Introduction

Human immunodeficiency virus (HIV) is the causative agent of the deadly disease AIDS, which is characterized by the progressive decline of CD4^+ T-cells. The general mechanisms that lead to this decline of HIV-infected CD4^+ T-cells include direct viral cytotoxicity and CTL (cytotoxic T-lymphocyte)-mediated killing of the HIV-infected cells. Virus-encoded protein(s) are known to mediate key signalling events that lead to cell death through several mechanisms, such as apoptosis, pyroptosis, autophagy, activation-induced cell death (AICD) or by the dysregulation of cytokine/chemokine profiles. In most viral infections, including HIV, apoptosis of the infected host cell undergoes modulation (McLean et al., 2008). Apoptosis is a form of programmed cell death leading to a regulated self-destruction of cells. Three well-established molecular pathways have been demonstrated to lead to apoptosis in mammalian cells – the extrinsic (receptor-mediated) pathway, the intrinsic (mitochondrial) pathway and the perforin/granzyme pathway (Czabotar et al., 2014). HIV targets distinct steps in the apoptotic pathway resulting in either activation or inhibition or even a delay in apoptosis of the infected cells (Cummins & Badley, 2014). The modulation of apoptosis during HIV infection results from the interplay between the viral and host cellular proteins. HIV-1-encoded proteins like gp120, Nef, Tat, Vpu, Vpr and protease exhibit anti-apoptotic and/or pro-apoptotic activities depending upon the host cell type and the stage of the HIV life cycle. During the early stages of the HIV life cycle, the viral proteins, particularly gp120, Tat, Nef and Vpr, inhibit apoptosis and favour viral replication, whereas during the advanced stages, these proteins induce apoptosis in both infected and uninfected bystander T-cells with marked host immune suppression (Abbas & Herbein, 2013). The role of various viral proteins is described in detail later in this review. By contrast, in latent reservoirs like resting central memory T-cells, transitional memory T-cells, macrophages, naïve T-cells, follicular dendritic cells, prominent plasma clones, haematopoietic progenitor cells, astrocytes and microglia, there is an inhibition of apoptosis. Hence, a pharmacological reactivation of HIV from the latent state alone is not a sufficient stimulus to push these infected cells towards death as most of these cells are resistant to apoptosis even after viral reactivation (Fernández Larrosa et al., 2008). Thus, latency remains a major challenge for HAART-mediated (highly active antiretroviral therapy) functional cure of HIV infection (Chun et al., 2015). A number of recent reviews have highlighted our current understanding of the molecular mechanisms underlying HIV latency, which is beyond the scope of the current review article (Archin et al., 2014; Dahabieh et al., 2015; Van Lint et al., 2013). The aim of our review is to provide an update on the current understanding of
HIV-mediated modulation of apoptosis

Many viruses trigger apoptosis for cell killing, while several others are known to target a variety of key steps during the apoptotic pathway that lead to a blockade or delay of apoptosis (Galluzzi et al., 2008; McLean et al., 2008). During HIV infections, there is an increased destruction of CD4+ T-cells by soluble or membrane-bound HIV proteins or host immune factors through either direct cytotoxicity of HIV-infected cells or apoptotic/non-apoptotic programmed cell death pathways (Cummins & Badley, 2013; Doitsh et al., 2014; Monroe et al., 2014). HIV infection reduces the circulating CD4+ T-cells by decreased production, increased destruction and effective redistribution. HIV infection contributes to thymic atrophy due to direct cytotoxicity of HIV-infected thymocyte precursors and apoptosis of uninfected immature thymocytes (Garda et al., 2012). HIV also mediates apoptosis of CD34+ multipotent haematopoietic stem cells resulting in a decreased progenitor cell input in the thymus (Carter et al., 2010). Increased apoptosis in all functional compartments of lymph nodes of HIV-infected persons depletes CD8+ and B-cells (van Grevenyenhe et al., 2011). The loss of circulating CD4+ T-cells could also be attributed to their sequestration in the secondary lymphoid tissues (Murooka et al., 2012), accelerated homing of the CD62+ CD4− resting T-cells to lymph nodes, and their exposure to enhanced pro-apoptotic signals (Green et al., 2009).

