Immunology Letters 196 (2018) 135–139

Contents lists available at ScienceDirect

Immunology Letters

journal homepage: www.elsevier.com/locate/immlet

The role of latency reversal agents in the cure of HIV: A review of current data

Kiandokht Bashiri[a,b], Nima Rezaei[a,b], Milena Nasi[c], Andrea Cossarizza[d,*]

[a] Research Center for Immunodeficiencies, Children's Medical Center, Tehran University of Medical Sciences, Dr. Qarib St, Keshavarz Blvd, Tehran, 14194, Iran
[b] Border of Immune Tolerance Education and Research Network (BITERN), Universal Scientific Education and Research Network (USERN), Tehran, Iran
[c] Department of Surgery, Medicine, Dentistry and Morphological Sciences, University of Modena and Reggio Emilia, Modena, Italy
[d] Department of Medical and Surgical Sciences for Children and Adults, University of Modena and Reggio Emilia, Modena, Italy

ARTICLE INFO

Keywords:
HDAC inhibitor
HIV
Latency
Resting CD4+ T Cells

ABSTRACT

The definitive cure for human immunodeficiency virus type-1 (HIV) infection is represented by the eradication of the virus from the patient's body. To reach this result, cells that are infected but do not produce the virus must become recognizable to be killed by the immune system. For this purpose, drugs defined "latency reverting agents" (LRA) that reactivate viral production are under investigation. A few clinical studies have been performed in HIV-infected patients treated with LRA and combined antiretroviral therapy (cART). The strategy is thus to combine cART and LRA to reactivate the virus and unmask latently infected cells that, because of cART, cannot produce a fully competent form of the virus. Unmasked cells can present viral antigens to the immune system, that ultimately recognizes and kills such latently infected cells. This review reports and discusses recent studies that have been published on this topic.

1. Introduction

Combined antiretroviral therapy (cART) controls replication of the human immunodeficiency virus type-1 (HIV) by affecting different stages of the viral life cycle, including cell entry, reverse transcription, integration, and assembling of the virion [1]. The use of cART has led to a prolongation of the lifespan of HIV-positive individuals lifespan and to an improvement in the quality of life, turning the concept of HIV infection as a life-threatening disease into a chronic infectious disease [2,3], characterized by a persistent activation of the immune system [4].

Even if cART puts a harness on viral replication and decreases plasma viremia in most treated patients [5,6], it is unable to completely eliminate infected cells. Thus, the presence of resting, memory $CD4^+$ T cells carrying proviral DNA remains a major obstacle for HIV eradication because, once re-activated, these latently infected cells are a potential source of viruses [7,8]. The genome of HIV integrates into the host DNA but cannot express itself significantly, because the activation of the proviral promoter long terminal repeat (LTR) requires the intervention of several cellular transcription factors. In the absence of adequate stimuli, these latent reservoirs are stable and resistant to different treatment regimens [9–12]. As respects, discovering integrated virus from the host's genome and accordingly target infected but not virus-producing cells is remarkably challenging.

The immune system cannot recognize latently infected cells, and such cells escape from both the attack of the immune system, and are not touched by cART [1]. There are two main strategies for trying to eradicate the infection, of for its cure, that include "sterilizing cure" and "functional cure". The sterilizing cure include treatments such as stem cell transplantation, genome editing, gene therapy and "shock and kill" strategy [13–15]. On the other hand, functional cure implies the long-term control of viral replication with the aim of preserving a normal CD4+ T cell count and undetectable level of viral replication [16].

In the last years, promising studies have identified drugs which are able to reverse latency without activating T cells and causing the production of new virions [17–19]. The aim of this strategy is to combine latency reverting agents (LRA) with cART so that LRA can activate the production of the virus by latentely infected cells: viral peptides are presented to the immune system that finally can recognize and kill infected cells. However, because of the presence of protease inhibitors among the drugs used in the cART, in theory the complete form of the virion does not have be produced, and thus viral load has to remain undetectable.

