Identification of myxobacteria-derived HIV inhibitors by a high-throughput two-step infectivity assay

Javier P Martinez1, Bettina Hinkelmann2, Eric Fleta-Soriano1, Heinrich Steinmetz3, Rolf Jansen3, Juana Diez4, Ronald Frank2, Florenz Sasse2 and Andreas Meyerhans1,5*

Abstract

Background: Drug-resistance and therapy failure due to drug-drug interactions are the main challenges in current treatment against Human Immunodeficiency Virus (HIV) infection. As such, there is a continuous need for the development of new and more potent anti-HIV drugs. Here we established a high-throughput screen based on the highly permissive TZM-bl cell line to identify novel HIV inhibitors. The assay allows discriminating compounds acting on early and/or late steps of the HIV replication cycle.

Results: The platform was used to screen a unique library of secondary metabolites derived from myxobacteria. Several hits with good anti-HIV profiles were identified. Five of the initial hits were tested for their antiviral potency. Four myxobacterial compounds, sulfangolid C, soraphen F, epothilon D and spirangien B, showed EC50 values in the nM range with SI > 15. Interestingly, we found a high amount of overlapping hits compared with a previous screen for Hepatitis C Virus (HCV) using the same library.

Conclusion: The unique structures and mode-of-actions of these natural compounds make myxobacteria an attractive source of chemicals for the development of broad-spectrum antivirals. Further biological and structural studies of our initial hits might help recognize smaller drug-like derivatives that in turn could be synthesized and further optimized.

Introduction

Current Human Immunodeficiency Virus (HIV) treatment comprises a combination of three or more anti-retroviral drugs, which often lead to drug-resistance and therapy failure due to drug-drug interactions and toxic effects, especially in patients with HIV-associated co-infections [1-5]. As such, there is a continuous need for the development of new and more potent anti-HIV drugs. Here we describe the establishment of a two-step high-throughput screening (HTS) platform to identify molecules against HIV infection. The assay is based on the highly permissive TZM-bl cell line [6]. These are modified HeLa cells expressing endogenous CD4, CXCR4 and CCR5 receptors, and an integrated Tat-dependent firefly luciferase gene. The TZM-bl cells in combination with HIV pseudoviruses have been extensively used in antibody neutralization tests with highly reproducible results [7] and in a previous siRNA screen [8]. Upon infection, the viral RNA genome is reversed transcribed into DNA and integrated into the host-cell as a provirus. Then, the proviral-produced Tat protein mediates the activation of the LTR-driven luciferase gene. Thus, the amount of luciferase signal is in direct relationship with the efficiency of infection and the antiviral activity of test compounds can be measured as a function of reductions in luciferase expression compared to un-treated or drug-solvent controls.

The two-step cell-based screen is shown in Figure 1 and described in Materials and Methods. Briefly, TZM-bl cells seeded in 384-well plates are incubated with test compounds and infected with HIVLAI at a multiplicity of infection (MOI) of 0.5. To monitor compound-related toxicity in parallel, TZM-bl cells are left uninfected and...
incubated with test compounds (Part 1 of the screen). 48 h after initial infection, virus-containing supernatants are used to infect fresh TZM-bl cells (Part 2). Cells from Part 1 are assayed for Tat-dependent luciferase expression and, in parallel, for compound-related toxicity. 48 h after re-infection, cells of Part 2 are assayed in the same manner (See text for details). The assay setup is similar to a previous anti-HIV assay using MAGI cells [9]. The effects of test compounds on infectivity and cell viability are quantified by normalizing the mean luciferase expression units to the solvent controls and hits are determined by calculating a robust Z-score as described [9] (see Materials and Methods).

**Results and discussion**

To validate the two-step screen, we tested the effect of three FDA-approved anti-HIV drugs: the nucleoside reverse transcriptase inhibitor Zidovudine, the fusion inhibitor Enfuvirtide and the protease inhibitor Indinavir.
on HIV infectivity and cell viability (Figure 2A). The assay yielded an excellent separation between solvent and drug controls (assay Z-factor of 0.9) [10]. The platform was then used to screen a small library of around 150 myxobacterial secondary metabolites from the Helmholtz Centre for Infection Research, Braunschweig, Germany [11,12]. Myxobacteria are among the top producers of natural products, matching those produced by marine bacteria [13]. These true microbial cell factories are known to manufacture highly active antimicrobials with novel chemical structures [14,15]. The library was screened using a single concentration of each test substance (and solvent controls) assayed in quadruplicate. Robust Z-score values were calculated from the mean of the replicates as described [9,16], and plotted against the % mean of the solvent controls. Hits are defined as compounds inhibiting 50% infectivity with Z-scores < 0 and, conversely, compounds having less than 70% toxic effects with Z-scores > 0 (Figure 2B-D).

