatively greater suppressive activity mounted by CD8+ T-cells in non-progressors, as opposed to the same response elicited by the progressors[5,32]. It is also accepted that long-term non-progressors elicit a broad immune response by CD8 T-cells against a variety of epitopes, inclusive of HIV[79]. It is equally possible that sustained HIV-specific CTL activity in non-progressors may be merely a manifestation of a preserved cellular immunity due to non-progressive infection, rather than evidence of sustained protective CTL responses[73]. Simply, this explains the loss of CTL activity in progressors as a result of HIV-induced immunosuppression and thus not a direct cause of disease. This postulation, however, was proved incorrect when Klein, et al. provided *in vivo* evidence that CD8+ T-cells play an important role in suppressing SIV replication, which was again confirmed by recent studies[80]. In addition, the decrease in CTL activity, known to associate with HIV progressing individuals, was found to have an inverse correlation with plasma viral load. Hence, to determine whether this association was

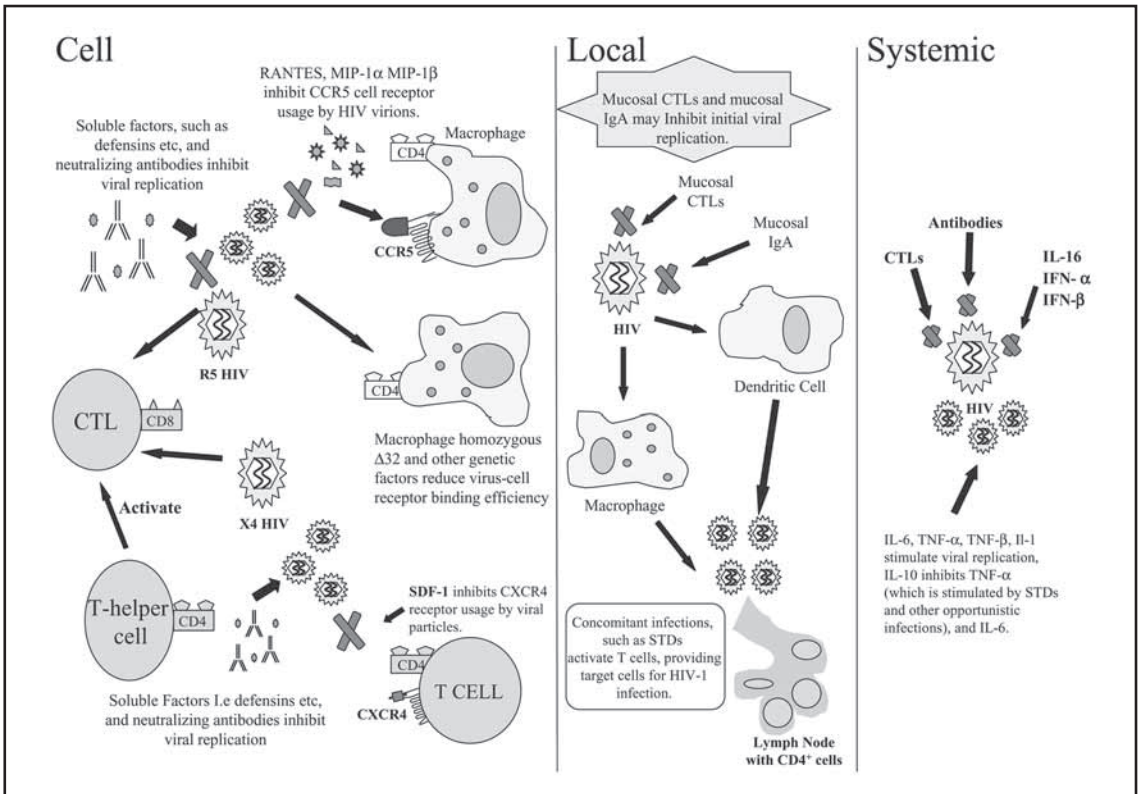

Figure 1. *Schematic overview of host responses to HIV at the cellular, local and systemic levels (adapted from Hogan, et al., 2001).*

due to suppression of HIV by CTLs or suppression of CTLs by HIV, investigations were carried out to further confirm whether increases in viral load were independent from CTL responses and therefore not responsible for CD8 T-cell suppression[81].