HIV infections are associated with marked alterations in the expression of several death-inducing ligands including FasL (Fas ligand), TRAIL (TNF-related apoptosis-inducing ligand) and TNF-α. The levels of both soluble and membrane-bound Fas and FasL are enhanced in HIV-infected patients and correlate with disease progression (Ipp et al., 2014). Fas expression is elevated in CD4+ and CD8+ T-cells, B-cells, monocytes, macrophages and NK cells (Milush et al., 2013). Studies have shown that an increased expression of Fas contributes to the bystander apoptosis of uninfected CD4+ T-cells (Nardacci et al., 2015). In addition, HIV-infected CD4+ T-cells exhibit a higher expression of both TRAIL and DR5 (death receptor 5), whereas in HIV-infected dendritic cells and macrophages, only the expression of TRAIL is enhanced (Chehimi et al., 2010; Kumar et al., 2013). TNF-α is a key player in the pathogenesis of HIV infection. It is secreted by activated macrophages and lymphocytes in symptomatic HIV patients and induces diverse responses including inflammation and apoptosis (Herbein & Khan, 2008). TNF-α directly or indirectly modulates uninfected bystander T-cell apoptosis through activation or inhibition of the members of the tumour necrosis family receptor (TNFR) superfamily such as TNFR1, TNFR2 and Fas. TNF-α also stimulates HIV replication in infected cells by activating the transcription factor NF-κB (Kumar et al., 2013). The role of these apoptosis modulator proteins in HIV infection has been depicted in Fig. 1.

Role of HIV-encoded proteins

HIV has evolved mechanisms to induce or inhibit apoptosis to facilitate its own survival. The viral-encoded proteins play a major role in modulating apoptosis depending on their expression levels during the different stages of the viral life cycle as well as the type of cells infected by HIV. In general, the viral proteins prevent apoptosis during early stages to favour viral replication at least until high levels of progeny virions are produced (Fig. 1a). Whereas during the late stages of HIV infection, viral proteins activate apoptosis leading to cell death and facilitate virus release from the infected cells (Fig. 1b). A number of drug candidates are currently being developed, which target the ability of viral proteins to modulate apoptosis. The roles of the viral proteins in manipulating apoptosis are briefly summarized below and as well as depicted in Fig. 1.

Role of gp120

gp120 is the HIV-1 envelope (Env) glycoprotein that binds with the CD4 receptor and the chemokine co-receptors (CXCR4 or CCR5) facilitating viral attachment, and along with gp41 mediates entry into the host cell. During early stages of HIV infection, gp120 inhibits the recycling of CD4 receptors to the cell surface by binding with them in the endoplasmic reticulum. This reduces the cell surface expression of CD4, thereby preventing superinfection. During later stages of HIV infection, binding of membrane-bound or soluble gp120 with CD4 leads to the apoptosis of HIV-infected and uninfected bystander CD4+ T-cells (Li & Pauza, 2013). In addition, soluble gp120 has been reported to induce apoptosis in a variety of cells such as CD8+ T-cells, neurons, human vascular endothelial cells, cardiomyocytes, oral keratinocytes and renal tubular cells (Chen et al., 2013; Février et al., 2011; Green et al., 2014; Vashistha et al., 2009) through several mechanisms as depicted in Fig. 1(b). These include the upregulation of expression of Fas, FasL, TNF-α (Poonia et al., 2009) and TRAIL receptors DR4 and DR5 (Herbeuval et al., 2006), or by acting as a molecular mimic of Fas (Silvestris et al., 1996). HIV Env can induce cell cycle arrest at the G2 phase (Kolesnikchenko et al., 1995) leading to the generation of reactive oxygen species (ROS) (Reshi et al., 2014). gp120 expression reduces both expression of BCL-2 (B-cell lymphoma 2) (Hashimoto et al., 1997) and the phosphorylation of mTOR (mammalian target of rapamycin) and p53 (Castedo et al., 2002). On the contrary, gp120 expression increases the expression of the PUMA (p53 upregulated modulator of apoptosis) protein (Perfetti et al., 2004) and activates p38 MAPK (Del Corno et al., 2001).
**Fig. 1.** Anti-apoptotic and pro-apoptotic functions of HIV-1-encoded proteins. (a) Anti-apoptotic functions. Path 1: Tat decreases susceptibility of cells to Fas, TRAIL and TNFα, upregulates expression of BCL-2 and c-FLIP, and downregulates expression of caspase 10. Path 2: Nef inhibits expression of ASK-1 and p53, and reduces activity of BAD by phosphorylation. Path 3: Vpr upregulates expression of BCL-2 and downregulates BAX expression. (b) Pro-apoptotic functions. Path 1a: fusion of the HIV-1 Env (gp120) with CD4 and/or chemokine receptors leads to an increase in the expression of both soluble and membrane-bound Fas and its ligand FasL; there is also a p53-dependent increase in the expression of BAX and PUMA and downregulation of the expression of BCL-2. Path 1b: Env increases expression of TRAIL and its receptors DR4 and DR5, and TNF-α, resulting in apoptosis through activation of caspase 8/3. Cleavage of BID by active caspase 8
Another apoptosis-inducing mechanism of membrane-bound gp120 involves its interaction with CD4/CXCR4 or CD4/CCR5, which are expressed on the membrane of uninfected HIV-1 target cells. These interactions can result in hemifusion of the two cells leading to syncytia formation that triggers apoptosis through the p53-dependent induction of mitochondrial outer membrane permeabilization (Nardacci et al., 2015). This phenomenon is further described in detail in the section on bystander apoptosis later in this review.

