

Selective Class I HDAC Inhibitors Based on Aryl Ketone Zinc Binding Induce HIV-1 Protein for Clearance

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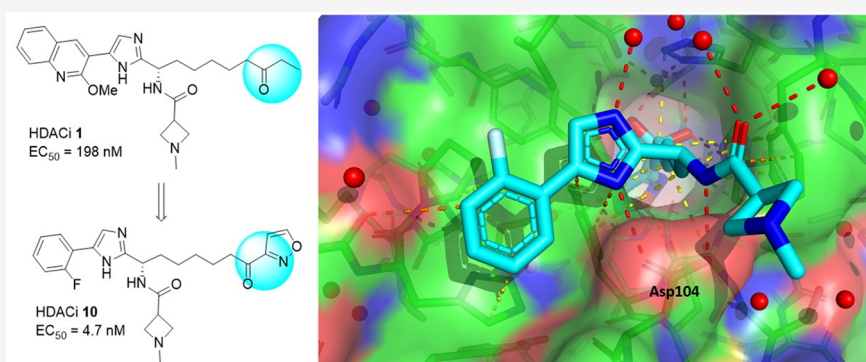
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ABSTRACT: HIV persistence in latently infected, resting CD4⁺ T cells is broadly considered a barrier to eradicate HIV. Activation of the provirus using latency-reversing agents (LRAs) followed by immune-mediated clearance to purge reservoirs has been touted as a promising therapeutic approach. Histone deacetylases (HDACs) and histone acetyltransferases (HATs) control the acetylation level of lysine residues in histones to regulate the gene transcription. Several clinical HDAC inhibitors had been examined as LRAs, which induced HIV activation in vitro and in vivo. Here we report the discovery of a series of selective and potent class I HDAC inhibitors based on aryl ketones as a zinc binding group, which reversed HIV latency using a Jurkat model of HIV latency in 2C4 cells. The SAR led to the discovery of a highly selective class I HDAC inhibitor **10** with excellent potency. HDACi **10** induces the HIV gag P24 protein in patient latent CD4⁺ T cells.

KEYWORDS: HDAC, inhibitors, HIV, latency, LRA

Human Immunodeficiency virus (HIV) has been an epidemic for over 30 years. By 2019, 74.9 million people had been infected with HIV since the beginning of the epidemic and 37.9 million people globally were living with HIV.¹ AIDS (acquired immune deficiency syndrome) caused by HIV used to be a universally fatal disease, but now can be managed as a chronic disease with the discovery and availability of combined antiretroviral therapy (cART). cART available in a single pill dosed once daily can control the virus in patients to below detectable levels, and various longer-acting combinations are in development. However, HIV is not curable with cART due to the establishment of a latent pool of HIV-infected resting memory CD4⁺ T cells that are transcriptionally silent.² These HIV latent CD4⁺ T cells have an extremely long biological half-life and may be activated, resurrecting HIV gene transcription and causing HIV rebound when cART treatment is interrupted.³ Many HIV patients have difficulties continuing the cART treatment due to long-term toxicities, side effects, and the need for daily compliance to

avoid viral resistance. Furthermore, life-long therapy presents a significant financial burden to global health care networks. It is also known that the low level of viremia in HIV patients contributes to chronic inflammation, immune dysfunction, and accelerated aging.⁴ Thus, there is a huge unmet medical need for an HIV cure treatment. One approach toward the HIV cure, often referred to as “shock and kill”,⁵ is to use pharmaceutical latency-reversing agents (LRAs) to reverse HIV-1 latency and turn on production of viral proteins in latently infected CD4⁺ T cells. This activation would expose these cells to killing by an immune-mediated mechanism or via cytopathic effects. LRAs that have been identified through