There is also evidence to support the role of CTL activity in the protection and prevention of HIV infection, from studies conducted on cell-mediated immune responses of individuals highly exposed to HIV, who have remained uninfected and with detectable HIV-specific CTL responses. This has been observed in a number of cohorts involving heterosexual partners of HIV infected individuals[82], infants of HIV-infected mothers[83], commercial sex workers in Gambia and Kenya[70], and amongst health care workers sustaining needlestick exposure to HIV-infected blood[84]. Therefore, cumulatively, the temporal correlation between vigorous CTL responses and initial control of HIV-1 replication during primary infection, the disproportionate presence of an active CTL response in long-term non-progressors compared with progressors, and evidence of CTL response in exposed yet uninfected persons, suggest a potentially protective role of CTLs in preventing and controlling HIV infection[85].

As seen in a recent study, CTLs were not prominent in the absence of viral replication in a rare true non-progressor[5]. This was consistent with observations of weak CTL activity in other non-progressors with undetectable viremia[86,87], but contrary to several other studies on LTNPs[32,34,66]. In a rare individual studied by Wang, et al. (2002), the high production of IFN-γ (a known marker for CTL), may alternatively suggest that the CD8+ T-cells have other antiviral functions apart from cytolysis. In addition, the CD8+ T-cell IFN-γ responses to some peptides containing immunodominant CTL epitopes were absent (e.g., HLA A2 p17 epitope SLYNTVATL; peptide 20), which further suggested that CTLs were not the major component of anti-HIV responses. Substitution of tyrosine for phenylalanine in position 3 of this immunodominant peptide[6] may have accounted for poor CTL recognition of *Gag* peptide 20. This natural escape variant results in reduced CTL recognition and is an antagonist to the wild-type epitope[88]. Thus, the reason for the lack of weak CTL responses in this subject[5] was the absence of viral evolution and infection with replication-incompetent strain over time[6]. These data were supported by a more pronounced response in another non-progressor, lacking IFN-γ response to all *Gag* peptides[5].

T-helper cell response

Another aspect of HIV-specific cell-mediated immunity is the T-helper cell response, which has been shown to be important in the maintenance of CD8+ T-cell responses in persistent viral infections[89], in addition to being antigen-specific. They are activated when they bind to recognizable viral epitopes presented in the context of MHC class II molecules. Once activated, they secrete interleukin-2 and other cytokines and enhance the CTL and humoral responses. Because HIV selectively infects activated CD4 cells, to some extent T-helper cells may be depleted early in the infection.

A recent study investigated T-helper responses and CTL activity in untreated HIV-infected persons with a wide range of viral loads. T-helper proliferative responses were positively correlated with *gag*-specific CTL activity and negatively correlated with viral load. In addition, levels of CTL precursors seemed to depend on the presence of T-helper function[90]. Thus, both HIV-specific T-helper cells and HIV-specific CTLs may be vital in controlling progression of disease, and persistence of functional CTLs may depend, in part, on preservation of the T-helper response. In the same study, CTLs were occasionally detected in the absence of detectable T-helper function, but these CTLs were not associated with control of viral replication. Thus, high levels of HIV-specific CTLs in persons with progressive disease may be suboptimally effective, partially because a sufficient HIV-specific T-helper response is lacking. This idea may help to explain the ability of HIV to actively replicate in the face of a vigorous CTL response[85].

Studies have shown that some non-progressors with undetectable viral loads demonstrate vigorous CD4-helper responses in addition to persistent CTL responses[91], which indicates the persistence of viral replication, and the presence of antigenic stimulation. Such investigations have proven beneficial by the incorporation of mechanisms maintaining and preserving HIV-specific T-helper cell response in the treatment of acute HIV infections[92]. This form of therapy has been previously reported to preserve HIV-specific T-helper function[93], which in some cases has been associated with preserved HIV-specific cytotoxic T-cell responses[94].