After discarding compounds without significant antiviral and/or toxic effects, we analyzed hit ranking by comparing their infectivity and viability Z-scores for both Part 1 and Part 2 of the screen (Figure 3). Among the top hits (i.e., compounds having infectivity Z-scores < -1) in Part 1, we found clustering of derivatives of the tubulin polymerization inhibitors disorazoles, polyketides isolated from Sorangium cellulosum [17], and tubulysins, unusual peptides isolated from Archangium gephyra [18]. In the primary screen both compounds inhibited HIV by 80 - 90% with infectivity and viability Z-scores of < -1.3 and > 0, respectively (Table 1). The tubulin polymerization enhancers epothilones, macrolides isolated from Sorangium cellulosum [19], were also found among the strongest inhibitors in Part 1 of the screen (Figure 3 and Table 1). Other hits were the ATPase inhibitors, apicularen and archazolid [18]. Due to their known function, compounds spirangien B and soraphen F were the most interesting hits from Part 1 of the screen. Spirangien B is an inhibitor of IκBα, a key regulator of the NF-κB signaling pathway [20]. Repressing IκBα has been also shown to inhibit HIV in Jurkat cells [21]. Soraphens are acetyl-CoA carboxylate inhibitors [22]. This enzyme is also suggested to play a role in HIV infection [23] and other viruses requiring fatty acids for replication [24].

In order to further analyze the results of the primary screen, we performed dose-response assays in TZM-bl cells with five of the initial hits and calculated their Selectivity Index (SI), i.e., antiviral potency, based on their respective effective concentration 50 (EC50) and cytotoxic concentration 50 (CC50). As shown in Figure 4 and Table 2 the non-nucleoside reverse transcriptase inhibitor Nevirapine, used as control, produced anti-HIV activity.

**Figure 2** Screen validation and hit identification. (A) The screen protocol was validated with the known HIV inhibitors Zidovudine (AZT), Enfuvirtide (T20) and Indinavir (IND). Values are plotted as % mean luciferase expression relative to solvent controls (B-D) The results of the myxobacterial library screen are shown for Viability (B), infectivity Part 1 (C) and re-infection Part 2 (D). For each test compound, the % mean RLU of the control is plotted vs. the calculated robust Z-score. Hits are defined as compounds inhibiting HIV by at least 50% of the untreated controls (infectivity Z-score < 0) and with less than 30% compound-related toxicity (viability Z-score > 0) (blue boxes). Error bars are standard deviations of quadruple measurements.
SI values similar to those previously described [25]. The myxobacterial compounds sulfangolid C and soraphen F showed EC50 and CC50 values comparable to each other, with SI ranging from 16 to 20. Both epothilon D and spirangien B showed lower EC50 values than Nevirapine, while spirangien exhibited the highest SI value of the five tested myxobacteria metabolites (>50) (Figure 4 and Table 2). Kulkenon was not selective in the follow-up, with a low SI of around 5.

The most interesting hit from Part 2 of the screen was rhizopodin, an actin inhibitor isolated from the myxobacterium Myxococcus stipitatus [26]. Actin filaments are known to be essential for virological synapse formation and HIV cell-to-cell transmission, the main route of HIV infection in naïve cells in vivo [27,28]. Another hit was leupyrrin B1, a macrodial with a unique pyrrole ring that has been shown to inhibit DNA, RNA and protein synthesis in yeast [29] (Figure 3 and Table 1). Interestingly, highly substituted pyroles have been shown to target the HIV reverse transcriptase [30] and, in our primary screen, leupyrrin B1 also showed a mild effect on Part 1 (data not shown). However, the studies needed to test the real potency of these late-acting hits are beyond this report.