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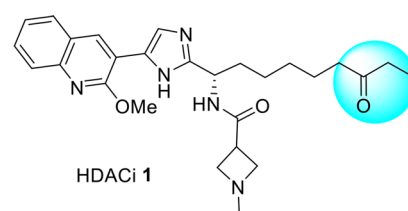
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mechanism-directed approaches include protein kinase C agonists, histone deacetylase inhibitors (HDACi), histone methylation inhibitors, DNA methyltransferase inhibitors, inhibitors of bromodomain and extra terminal domain proteins, and unclassified agents such as disulfiram.⁶

Histone deacetylases (HDACs) and histone acetyl transferases (HATs) together regulate the acetylation level of lysine residues of histones. Histone acetylation diminishes the charge–charge interaction between nucleic acids in DNA, and this relaxed conformation induces gene expression.^{7,8} Eighteen unique isoforms of HDACs have been discovered and are separated into four distinct classes.⁹ Class I (HDACs 1, 2, 3, and 8), Class II (HDACs 4, 5, 6, 7, 9, and 10), and Class IV (HDAC11) are Zn²⁺-dependent, and Class III are the NAD⁺-dependent Sirtuins (SirT1–7). HDAC inhibitors have been developed for oncology, and recently these HDACs have been broadly explored as LRAs. The clinical studies have shown that single and multiple administrations result in induced RNA transcription in CD4⁺ T cells from cART suppressed HIV⁺ subjects^{10–12} using vorinostat, panobinostat, and romidepsin. We recently reported that clinical administration of histone deacetylase inhibitors (vorinostat and panobinostat) induced HIV gag p24, and ex vivo stimulation produced sufficient viral antigen to elicit immune mediated cell killing using anti-gp120/CD3 bispecific antibody.¹³ However, these repurposed HDACis tested in clinical trials are pan-HDAC inhibitors without any subtype selectivity, which cause significant adverse effects (AEs) in clinical tests.^{14–16} The AEs may be acceptable for short-term cancer chemotherapies. However, the AEs would limit the doses for potential chronic treatment of HIV patients. Subtype selective HDAC inhibitors may be helpful to increase the therapeutic window for HIV activation.

Our collaborators at IRBM recently disclosed¹⁷ a series of selective HDAC inhibitors based on ethyl ketone, exemplified by HDACi 1 shown in Figure 1. HDACi 1 is a class I HDAC inhibitor with potent inhibitory activity for HDAC1–3 enzymes. We also profiled HDACi 1 for the ability to reverse HIV latency, using a Jurkat model of HIV latency (2C4 cells), which was produced in the same manner as a similar Jurkat T-cell line that utilized an eGFP reporter gene.¹⁸ In this cell line, reactivation of a quiescent HIV provirus is measured by quantification of a luciferase reporter gene in the HIV provirus. HDACi 1 shows moderate activation efficacy with EC₅₀ at 198 nM in this cell assay. We obtained the X-ray crystal structure of a complex of HDACi 1 with the HDAC2 enzyme at high resolution of 1.6 Å. Several key interactions between HDACi 1 and the enzyme are observed. The ketone exists as the hydrate and forms bidentate chelation with the metal zinc ion. The linear alkane chain linker goes through the narrow hydrophobic tunnel and connects the surface binding group and the ketone. The amide NH and the imidazole form a bidentate chelation with the enzyme Asp104 side chain carboxylic acid. The imidazole and the amide carbonyl form hydrogen bonds with water molecules in the water network. The bicyclic heteroaromatic methoxy quinoline has van der Waals interactions with the protein surface. The ketone is presumed to become hydrated with the water bound to zinc in the HDAC enzyme after binding to the pocket. This X-ray crystal structure shows that there is a foot pocket under the zinc binding ketone, which has also been reported previously.¹⁹ Replacement with an aromatic ring for the ethyl group is possible to fit in the foot pocket to potentially improve the selectivity for class I HDACs, and at the same time enhance



HDACi 1
 HDAC1: IC₅₀ = 13 nM
 HDAC2: IC₅₀ = 18 nM
 HDAC3: IC₅₀ = 12 nM
 2C4 Cell: EC₅₀ = 198 nM (131%)

