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Review

Cross-talk between innate and adaptive immunity in HIV infection: A review of literature

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ABSTRACT

HIV infection is a highly complex phenomenon involving different actors of the immune system like cells and molecules. All the actors interact each other in order to shut down viral replication in infected cells and also to promote the activation of the immune response. Natural Killer (NK) cells represent a key cells in HIV infection by mediating anti-viral immunity through lysing infected cells, producing pro-inflammatory cytokines and modulating adaptive immunity. Dendritic cells (DCs) also are crucial in the generation and regulation of adaptive immunity and are key regulators of the host response to human immunodeficiency virus-1 (HIV-1) infection by driving the activation of T lymphocyte cells and the production of antibodies. When activated, adaptive immunity modulates the innate immune response against HIV through Tregs cells, which have the potential to limit excessive inflammatory immune responses and to reduce tissue damage. Tregs cells can also suppress antimicrobial immune responses and promote pathogen persistence. This crosstalk between innate and adaptive immunity helps to "contain" the infection for many years without symptoms.

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1. Introduction

Global statistics from UNAIDS (United Nations program on HIV and AIDS) indicate nearly 35 million people are currently infected with human immunodeficiency virus (HIV), the causative agent of acquired immunodeficiency syndrome (AIDS) with 2 million people infected each year. Since the first identification of the disease in the early 1980s, over 25 million people have died from AIDS [1].

While therapeutic interventions and prevention education efforts are ongoing, there remains a pressing need for an in-depth understanding of the cooperation between the different actors of immune system and the development of an effective vaccine in order to control this global pandemic.

AIDS is the obvious consequence of HIV-1 infection. Despite different studies on this topic our understanding of the interaction of virus with the immune system and the consequences of their relation are still limited. HIV-1 infection is strongly associated with activation of the immune system. Different studies have shown that the main clinical features of AIDS are CD4+ T cell reduction, excessive immune activation, and rapid

increase in HIV viral load [2, 3]. The defining features of the acquired immunodeficiency are the persistent and profound selective decrease in the function as well as number of T lymphocytes of the helper/inducer subset and a possible activation of the suppressor/cytotoxic subset. Pathogenesis of the disease is considered multifactorial, in that no unique immune alteration has been identified that can fully explain the plethora of dysregulations described so far.

This review presents the various aspects of the HIV-1-associated immune dysregulation that we consider critical for pathogenesis. Some of these immune dysregulations may suggest new directions for immune-based therapy and vaccines.

2. Innate responses in HIV infection

2.1. Protective role of the innate response in HIV-infected patients

Innate immunity against HIV is induced when pathogen recognition receptors of the host cell sense viral products including nucleic acid as "non-self". The immune response against viral infection begins then with the detection of Pathogen-Associated Molecular Patterns (PAMPs) in viral products using host cell Pathogen Recognition Receptors (PRR). This interaction triggers an intrinsic innate immune response that directs antiviral defenses and viral restriction [4].

Different subsets of cells have been described as effectors for innate immune protection in HIV-infected patients. All cells derived from the bone marrow express distinct sets of innate immune receptors, giving them a unique capacity to respond to incoming pathogens. These cells include monocytes/macrophages that are important for antigen clearance; Natural Killer (NK) cells that are responsible for the destruction of the pathogen or pathogen-infected cells; and Dendritic Cells (DCs), the professional antigen-presenting cells, which aim to capture foreign antigens and to present them to the adaptive immune system for the induction of immunological memory [5, 6].

NK cells are the critical antiviral effectors of the innate immune system that can recognize signals of stress, transformation or infection with immediate effector function. These cells can recognize and kill virus-infected cells through different mechanisms. Among these mechanisms there is the direct recognition of viral proteins or virus-induced stress ligands by the activating NK cell receptors, the loss of NK cell inhibitory signals resulting from virus-mediated downregulation of HLA class I molecule in infected cells and the process of antibody-dependent cell-mediated cytotoxicity (ADCC). Moreover, NK cells express Killer Immunoglobulin-like Receptors (KIRs) that heavily influence NK cell activation governed by the integration of activating and inhibitory signals *via* the Human Leukocyte Antigen (HLA) molecules. The interactions between KIRs and HLA molecules set a threshold for NK activity and have been associated with the pace of disease progression [7, 8] and protection against disease acquisition [9, 10]. For HIV-1 infection, pathogen sensing and innate immune induction typically occur in CD4+ target cells, including innate immune cells and CD4+ T cells.

HIV-1 infection results in increased expression of stress signal on infected cells and reduced expression of some HLA class I molecules rendering infected cells more susceptible to NK cell–mediated lysis *via* the interaction between KIR and HLA molecules [11, 12]. In addition to KIRs, NK cells express the FcRIIIA receptor (CD16) that binds to the constant (Fc) domain of IgG antibodies. CD16 engagement is a strong activator of NK cell function, and allows antigen-specific recruitment of NK responses for ADCC mechanism that have been shown to be associated with slower disease progression and the control of HIV-1 infection [13].

NK cells can also be activated *via* the cytokine cascade such as Interleukin IL-12, IL-15 and IL-2. In fact, IL-15 is a master regulator of NK cell maturation, survival, and functional competence. IL-15 preferentially expands cytotoxic NK subsets, with less proliferative stimulus for regulatory T cells than IL-2, shifting the net balance towards activation. This cytokine is secreted primarily by monocytes and macrophages activated by products of viral infection. DCs can also be induced to secrete IL-15 *in vitro* by engagement of CD40 molecules. This IL-15 activity is relevant to HIV cure strategies and enhancing innate surveillance by NK cells [14].