Humoral immunity

The exact role of humoral immune response in delaying disease progression, or preventing HIV infection, has not yet been fully elucidated. However, the available evidence found in published reports indicates that this arm of the immune system is not fully protective, and that cell-mediated immunity, or a combination of cellular and humoral immunity, is crucial in delaying HIV disease. Partial virions, intact virions, and HIV-infected cells appear to be able to stimulate the formation of neutralizing antibodies[95], the titers of which do not correlate to a certain disease status, and hence are not suggestive of protection from AIDS development. Though controversy surrounds the actual mechanisms and function of these neutralizing antibodies, they have been reported to exist in increased frequency and breadth in long-term survivors; however their ability to neutralize clinically-relevant primary or autologous viral isolates varies[96]. Other investigations have revealed no significant difference in relation to breadth and frequency of neutralizing antibodies between progressors and non-progressors[33]. This, however, was in contrast with studies conducted by Harrer, et al. that show non-progressors eliciting weaker or even undetectable neutralizing antibody re-

sponses in comparison to their progressing counterparts[66]. In addition, although exposed yet uninfected individuals have been shown to develop HIV-specific CTL responses, they have not been shown to develop neutralizing responses.

The general mechanism of function for neutralizing antibodies is the protection from pathogens such as viruses or pathogenic toxins through direct interaction by binding and hence neutralizing the virus by blocking access for infection and consequent death of cells. Other functions have also been attributed to this arm of the immune response, including destruction of virions by complement-antibody interactions[97]. Antibody-dependant cell-mediated cytolysis, a method of eliminating virus-infected cells, has been temporally correlated with the control of viremia during acute infection[98], and has been shown *in vitro* to protect PBMCs from infection by cell-associated HIV[99]. Overall, there are conflicting results showing high variability in neutralizing antibodies present in non-progressors, as opposed to their progressor counterparts. Some non-progressors produce significantly higher titers of neutralizating antibodies[100], while others provide no evidence of this activity[101,102].

Chemokines

A breakthrough in our understanding of HIV pathogenesis, and thus in our understanding of host factors that can affect disease progression and susceptibility to HIV-1 infection, was the identification of chemokine coreceptors as being necessary for HIV entry into CD4+ cells. Chemokines are chemoattractant substances secreted at sites of infection or injury[103]. In addition, it was known that CD8 cells secrete substances that interfere with the ability of HIV to infect cells. In 1995, Cocchi, et al. identified these substances as RANTES (regulated on activation, normal T expressed and secreted), MIP-1α (macrophage inflammatory protein-1α) and MIP-1β[104]. It was later confirmed that such substances bound to the same cell receptor which HIV requires for cell entry. The HIV virions partly require chemokine receptors for cellular fusion and consequent entry into the host cell. These chemokine receptors are located on macrophages, monocytes and some T-cells. Corresponding infectious viral strains are referred to as M-tropic strains, which infect cells using CD4 and CCR5, as opposed to T-tropic strains using CD4 and the CXCR4 coreceptor[105,106]. The interaction between the virus envelope protein gp120 and CD4 induces a conformational change that allows interaction between the virus and the chemokine receptor, ultimately leading to fusion of the virus with the host-cell membrane[107].

The ligands for chemokine receptors can block viral entry by interfering with viral binding to the receptor, or by down-regulating the receptor itself[108]. The CCR5-using chemokines-RANTES, MIP-1α, and MIP-1β can block M-tropic viral strains, whereas SDF-1 (stromal cell-derived factor-1: a CXCR4 receptor ligand) blocks T-tropic strains (Fig. 1). CD4 T-cells from exposed yet uninfected individuals have been shown to produce increased levels of RANTES, MIP-1α, and MIP-1β, in addition to significantly suppressing replication of M-tropic strains of HIV-1[109]. Several investigations into such chemokines and coreceptors have revealed their correlation with differing rates of disease progression. The best characterized of these genetic traits is the homozygous and heterozygous CCR5-Δ32 mutation, which was identified in 1996[110]. This mutation results in a truncated protein and both homo- and heterozygosity of this CCR5 mutation correlate well with slow progression of HIV disease. HIV-infected individuals homozygous for the CCR5-Δ32 allele are rare. There was an estimated 3.6% prevalence in an uninfected, but at risk, group in a Multicenter AIDS Cohort Study with a reduced 1.4% prevalence in blood samples from random Caucasian men, and 0% prevalence among randomly collected HIV-infected men[111]. With few exceptions[106,112], most reports have suggested that, although HIV-infected individuals who are CCR5-Δ32 heterozygotes show no difference in susceptibility to infection, they do however show a longer progression period to AIDS or death[111,113,114]. Wang, et al. showed only a partial correlation of CCR5 heterozygosity with disease progression in an epidemiologically-linked mother-child pair[30].