**Role of Tat**

HIV-1 Tat is also produced early in the HIV life cycle and gets secreted by the infected cells. Encoded by two exons, this regulatory protein has a variable length of 86–104 aa. The effect of Tat on apoptosis depends on the levels of the protein. At low levels, Tat inhibits apoptosis in the infected cells during early stages of infection by decreasing their susceptibility to TRAIL, TNF-α and Fas, as well as upregulating expression of BCL-2 and c-FLIP [cellular FLICE (FADD-like IL-1β-converting enzyme)-inhibitory protein] with concomitant downregulation of caspase 10 expression (López-Huertas et al., 2013). Tat also decreases p53 expression, thereby promoting cell cycle progression and cell transformation (Fig. 1a).

In contrast, during the advanced stages of HIV infection, a high level of Tat protein is produced. Tat secreted from the infected cells is taken up by uninfected bystander cells via clathrin-mediated endocytosis (Vendeville et al., 2004). Tat induces apoptosis in infected and uninfected bystander cells by upregulating the expression of FasL, BAX (BCL-2-associated X), caspase 8 and RCAS1 (receptor binding cancer antigen expressed on SiSo cells 1) (Bartz & Emerman, 1999), thereby leading to increased oxidative stress (Nicolini et al., 2001). In addition, binding of Tat with tubulin results in microtubule alteration and BIM (B-cell lymphoma 2 interacting mediator of cell death)-mediated intrinsic apoptosis (de Mareuil et al., 2005) (Fig. 1b).

**Role of Vpu**

The HIV-encoded accessory protein Vpu downregulates the expression of CD4 receptors by binding to the CD4 polypeptide in the endoplasmic reticulum and targeting it to the proteasomes for degradation (Tanaka et al., 2003). Thus, like gp120, Vpu also prevents the superinfection of infected cells and allows efficient budding of newly produced virions through Vpu-mediated antagonism of BST-2 during early stages of viral infection. However, during later stages, the cation-selective ion channel property of Vpu makes cells more responsive to apoptotic stimuli through a decrease in potassium levels in T-cells. As can be seen in Fig. 1(b), expression of Vpu increases the susceptibility of infected cells to Fas-mediated apoptosis by inhibiting NF-κB-mediated expression of anti-apoptotic genes (Nomaguchi et al., 2008; Strebel, 2014).

**Role of Nef**

HIV-1 Nef is also expressed early in the life cycle of HIV and inhibits apoptosis giving a chance for viral particles to mature. Nef prevents apoptosis by the endocytosis of the CD4 receptor and class I major histocompatibility complex (MHC I), enhancement of viral infectivity and inhibition of T-cell activation (Stoddart et al., 2003) (Fig. 1a). Nef also inhibits expression of p53 (Geleziunas et al., 2001) and the pro-apoptotic serine/threonine kinase ASK1 (apoptosis signal-regulating kinase-1) (Fig. 1a). Nef reduces the activity of the pro-apoptotic protein BAD (BCL-2-associated death promoter) by PAK (p21-activated kinase)-mediated phosphorylation (Wolf et al., 2001) (Fig. 1a).

In contrast, Nef induces apoptosis of HIV-infected cells during the advanced stages of HIV infection by upregulating Fas/FasL and downregulating BCL-2/BCL-XL (Rasola et al., 2001). It increases expression of PD-1 (programmed cell death) (Muthumani et al., 2008) and lysosomal permeabilization with release of cathepsin-D into the cytosol leading to outer mitochondrial membrane rupture followed by cell death (Laforge et al., 2007) (Fig. 1b).