The synergistic effect of IL-15 and IL-12 associated to IL-18 can induce high levels of interferon (IFN)-gamma production by NK cells [15]. IFN-gamma production by NK cells plays a central role in the regulation of immune response, inducing the development of type 1 T cell response. Type 1 T cells, in turn, secrete IL-2 and TNF-alpha [16]. IFN-gamma is in fact the main cytokine produced by NK cells early in the immune response. Although cytokines produced by macrophages and DCs are potent inducers of IFN-gamma secretion by NK cells, these cells can also produce IFN-gamma upon their activation, independently from cytokine stimulation [17]. IFN-gamma is also secreted by T lymphocytes under certain conditions of activation and was originally defined as an antiviral agent. But, its activity is much more general, modulating several aspects of the immune response, including antigen presentation, development of specific activation of NK cells, cell proliferation and apoptosis [18].

Although NK cells can be activated by PRR-PAMPs, KIR-HLA interaction and the cytokine pathway, it seems that the activation of NK cells by virus-encoded PAMPs depends on both the presence and the activation status of macrophages and/or DCs [19].

DCs are professional antigen-presenting cells required for generation of adaptive immunity. They are motile cells, migrating continuously around the body even under steady-state conditions [20-22]. These cells express the CD4 receptor and co-receptors such as CCR5, CXCR4 that are required for HIV fusion and entry into a cell, making them one of the initial target cells for HIV-1 infection at the site of transmission [23, 24].

The role of DCs in HIV pathogenesis is currently an issue of much debate but their motility likely allows virus that has been captured by them to rapidly move to lymphoid tissues, where virus can be efficiently amplified upon encountering permissive target cells, like CD4 T cells and macrophages [25]. At least two major subsets of DCs have been identified; myeloid DCs (mDCs) that originate from myeloid precursors and the plasmacytoid DCs (pDCs), which are thought to share precursor cells with lymphocytes. While mDCs are more frequently found and secrete high levels of IL-12, pDCs have the unique ability to produce high levels of Type I IFN (IFN-alpha) in response to microbial stimuli that are potent inhibitor of early and late stages of HIV replication. pDCs are activated upon recognition of common structural patterns of viruses, particularly single-stranded viral RNA and unmethylated CpG-rich DNA which trigger Toll-like receptors (TLR) 7 and 9, respectively [26]. The response of mDCs and pDCs to HIV differs greatly, as pDCs secrete very high amounts of IFN-alpha in response to HIV and partially mature, whereas mDCs secrete little if any type I IFN and do not mature.

HIV replication is severely limited in DCs and several restrictions factors have been shown to block HIV replication at different stages of infection in DCs [27]. The major component of DCs' responses triggered by viral infections is the production of type I IFN. Type I IFN creates a cellular environment that is hostile to the virus by increasing the degradation of virus RNA, arresting progression through the cell cycle and eventually favouring the apoptotic death of the target cell. Furthermore, type I IFN directly induces the expression of major histocompatibility complex and co-stimulatory molecules on antigen-presenting cells, thus promoting efficient antigen presentation and the priming of adaptive immune responses [28]. Despite the multiple blocks to infection by the innate immune system, HIV was shown to exploit an innate immune signalling pathway to facilitate productive infection of DCs and monocytes/macrophages. Immunodeficiency viruses have taken advantage of a unique capacity of antigen-presenting cells to efficiently capture pathogens and present them to the adaptive immune system to facilitate the infection and the dissemination of the virus to surrounding permissive cells.

2.2. Dysregulation of the innate response in HIV-infected patients

Activation of NK cells and production of type I IFN by pDCs are the main effector arms of innate antiviral responses. Both mechanisms are severely affected during HIV-1 infection, with potential consequences for pathogenesis [29].

2.2.1. Dysregulation of NK cells

Chronic HIV-1 infection alters maturation, distribution and functional capacity of NK cells.

Based on their expression of CD56 molecules, NK cells are divided into subsets and these subsets are thought to represent stages in NK cell differentiation. In healthy subjects, NK cells are divided into two subsets: CD56bright, which represents a minor population with relatively limited cytotoxic capacity and CD56dim, which represents about 90% of the total population of NK cells with strong production of cytokines, less proliferative potential, increased cytotoxic capacity, and progressive acquisition of KIRs and other markers including CD57 during differentiation. A third subset is present in negligible frequencies: the dysfunctional CD56neg NK cell subset [30]. These cells have been difficult to characterize with their limited expression of lineage markers [31]. In immunodeficiency syndromes both CD56bright and CD56dim populations contribute to antiviral immunity. Dysfunctional CD56neg NK cells are defective in the production and secretion of important immune regulatory cytokines such as IFN-gamma, TNF-α and Granulocyte-macrophage colony-stimulating factor (GM-CSF) [19, 32]. The chronic HIV-1 infection and the prolonged cellular activation represent the major factors likely driving the expansion of the dysfunctional CD56neg. These NK cells have a strong negative impact on their interplay with DCs. The expansion of CD56neg NK cells is associated with a reduced ability of NK cells to induce an optimal maturation of DCs, an impaired NK cell-mediated clearance of HIV-1 infected and immature DCs [32, 33]. In addition to the abnormal expression of CD56, chronic exposure to the HIV-1 also induced changes in the NK cell expression of KIRs. The impact of viral replication in inducing these phenotypic abnormalities has also been confirmed by experimental evidence [34]. Thereby, NK cells from those HIV-1 infected patients with low/undetectable levels of viral replication are similar and undistinguishable from the ones from uninfected healthy individuals [13].

NK cells also actively participate in the control of viral replication by releasing β-chemokines, which represent the ligand for the co-receptor CCR5 that facilitates the entry process of HIV in target cell. These β-chemokines could inhibit the entry of HIV-1 in the target cells by preventing the binding of CCR5 with viral envelope [35, 36]. The NK cells from HIV-infected patients secrete low amount of β-chemokines that actively participate in the control of viral replication by inhibiting the entry of HIV-1 into the target cells. This effector function is highly impaired in active and chronic HIV-1 infection [13].

In chronic HIV-1 infection, NK cells also express lower levels of CD16 together with an impaired downstream signal pathway of this FcγRIII. This was directly correlated with a decrease of the ADCC mechanism [37, 38]. Nevertheless, the role of NK cell-mediated ADCC in the pathogenesis of HIV-1 remains controversial and a few studies have shown that NK cells in HIV-1-infected patients remain capable to mediate ADCC [39, 40].