LRA include disulfiram and the histone deacetylase (HDAC) inhibitors, such as vorinostat (suberoylanilide hydroxamic acid or SAHA) [20–22]. The molecular mechanism of latency reactivation induced by

* Corresponding author at: University of Modena and Reggio Emilia, via Campi, 287, 41125 Modena, Italy.
E-mail address: andrea.cossarizza@unimore.it (A. Cossarizza).

https://doi.org/10.1016/j.imlet.2018.02.004
Received 12 January 2018; Accepted 6 February 2018
Available online 07 February 2018

disulfiram is unclear [23]. The role of HDAC is to remodel chromatin during transcription, leading to the prevention of LTR promoter expression, thereupon repressing viral replication. HDAC inhibitors disrupt the aforementioned process, so causing LTR activation [24].

Previous studies showed that three HDAC inhibitors, including vorinostat (Zolinza™), romidepsin (depsipeptide, FK228, FR901228), and panobinostat (LBH589, Farydak™) have enhanced the level of plasma viral RNA in aviremic patients on cART, indicating successful reactivation of latent cells that unfortunately produce intact virions [25–29]. Considering the performance of each inhibitor as far as the plasma level of HIV is concerned, a survey summarizing and comparing the role of each drug in treating infected patients can be of interest. Thereby, the current paper briefly reviews and assesses the clinical trials performed by using the HDAC inhibitors Vorinostat, Panobinostat and Romidepsin.

2. Material and methods

After literature search, original articles were selected and included if they met the following criteria: (1) all HIV-infected people were on cART; (2) HDAC inhibitors included Vorinostat, or Panobinostat, or Romidepsin; (3) the study was designed to measure the increase of intracellular HIV-1 mRNA in CD4+ T cells of patients irrespective of age, gender or race. We performed a search in different sources: Medline, Scopus, DART, OpenGrey and ProQuest, without applying any language filter, up to March 2017. Two of the authors independently reviewed search results for irrelevant and duplicate studies and extracted data from relevant studies. Our search strategy was ((Panobinostat) OR (Farydak) OR (LBH-589) OR (LBH589) OR (Romidepsin) OR (Istodax) OR (Vorinostat) OR (suberanilohydroxamic acid) OR (SAHA) OR (Zolinza)) AND ((Human immunodeficiency virus) OR (Human immune deficiency virus) OR (HIV-1) OR (human immunodeficiency type 1) OR (HIV-1) OR (HIV-infected) OR (Acquired Immunodeficiency Syndrome) OR (Acquired immune deficiency syndrome) OR (AIDS) OR (HIV/AIDS)).

Then, we imported citations from all databases into an EndNote library (version X6, Thomson Reuters, New York, NY, USA). In the EndNote library, we identified duplicates among citations using the “Find Duplicates” feature of the EndNote software. Then two of the authors independently reviewed the title and abstract of the remainder of the search results for irrelevant studies, that were excluded. We collected the full text of the included articles for further screening and data collection process.

We extracted the following data from each included publication; first-named author, year of publication, the administration regimen of medication, number of subjects, increases in level of intracellular HIV-1 mRNA in patients.

3. Results

We found 905 references by recruiting the search strategy in five above-mentioned databank. We did not retrieve any new studies in the reference lists of the main articles. After discarding duplicates, we identified 745 publications. In a primary screening of titles and abstracts, 731 articles were further excluded because of irrelevancy of the topics. In a secondary screening of full texts, 7 studies with a total number of 94 HIV-1 positive patients were eligible to be included in this review (Table 1).

We found one study with 15 enrolled patients treated solely with Panobinostat, three studies with 33 enrolled patients treated solely with Vorinostat, and three studies with 46 enrolled patients treated with Romidepsin.