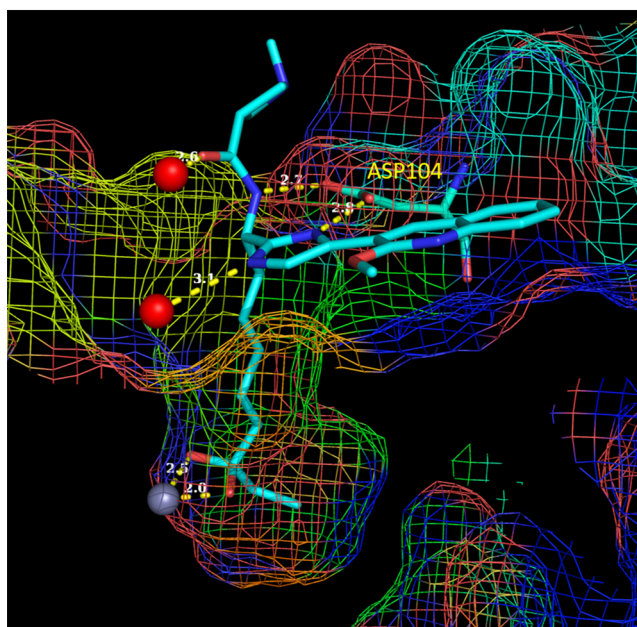
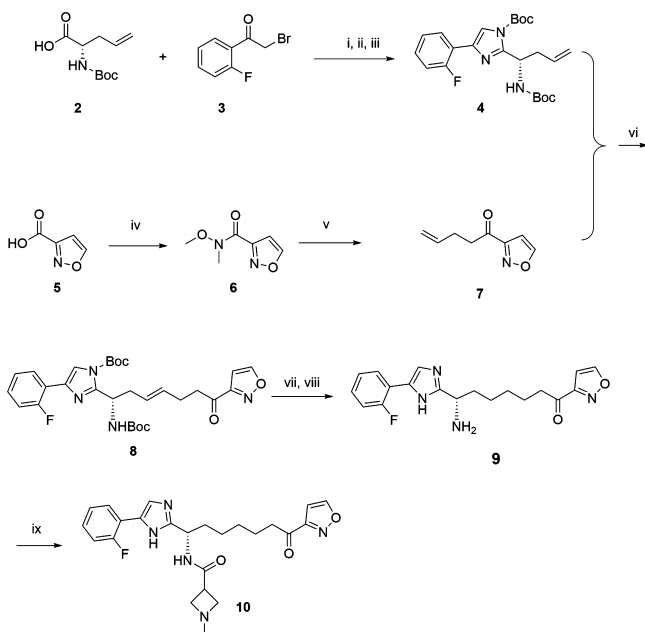


Figure 1. X-ray crystal structure of HDACi 1 bound to HDAC2 enzyme.

the potency and physicochemical properties. Here we report a series of potent and selective class I HDAC inhibitors based on aryl ketones as the zinc binding group for reversing HIV latency.

A synthetic route was designed for the quick exploration of the different aryl ketones as zinc binding groups, depicted in Scheme 1. We prepared two building blocks with terminal double bonds for cross metathesis to quickly explore the structure activity relationships (SAR) of different heteroaryl ketones as the zinc binding group and different substitutions on the amide, the imidazole replacements, and the aryl substitutions on the imidazole. This synthesis is exemplified by the preparation of compound 10. (*S*)-2-Boc-amino-4-pentenoic acid 2 was converted to an ester quantitatively by 2-fluoro-2'-bromoacetylphenone 3 in DMF with cesium carbonate as the base. This ester then condensed with ammonium acetate in toluene under heating to form the imidazole ring, which was then protected with Boc anhydride catalyzed with dimethyl aminopyridine to afford compound 4 in excellent yield. The isoxazole-3-carboxylic acid 5 was converted to the Weinreb amide 6, which later reacted with but-3-en-1-yl magnesium bromide in THF at elevated temperature to provide the aryl ketone compound 7 in good yield. The cross-metathesis reaction between compounds 4 and 7 was successfully carried out in toluene catalyzed by Umicore M71 SIPr ruthenium catalyst²⁰ to form the coupled product 8 with good yield. We experienced much poorer yield for the cross metathesis if the