**Role of Vpr**

Vpr is an accessory protein having several functions including the nuclear import of the HIV pre-integration complex, modulation of T-cell apoptosis, induction of G2 cell cycle arrest and regulation of NF-κB activity by transcriptional co-activation of viral and host genes (Abbas & Herbein, 2013; Kogan & Rappaport, 2011). Vpr plays different roles in apoptosis, based on its level of expression. Early during
the life cycle of HIV, the low-level expression of Vpr is anti-apoptotic because it upregulates expression of BCL-2 and downregulates expression of BAX along with suppression of NF-κB-dependent pro-inflammatory cytokine production (Fig. 1a). But later, after G2 cell arrest, Vpr shows pro-apoptotic properties by inducing apoptosis by binding to either BAX or ANT (adenine nucleotide translocase) and VDAC (voltage-dependent anion channel) in the mitochondrial membrane and causing release of cytochrome c to activate caspases 9 and 3 (Andersen et al., 2008) (Fig. 1b). Increased expression of NkG2D (natural killer group 2D) ligands in CD4+ T-cells mediated by expression of Vpr also enhances the susceptibility of infected cells to NK-cell-mediated killing (Richard et al., 2010).

Role of protease
In addition to its role in viral maturation, the HIV-1 protease packaged in the virus can induce apoptosis by cleaving cellular targets like anti-apoptotic BCL-2 and procaspase 8 after infection in target cells (Rumlová et al., 2014). Protease-mediated cleavage of BCL-2 leads to oxidative-stress-dependent activation of NF-κB and apoptosis of infected cells (Blanco et al., 2003). Meanwhile, the HIV protease-mediated cleavage of procaspase 8 generates a pro-apoptotic cleavage fragment, Casp8p41, which induces apoptosis in infected CD4+ T-cells via the intrinsic pathway (Nie et al., 2008; Sainski et al., 2014) (Fig. 1b).

Compared with HIV-1 infection, HIV-2 infection is characterized by higher CD4+ T-cell counts and lower viral RNA levels with a slower disease progression. Similarly, natural simian immunodeficiency virus (SIV) infections of African primates are characterized with marked virus replication with no disease progression. Although not fully understood, differences in the viral proteins and immune activation mechanisms may be responsible for these distinct phenomena (Campbell-Yesufu & Gandhi, 2011; Klatt et al., 2012; Nyamweya et al., 2013). The key factors responsible for non-progressive SIV infection may include low levels of CD4+ T-cell activation and apoptosis, SIV Nef-mediated downregulation of CD3-TCR complex, lack of viral replication and preserved CD4+ T-cell homeostasis (Chahroudi et al., 2012; Klatt et al., 2012). In the case of HIV-2, the Env (gp105) inhibits T-cell proliferation and upregulates the co-stimulatory molecules CD40 ligand and OX40, which are associated with reduced level of apoptosis (Cavaleiro et al., 2000). These findings from the non-pathogenic model of SIV and the attenuated disease model of HIV-2 highlight the role of viral-encoded proteins in the inhibition of host cell apoptosis.

Bystander apoptosis
HIV infection also induces apoptosis in uninfected CD4+/CD8+ T-cells or bystander cells present in the vicinity of infected cells. Bystander T-cell apoptosis is mediated by secreted HIV viral proteins like gp120, Nef and Tat through several mechanisms. Apoptosis of uninfected CD4+ T-cells by infected macrophages, monocytes, PBMCs, CD4+ T-cells and CD8+ T-cells from HIV-infected patients is mainly mediated through the envelope glycoproteins and/or Fas/FasL system (Cummins & Badley, 2010). In addition, envelope glycoproteins expressed on the surface of infected cells can interact with CD4 and/or chemokine co-receptors expressed on neighbouring uninfected immune cells and can induce apoptotic signalling via the receptors. These interactions can also cause cell-to-cell fusion resulting in formation of multinucleated syncytia leading to apoptosis by the intrinsic mitochondrial pathway (Nardacci et al., 2015). Secreted Nef binds to CD4 and/or chemokine co-receptor molecules on uninfected CD4+ T-cells and activates the Fas/FasL signalling pathway resulting in induction of apoptosis (Lenassi et al., 2010). Furthermore, HIV-specific CD8+ T-cells are also highly sensitive to apoptosis (Petrovas et al., 2004). TNF-α is released as a soluble factor or expressed in a membrane bound form on HIV-infected macrophages, which stimulates TNFR2, thereby triggering apoptosis of CD8+ T-cells (Herbein et al., 1998). Tat secreted from infected cells also mediates apoptosis of uninfected bystander cells by upregulating expression of FasL (Vendeville et al., 2004).