HDAC inhibitors are anticancer agents with different structures and some of them are currently in clinical development. The effectiveness of Vorinostat in breaking HIV-1 latency was first evaluated by Archin and colleagues in 2012. The study population was composed by 8 HIV-positive patients who were on ART with plasma HIV-1 RNA less than 50 copies/ml for at least 6 months and a CD4+ T cell count higher than 300 cells/μl. The main outcomes of interest, *i.e.* HIV-1 RNA copy number in resting CD4+ T cells and histone acetylation in peripheral blood mononuclear cells, were measured at multiple time points 24 h after the administration of 400 mg Vorinostat. In each patient, a single dose of Vorinostat increased HIV-1 RNA expression in latently infected CD4+ T cells (mean increase: 4.8 fold); similarly, an increment in biomarkers of cellular acetylation was observed [25]. Among these 8 patients, 5 were selected for second study by Archin et al. in 2014 with the same inclusion criteria as the precedent study. The study covers white males, with a mean age of 54 years. Patients received 22 cyclical doses of Vorinostat during a period of 12–16 weeks. HIV-1 RNA in resting CD4+ T cells increased significantly in 3 of 5 patients after dose 11 or dose 22, and the magnitude of increase in the resting CD4+ T-cell was much declined compared with that after a single dose. In contrary to the previous study, changes in histone acetylation were blunted. No change was observed in viral outgrowth [30].

Elliot and colleagues administered 400 mg Vorinostat daily for 2 weeks to 20 HIV-infected patients maintained on cART. At the end of second week, CD4+ T cell-associated unspliced HIV-1 RNA enhanced significantly in 18/20 (90%) participants (median increase: 7.4 fold). Changes in plasma HIV-1 RNA, concentration of HIV-1 DNA, integrated DNA and markers of T-cell activation were blunted [28].

In all of these three studies Vorinostat was safe and well tolerated, and the presence of adverse effects (*i.e.*, mild gastrointestinal symptoms and transient headache) did not affect the study. Likewise Vorinostat causes a significant increase in expression of HIV-1 RNA in CD4+ T cells.

Previous studies on anticancer effects of HDAC inhibitors showed that Panobinostat was at least 10-fold more potent than Vorinostat, and might become the most potent HDAC inhibitor to be used in trials [31]. In 2014, Rasmussen et al. administered Panobinostat 20 mg 3 times/week orally for 8 weeks to 15 patients on cART. The CD4+ T cell-associated unspliced HIV-1 RNA increased significantly (median maximal fold increase: 3.5) and remained elevated 1 month post-Panobinostat (fold-increase 1.60; 95% CI: 1.17-2.19; $p = .003$). The most common adverse effect of Panobinostat among participants was fatigue [32].

The study performed *in vitro* by Wei et al. with Romidepsin showed a greater magnitude and continuity of HIV-1 RNA expression compared to Vorinostat, and was able to induce the release of HIV-1 virions from latently infected cells. Intracellular pharmacology and interactions of Romidepsin with HDAC enzymes may be responsible for such long effects, especially if compared to Vorinostat. Intramolecular disulfide bond of Romidepsin undergoes reduction upon entering cells and turns into free sulfhydryl groups, then interacts with the zinc ion in the active site of different target HDAC isoforms. This inhibitory mechanism does not apply for Vorinostat or Panobinostat [33,34]. In 2015, Sogaard et al., administered Romidepsin 5 mg/m^2 intravenously 4 h per week until 3 weeks to 6 HIV-infected patients, and found a significant increase in cell associated unspliced HIV-1 RNA (median fold increase: 3.4; $p = .030$) [29]. Notwithstanding the use of cART, plasma HIV-1 RNA increased from < 20 copies/mL at baseline to readily quantifiable levels in 5 of 6 patients who received Romidepsin (range: 46–103 copies/mL following the second infusion, $p = .040$). The most frequently reported adverse effects of Romidepsin were gastrointestinal symptoms (*i.e.*, nausea, increased bowel sounds and abdominal pain). Albeit increasing in virus transcription has been observed, no changes were observed in amount of latent reservoirs *in vivo* [28,29].

4. Discussion

LRA are interesting drugs for pursuing the shock and kill strategy, aimed at eradicating HIV infection. Indeed, in order to kill infected cells that do not produce spontaneously the virus, the immune system has to

Table 1
Main characteristics of the selected studies.