Scheme 1^a

^aReagents and conditions: (i) Cs_2CO_3 , DMF, 2 h, 100%; (ii) NH_4OAc , toluene, 110 °C, 3 h; (iii) Boc_2O , DMAP, CH_2Cl_2 , rt, 3 h, 92% two steps; (iv) EDCl, HOBT, MMP, $\text{MeONHMe}\cdot\text{HCl}$, DMF/DCM, overnight, 72%; (v) $\text{CH}_2=\text{CHCH}_2\text{CH}_2\text{MgBr}$, THF, 60 °C, 3 h, 100%; (vi) UMICOREM71 SIPR, toluene, 55 °C, 3 h, 78%; (vii) TFA/ CH_2Cl_2 , rt, 92%; (viii) Pd/C, H_2 , MeOH, 92%; (ix) 1-methylazetidine-3-carboxylic acid, MMP, COMU, $\text{CH}_2\text{Cl}_2/\text{DMF}$, 79%.

imidazole in compound 4 is not protected by Boc. The Boc protection on both the imidazole and the amino groups in compound 8 was then removed by treatment of trifluoroacetic acid in methylene chloride, followed by double bond reduction on the resulting product using palladium on carbon catalyzed hydrogenation to give compound 9. The free amine was finally coupled with 1-methylazetidine-3-carboxylic acid using (1-cyano-2-ethoxy-2-oxoethylideneaminoxy)dimethylamino-morpholino-carbenium hexafluorophosphate in DMF/DCM with *N*-methylmorpholine as base to generate compound 10 in good yield.²¹

In the similar synthetic sequence depicted in Scheme 1, the compounds in Table 1 were prepared applying different starting materials for the substitutions Ar_1 , Ar_2 , and R in the general structures. These compounds were evaluated in the inhibitory activity and selectivity for HDAC1, 2, 3, 6, and 8, as well as in a cellular assay for activation of HIV latency. For this initial SAR study searching for a heteroaryl ketone, several five-membered hetero aryls and the pyridinyl were prepared. The 3-isoxazolyl ketone in compound 10 shows the highest potency with subnanomolar inhibition to HDAC1, 2, and 3. This inhibition translated to exceptionally high potency in the cellular HIV latency activation assay with EC_{50} of 4.7 nM. Compound 10 also has excellent selectivity over HDAC6 and moderate selectivity over HDAC8 (37 fold to HDAC1). The 2-oxazolyl (Compound 11) and 2-thiazolyl (Compound 12) are less potent by at least 10-fold in the inhibitory assay compare to 10. 4-Methyl substitution on the oxazolyl (Compound 13) causes further loss of inhibitory activity, while the 5-methyl substitution on the 3-isoxazolyl (Compound 14) is more tolerated with only less than 2-fold loss of

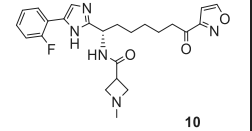
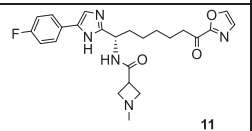
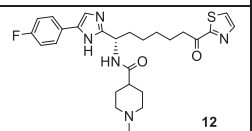
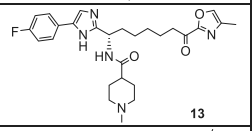
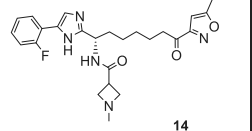
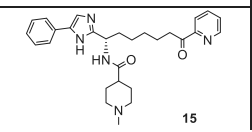
potency. On the other hand, six-membered aryl group such as 2-pyridinyl (Compound 15) is much weaker in the inhibitory assays, possibly due to the larger aromatic ring size and its unfit shape.