2.2.2. Dysregulation of DCs

There is increasing evidence that HIV selectively modulate immature DCs' function to favor virus spread. In HIV-infected patients, during the acute phase of HIV-1 infection and phases of high viremia, the frequencies of both mDCs and pDCs in blood decrease. The depletion of mDCs in blood correlates closely with the increase of plasma virus load, whereas this correlation is not so clear for pDCs [41, 42]. There are also evidences that frequencies of mDCs in the skin and in the mucosa are also reduced during HIV-1 infection [43-45]. However, in patients with recent HIV infection, an increase in the number of both subtypes of DCs after 12 months of anti-retroviral treatment (ART) was observed. But, in previously treated patients for a median of 54 months with undetectable HIV load, both mDC and pDC and the expression of IL-12 and IFN-alpha were significantly lower when compared with those controlled after only 12 months of ART suggesting a decrease of DCs after a prolonged period of viral load undetectability [46]. These findings together point to a redistribution of DCs during active stages of the infection. Moreover, during acute HIV-1 infection, DCs are found to have reduced expression of the co-stimulatory molecules CD80 and CD86 [44] that may be a direct effect of HIV-1 on DCs or the outcome of lower expression of CD40 ligand, CD28 and/or other receptors on T cells whose interactions are essential for the induction of full maturation of DCs.

Therefore, a downregulation of co-stimulatory molecules could negatively affect antigen presentation and generation of T cell immunity [44].

Considering CD4 expression and cytokine production, infected DCs exhibit a downregulation of CD4 and changed cytokine production profile compared to uninfected DCs [47]. The downregulation of CD4 expression were also found *in vitro* in mDCs exposed to HIV-1 [48]. Moreover chronic immune activation is a hallmark of progressive HIV infection and it was shown that in HIV-infected patients, pDCs accumulate in the gut mucosa and associated lymphoid tissue and contribute to immune activation by secreting inflammatory cytokines [49, 50]. This is characterized by a systemic increase in inflammatory cytokines and high levels of viral replication [51]. Inflammatory cytokine secretion by DCs as type I IFN probably plays a major role in disease progression in HIV-infected patients. Indeed, high plasma titers of type I IFN in acute and chronic infection correlate with disease progression [52], and lymphoid tissue of progressor patients express higher levels of IFN alpha [53]. Importantly, it has been shown that prolonged type I IFN signalling in chronic viral infection induces the expression of inhibitory surface molecules on DCs, secretion of suppressive cytokines and decreased IFN-alpha secretion by CD4+ T cells. Consequently, blockade of type I IFN signaling helped resolve persistent infection [54, 55].

Functional analysis showed that mDC infected by HIV stimulated lower T cell responses [45, 56]. Furthermore, both mDC and pDC from chronically infected individuals were less responsive to toll-like receptor (TLR 7/8) stimulation that play a key role in the innate immune system by ensuring maturation marker upregulation and IFN-alpha secretion [57]. In addition, viral replication leads to T cell death, either *via* infection or indirect mechanisms [58]. This cell death leads to the appearance of apoptotic microparticles in the plasma [51] leading to immunologically active environment that can potently affect DC function.

3. Adaptive immunity in HIV infection

HIV infects and deletes CD4 T cells that normally coordinate the adaptive T cell and B cell response to defend against intracellular pathogens leaving a damaged immune system to contend with a lifelong infection.

3.1. Adaptive immunity against HIV

Antiviral CD8 T cells were first identified as T cells that mediate lysis of virus-infected cells and are often referred to as Cytotoxic T Lymphocytes (CTLs) [59]. During HIV infection, CTLs play a central role in the clearance of infected cells and the control of virus replication. These cells play an important role in control of HIV-1 by direct cytolysis of infected cells and the secretion of factors that suppress viral replication. During acute infection, plasma viral load initially increases followed by a period of control, which directly coincides with the initiation of an HIV-specific CD8+ T cell response. Non-human primate models confirm the critical role of CD8+ T cells in controlling HIV disease progression. SIV (simian's immunodeficiency virus)-infected rhesus macaques with CD8+ T cell depleted present immediately uncontrolled viremia. Upon the reappearance of CD8+ T cells, viral load decreased [60]. This pattern was consistent in both acute and chronic SIV-infected animals. The CD8+ T cell functional properties responsible for control of HIV remain unclear, especially whether the cytokines produced from CD8+ T cells play an active role in limiting HIV replication. In addition of the activity on infected cells lysis, CTLs can use other effector mechanisms including production of IFN-gamma, IL-2, and TNF-alpha [61, 60]. But all CD8+ T cells do not display all functions at all times [62]. Thereby, naive CD8+ T cells are quiescent and require days of antigen stimulation to show these functions [63, 64]; in contrast memory CD8+ T cells respond rapidly producing IFN-gamma within a few hours [65]. The expression of the different functions of CTLs depends also on the state of the infection. For example, the lysis of infected cells is the most important antiviral function of the CD8+ T cells in acute infection whereas during chronic infection, once viral set point is established; the other functions of CD8+ T cells may become more important, although lytic potential may still be essential [66, 67].

Generally, in HIV-1 infected individuals, T cell responses are dominated by CD8 + T cells

compared to CD4+ T cells. CD8+ T cell responses are much stronger than CD4+ T-cell responses, which are damaged by the virus. But persons who progress very slowly to AIDS, in the absence of treatment, make stronger CD4+ T-cell responses and there is a correlation between the strength of CD4+ response and slow progression of disease [68]. In murine models with CD4+ T cells depleted, CD8+ T cell responses are greatly impaired [69-71]. Likewise, cross-sectional data in chronically infected persons indicate a link between strong CD4+ T-cell responses and effective CD8+ T-cell responses [72]. This has been confirmed by more recent data showing that CD4+ T cells are particularly important in maintaining CD8+ responses [73].