Author	HDAC inhibitor	Number of Participants	HDAC inhibitor's Regimen	Changes in Resting CD4+ T cells HIV RNA	Other outcomes
Archin et al. [28]	Vorinostat	5	Daily Vorinostat Monday through Wednesday for 8 weekly cycles	After dose 11 (second dose of cycle 4) or dose 22 (second dose of cycle 8) increased significantly in only 3 of the 5 participants, and the magnitude of the increase was much reduced compared with that after a single dose	Changes in histone acetylation were blunted. Quantitative viral outgrowth and total cellular HIV DNA were unchanged
Archin et al. [25]	Vorinostat	8	A single oral 200 mg dose for assessing tolerability, then 400 mg dose 4 (or more) weeks later	An increase of 1.5- to 10.0-fold (mean 4.8) in expression of unspliced HIV-1 *gag* RNA within resting CD4+ T cells was measured in seven patients	
Elliott et al. [28]	Vorinostat	20	Vorinostat was administered 400 mg orally once daily for 14 days while maintaining ART	Cell associated unspliced HIV RNA in blood increased significantly in 18/20 patients (90%) with a median fold change from baseline to peak value of 7.4 (IQR 3.4, 9.1).	There were no statistically significant changes in plasma HIV RNA, concentration of HIV DNA, integrated DNA, inducible virus in CD4+ T-cells or markers of T-cell activation.
Leth et al. [35]	Romidepsin	20	Participants received 6 therapeutic intradermal HIV-1 immunizations with 12 mg/mL Vacc-4 x and 0·6 mg/mL rhuGM-CSF over 12 weeks (at 0 weeks, 1 week, 2 weeks, 3 weeks, 11 weeks, and 12 weeks) before receiving 5 mg/m2 intravenous Romidepsin once a week for 3 weeks.	No major changes in the CD4 T-cell compartment during Romidepsin infusions	Total HIV-1 DNA declined from screening to 6 weeks after Romidepsin treatment (mean reduction 39.7%, 95% CI −59.7 to −11.5; p = .012). The decrease in integrated HIV-1 DNA from baseline to 8 weeks after Romidepsin treatment was not significant between four (24%) and eight (47%) of 17 patients had detectable plasma HIV-1 RNA throughout the course of the study t (19.2%, −38.6 to 6.3; p = .123).
Rasmussen, [32]	Panobinostat	15	Oral Panobinostat 20 mg 3 times/week every other week for 8 weeks while maintaining combination antiretroviral therapy (cART)	Levels of CA-US RNA increased significantly during Panobinostat treatment (p < .0001) with significant increases on time points on-treatment as compared to baseline. The median maximal fold-increase in CA-US RNA was 3.5 (range 2.1–14.4). Levels of CA-US RNA remained elevated 4 weeks post-Panobinostat (fold-increase 1.60; 95% CI: 1.17–2.19; P = .003)	Using a transcription mediated amplification-based semi-quantitative assay (Procleix Ultrio Plus, 59% analytic sensitivity of 3.6 copies/mL), HIV-RNA in plasma was detected more frequently during Panobinostat administration with an odds ratio of 10.5 (95% CI: 2.2–50.3) for a positive test on-treatment compared to baseline
Sogaard et al. [29]	Romidepsin	6	One 4 h Romidepsin infusion (5 mg/m2) per week for three consecutive weeks and were followed for up to 70 days after the last infusion	HIV-1 transcription quantified as copies of cell-associated un-spliced HIV-1 RNA increased significantly from baseline during treatment (range of fold-increase: 2.4–5.0; p = .03).	Plasma HIV-1 RNA increased from < 20 copies/mL at baseline to readily quantifiable levels at multiple post-infusion time-points in 5 of 6 patients (range 46–103 copies/mL following the second infusion, p = .04).
Tapia [37]	Romidepsin	20	Six Vacc-4 x (1.2 mg) intradermal immunizations using rhuGM-CSF (60 μg) as adjuvant were followed by 3 weekly intravenous infusions of romidepsin (5 mg/m^2).		Participants with CD8+ T-cell proliferation assay positivity post-vaccination showed reductions in total HIV DNA post-vaccination (p = .006; q = 0.183), post-latency reversal (p = .005; q = 0.183), and CA-RNA reductions post-vaccination (p = .015; q = 0.254). Participants (40%) were defined as proliferation 'Responders' having ≥2-fold increase in assay positivity post-baseline. Robust ELISpot baseline responses were found in 87.5% participants. No significant changes were observed in the proportion of polyfunctional CD8+ T-cells to HIVGag by ICS. There was a trend towards increased viral inhibition from baseline to post-vaccination (p = .08).

recognize viral antigens, and LRA are able to unmask latently infected cells, allowing the presentation of viral peptides on MHC class I. *In vitro* studies showed that Romidepsin is the most potent LRA tested, introducing it as a "prototype" drug that can be used as a reference for developing and testing new LRA.