Activation-induced cell death
HIV-infected patients show generalized lymphadenopathy, increased circulating levels of activated T-lymphocytes, NK cells, B-lymphocytes, antigen presenting cells and other markers characteristic of chronic activation of the immune system (Cha et al., 2014; Naranbhai et al., 2013; Piguet et al., 2014). The CD4+ T-cell decline observed during HIV pathogenesis is also attributed to the AICD of CD4+ T-cells (Haas et al., 2011). The mechanism of HIV-associated AICD can be either Fas/FasL dependent or independent (Cummins & Badley, 2010). The characteristic features suggestive of Fas/FasL-dependent AICD include activated T-cell phenotypes like increased expression of CD38, Ki67, HLA-DR and CD25 along with upregulation of Fas/FasL expression in T-cells, leading to an enhanced susceptibility to Fas-mediated death (Ara-kaki et al., 2014). Whereas, the Fas independent pathway is mediated through activation of Toll-like receptors (TLR). Depletion of CD4+ T-cells in the gastrointestinal tract results in the translocation of microbial cellular components such as bacterial DNA and lipopolysaccharide into the systemic circulation leading to activation of TLR signalling, which triggers both AICD and CD4+ T-cell apoptosis outside the gastrointestinal tract (Funderburg et al., 2008; Marchetti et al., 2013).

HIV latency and apoptosis
Viral latency has been the biggest hurdle in our efforts to treat HIV (Tyagi & Bukrinsky, 2012). The partial or complete blockade of the viral life cycle prior to integration of
XIAP (X-linked inhibitor of apoptosis protein) is expression of TSG101 required in the ESCRT signalling p24 capsid protein into lipid rafts and inhibiting the assembly by blocking the trafficking of gp120 and Gag by increasing the expression of viral restriction factors et al. (2003). HIV latency in vivo is a complex mechanism regulated both at the transcriptional as well as post-transcriptional level. Transcriptional repression of HIV depends on several elements including the site of integration into the host genome (Rezaei & Cameron, 2015) and the chromatin environment (Gallastegui et al., 2011). Resting central memory T-cells and transitional memory T-cells are thought to be the prominent reservoirs of the latent virus (Chomont et al., 2009; Jaafoura et al., 2014) due to the absence of transcription factors, NF-κB and NFAT (nuclear factor of activated T-cells) (Bosque & Planelles, 2009; Williams et al., 2007), and increased expression of transcriptional repressors like C/EBPβ, DSIF (6-dichloro-1-β-D-ribofuranosylbenzimidazole sensitivity-inducing factors) and NELF (negative elongation factor), and the TRIM (tripartite motif) family of proteins (Nisole et al., 2005; Ping & Rana, 2001). Additional factors influencing latency include repressive nucleosome (nuc-1) (Van Lint et al., 1996), epigenetic silencing of HIV promoters, HEXIM-1-mediated inactivation of cellular P-TEFb (positive transcription elongation factor b) (Tyagi et al., 2010) and the absence of viral trans-activator protein Tat (Donahue et al., 2005). In addition, infected monocytes, macrophages, naïve T-cells, follicular dendritic cells, haematopoietic progenitor cells, astrocytes and microglia in the central nervous system can also act as viral reservoirs (Coleman & Wu, 2009; Gray et al., 2014; McNamara & Collins, 2011; Wightman et al., 2010).