Studies incorporating viral phenotype have suggested that the protective effect of CCR5-Δ32 heterozygosity against disease progression is lost when the infecting strain is syncytium-inducing or T-tropic[115]. Studies conducted by two independent laboratories have shown that CD4+ T-cells from patients who are homozygous or heterozygous for ΔCCR5, can be effectively superinfected with T-tropic or CXCR4 using strains[5,116].

Other mutations have also been identified in the CCR5 receptor and associated with slow and long-term non-progressive HIV disease. One such mutation includes a point mutation involving a T-to-A substitution at position 303, which encodes a truncated protein when found in the compound heterozygotic state with the Δ32 deletion. This produces a phenotype of resistance to HIV-1 primary isolates of the virus *in vitro*[117]. The density of CCR5 receptor has also been an important prognostic indicator, as it reflects rate of HIV-1 infectability, determines *in vivo* HIV production and the rate of CD4 cell decline. Consequently, CCR5 density quantitation could be a new valuable prognostic tool in HIV-1 infection, emphasizing the therapeutic potential of treatments that reduce functional CCR5 density[118]. Findings revealed by Walli, et al. strengthen the hypothesis of a favorable influence of CCR5/delta32-CCR5 genotype on progression of HIV-1 infection, by revealing significantly lower plasma viral loads in CCR5/delta32-CCR5 heterozygous long-term slow progressors relative to that seen in CCR5/CCR5 long-term slow progressors[119]. Recent studies have also suggested an association between slow disease progression and genetic polymorphisms located in the CCR5 regulatory or promoter regions, such as the CCR5 allele 59029-G in combination with homozygosity for 59356-T, a polymorphism more frequently en-

countered in black Africans and Hispanic individuals. Although it appears to be associated with an increased rate of perinatal HIV-1 transmission, there is no biological proof validating this[120].

In addition, another genetic mutation in the CCR2 coreceptor gene (V641) has been found to be associated with a delayed course of HIV disease, but its functional significance remains to be elucidated. Though the CCR2-V641 mutation results in normal CCR2 receptor expression, both heterozygotes and homozygotes appear to affect clinical progression at a much delayed rate as opposed to their progressing counterparts[114,121]. This, however, is preliminary data which was not seen in other similar studies and hence needs to be confirmed in the future[122,123]. The CCR2-V641 allele is found in 10 to 25% of all ethnic groups, and has helped in explaining the slow progression in 21 to 46% of slow progressors seen among commercial sex workers in South Africa. This is distinct from the CCR5-Δ32 mutation, which is predominantly seen in Caucasian men[121].

A third genetic trait that may affect the progression of HIV disease involves the stromal cell derived factor-1 (SDF-1), which is the main ligand for CXCR4, and has been associated as a main blocking factor to infection[124]. The most commonly encountered mutation is that of SDF-1 3'α. This involves a mutation in an untranslated region of the gene, and may up-regulate the synthesis of SDF-1 3'α, allowing competitive inhibition of T-tropic strains of HIV from binding (Fig. 1). Individuals with HIV infection who are homozygous for this mutation have been shown to experience delayed progression to AIDS, but do not exhibit decreased susceptibility to infection with HIV[122,125]. In contrast, other studies have revealed an association with accelerated progression or, in other cases, no particular correlation to clinical outcome for patients with the same polymorphisms[114,126].

Though little can be concluded from chemokine-associated polymorphisms and their correlation to disease progression, one can still elucidate some contribution to the delay of clinical disease outcome, and hence the rate of AIDS progression.