To investigate the interaction of compound 10 with the HDAC enzyme, we obtained an X-ray crystal structure of HDACi 10 bound to the HDAC2 enzyme with a resolution of 1.6 Å (Figure 2).²³ HDACi 10 and HDACi 1 have similar interactions with the HDAC2 protein at the surface, the hydrophobic tunnel, the zinc, and the catalytic residues His141, His142, and Tyr304. The key interactions include the bidentate hydrogen bond of the amide NH and imidazole with the side chain of Asp104 and the van der Waal's interaction between the aromatic substitution on the imidazole and the HDAC2 surface (Figure 2A). The amide carbonyl oxygen and the imidazole 3-position nitrogen each have a hydrogen bond interaction with one water molecule in the water network, which was also observed in the crystal structures of HDACi 1 with the HDAC2 enzyme. The ketone hydrate makes bidentate inner coordination sphere contacts to the catalytic zinc ion at the bottom of the tunnel. The enhanced binding potency is introduced by the exceptional fit of the isoxazole ring in the foot pocket where it contacts Met31, Gly301, Gly302, Tyr304, Leu140, His141, and Cys152 (Figure 2B). The structure suggests that substitutions on the isoxazole, or replacement of the isoxazole with larger aryl ring systems, would likely disrupt these optimal contacts and result in steric clashes with the HDAC2 foot pocket.

The favorable interactions of HDACi 10 in the foot pocket with the enzymes led to very potent HDAC 1/2/3 inhibition and the most potent activity in the HIV latency assay. We thus evaluated the intermediate compound 9 in these in vitro assays. Even without the amide, compound 9 still shows excellent HDAC inhibitory activity with an IC_{50} of 8.5 nM against HDAC2 and cellular potency in the HIV latency activation with EC_{50} of 178 nM. This 18-fold loss in potency compared to compound 10 presumably is caused by the loss of a hydrogen bond interaction between the amide carbonyl oxygen and the water network, and the weaker interaction of the amine nitrogen to Asp-104 side chain acid. These results encouraged us to continue the SAR exploration using the amine analogs. HDAC inhibitors (Table 2) using additional five membered heterocycle replacements were synthesized applying the similar chemistry as in Scheme 1 and evaluated in the in vitro assays. The 4-oxazolyl (16), 3-pyrazolyl (17), and 2-imidazolyl (18) all resulted in over 100-fold loss of inhibitory potency compared to compound 9, with no cellular activity. The methyl substitution on the 4-isoxazolyl and 5-hydroxymethyl-2-oxazolyl also led to over 138-fold loss of inhibitory activity. Based on this SAR result, we selected the 3-isoxazolyl ketone as the zinc binding group for further SAR investigation.

We also carried out the SAR to search for heterocyclic replacements to the imidazole ring using the same synthetic strategy as depicted in Scheme 1 for the synthesis of compounds 9 and 10. We recently reported our work on HDAC inhibitors using ethyl ketone as the zinc binding group, in which we described the SAR for the replacement of the surface imidazole.^{24,25} We aimed to reduce a hydrogen bond donor in an attempt to improve the permeability of the compound. The isoxazole in place of imidazole (21) is well tolerated with only a 2-fold loss of inhibitory activity. However, the *N*-aryl triazole (22) and the two *N*-aryl pyrazole (23, 24) all cause significant loss of HDAC inhibitory potency (Table

Table 1. HDAC Inhibitors with Aryl Ketones as the Zinc Binding Groups

Struct & Comp #	HDAC1 IC ₅₀ (nM) ^a	HDAC2 IC ₅₀ (nM)	HDAC3 IC ₅₀ (nM)	HDAC6 IC ₅₀ (nM)	HDAC8 IC ₅₀ (nM)	Cell Assay (5%NHS) ²² EC ₅₀ (nM) (%Act @max dose)
 10	0.10	0.47	0.087	>45000	3.0	4.7 174% @ 2.5 μM
 11	1.1	7.1	3.1	>45000	100	39 138% @0.63 μM
 12	5.8	24	2.0	>45000	380	2800 152% @10 μM
 13	8.3	50	50	>15000	8000	2300 147% @5 μM
 14	0.16	0.76	0.07	>45000	16	20 208% @78 nM
 15	150	240	NT ^b	>1000	NT	NT

^aReported values are the average of ≥ 2 independent measurements with standard deviation less than 3-fold of the reported mean. ^bNT = not tested.