3.2. Dysregulation of adaptive immunity in HIV-infected patients

CD4+ T cell lineages are classically divided into either Th1 cells, which initiate a cellular immune response, or Th2 cells, which initiate a humoral immune response. However, CD4+T cells can also become IL-17-producing Th17 cells, immunosuppressive T regulatory cells (Treg), or T follicular helper cells (Tfh). Each of these subsets likely plays an important role in nearly every normal immune response, and in the context of HIV, each of these subsets has probably been shown at some level to be protective, defective, or pathogenic. CD4+ T-cell responses, in particular anti-HIV-1 CD4+ T-cell responses are profoundly altered in HIV-infected patients. Underlying mechanisms include both cellular depletion and functional abnormalities [74]. In acute HIV-1 infection, memory CD4+ T cells are massively depleted from the lymphoid system, particularly in the gut involving both direct targeting by the virus and bystander activation-induced cell death [75, 76]. In addition, impaired production of new cells may also be implicated, as evidenced by defective thymocyte proliferation [68].

Functional CD4+ T-cell abnormalities are well described in HIV-infected individuals. These defects occur early in the course of HIV-1 infection, prior to the decline of circulating CD4+ T-cell numbers [77]. Several abnormalities have been defined *ex vivo* such as decreased polyfunctionality of HIV-specific T CD4+ cells. Activated CD4+ T cells in HIV-infected patients also exhibit decreased proliferation and production of IL-2, but not IFN-gamma and tumor necrosis factor (TNF)-α, and defective upregulation of the activation marker CD40 ligand cytotoxic T-lymphocyte-associated protein 4 (CTLA-4) and programmed cell death protein 1 (PD-1) [78]. There is also evidence that expression of apoptosis markers and the percentage of CD4+ T cells entering apoptosis have increased [53].

T-cell signaling induced by T-cell receptor (TCR) cross-linking, IL-2 or phorbol esters/ionomycin is also attenuated in HIV infected patients. Interestingly, there is a discrepancy between the *ex vivo* picture, in which CD4+ T cells from HIV-infected patients exhibit increased expression of many activation markers, including increased basal level of kinase phosphorylation and CD40L, and the *in vitro* picture, in which patients respond poorly to stimulation [79, 80].

Loss of *in vitro* CD4+ T-cell responses and of *in vivo* delayed-type hypersensitivity is predictive of disease progression and time to death in untreated HIV-1 infection [81]. Though greatly depleted, CD4+ T cells in HIV-infected patients are not entirely absent, but abnormalities in the development of CD8+ T-cell responses could be consistent with partial loss of CD4+ T-cell help, or impaired function of what cells remain [82].

Defect of CD4+ T Cell function may affect the different lineage of T cell lymphocytes. For example, regulatory T (Tregs) cells, identified by expression of CD25 and FOXP3, are important for maintaining homeostasis in the immune system [83]. They help to prevent autoimmunity and limit tissue damage during infection by suppressing activation and effector functions, mainly through expression of IL-10 and TGF-β [84]. In HIV infection, Treg cells can be both helpful and harmful. Tregs have been shown to suppress general immune activation, which has been closely linked to HIV disease progression. However, strong Treg responses may contribute to HIV pathogenesis by suppressing HIV-specific immune responses, particularly effector T cells [85].

Tregs may also contribute to HIV-related gut damage by inhibiting Th17 cell recovery. Examination of T cell populations in the rectal

mucosa show increased percentages of Tregs in chronic progressors compared to elite controllers, which positively correlate with viral loads and immune activation but Treg CD4+ T cell suppression can be blocked by inhibition of IL-13. The deregulation of regulatory T cells (Treg) and Th17 cells in blood and mucosal tissues of HIV-infected patients may result in the breakdown of the mucosal barrier and microbial translocation [86, 87]. On the other hand, T follicular helper cells (Tfh) are found in lymphoid germinal centers and are responsible for driving B cell memory differentiation and plasma cell formation. Tfh are classified by their expression of the transcription factor Bcl-6, CXCR5, ICOS, PD-1 and production of large amounts of IL-21, which is necessary for B cell differentiation [87]. These helper T cells are likely targets for HIV infection given their location in a tissue that undergoes significant remodeling during HIV infection. During HIV infection, IL-21-producing CD4+ T cells are upregulated in the Peripheral Blood Mononuclear Cell population and have been associated with maintenance of the CD8+ T cell pool and control of viremia, but whether these cells are related to Tfh is unclear.

4. The innate immune system modulates adaptive immunity against HIV

NK cells and DCs have a central role in antiviral immunity by modulating the adaptive immune response and hence in HIV infection. These two cells' crosstalk results in maturation of DC. After maturation, DC in turn up regulates NK cell effector function. This is due to the production of cytokine and cell-to-cell contact. IL-12 and IL-18 are conventionally produced cytokines by activated DCs, and these cytokines promote Th1 response with the induction of IFN-gamma production by NK cells [88-91].

Immature DC can also produce type I IFN, which promotes NK cell proliferation and cytotoxicity leading to infected cells lysis. Lysis of infected cells by NK cells provides a microenvironment with apoptotic bodies for immature DCs that will lead to their maturation and promote viral antigen presentation to T cells [92].

With focus on NK cells, recent studies have shown a critical role for NK cells in the shaping of adaptive immune responses. NK cells have emerged as multifunctional effector cells with the potential to control infections and shape adaptive immune responses. NK cells exert immune pressure on HIV and contribute to protective vaccine responses and some phenotypes of immune control [91]. Thus the HIV vaccine literature has identified the critical importance of ADCC, mediated predominantly by NK cells, *via* non-neutralizing antibodies as a pathway to protective vaccine responses [93].

NK cells have also been shown to shape the induction of antibodies that are involved in ADCC mechanism. Indeed, in the lymph node, it has been shown that the elimination of Tfh cells by perforin produced by NK cells disturbs the formation of the germinal center, thus limiting the development of immune memory. Studies carried out in mouse models have shown that in early moments of infection, NK cells can inhibit the generation of long-lived specific memory T and B cells, as well as specific antibodies by eliminating CD4+ activated T cells and Tfh [94]. In fact, in mouse models, the NK cell depletion results in higher numbers of CD4+ and CD8+ memory T cells with increased polyfunctionality during acute infection and higher numbers of antibody-secreting cells. However the control mice showed reduced number of antibody-secreting cells [95]. In addition, the cytotoxic activity of NK cells and their IFN-gamma production can enhance immunoglobulin class switching [96].