In summary, we have established a two-step HTS platform based on the TZM-bl cell line for the screening of novel HIV inhibitors. The advantage of this method is that it can distinguish between compounds acting on early or late events of the HIV replication cycle so that initial hints on the mode-of-action can be already obtained from primary screens. With this assay, a small library of secondary metabolites derived from myxobacteria was screened and interesting primary hits were found. In general, the intrinsic antiviral potency was lower in the follow-up experiments. Unfortunately, these observations are common in HTS campaigns, from which few initial hits are confirmed in subsequent assays [31,32]. In particular, comparative low SI values have been commonly observed for natural products of diverse origin [33-38], and it has also been noted that a compound’s biological efficacy is not due to in vitro toxicity when the SI > 10 [39]. In our experiments, even when dose response assays with five of the initial hits showed a rather low SI compared to a standard FDA-approved drug, four compounds exhibited SI values >15. The screening for natural compounds allows for the discovery of molecules with good effectiveness, which could be further optimized by synthetic methods. The myxobacteria secondary metabolism comprises compounds with large chemical components that might “mask” the actual antiviral pharmacophore (see structures in Table 2). Therefore, further structural studies of our initial hits might help recognize smaller drug-like derivatives that in turn could be synthesized and further optimized.

Conclusions
We have established a robust high-throughput anti-HIV assay that allows, already from the primary screen, to discriminate compounds acting on early or late steps of the virus replication cycle. We identified several secondary metabolites derived from myxobacteria with good anti-HIV profiles. Remarkably, compared to a previous
screen for Hepatitis C Virus (HCV) using the same library, we found a high amount of overlapping hits (Table 1 and [40]), suggesting that these compounds may target commonly used host factors or pathways necessary for viral replication. Although the intrinsic antiviral potency of most of these compounds remains to be elucidated, the unique structures and mode-of-actions of these natural compounds make myxobacteria an attractive source of chemicals for the development of broad-spectrum antivirals [14].

### Materials and methods

#### Cells and culture medium

TZM-bl, a modified HeLa cell-line susceptible to infection with different HIV-1 isolates, was obtained from the NIH AIDS Research and Reference Reagent Program, Cat# 8129) and maintained in Dulbecco’s modified Eagle’s medium (DMEM; Invitrogen, Karlsruhe, Germany) supplemented with 10% heat-inactivated FCS, HEPES 25 mM and 0.5% gentamycin. PM1 cells (NIH AIDS Research and Reference Reagent Program, Cat# 3038) were