Among studies in HIV-infected patients, one failed to show any significant variation of plasma viral load in almost half of patients treated with Romidepsin and Vacc-4 x in combination therapy [35,36]. The following reasons may justify ineffectiveness of the LRA in this study: first, stimulation of HIV-1 expression did not lead to the death of the cells. Secondly, these drugs are incapable to cross obstacles such as blood brain barrier. Moreover, neither ART nor LRA are reluctant to restore the impaired cytotoxic activity of CD8+ T cells. Likewise, following the "shock" strategy, a boosted immune system is required to eliminate HIV-infected cells [36,37]. Thus, a more complex strategy could be envisaged that likely could include molecules able to activate immune responses, such as cytokines. Another study by Tapia. et. al [37] that may support the results of the *in vitro* studies, showed reductions in total HIV DNA after using Vacc-4 x followed by Romidepsin administration. Also in this case, it is likely that a more sophisticated combination approach may be needed to effectively eliminate the pool of latent reservoirs.

Vorinostat affects gene regulation by several mechanisms consistent with ability of HDAC enzymes to target both histones and non-histone proteins, including HDAC1, signaling mediators and chaperones [38]. This drug could increase HIV-RNA significantly in human studies, similar to its effects during *in vitro* trials [25,28,30]. Panobinostat as a highly potent HDAC inhibitor had been previously used in treatment of multiple myeloma. Therapeutic concentrations of Panobinostat induce viral production in latently infected cells *in vitro* [36].

Overall, based on the current studies, it is pivotal to consider latently infected cells as a crucial target of the strategy to reduce the size of HIV reservoir. As a future investigation, it is worth to study innate immune activity, which, based on a work by Olesen et al. [39], can modify the effects of LRAs on quiescent cells.

5. Conclusions

To date, HDAC inhibitors have shown the proof-of-concept, interfering with persistent infection. Molecules like vorinostat, panobinostat, or romidepsin have yet to be tested in patients with persistently low CD4+ counts. They seem to be effective in those on stable ART for long time, and likely could be utilized in addition to ART. In any case, more investigation is needed to better understand other major aspects of the impact of HDAC inhibitors on the strategy required to eradicate HIV.

Funding

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

Acknowledgement

We would like to express our very great appreciation to Professor Sadegh Poozesh for his valuable suggestions during the planning of this paper.