5. Adaptative Immunity modulates innate response in HIV patients: Focus on TReg cells

Treg cells have been shown to be essential for the development and the maintenance of peripheral tolerance and immune homeostasis. Indeed, Treg dysfunction is associated with allergy, autoimmunity, cancer or early graft rejection [97]. In the context of infectious diseases, Tregs have the potential to limit excessive inflammatory immune responses, thereby reducing tissue damage, but can also suppress antimicrobial immune responses and promote pathogen persistence [98].

Regulatory T cells have been associated with several roles in HIV infection, which may occur at different times during the infection process and may be affected by ongoing therapy. The negative

roles of Tregs in HIV infection include inhibitory effects on effector T cells during early infection; may serve as possible targets for HIV replication; and may have the ability to suppress HIV-specific responses that can lead to inhibition of T cell responses to HIV and increase viral persistence, leading to immune exhaustion. Possible beneficial roles of Tregs may be their ability to reduce immune activation, particularly in situations of increased lipopolysaccharide (LPS) concentrations, and this restriction of activation of CD4 T cells could limit their loss [99].

According to the peripheral CD4 T-cell depletion associated with progressive HIV infection, absolute numbers of peripheral CD4 Tregs and proportions of Tregs among total CD3 T cells were mainly found to be decreased in HIV-infected patients compared with healthy donors [100]. Thus, results of Treg numbers or frequencies among total T cells could be solely interpreted as a direct consequence of changes in CD4 T-cell counts. In contrast, most studies that assessed the frequency of Tregs among CD4 T cells found an expansion of peripheral Tregs [101].

However little data on Treg cells is available during acute and chronic untreated infection. Treg proportions among CD4 T cells were reported to be lower compared with healthy controls [102]. In contrast, an increase in Treg frequencies was consistently reported in untreated patients with chronic infection compared with healthy donors when Tregs were assessed by flow cytometry [103]. The increased frequency of peripheral and mucosal Tregs, which seems to be a characteristic feature of untreated HIV infection, triggers various effects that are either beneficial or detrimental. The influence of natural Treg cells on disease outcome depends on the equilibrium between a balanced Treg to effector T-cell response and immune activation.

Although Tregs are able to suppress HIV-specific cellular immune responses *in vitro*, there is no clear evidence that these responses are suppressed *in vivo*, at least in viremic patients who exhibit high CD8 T-cell responses. Regarding the control of immune activation, Tregs seem to be somewhat efficient in controlling residual immune activation in patients with ART-mediated viral suppression [104].

6. Conclusion

A broader understanding in the HIV immunology field over the last few years implies the need to study the immune compartments and attributes importance to HIV pathogenesis. Monitoring cellular immune markers that are relevant for control of replication, immune activation and inflammation, and the alteration or exhaustion of the immune functions remains essential. The failure of recent HIV-1 vaccine trials to induce protective immunity in humans has highlighted our lack of understanding of the correlates of immune protection in HIV-1 infection.

AUTHORS' CONTRIBUTIONS

HSR, YHE, and SSG, drafted the sections of the text. NI, KDS, and KE contributed to the discussion. SA and TY read and approved the final manuscript.

CONFLICT OF INTEREST STATEMENT

The authors declare that they have no conflict of interests.

REFERENCES

1. United Nations Program on HIV/AIDS (UNAIDS), Global HIV Statistics. Fact sheet - World AIDS Day. 2019, https://www.unaids.org/en/resources/documents/2019/UNAIDS_FactSheet
2. Petrovas, C., Yamamoto, T., Gerner, M. Y., Boswell, K. L., Wloka, K., Smith, E. C., Ambrozak, D. R., Sandler, N., Timmer, K., Sun, X., Li, P., Poholek, A., Rao, S., Brenchley, J. M., Alam, M., Tomaras, G. D., Roederer, M., Douek, D., Seder, R., Germain, R. N., Haddad, E. K. and Koup, R. A. 2012, J. Clin. Invest., 122(9), 3281-3294.
3. Li, Y. and Sun, W. 2018, Exp. Ther. Med., 15(3), 2879-2883.
4. Rustagi, A. and Gale, M. 2014, J. Mol. Biol., 426(6), 1161-1177.
5. Carrington, M. and Alter, G. 2012, Cold Spring Harbor Perspectives in Medicine, 1, 2(7), a007070.
6. Altfeld, M. and Gale, Jr. M. 2015, Nat. Immunol., 16(6), 554-562.