### Table 1 Robust Z-score ranking of the myxobacterial hits for both part 1 (upper) and part 2 (bottom) of the screen

| Name         | Abbr. | Z-score | Infectivity | Viability |
|--------------|-------|---------|-------------|-----------|
|              |       |         | % Mean | SD | % Mean | SD |
| **Hits from part 1** | | | | | | |
| Tubulysin | Tsl | -2.47 | 3.70 | 1.27 | -0.08 | 92.04 | 4.38 |
| Kulkennon | Kulk | -2.41 | 5.15 | 1.09 | 0.03 | 93.01 | 18.62 |
| Epothion A | EpoA | -2.38 | 0.49 | 0.15 | 0.24 | 87.72 | 10.27 |
| Spirangien B | SpIB | -2.38 | 1.09 | 0.19 | 0.08 | 85.71 | 4.14 |
| Epothion C | EpoC | -2.37 | 0.54 | 0.14 | -0.25 | 81.63 | 4.07 |
| Epothion D | EpoD | -2.37 | 1.52 | 0.54 | 1.00 | 97.33 | 11.03 |
| Terrestrisbiamid A | TerA | -2.35 | 6.70 | 0.26 | 0.34 | 95.99 | 10.58 |
| Enfuvirtide (Control) | T20 | -2.30 | 2.26 | 0.18 | 1.36 | 97.21 | 0.05 |
| Sulfangolid C | SulfC | -2.29 | 8.13 | 1.26 | 1.73 | 109.20 | 13.03 |
| Zidovudine (Control) | AZT | -2.24 | 4.29 | 0.21 | 1.59 | 98.62 | 1.59 |
| Apicularen B* | ApiB | -1.93 | 13.77 | 2.40 | -0.16 | 82.77 | 4.37 |
| Disorazol A4* | DszA4 | -1.78 | 20.37 | 2.01 | 0.21 | 94.79 | 14.99 |
| Tubulysin B* | TlsB | -1.61 | 24.04 | 1.01 | 0.38 | 89.55 | 10.43 |
| Disorazol B1* | DszB1 | -1.55 | 25.89 | 3.60 | 1.25 | 104.59 | 15.96 |
| Disorazol E1* | DszE1 | -1.54 | 26.22 | 3.20 | -0.13 | 91.48 | 18.83 |
| Stigmatelatin A | StgA | -1.43 | 29.56 | 2.80 | -0.34 | 80.43 | 3.79 |
| Tubulysin A* | TlsA | -1.42 | 29.95 | 4.87 | -0.11 | 83.33 | 9.78 |
| Archazolid A* | ArcA | -1.40 | 30.47 | 4.01 | -0.35 | 80.30 | 4.86 |
| Tubulysin G* | TlsG | -1.34 | 32.46 | 3.73 | 0.34 | 88.98 | 2.54 |
| Sorapheen F | SorphF | -1.32 | 31.43 | 7.72 | 1.58 | 107.80 | 6.05 |
| Disorazol E3* | DszE3 | -1.31 | 31.74 | 4.50 | -0.05 | 92.32 | 3.12 |
| Maltepolid B | MalB | -1.16 | 35.34 | 8.71 | -0.11 | 91.76 | 7.42 |
| Crocacin A | CroA | -1.06 | 41.32 | 6.39 | 0.15 | 86.66 | 2.43 |
| Noricumazol B | NoricB | -1.01 | 43.03 | 6.34 | 0.86 | 95.47 | 5.54 |
| Socein A | SoceinA | -0.97 | 40.09 | 7.31 | 0.68 | 99.20 | 13.69 |
| **Hits from part 2** | | | | | | |
| **Name** | **Abbr.** | **Z-score** | **Infectivity** | **Viability** |
|              |       |         | % Mean | SD | % Mean | SD |
| Rhizopodin A | RhiA | -2.11 | 0.00 | 0.00 | -1.38 | 67.39 | 11.37 |
| Indinavir (control) | IND | -1.93 | 2.73 | 0.10 | 1.43 | 91.21 | 8.50 |
| Leupyrin B1 | LeuB1 | -1.85 | 3.12 | 0.24 | -0.01 | 67.54 | 11.23 |
| Crocapeptin B* | CrpB | -1.80 | 11.38 | 1.78 | -1.40 | 67.17 | 6.95 |
| Soraphen A | SorA | -1.49 | 22.24 | 2.69 | 0.15 | 86.61 | 6.68 |
| Cruentaren A | CruA | -1.25 | 31.05 | 5.38 | 0.66 | 93.01 | 5.04 |

*: Compounds with anti-HCV activities according to [40].

Corresponding % relative infectivity and cell viability is shown. Compounds are listed by infectivity Z-scores. Control drugs are shown for comparison.
maintained with RPMI medium supplemented with 10% heat-inactivated FCS and 1% of penicillin-streptomycin. Both cell-lines were cultured at 37°C, 5% CO₂.

**Virus stocks and infections**

HIV-1LAI isolate was obtained from the Centre for AIDS Reagents, NIBSC, UK. Virus was propagated in PM1 cells and titrated in TZM-bl cells as described [41]. 1 mL aliquots of virus stocks were stored at -80°C until use. Infection experiments in TZM-bl cells were performed in quadruple at a multiplicity of infection (MOI) of 0.5. For drug-response assays TZM-bl cells were plated (10⁴ cells/well) in Nunc® MicroWell 96 well optical bottom plates (Sigma) and incubated for 1 h with increasing concentrations of test compounds in 10-fold dilutions or with the corresponding vehicle (DMSO or MeOH) as negative control in triplicates. After drug incubation, cells were infected with HIVLAI and 48 h after infection luciferase activity was measured using Britelite Plus™ (PerkinElmer, Waltham, USA). In parallel, cell viability of TZM-bl cells was determined with an ATP quantification method using the commercial kit CellTiter-Glo® Luminescent Cell Viability Assay (Promega, Madison, USA). ATP is a marker of the presence of metabolically active cells [42]. Therefore, the ATP levels relative to the untreated control are a measure of drug-induced cytotoxicity. Mean luciferase values were normalized to untreated controls and Effective Concentration 50 (EC₅₀) and Cytotoxic Concentration 50 (CC₅₀) were calculated in GraphPad Prism (GraphPad Software, San Diego, CA, USA) by analyzing the log-dose vs. normalized response. The Selectivity Index (SI) refers to the antiviral potency of a drug and is calculated as the ratio of CC₅₀ to EC₅₀ [43,44].