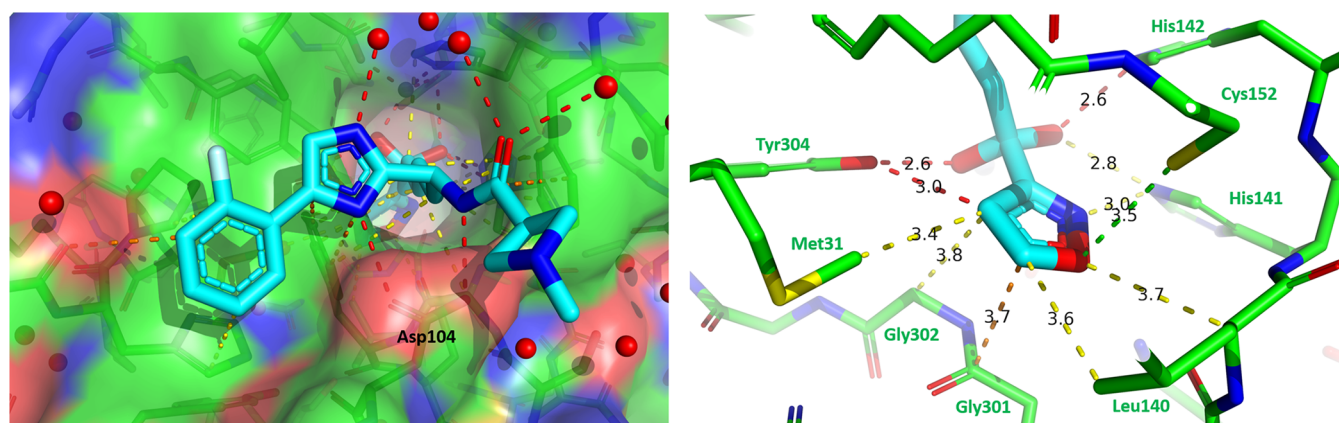


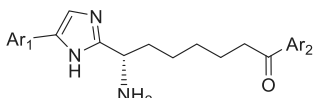
Figure 2. X-ray crystal structure of HDACi 10 bound to HDAC2 enzyme. (A) Top view; (B) Foot pocket. Yellow dashes = vdW contacts; Red dashes = Hydrogen bonds; Green dashes = Hydrogen bond donor- π interactions; Orange dashes = π - π interactions.

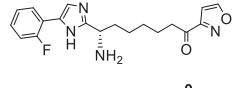
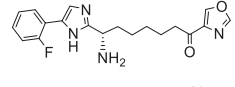
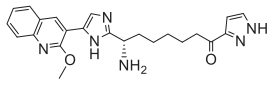
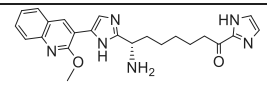
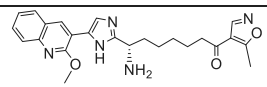
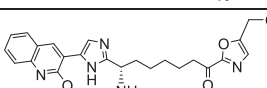
3). We used oxazole and oxadiazole in the ethylketone HDAC inhibitors, and a similar impact on the inhibitory potency was observed.²⁴ Compound 25 with an oxazole replacement for imidazole showed about 2-fold loss of activity compared to

compound 10. By contrast, the oxadiazole replacement compound 26 had over 500-fold loss of inhibitory activity.