References

[1] K. Barton, B. Burch, N. Soriano-Sarabia, D.M. Margolis, Prospects for treatment of latent HIV, Clin. Pharmacol. Ther. 93 (1) (2013) 46–56.
[2] S.G. Deeks, S.R. Lewin, D.V. Havlir, The end of AIDS: HIV infection as a chronic disease, Lancet 82 (9903) (2013) 1525–1533.
[3] J. Stein, M. Storcksdieck Genannt Bonsmann, H. Streeck, Barriers to HIV cure, HLA 88 (4) (2016) 155–163.
[4] M. Nasi, M. Pinti, C. Mussini, A. Cossarizza, Persistent inflammation in HIV infection: established concepts, new perspectives, Immunol. Lett. 161 (2) (2014) 184–188.
[5] R.M. Gulick, J.W. Mellors, D. Havlir, J.J. Eron, C. Gonzalez, D. McMahon, D.D. Richman, F.T. Valentine, L. Jonas, A. Meibohm, Treatment with indinavir, zidovudine, and lamivudine in adults with human immunodeficiency virus infection and prior antiretroviral therapy, N. Engl. J. Med. 337 (11) (1997) 734–739.
[6] S.M. Hammer, K.E. Squires, M.D. Hughes, J.M. Grimes, L.M. Demeter, J.S. Currier, J.J. Eron Jr., J.E. Feinberg, H.H. Balfour Jr., L.R. Deyton, A controlled trial of two nucleoside analogues plus indinavir in persons with human immunodeficiency virus infection and CD4 cell counts of 200 per cubic millimeter or less, New Engl. J. Med. 337 (11) (1997) 725–733.
[7] D. Finzi, M. Hermankova, T. Pierson, L.M. Carruth, C. Buck, R.E. Chaisson, T.C. Quinn, K. Chadwick, J. Margolick, R. Brookmeyer, Identification of a reservoir for HIV-1 in patients on highly active antiretroviral therapy, Science 278 (5341) (1997) 1295–1300.
[8] L. Gibellini, S. Pecorini, S. De Biasi, E. Bianchini, M. Digaetano, M. Pinti, G. Carnevale, V. Borghi, G. Guaraldi, C. Mussini, A. Cossarizza, M. Nasi, HIV-DNA content in different CD4+ T-cell subsets correlates with CD4+ cell: CD8+ cell ratio or length of efficient treatment, AIDS 1 (10) (2017) 1387–1392.
[9] A.M. Crooks, R. Bateson, A.B. Cope, N.P. Dahl, M.K. Griggs, J.D. Kuruc, C.L. Gay, J.J. Eron, D.M. Margolis, R.J. Bosch, Precise quantitation of the latent HIV-1 reservoir: implications for eradication strategies, J. Infect. Dis. 212 (9) (2015) 1361–1365.
[10] D. Finzi, J. Blankson, J.D. Siliciano, J.B. Margolick, K. Chadwick, T. Pierson, K. Smith, J. Lisziewicz, F. Lori, C. Flexner, Latent infection of CD4+ T cells provides a mechanism for lifelong persistence of HIV-1, even in patients on effective combination therapy, Nat. Med. 5 (5) (1999) 512–517.
[11] J.D. Siliciano, J. Kajdas, D. Finzi, T.C. Quinn, K. Chadwick, J.B. Margolick, C. Kovacs, S.J. Gange, R.F. Siliciano, Long-term follow-up studies confirm the stability of the latent reservoir for HIV-1 in resting CD4+ T cells, Nat. Med. 9 (6) (2003) 727–728.
[12] L. Gibellini, S. Pecorini, S. De Biasi, M. Pinti, E. Bianchini, A. De Gaetano, M. Digaetano, R. Pullano, D. Lo Tartaro, A. Iannone, C. Mussini, A. Cossarizza, M. Nasi, Exploring viral reservoir: the combining approach of cell sorting and droplet digital PCR, Methods 134-135 (2018) 98–105.
[13] C. Spragg, H.D.S. Feelixge, K.R. Jerome, Cell and gene therapy strategies to eradicate HIV reservoirs, Curr. Opin. HIV AIDS 11 (4) (2016) 442–449.
[14] D.H. Barouch, S.G. Deeks, Immunologic strategies for HIV-1 remission and eradication, Science 45 (6193) (2014) 169–174.