Cytokines

Chemokines are a small subset of the broader more complex network of cytokines, which are polypeptides secreted by cells of the immune system and are hence involved in immunoregulation[85]. Such an array of cytokines, whether stimulatory or inhibitory or both, helps to determine the extent of viral replication occurring within the host. Examples of such cytokines include tumor necrosis factor-α, tumor necrosis factor-β, interleukin-1, and interleukin-6, all of which are proinflammatory cytokines whose levels are elevated in HIV-infected individuals. Perhaps the most important and potent of all the HIV-inducing cytokines activates NF-kβ, which is a cellular transcription factor that induces the expression of HIV[127]. In contrast, interferon-α, interferon-β, and interleukin-16 are known for their ability to suppress HIV replication[128]. Other cytokines, such as interleukin-2, interleukin-4, interleukin-10, transforming growth factor-β, and interferon-γ, have been shown to induce or suppress HIV expression, depending on the experimental conditions[91,129] (Fig. 1). Recent investigations into the release of cytokines, in particular IFN-γ and IL-2, in association with the CD28 cell subset of CD4 T-cells, have been shown to be associated with long-term non-progression of HIV-1 disease[130].

New research is currently being conducted, investigating a broad range of cytokine production and comparisons between the quantities of immune response between differing groups of progressing patients. One such account examined the level of thymic production of T-cells by assessing levels of TRECs (T-cell-rearrangement excision circles) and IL-7 production in HIV-infected children[131]. Both modulators of the host immune system were present in significantly higher levels in the non-progressing patients as opposed to progressors[131,132].

Due to the discrepancy in experimental parameters used, the exact role of cytokines and chemokines, and the mechanism used for disease control *in vivo,* is not yet fully understood. However, it is acknowledged that interhost variations in the balance of these endogenous cytokines may alter the rate of progression to HIV-related disease. Though no distinct consensus has been reached as to the breadth and quantity of cytokines secreted by long-term non-progressors, they are nevertheless known to play a vital role in the control between protection of the host immune system and viral replication. As most studies to date have analyzed the role of one or two cytokines at a time to study disease progression, a study with simultaneous analysis of multiple cytokines and their genes seems warranted in unveiling a clear picture of the involvement of cytokines in HIV disease.

Other biological and antiviral factors associated with LTNP

In 1986, it was noted by researchers that the depletion of CD8+T cells obtained from peripheral blood mononuclear cells derived from HIV-infected patients resulted in a marked increase in viral replication in the remaining CD4+T cells[85]. This was attributed to soluble suppressive factors that are termed CD8 antiviral factors or simply "CAF"[133]. Investigations were then carried out, revealing that the suppressive activity of these CAF mediators was not fully attributable to the effects of RANTES, MIP-1α, and MIP-1β, and there remains uncertainty as to the elements responsible for this activity[134,135]. Until late 2002, much of the controversy existed over the identification and role of such noncytolytic, non-HLA-restricted CD8-mediated soluble factors in the prevention of HIV acquisition and control over governing the rate of disease progression[134,136]. However, recent work conducted in Dr. David Ho's laboratory indicated that α-defensin 1, 2 and 3 collectively account for much of the anti-HIV-1 activity of CAF that is not attributable to β-chemokines, and which are largely peptide antibiotics secreted principally

by human neutrophils[137]. The inhibitory activity of these moderators occur irrespective of viral phenotype or tropism and these factors are known to be heat and acid stable[138]. Though additional studies are necessary to define the true antiviral potency of α-defensins, their identification as a major contributor to the soluble activity of CD8 in delaying HIV infection in LTNPs warrants further investigations.

Interactions between HIV-1 and the host immune system highlight several complex immune compartments, including the role of heat-shock protein receptors, retinoids, cell-associated ceramide, apoptotic markers, interactions with the complement system and human genes that inhibit viral replication, all of which contribute to determining the rate of HIV disease progression. Heat-shock proteins (HSPs) have been implicated in the cross-presentation of antigens *in vivo*[139]. It has been shown that their receptors, in particular CD91 (also called α_2-macroglobulin), are found in greater quantities in LTNPs. These data suggest an enhanced cross-presentation of HIV, and a consequent stimulation of activated anti-HIV CTLs in these individuals[75,79]. The up-regulation of the CD91 receptor in true long-term non-progressors suggests that further investigations in this field may be useful in future therapeutics and perhaps preventative strategies against HIV[140].