**Test compounds**

The library of 154 myxobacterial secondary metabolites used for the screening belongs to a collection of natural compounds isolated at the Helmholtz Centre for Infection...
Research, Braunschweig, Germany [11,12,45]. Compounds of > 95% purity as measured by LC-MS were provided in 96-well screening plates in a concentration of 1 mM in puriss.p.a. dimethyl sulfoxide (DMSO) or methanol (MeOH). The fusion inhibitor Enfuvirtide (Fuzeon, Roche, Basel Switzerland) and the nucleoside reverse transcriptase inhibitor Zidovudine (NIH AIDS Research and Reference Reagent Program, Cat# 3485) were used as positive controls at final concentrations of 1 μM.

**Two-step TZM-bl based high-throughput screening assay**

For the Part 1 of the screen, TZM-bl cells were seeded in 384-well plates at a density of 2500 cells per well in 30 μL of culture medium and incubated overnight at 37°C and 5% CO₂. After incubation, 50 to 70 nL of the test compounds and DMSO and/or MeOH controls from the 96-well screening plates were dispensed in quadruple to the cells with the PinTool of an Evolution P3 pipetting platform (PerkinElmer, Zaventem, Belgium). Final concentration of compounds was around 1.5 to 2 μM with a content of around 0.2% of DMSO or MeOH in all cases. DMSO or MeOH alone were used as controls. 2 h after addition of compounds, cells were infected with 50 μL of HIV LAI at a MOI of 0.5. In parallel, duplicate plates were left uninfected for quantification of compound-related toxicity. Plates were incubated at 37°C and 5% CO₂.

| Compound name | Structure | EC₅₀ (μM) | CC₅₀ (μM) | SI* |
|---------------|-----------|-----------|-----------|-----|
| Nevirapine    | ![Structure](image1.png) | 0.07      | 8.18      | >1000 |
| Sulfangolid C | ![Structure](image2.png) | 0.41      | 8.18      | 20.2 |
| Soraphen F   | ![Structure](image3.png) | 0.30      | 5.02      | 16.5 |
| Epothilon D  | ![Structure](image4.png) | 0.0005    | 0.012     | 24.4 |
| Spirangien B | ![Structure](image5.png) | 0.007     | 0.35      | 52   |
| Kulkenon      | ![Structure](image6.png) | 0.07      | 0.36      | 5.3  |

*SI: Selectivity Index.*
Data analysis and statistics
Data was analyzed by obtaining the % mean luciferase values normalized to solvent controls (% control mean RLU) and by calculating the assay Z-factor (a measure for assay quality) and samples (robust) Z-scores (a measure of hit quality) as described [9,10,16]. Briefly, background levels were subtracted from the mean luciferase of samples and values were normalized to those of the solvent controls (set to 100% expression). The assay Z-factor was calculated by dividing 3X standard deviations of controls by the sum of their means as described (see Table 1 in [10]). For hit determination, we adapted a previously described robust Z-score calculation [9] to adjust for interplate variation by dividing the absolute deviation of the mean of the quadruple data points (4 wells for each compound) by the median absolute deviation of each plate. Unless stated otherwise, errors are given as ± SD.

Competing interest
The authors declare that they have no competing interests.

Authors’ contributions
JPM and BH established and performed the screening assay; JPM and EFS performed drug-response assays; HS and RJ provided the myxobacterial library and additional compounds; JPM, JD, RF, FS and AM designed the experiments and wrote the manuscript. All listed authors read and approved the final manuscript.

Acknowledgements
The research is supported by grants from the Bill and Melinda Gates Foundation, Institució Catalana de Recerca i Estudis Avançats (ICREA), the Spanish Ministry of Science and Innovation (SAF2010-21336 and BFU2010-20803), FPI grant number BES-2011-048569.

Author details
1Infection Biology Group, Department of Experimental and Health Sciences, Universitat Pompeu Fabra, Barcelona, Spain. 2Department of Chemical Biology, Helmholtz Centre for Infection Research, Braunschweig, Germany. 3Department of Microbial Drugs, Helmholtz Centre for Infection Research, Braunschweig, Germany. 4Molecular Virology Group, Department of Experimental and Health Sciences, Universitat Pompeu Fabra, Barcelona, Spain. 5Institució Catalana de Recerca i Estudis Avançats (ICREA), Barcelona, Spain.