[15] J. Lisziewicz, E. Rosenberg, J. Lieberman, H. Jessen, L. Lopalco, R. Siliciano, B. Walker, F. Lori, Control of HIV despite the discontinuation of antiretroviral therapy, N. Engl. J. Med. 340 (21) (1999) 1683.
[16] Z. Kmietowicz, Early HIV treatment led to functional cure in 14 patients, report researchers, Br. Med. J. 18 (March (346)) (2013) f1759, http://dx.doi.org/10.1136/bmj.f1759.
[17] H.-C. Yang, S. Xing, L. Shan, K. O'Connell, J. Dinoso, A. Shen, Y. Zhou, C.K. Shrum, Y. Han, J.O. Liu, Small-molecule screening using a human primary cell model of HIV latency identifies compounds that reverse latency without cellular activation, J. Clin. Invest. 119 (11) (2009) 3473–3486.
[18] S. Xing, C.K. Bullen, N.S. Shroff, L. Shan, H.-C. Yang, J.L. Manucci, S. Bhat, H. Zhang, J.B. Margolick, T.C. Quinn, Disulfiram reactivates latent HIV-1 in a Bcl-2-transduced primary CD4+ T cell model without inducing global T cell activation, J. Virol. 85 (12) (2011) 6060–6064.
[19] S. Xing, S. Bhat, N.S. Shroff, H. Zhang, J.A. Lopez, J.B. Margolick, J.O. Liu, R.F. Siliciano, Novel structurally related compounds reactivate latent HIV-1 in a bcl-2-transduced primary CD4+ T cell model without inducing global T cell activation, J. Antimicrob. Ther. 67 (2) (2012) 398–403.
[20] A. Bosque, V. Planelles, Induction of HIV-1 latency and reactivation in primary memory CD4+ T cells, Blood 13 (1) (2009) 58–65.
[21] X. Contreras, M. Schweneker, C.-S. Chen, J.M. McCune, S.G. Deeks, J. Martin, B.M. Peterlin, Suberoylanilide hydroxamic acid reactivates HIV from latently infected cells, J. Biol. Chem. 284 (11) (2009) 6782–6789.
[22] N.M. Archin, A. Espeseth, D. Parker, M. Cheema, D. Hazuda, D.M. Margolis, Expression of latent HIV induced by the potent HDAC inhibitor suberoylanilide hydroxamic acid, AIDS Res. Hum. Retrovir. 25 (2) (2009) 207–212.
[23] E. Petersen, The pharmacology and toxicology of disulfiram and its metabolites, Acta Psych. Scand. 69 (S369) (1992) 7–13.
[24] G. He, D.M. Margolis, Counterregulation of chromatin deacetylation and histone deacetylase occupancy at the integrated promoter of human immunodeficiency virus type 1 (HIV-1) by the HIV-1 repressor YY1 and HIV-1 activator Tat, Mol. Cell. Biol. 22 (9) (2002) 2965–2973.
[25] N.M. Archin, A. Liberty, A.D. Kashuba, S.K. Choudhary, J. Kuruc, A. Crooks, D. Parker, E. Anderson, M. Kearney, M. Strain, Administration of vorinostat disrupts HIV-1 latency in patients on antiretroviral therapy, Nature 487 (7408) (2012) 482–485.
[26] K.M. Barton, T.A. Rasmussen, M. Tolstrup, W. Shao, B. Hiener, A. Solomon, L. Østergaard, S.R. Lewin, O. Søgaard, S.E. Palmer, Panobinostat broadly activates latent HIV-1 proviruses in patients, Top. Antivir. Med. 23 (2015) (p. 46).
[27] A.B. Christensen, A. Dige, J. Vad-Nielsen, C.R. Brinkmann, M. Bendix, L. Østergaard, M. Tolstrup, O.S. Søgaard, T.A. Rasmussen, J.R. Nyengaard, J. Agnholt, P.W. Denton, Administration of panobinostat is associated with increased IL-17A mRNA in the intestinal epithelium of HIV −1 patients, Med. Inflamm. (2015) 120605, http://dx.doi.org/10.1155/2015/120605.
[28] J.H. Elliott, F. Wightman, A. Solomon, K. Ghneim, J. Ahlers, M.J. Cameron, M.Z. Smith, T. Spelman, J. McMahon, P. Velayudham, Activation of HIV transcription with short-course vorinostat in HIV-infected patients on suppressive