The mechanism by which retonic acids inhibit T-cell receptor (TCR)-mediated death of T-lymphocytes was first demonstrated by Ashwell's group, which provided evidence of a blocking mechanism to the production of the TCR-induced apoptosis peptide Fas-Ligand (FasL)[141]. There are also reports indicating that high *in vitro* concentrations of retinoids selectively inhibit CD4 apoptosis without affecting CD8 apoptotic pathways, which proposes retinoids as possible future apoptotic modulators in HIV disease[142]. Because apoptotic cell death is regarded as the principal mechanism for the depletion of host T-cells during the progression of HIV infection into clinical AIDS, identification and modification of the mediators responsible in this cell killing is vital for further attempts in delaying disease onset. In addition, it was recently shown that ceramide enhances HIV infection, and is up-regulated during *in vitro* infection of cells by HIV-1[143,144]. Hence, as indicated by De Simone, et al., by inhibiting cell-associated ceramide one is able to impair HIV replication and consequently delay progression of HIV disease[143].

Moretti, et al. conducted investigations comparing apoptosis and apoptosis-associated perturbations of peripheral blood lymphocytes during HIV infection between AIDS patients and asymptomatic LTNPs. These studies revealed lower frequencies of apoptosis-related receptors and ligands (Fas, and FasL), with disrupted mitochondrial transmembrane potential, and increased superoxide generation in LTNPs than their progressing counterparts. All such abnormalities were associated with lower levels of caspase-1 activation in LTNPs[145]. Lymphocytes undergoing apoptotic pathways suffer a sequential dysregulation of their mitochondrial function early during the apoptotic process, and this implicates some functional influence of the mitochondrial organelles on the regulation of apoptotic cell death. In addition, lower impairment of these cellular granules, which are responsible for oxidation, will consequently lead to a lower degree of superoxide anion generation. This appears to be a possible contributor to delayed AIDS progression and the improvement of the long-term outcome of HIV infection through protection of host-cell lymphocytes[146]. Other studies investigating the efficiency of function of these apoptotic markers revealed a decrease in function of Fas displayed in patients with delayed disease progression, probably encountered as a consequence of inherited alterations in the Fas-signaling pathway, which may be a novel factor in delayed AIDS progression[147].

Activation of the complement system is initiated under any form of infection by foreign particles. This results in the disposition of C3 molecular fragments on the viral surface, which leads to membrane lysis by attack complexes. For its own survival, HIV has developed a means of resistance through the use of opsonisation with complement fragments for its own advantage. These opsonised virions interact with complement receptor-expressing cells, which are subsequently either infected or retain viral particles on their surface as antigen presenters, promoting virus transmission to other permissive cells. This pathway may be interrupted by the development of complement-derived anaphylatoxins that will consequently, in the future, disrupt complement activation and eventual HIV disease progression[97,148].

Human genes and control of HIV replication

One unique investigation into the role of human genes in inhibiting HIV-1 infection was that conducted by Malim, et al., which revealed the presence of an efficient and specific inhibitor termed CEM15 of HIV-1/Δ *vif* infectivity. This suppressive ability is readily overcome by normal levels of *Vif* during normal HIV-1 replication[149]. CEM15 is a human gene expressed by human T-lymphocytes, which has shown similar homology only to phorbolin-1 and apobec-1, which is a deaminase that is the catalytic subunit of the mammalian apolipoprotein B mRNA editing enzyme. The sequence similarity between CEM15 and cytosine deaminase suggests that CEM15 elicits its response through interaction with HIV-1 RNA[149]. In addition, the dependence of the *vif* protein on the viral genomic RNA for the production of normal replicative-competent virions, is the hypothesized method by which CEM15 may affect Δ *vif* virions. Hence, elevating the suppressive activity of CEM15 by *Vif* may prove promising as a potential candidate for innate antiviral resistance against HIV replication. Its analysis and possible role in non-progressive HIV disease is lacking at this time.