The mechanism of HIV-mediated apoptosis evasion in latently infected cells is not completely understood. Several studies have reported that in latently infected cells, there is decreased HIV replication due to the increased expression of anti-apoptotic proteins (e.g. BCL-2, cFLIP, Mcl-1) (Aillet et al., 1998; Berro et al., 2007; Tan et al., 2013) or downregulation of pro-apoptotic proteins (e.g. BAX, FADD) (Badley et al., 2013; Wang et al., 2011). The increase in BCL-2 leads to enhanced levels of cellular antioxidant molecules like glutathione and thioredoxin, thereby protecting cells from oxidative-stress-induced apoptosis (Aillet et al., 1998). Similarly, increased cFLIP expression in latent cells downregulates HIV expression by increasing the expression of viral restriction factors (TRIM5, APOBEC3G and BST-2/tetherin) and decreasing expression of nuclear factor 1C. cFLIP also prevents HIV assembly by blocking the trafficking of gp120 and Gag p24 capsid protein into lipid rafts and inhibiting the expression of TSG101 required in the ESCRT signalling pathway (Tan et al., 2013). In addition, the anti-apoptotic XIAP (X-linked inhibitor of apoptosis protein) is upregulated in latently infected T-cell lines, macrophage/monocytic cell lines and PBMCs. It was reported that chemical inhibition of XIAP sensitizes these cells to apoptosis (Berro et al., 2007). All these evidences suggest that apoptosis is inhibited during latency. Triggering apoptosis by addition of cytotoxic drugs such as doxorubicin, etoposide, fludarabine phosphate or vincristine in latently infected pro-monocytic and T-cell lines has been shown to activate HIV replication in a caspase 3 or 8 dependent manner (Khan et al., 2015). Understanding the mechanism of apoptosis evasion in latently infected cells might help in devising strategies to deplete long-lived HIV reservoirs (Cummins & Badley, 2013).

**Modulation of apoptosis as a therapeutic approach**

HAART has significantly reduced AIDS-related morbidity and mortality. But, persistent residual viraemia in HAART-treated HIV-infected patients due to latent viral reservoirs and/or residual ongoing viral replication has been a limitation for functional cure of HIV-infected individuals (Fig. 2). Several approaches have been proposed as curative strategies against HIV including gene therapy, immune-based therapies, cytotoxic approaches, reactivation and purging of HIV latent reservoirs.

HIV gene therapy mainly relies on three basic approaches. Firstly, gene knockdown of host proteins like CCR5 and CXCR4 essential for the HIV virus to complete its life cycle (e.g. use of zinc finger nucleases to degrade CCR5 and CXCR4 mRNAs) (Didigu et al., 2014; Goila & Banerjea, 1998; Sasso & Kelleher, 2014). Secondly, overexpression of proteins which could limit HIV replication, e.g. use of chimeric TRIM5α representing human/rhesus fusions and overexpression of broadly neutralizing antibodies (Anderson, 2013; Caskey et al., 2015), and finally the expression of chimeric antigen receptors to activate T-cells (Lam & Bollard, 2013; Liu et al., 2015). Recently, a novel genome-editing method based on the clustered regularly interspaced short palindromic repeat (CRISPR)/CRISPR-associated (Cas)-9 protein system was found to be able to edit the HIV-1 genome and suppress its expression in Jurkat cell line based latency models after stimulation with TNF-α (Ebina et al., 2013). Immune-based therapies include therapeutic vaccination (Excler et al., 2014; Iaria et al., 2014), administration of recombinant cytokines like IL-2, IL-7, IL-15 and IL-21 (Katsikis et al., 2011), and the use of anti-inflammatory agents – statins, chloroquine derivatives, pre- and probiotics, and growth hormone (Ganesan et al., 2011; Hummelen et al., 2010; Klatt et al., 2013; Murray et al., 2010; Napolitano et al., 2008). Autologous and allogeneic stem cell transplantations are other potential approaches (Bauer & Anderson, 2014; Zaia & Forman, 2013). Allogeneic bone marrow transplant from a donor with CCR5 delta 32 mutation to a HIV-1-infected individual in Berlin provided a cure for HIV infection (Allers et al., 2011). Long-term reduction
of peripheral blood HIV-1 reservoirs was also reported following allogeneic haematopoietic stem cell transplantation in two HIV-1-infected individuals (Henrich et al., 2013). Tat inhibitor didehydro-cortistatin A has been reported to suppress HIV reactivation upon CD3/CD28 or prostratin stimulation of latently infected CD4+ T-cells from HIV-infected individuals under suppressive antiretroviral therapy. This may open a new avenue for the functional cure of HIV infection as Tat inhibitor plus combined antiretroviral therapy (cART) may contribute to a deep state of latency from which the ability of the virus to reactivate is permanently abolished (Mousseau et al., 2015).