antiretroviral therapy, PLoS Pathog. 10 (11) (2014) e1004473.
[29] O.S. Søgaard, M.E. Graversen, S. Leth, R. Olesen, C.R. Brinkmann, S.K. Nissen, A.S. Kjaer, M.H. Schleimann, P.W. Denton, W.J. Hey-Cunningham, The depsipeptide romidepsin reverses HIV-1 latency in vivo, PLoS Pathog. 11 (9) (2015) e1005142.
[30] N.M. Archin, R. Bateson, M.K. Tripathy, A.M. Crooks, K.H. Yang, N.P. Dahl, M.F. Kearney, E.M. Anderson, J.M. Coffin, M.C. Strain, D.D. Richman, K.R. Robertson, A.D. Kashuba, R.J. Bosch, D.J. Hazuda, J.D. Kuruc, J.J. Eron, D.M. Margolis, HIV-1 expression within resting CD4+ T cells after multiple doses of vorinostat, J. Infect. Dis. 210 (5) (2014) 728–735.
[31] W. Shao, J. Growney, Y. Feng, P. Wang, Y. Yan-Neale, G. O'Connor, P. Kwon, Y. -m. Yao, S. Fawell, P. Atadja, Potent anticancer activity of a pan-deacetylase inhibitor panobinostat (LBH589) as a single agent in in vitro and in vivo tumor models, 99th American Association of Cancer Research Annual Meeting, April 12–16, 2008 (Abstract 6244).
[32] T. Rasmussen, Panobinostat induces HIV transcription and plasma viremia in HIV patients on suppressive cART, Top. Antivir. Med. 22 (2014) (pp. 207).
[33] D.G. Wei, V. Chiang, E. Fyne, M. Balakrishnan, T. Barnes, M. Graupe, J. Hesselgesser, A. Irrinki, J.P. Murry, G. Stepan, Histone deacetylase inhibitor romidepsin induces HIV expression in CD4 T cells from patients on suppressive antiretroviral therapy at concentrations achieved by clinical dosing, PLoS Pathog. 10 (4) (2014) e1004071.
[34] R. Furumai, A. Matsuyama, N. Kobashi, K.-H. Lee, M. Nishiyama, H. Nakajima, A. Tanaka, Y. Komatsu, N. Nishino, M. Yoshida, FK228 (depsipeptide) as a natural prodrug that inhibits class I histone deacetylases, Cancer Res. 62 (17) (2002) 4916–4921.
[35] S. Leth, M.H. Schleimann, S.K. Nissen, J.F. Højen, R. Olesen, M.E. Graversen, S. Jørgensen, A.S. Kjær, P.W. Denton, A. Mørk, M.A. Sommerfelt, K. Krogsgaard, L. Østergaard, T.A. Rasmussen, M. Tolstrup, O.S. Søgaard, Combined effect of Vacc-4x, recombinant human granulocyte macrophage colony-stimulating factor vaccination, and romidepsin on the HIV-1 reservoir (REDUC): a single-arm, phase 1B/2A trial, Lancet HIV 3 (10) (2016) e463–e472.
[36] T.A. Rasmussen, M. Tolstrup, C.R. Brinkmann, R. Olesen, C. Erikstrup, A. Solomon, A. Winckelmann, S. Palmer, C. Dinarello, M. Buzon, M. Lichterfeld, S.R. Lewin, L. Ostergaard, O.S. Sogaard, Panobinostat, a histone deacetylase inhibitor, for latent-virus reactivation in HIV-infected patients on suppressive antiretroviral therapy: a phase 1/2, single group, clinical trial, Lancet HIV 1 (1) (2014) e13–e21.
[37] G. Tapia, J.F. Hojen, M. Okvist, R. Olesen, S. Leth, S.K. Nissen, D.J. VanBelzen, U. O'Doherty, A. Mork, K. Krogsgaard, O.S. Sogaard, L. Ostergaard, M. Tolstrup, G. Pantaleo, M.A. Sommerfelt, Sequential Vacc-4 x and romidepsin during combination antiretroviral therapy (cART): immune responses to Vacc-4 x regions on p24 and changes in HIV reservoirs, J. Infect. 75 (6) (2017) 555–571.
[38] P.A. Marks, W.S. Xu, Histone deacetylase inhibitors: potential in cancer therapy, J. Cell Biochem. 07 (4) (2009) 600–608.
[39] R. Olesen, S. Vigano, T.A. Rasmussen, O.S. Søgaard, Z. Ouyang, M. Buzon, A. Bashirova, M. Carrington, S. Palmer, C.R. Brinkmann, X.G. Yu, L. Østergaard, M. Tolstrup, M. Lichterfeld, Innate immune activity correlates with CD4 T cell-associated HIV-1 DNA decline during latency-reversing treatment with panobinostat, J. Virol. 89 (20) (2015) 10176–10189.