Though the role of the immune system in combating infection, replication and eventual dissemination of HIV in the host is complex, and not yet fully understood. Exploring such different avenues and their effect on HIV disease progression may in future prove beneficial in attenuating viral effects on the host. By disrupting the complement system and its

role in viral incorporation by susceptible cells, and increasing host-derived antiviral factors including cytokines, chemokines, and other soluble factors such as defensins, retinoids, and cell-associated ceramides, one can develop a multi-targeted approach to delaying the onset of clinical AIDS. This will also significantly contribute to the future development of HIV vaccines. Studies have indicated that co-administration of HIV vaccine vectors with vaccina viruses expressing IL-15, but not IL-2, was found to induce longer-lasting cellular immunity[150]. This has provided a promising hope for future clinical trails and development of anti-HIV therapeutics, including a form of innate antiviral resistance through the usage of the human gene CEM15. The anti-viral effects of IL-15 were also reported by Muelerr, et al., indicating that IL-15 potently enhances the survival and effector function of HIV-specific CD8+ T-cells and, therefore, may prove useful in augmenting the antiviral function of these cells[151]. Thus, together, the HIV-infected therapy-naive non-progressors may provide possible clues to a new line of anti-HIV therapeutics.

Conclusions

HIV-infected non-progressors harbor clues to natural therapeutics for HIV. Twenty years after the discovery of the AIDS virus, we have entered a new era of understanding the pathogenesis of HIV infection and appreciate the complexity of its molecular and immune correlates. A trend is becoming apparent that some therapy-naive non-progressing HIV-infected individuals have controlled virus for >20 years through their superior qualitative and quantitative immune and antiviral responses (CAF and defensins), as opposed to their slow and rapidly progressing counterparts. Further, this non-progressive state of infection may be caused by infection with less pathogenic strains. Alternatively, the evolution of a pathogenic variant into a non-pathogenic one can also occur under the active control of the host immune system. Thus, both viral and host factors appear to be crucial for the prolonged remission of HIV disease in non-progressors. As this subset of individuals has the ability to control the virus in the complete absence of antiretroviral therapy, the characterization of natural antiviral factors and the viral strains which infect them may lead to the development of therapeutics and vaccines facilitating permanent remission from HIV disease. This new knowledge may reinforce our conviction that, in order to develop effective HIV therapeutics and vaccines, a clear understanding of viral and immune correlates is imperative. The therapy-naive non-progressors are models for understanding these correlates. Whether non-progressors are genetically programmed to efficiently and successfully control HIV remains unknown. Nonetheless, if we amass a clear understanding of the role of host genetic background in determining HIV disease progression in non-progressors, then immune reconstitution to rebuild a robust immune system using allogenic bone marrow and thymus grafts may be a possible theoretical option.

A critical understanding of the breadth of cellular and humoral responses is needed. Protective anti-HIV neutralization antibodies in non-progressors are extremely important, as this is a key biological basis of antiviral responses against all clades of HIV-1. Moreover, such responses can be translated to commercial applications of such antibodies for other progressing HIV patients.

Although the use of attenuated viral vaccines to control HIV infection has remained controversial, it should be emphasized that a clear understanding and characterization of viral genomes from non-progressing individuals is needed. If such individuals have a common feature, which is represented in strains isolated from such individuals, there may be a stronger case to rekindle the issue of live-attenuated HIV vaccines. Long-term priming of the immune system in such individuals who remain below detection for plasma viremia is an important issue. This may reveal where low, but active, viral replication actually occurs in the body and primes the immune system. As many of these individuals may harbor defective proviral genomes, it is possible that viral antigen coupled with memory cells may drive sustained anti-HIV responses. How this persistent presentation of viral antigens occurs in such individuals may provide deeper insights into future vaccine strategies for the control of HIV. There are no techniques which can unambiguously establish true aviremia in non-progressors (a phenomenon never shown), due to the limitation of most commercially available detection systems. Sensitive techniques to detect a single copy are needed. A clear understanding of the true state of viremia in non-progressors may further facilitate future vaccine design, as it will reveal whether the long-term immune responses in such chronically-infected individuals are due to low viremia or the entrapped antigens.

Overall, the truly HIV non-progressing individuals could serve as important guideposts for future therapeutic and prophylactic efforts against disease development in progressing individuals.

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