7. Martin, M. P., Gao, X., Lee, J-H., Nelson, G. W., Detels, R., Goedert, J. J., Buchbinder, S., Hoots, K., Vlahov, D., Trowsdale, J., Wilson, M., O'Brien, S. and Carrington, M. 2007, Nat. Genet., 39(6), 733-740.
8. Martin, M. P., Gao, X., Lee, J-H., Nelson, G. W., Detels, R., Goedert, J. J., Buchbinder, S., Hoots, K., Vlahov, D., Trowsdale, J., Wilson, M., O'Brien, S. J. and Carrington, M. 2002, Nat. Genet., 31(4), 429-434.
9. Boulet, S., Kleyman, M., Kim, J. Y. J., Kamya, P., Sharafi, S., Simic, N., Bruneau, J., Routy, J. P., Tsoukas, C. M. and Bernard, N. F. 2008, AIDS, 22(12), 1487-1491.
10. Boulet, S., Sharafi, S., Simic, N., Bruneau, J., Routy, J-P., Tsoukas, C. M. and Bernard, N. 2008, AIDS, 22(5), 595-599.
11. Cohen, J. 2007, Science, 317(5838), 612-614.
12. Norman, J. M., Mashiba, M., McNamara, L. A., Onafuwa-Nuga, A., Chiari-Fort, E., Shen, W. and Collins, K. L. 2011, Nat. Immunol., 12(10), 975-983.
13. Mikulak, J., Oriolo, F., Zaghi, E., Di Vito, C. and Mavilio, D. 2017, AIDS, 31(17), 2317-2330.
14. Scully, E. and Alter, G. 2016, Curr. HIV/AIDS Rep., 13(2), 85-94.
15. Lusty, E., Poznanski, S. M., Kwofie, K., Mandur, T. S., Lee, D. A., Richards, C. D. and Ashkar, A. 2017, Mol. Immunol., 88, 138-147.
16. Vitale, M., Caruso, A., Licenziati, S., Rodella, L., Fiorentini, S., Zauli, G., Castelli, F., Manzoli, F. and Turano, A. 2000, Cytometry, 39, 189-194.
17. Montoya, C. J., Pollard, D., Martinson, J., Kumari, K., Wasserfall, C., Mulder, C. B., Maria, T., Rugeles, M. T., Atkinson, M. A., Landay, A. L. and Brian, S. W. 2007, Immunology, 122(1), 1-14.
18. Schoenborn, J. R. and Wilson, C. B. 2007, Advan. Immunol., 9, 41-101.
19. Alter, G., Suscovich, T. J., Teigen, N., Meier, A., Streeck, H., Brander, C. and Altfeld, M. 2007, J. Immunol., 178(12), 7658-7666.
20. Romani, N., Ratzinger, G., Pfaller, K., Salvenmoser, W., Stössel, H., Koch, F. and Stoitzner, P. 2001, Internat. Rev. Cytol., 207, 237-270.
21. Randolph, G. J. 2001, Semin. Immunol., 13(5), 267-274.
22. Penna, G., Vulcano, M., Sozzani, S. and Adorini, L. 2002, Hum. Immunol., 63(12), 1164-1171.
23. Oliva, A., Kinter, A. L., Vaccarezza, M., Rubbert, A., Catanzaro, A., Moir, S., Monaco, J., Linda, E. L., Mizell, S., Jackson, R., Li, Y., Romano, J. W. and Fauci, A. S. 1998, J. Clin. Invest., 102(1), 223-231.
24. Granelli-Piperno, A., Moser, B., Pope, M., Chen, D., Wei, Y., Isdell, F., O'Doherty, U., Paxton, W., Koup, R., Mojsov, S., Bhardwaj, N., Clark-Lewis, I., Baggiolini, M. and Steinman, R. 1996, J. Experiment. Med., 184(6), 2433-2438.
25. Teleshova, N., Frank, I. and Pope, M. 2003, J. Leuk. Biol., 74(5), 683-690.
26. Manches, O., Frleta, D. and Bhardwaj, N. 2014, Tren. Immunol., 35(3), 114-122.
27. Iwasaki, A. 2012, Immunity, 37(3), 389-398.
28. Gessani, S., Conti, L., Del Cornò, M. and Belardelli, F. 2014, Toxins, 6(6), 1696-1723.
29. Towers, G. J. and Noursadeghi, M. 2014, Cell Host & Microbe, 16(1), 10-18.
30. Björkström, N. K., Ljunggren, H-G. and Sandberg, J. K. 2010, Tren. Immunol., 31(11), 401-406.
31. Hens, J., Jennes, W. and Kestens, L. 2016, AIDS Res. Ther., 13(1), 15.
32. Mavilio, D., Lombardo, G., Benjamin, J., Kim, D., Follman, D., Marcenaro, E., O'Shea, M. A., Kinter, A., Kovacs, C., Moretta, A. and Fauci, A. S. 2005, Proceedings of the National Academy of Sciences, 102(8), 2886-2891.
33. Tasca, S., Tambussi, G., Nozza, S., Capiluppi, B., Zocchi, M. R., Soldini, L., Veglia, F., Poli, G., Lazzarin, A. and Fortis, C. 2003, AIDS, 17(16), 2291-2298.
34. Mavilio, D., Benjamin, J., Daucher, M., Lombardo, G., Kottilil, S., Planta, M. A., Marcenaro, E., Bottino, C., Moretta, L.,