Viral reactivation from the pools of latent reservoirs and the subsequent killing of these infected cells has become one of the areas of intensive research and many compounds that can activate latent infection are being evaluated through various clinical trials (Sebastian & Collins, 2014). Some of the strategies to activate latent HIV reservoirs and induce their death may include immune activation therapies using IL-2 and/or IL-7 (Brooks et al., 2003; Wang et al., 2005), and other mitogenic stimuli in combination with HAART (Shen et al., 2007). Although IL-2 therapy increased CD4+ T-cell number, a rapid rebound in virus was reported among the individuals receiving cART plus IL-2 after cessation of therapy (Chun et al., 2015). Furthermore, IL-7 mediates survival and expansion of HIV reservoirs (Yin et al., 2015). Compounds like valporic acids, vorinostat (SAHA), panobinostat and romidepsin, which are histone deacetylase inhibitors, have been found to upregulate

![Diagram](http://jgv.microbiologyresearch.org)
cellular transcription to induce HIV gene expression (Barton et al., 2014; Shirakawa et al., 2013). These compounds are capable of reactivating latent viral reservoirs without global T-cell activation. Protein kinase C inducers like bryostatin (Mehla et al., 2010), NF-κB activators like prostratin (Williams et al., 2004), Akt/HEXIM-1 modulators like disulfiram (Spivak et al., 2014; Xing et al., 2011) and histone methylation inhibitors like 5-azacytidine are other compounds being evaluated for reactivation of latent HIV virus (Bouchat et al., 2012). A combination of different classes of latency reversing agents (LRAs) might be more effective in reactivating latent HIV compared with the use of a single LRA at a time (Laird et al., 2015). A few recent studies have reported the selective induction of apoptosis in apoptosis-resistant cells like productively infected cells (Greenberg et al., 2014). The iron-chelator deferiprone and topical antifungal ciclopirox cause selective death of HIV-infected H9 cells, PBMCs and latently infected cells (Greenberg et al., 2014). The iron-chelator deferiprone and topical antifungal ciclopirox cause selective death of HIV-infected H9 cells, PBMCs and latently infected Jurkat 3C9 cells in vitro by therapeutic reclamation of apoptotic proficiency (Hanauske-Abel et al., 2013). However, recent studies have shown that an increase in viral RNA induced by these compounds in reactivated cells is not sufficient to lead to viral cytopathicity and/or elimination by cytotoxic T-cells (Shan et al., 2012). Indeed, other interventions, like boosting of CTL responses, may be required to trigger apoptosis of the reactivated cells (Deng et al., 2013). As reviewed recently (Badley et al., 2013), the specific approaches to trigger apoptosis of these cells after reactivation include priming reactivated cells by chemosensitization using agents that can act on the mitochondrial permeability transition pore complex (PTPC) (Fulda et al., 2010; Le Bras et al., 2006), BCL-2 inhibitors (Fulda et al., 2010; Thomas et al., 2013), IAP inhibitors (Fulda, 2014), Akt inhibitors (Doyon et al., 2013; Pal et al., 2010), FasL and TRAIL sensitizers (Lemke et al., 2014), TLR agonists (Novis et al., 2013; Winckelmans et al., 2013), p53 inducers (Khoo et al., 2014; Zawacka-Pankau & Selivanova, 2015) or co-stimulatory agents like anti-CD28 ( Peggs et al., 2009). Inducing HIV-infected macrophages and latently infected T-cells to undergo selective apoptosis is one of the promising approaches to achieve viral eradication (Fig. 2). In order to effectively eliminate viral reservoirs, there is a need to search for novel compounds that are capable of reactivating and killing the latent cells. These compounds should be non-toxic to other cells, unable to induce global immune activation and compatible with HAART components.

**Conclusion**

HIV infection is characterized by the pleiotropic regulation of apoptosis. The majority of CD4⁺ T-cells and other HIV-infected cells are effectively killed during productive infections whereas latent viral reservoirs remain resistant to death and allow for a long-term persistence of HIV. These long-lived latent viral reservoirs have become a major challenge for the functional cure of HIV. In-depth understanding of the mechanisms of HIV-mediated apoptosis evasion may help in selective reactivation of latent viral reservoirs and in triggering the latent (or reactivated) cells towards apoptosis, which could help in revealing a mechanism for the successful cure of HIV.

**References**


Modulation of apoptosis and viral latency


DNA-enzyme: inhibition of the coreceptor function by DNA-enzyme. 


http://jgv.microbiologyresearch.org