References
1. Arts EJ, Hazuda DJ: HIV-1 antiretroviral drug therapy. Cold Spring Harbor perspectives in medicine 2012, 2(4):a007161.
2. Back D: New drug interactions in HIV and HCV. Retrovirology 2012, 9:88.
3. de Maat MM, Elhart GC, Hutema AD, Roos CH, Mulder JW, Beijnen JH: Drug interactions between antiretroviral drugs and comedicated agents. Clinical pharmacokinetics 2003, 42:223–282.
4. McIleron H, Meintjes G, Burman WJ, Maatens G: Complications of antiretroviral therapy in patients with tuberculosis: drug interactions, toxicity, and immune reconstitution inflammatory syndrome. J Infect Dis 2007, 196:563–575.
5. Tseng A, Fosy M: Important drug-drug interactions in HIV-infected persons on antiretroviral therapy: an update on new interactions between non-HIV and non-HIV drugs. Current infectious disease reports 2012, 14:67–82.
6. Wei X, Decker JM, Liu H, Zhang Z, Aran RB, Klby JM, Saag MS, Wu X, Shaw GM, Kappes JC: Emergence of resistant human immunodeficiency virus type 1 patients receiving fusion inhibitor (T-20) monotherapy. Antimicrob Agents Chemother 2002, 46:1896–1905.
7. Montefiori DC: Evaluating neutralizing antibodies against HIV, SIV, and SHIV in luciferase reporter gene assays. Curr Protoc Immunol 2005, Chapter 12:Unit 12–11.
8. Brass AL, Dykhoorn DM, Benita Y, Yan N, Engelman A, Xavier RJ, Lieberman J, Elledge SJ: Identification of host proteins required for HIV infection through a functional genomic screen. Science 2008, 319:921–926.
9. Tan X, Hu L, Luquette Li 3rd, Gao G, Liu Y, Ou H, Xi R, Lu Z, Park PJ, Elledge SJ: Systematic identification of synergistic drug pairs targeting HIV. Nat Biotechnol 2012, 30:1125–1130.
10. Zhang Jh, Chung TD, Oldenburg KR: A simple statistical parameter for use in evaluation and validation of high throughput screening assays. J Biomol Screen 1999, 4:67–73.
11. Reichenbach H: Myxobacteria, producers of novel bioactive substances. J Ind Microbiol Biotechnol 2001, 27:149–156.
12. Reichenbach H, Höfte G: Myxobacteria as producers of secondary metabolites. In Drug discovery from nature. 1999:149–179.
13. Pandey S, Sree A, Dash SS, Sethi DP, Chowdhury L: Diversity of marine bacteria producing beta-glucosidase inhibitors. Microb Cell Fact 2013, 12:55.
14. Diez J, Martinez JP, Mestres J, Sasse F, Frank R, Meyerhans A: Myxobacteria: natural pharmaceutical factories. Microb Cell Fact 2012, 11:42.
15. Huttel S, Muller R: Methods to optimize myxobacterial fermentations using off-gas analysis. Microb Cell Fact 2012, 11:59.
16. Malo N, Hanley JA, Cereauzzi S, Pelletier J, Nador R: Statistical practice in high-throughput screening data analysis. Nat Biotechnol 2006, 24:167–175.
17. Hopkins CD, Wipf P: Isolation, biology and chemistry of the disorazoles: new anti-cancer macrodiolides. Nat Prod Rep 2009, 26:585–601.
18. Weissman KJ, Muller R: Discovery and development of the epothilones: a novel class of antineoplastic drugs. Drugs R D 2008, 9:1–10.
19. Rebolli MR, Ritter B, Sasse F, Niggemann J, Frank R, Nourbakhsh M: The myxobacterial compounds spirangien a and spirangien M522 are potent inhibitors of IL-8 expression. ChemBioChem 2012, 13:409–415.
20. Kwon H, Pelletier N, DeLuca C, Genin P, Cisternas S, Lin R, Wainberg MA, Hiscott J: Inducible expression of IpappaAlpha repressor mutants interferes with NF-kappaB activity and HIV-1 replication in Jurkat T cells. J Biol Chem 1998, 273:7431–7440.
21. Jump DB, Torres-Gonzalez M, Olson UK: Soraphen A, an inhibitor of acetyl CoA carboxylase activity, interferes with fatty acid elongation. Biochemical pharmacology 2011, 81:649–660.
22. Steger DJ, Eberharder A, John S, Grant PA, Workman J: Purified histone acetyltransferase complexes stimulate HIV-1 transcription from preassembled nucleosomal arrays. Proc Natl Acad Sci 1998, 95:12924–12929.