Moretta, A. and Fauci, A. S. 2003, Proceedings of the National Academy of Sciences, 100(25), 15011-15016.
35. Fauci, A. S., Mavilio, D. and Kottilil, S. 2005, Nat. Rev. Immunol., 5(11), 835-843.
36. Kottilil, S., Chun, T., Moir, S., Liu, S., McLaughlin, M., Hallahan, C. W., Maldarelli, F., Corey, L. and Fauci, A. S. 2003, J. Infect Dis., 187(7), 1038-1045.
37. Forthal, D. N. and Finzi, A. 2018, AIDS, 32(17), 2439-2451.
38. Lichtfuss, G. F., Cheng, W-J., Farsakoglu, Y., Paukovics, G., Rajasuriar, R. and Velayudham, P. 2012, J. Immunol., 189(3), 1491-1499.
39. Katz, J. D., Nishanian, P., Mitsuyasu, R. and Bonavida, B. 1988, J. Clin. Immunol., 8(6), 453-458.
40. Johansson, S. E., Rollman, E., Chung, A. W., Center, R. J., Hejdeman, B., Stratov, I., Hinkula, J., Wahren, B., Kaerre, K., Kent, S. J. and Berg, L. 2011, Viral Immunol., 24(5), 359-368.
41. Donaghy, H., Pozniak, A., Gazzard, B., Qazi, N., Gilmour, J., Gotch, F. and Patterson, S. 2001, Blood, 98(8), 2574-2576.
42. Chehimi, J., Campbell, D. E., Azzoni, L., Bacheller, D., Papasavvas, E., Jerandi, G., Mounzer, J., Kostman, J., Trinchieri, G. and Montaner, L. J. 2002, J. Immunol., 168(9), 4796-4801.
43. Spinillo, A., Tenti, P., Rao, S., Zappatore, R., Romagnoli, S. and Pizzoli, G. 1994, Amer. J. Obstret. Gynecol., 171(3), 773-777.
44. Lore, K. and Larsson, M. 2003, APMIS, 111(7-8), 776-788.
45. Macatonia, S. E., Lau, R., Patterson, S., Pinching, A. J. and Road, W. 1990, Immunology, 71(8), 38-45.
46. Márquez-Coello, M., Montes de Oca Arjona, M., Martín-Aspas, A., Guerrero Sánchez, F., Fernández-Gutiérrez del Álamo, C. and Girón-González, J. A. 2019, Sci. Rep., 9(1), 11654.
47. Smed-Sörensen, A., Loré, K., Walther-Jallow, L., Andersson, J. and Spetz, A-L. 2004, Blood, 104(9), 2810-2817.
48. Patterson, S., Rae, A., Hockey, N., Gilmour, J. and Gotch, F. 2001, J. Virol., 75(14), 6710-6713.
49. Kwa, S., Kannanganat, S., Nigam, P., Siddiqui, M., Shetty, R. D., Armstrong, W., Ansari, A., Bosinger, S. E., Silvestri, G. and Amara, R. R. 2011, Blood, 118(10), 2763-2773.
50. Reeves, R. K., Evans, T. I., Gillis, J., Wong, F. E., Kang, G., Li, Q. and Johnson, R. P. 2012, J. Infect. Dis., 206(9), 1462-1468.
51. McMichael, A. J. 2010, Nat. Rev. Immunol., 10(13), 11-23.
52. von Sydow, M., Sönnerborg, A., Gaines, H. and Strannegård, Ö. 1991, AIDS Res. Hum. Retrovir., 7(4), 375-380.
53. Herbeuval, J-P. 2005, Blood, 106(10), 3524-3531.
54. Wilson, E. B., Yamada, D. H., Elsaesser, H., Herskovitz, J., Deng, J., Cheng, G., Aronow, B. J., Karp, C. L. and Brooks, D. G. 2013, Science, 340(6129), 202-207.
55. Teijaro, J. R., Ng, C., Lee, A. M., Sullivan, B. M., Sheehan, K. C. F., Welch, M., Schreiber, R. D., de la Torre, J. C. and Oldstone, M. B. 2013, Science, 340(6129), 207-211.
56. Roberts, M., Gompels, M., Pinching, A. J. and Knight, S. C. 1994, Immunol. Lett., 43(1-2), 39-43.
57. Martinson, J. A., Roman-Gonzalez, A., Tenorio, A. R., Montoya, C. J., Gichinga, C. N., Rugeles, M. T., Mark, T. M., Kriege, A. M., Ghanekarf, S., Bauma, L. L. and Landay, A. L. 2007, Cell. Immunol., 250(1-2), 75-84.
58. Doitsh, G., Cavrois, M., Lassen, K. G., Zepeda, O., Yang, Z., Santiago, M. L., Hebbeler, A. M. and Greene, W. C. 2010, Cell, 143(5), 789-801.
59. Plata, F., Cerottini, J-C. and Brunner, K. T. 1975, Eur. J. Immunol., 5(4), 227-233.
60. Betts, M. R. 2006, Blood, 107(12), 4781-4789.
61. Streeck, H., Brumme, Z. L., Anastario, M., Cohen, K. W., Jolin, J. S., Meier, A., Brumm, C. J., Rosenberg, E. S., Alter, G., Allen, T. M., Walker, B. D. and Altfeld, M. 2008, PLoS Med., 5(5), 15.

62. Ferrari, G., Korber, B., Goonetilleke, N., Liu, M. K. P., Turnbull, E. L., Salazar-Gonzalez, J. F., Hawkins, N., Watson, S. S. S., Betts, M. R., Gay, C., McGhee, K., Pellegrino, P., Williams, I., Tomaras, G. D., Haynes, B. F., Gray, C. M., Borrow, P., Roederer, M., McMichael, A. J. and Weinhold, K. J. 2011, PLoS Pathog., 7(2), e1001273.
63. Veiga-Fernandes, H., Walter, U., Bourgeois, C., McLean, A. and Rocha, B. 2000, Nat. Immunol., 1(1), 47-53.
64. Peixoto, A., Evaristo, C., Munitic, I., Monteiro, M., Charbit, A., Rocha, B. and Veiga-Fernandes, H. 2007, J. Experiment. Med., 204(5), 1193-1205.
65. Lalvani, A., Brookes, R., Hambleton, S., Britton, W. J., Hill, A. V. S. and McMichael, A. J. 1997, J. Exp. Med., 186(6), 859-865
66. Betts, M. R. and Harari, A. 2008, Curr. Opin. HIV AIDS, 3(3), 349-355.
67. Walker, B. and McMichael, A. 2012, Cold Spring Harbor Perspectives in Medicine, 2(11), a007054.
68. Rosenberg, E. S., Altfeld, M., Poon, S. H., Phillips, M. N., Wilkes, B. M., Eldridge, R. L., Robbins, G. K., D'Aquila, R. T., Goulder, P. J. and Walker, B. D. 2000, Nature, 407(6803), 523-526.
69. Shedlock, D. J. 2003, Science, 300(5617), 337-339.
70. Sun, J. C. 2003, Science, 300(5617), 339-342.
71. Janssen, E. M., Lemmens, E. E., Wolfe, T., Christen, U., von Herrath, M. G. and Schoenberger, S. P. 2003, Nature, 421(6925), 852-856.
72. Kalams, S. A., Buchbinder, S. P., Rosenberg, E. S., Billingsley, J. M., Jones, N. G., Shea, A. K., Trocha, A. K. and Walker, B. D. 1999, J. Virol., 73, 6.
73. Chevalier, M. F., Julg, B., Pyo, A., Flanders, M., Ranasinghe, S., Soghoian, D. Z., Kwon, D. S., Rychert, J., Lian, J., Muller, M. I., Cutler, S., McAndrew, E., Jessen, H., Pereyra, F., Rosenberg, E. S., Altfeld, M., Walker, B. D. and Streeck, H. 2011, J. Virol., 85(2), 733-741.
74. Dion, M-L., Poulin, J-F., Bordi, R., Sylvestre, M., Corsini, R., Kettaf, N., Routy, J. P., Dalloul, A., Boulassel, M. R., Patrice, D. Z. G., Rafick-Pierre, S. and Rémi, C. 2004, Immunity, 21(6), 757-768.
75. Mattapallil, J. J., Douek, D. C., Hill, B., Nishimura, Y., Martin, M. and Roederer, M. 2005, Nature, 434, 1093-1097.
76. Douek, D. C., Roederer, M. and Koup, R. A. 2009, Annu. Rev. Med., 60, 471-484.
77. Shearer, G. M. 1998, Immunity, 9(5), 587-593.
78. Zhang, R., Fichtenbaum, C. J., Hildeman, D. A., Lifson, J. D. and Chougnet, C. 2004, J. Immunol., 172(4), 2678-2686.
79. Alfano, M. and Poli, G. 2001, Curr. Pharm. Desig., 7, 993-1013.
80. Abbas, W. and Herbein, G. 2013, Open Virol. J., 7(1), 57-71.
81. Schweneker, M., Favre, D., Martin, J. N., Deeks, S. G. and McCune, J. M. 2008, J. Immunol., 180(10), 6490-6500.
82. Pitcher, C. J., Quittner, C., Peterson, D. M., Connors, M., Koup, R. A., Maino, V. C. and Picker, J. L. 1999, Nat. Med., 5(5), 518-525.
83. Fontenot, J. D., Gavin, M. A. and Rudensky, A. Y. 2003, Nat. Immunol., 4(4), 330-336.
84. Belkaid, Y. and Rouse, B. T. 2005, Nat. Immunol., 6(4), 353-360.
85. Chase, A. J., Yang, H-C., Zhang, H., Blankson, J. N. and Siliciano, R. F. 2008, J. Virol., 82(17), 8307-8315.
86. Allers, K., Puyskens, A., Epple, H-J., Schürmann, D., Hofmann, J., Moos, V. and Schneider, T. 2016, Mucosal Immunol., 9(1), 265-274.
87. Del Alcazar, D., Wang, Y., He, C., Wendel, B. S., Del Río-Estrada, P. M., Lin, J., Ablanedo-Terrazas, Y., Malone, M. J., Hernandez, S. M., Frank, I., Naji, A., Reyes-Terán, G., Jiang, N. and Su, L. F. 2019, Cell Reports, 28(12), 3047-3060.e7.
88. Flórez-Álvarez, L., Hernandez, J. C. and Zapata, W. 2018, Front. Immunol., 9, 2290.
89. Borg, C., Jalil, A., Laderach, D., Maruyama, K., Wakasugi, H., Charrier, S., Ryffel, B., Cambi, A., Figdor, C., Vainchenker, W., Galy, A., Caignard, A. and Zitvogel, L. 2004, Blood, 104(10), 3267-3275.

90. Gerosa, F., Baldani-Guerra, B., Nisii, C., Marchesini, V., Carra, G. and Trinchieri, G. 2002, J. Experiment. Med., 195(3), 327-333.
91. Elemans, M., Boelen, L., Rasmussen, M., Buus, S. and Asquith, B. 2017, PLoS Pathog., 13(6), e1006361.
92. Alloatti, A., Kotsias, F., Magalhaes, J. G. and Amigorena, S. 2016, Immunol. Rev., 272(1), 97-108.
93. Yang, K., Lan, J., Shepherd, N., Hu, N., Xing, Y., Byrd, D., Amet, T., Jewell, C., Gupta, S., Kounga, C., Gao, J. and Yu, Q. 2015, J. Virol., 89(18), 9393-9406.
94. Alter, G., Teigen, N., Davis, B. T., Addo, M. M., Suscovich, T. J., Waring, M. T., Streeck, H., Johnston, M. N., Staller, K. D., Zaman, M. T., Yu, X. G., Lichterfeld, M., Basgoz, N., Rosenberg, E. S. and Altfeld, M. 2005, Blood, 106(10), 3366-3369.
95. Rydyznski, C., Daniels, K. A., Karmele, E. P., Brooks, T. R., Mahl, S. E., Moran, M. T., Li, C., Sutiwisesak, R., Welsh, R. M. and Waggoner, S. N. 2015, Nat. Commun., 6(1), 6375.
96. Krebs, P., Barnes, M. J., Lampe, K., Whitley, K., Bahjat, K. S., Beutler, B., Edith, J. and Hoebe, K. 2009, Blood, 113(26), 6593-6602.
97. Sakaguchi, S. 2005, Nat. Immunol., 6(4), 345-352.
98. Siewe, B., Stapleton, J. T., Martinson, J., Keshavarzian, A., Kazmi, N., Demarais, P. M., French, A. L. and Landay, A. 2013, J. Leuk. Biol., 93(5), 811-818.
99. Sauce, D., Elbim, C. and Appay, V. 2013, AIDS, 8(2), 125-131.
100. McBrien, J. B., Kumar, N. A. and Silvestri, G. 2018, Eur. J. Immunol., 48(6), 898-914.
101. Kinter, A. L., Hennessey, M., Bell, A., Kern, S., Lin, Y., Daucher, M., Planta, M., McGlaughlin, M., Jackson, R., Ziegler, S. F. and Fauci, A. S. 2004, J. Experiment. Med., 200(3), 331-343.
102. Khaitan, A., Kravietz, A., Mwamzuka, M., Marshed, F., Ilmet, T., Said, S., Ahmed, A., Borkowsky, W. and Unutmaz, D. 2016, J. Acq. Imm. Def. Synd., 72(5), 474-484.
103. Kared, H., Lelièvre, J-D., Donkova-Petrini, V., Aouba, A., Melica, G., Balbo, M., Weiss, L. and Lévy, Y. 2008, AIDS, 22(18), 2451-2460.
104. Chevalier, M. F. and Weiss, L. 2013, Blood, 121(1), 29-37.