We also investigated the binding inhibitory kinetic profile of HDACi 1 and 10, to understand the enormous improvement

Table 2. HDAC Inhibitors with Aryl Ketones as the Zinc Binding Groups



Struct & Comp #	HDAC1 IC ₅₀ (nM) ^a	HDAC2 IC ₅₀ (nM)	HDAC3 IC ₅₀ (nM)	HDAC6 IC ₅₀ (nM)	HDAC8 IC ₅₀ (nM)	Cell Assay (5%NHS) EC ₅₀ (nM) ²² %Act @max dose
 9	1.4	8.5	1.2	>45000	280	178 132% @1.25 μM
 16	170	740	150	>45000	>1000	>40000 21% @20 μM
 17	250	700	230	NT ^b	NT	17000 69% @ 40 μM
 18	1600	4900	1200	NT	NT	NT
 19	207	1300	90	>45000	5000	NT
 20	194	470	120	NT	NT	>20000 107% @ 40 μM

^aReported values are the average of ≥ 2 independent measurements with standard deviation less than 3-fold of the reported mean. ^bNT = not tested.

of inhibitory potency of the 3-isoxazolyl ketone compared to the ethyl ketone. We tested the kinetics of the binding and inhibitory activity to the HDAC3 enzyme applying a global progress curve analysis (GPCA, see the [Supporting Information](#) for details). The ethyl ketone HDACi **1** is a fast on ($k_{on} = 1.70 \times 10^6$ /nM/min) and fast off inhibitor with a residence time of only 5.6 min. The 3-isoxazolyl ketone HDACi **10** is a relatively slower on ($k_{on} = 1.82 \times 10^{-2}$ /nM/min) inhibitor, which still reaches full inhibition within an hour. However, HDACi **10** is an inhibitor with very slow off rate having a residence time of 35 h. This long residence time is presumed to be caused by the tight fit of the isoxazole in the foot pocket. We hypothesize that this long residence time of HDACi **10** may provide prolonged efficacy.

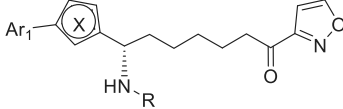
The HDACi **10** was then selected for counter screen against other subtype HDAC enzymes. It is a potent inhibitor to class I HDAC enzymes HDAC1, 2, 3, and has excellent selectivity against other HDACs, except moderate selectivity over HDAC8 (38 fold over HDAC1) (Table 4). These selectivities are the results of the specific interactions described before between compound **10** and the HDAC enzymes.

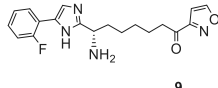
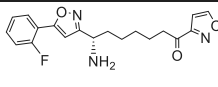
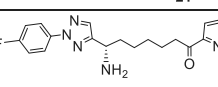
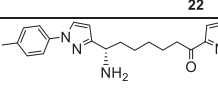
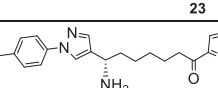
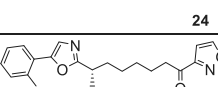
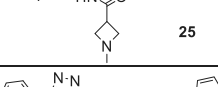
The HDACi **10** was evaluated in pharmacokinetic tests in several preclinical species for predicting the dose of in vivo assays. This compound has quite high total clearance in all the tested preclinical species mouse, rat, and dog, however with reasonable half-life because of its quite high volume of distribution (Table 5). The oral bioavailabilities in mouse,

rat, and dog are moderate with F% as 18, 16, and 7.2, respectively. The plasma protein binding fractions of compound **10** are also listed in Table 5 for these preclinical species. The plasma protein binding fraction in human plasma is 91.5%. Thus, compound **9** has reasonable free fraction in plasma from all species. HDACi **10** demonstrated its in vivo efficacy by elevating the H4 acetylation level and the H1FO gene expression when dosed orally to rats, which will be reported in detail elsewhere.²⁶

We recently reported a robust method to detect HIV *gag* p24 in CD4⁺ T cell lysates from aviremic HIV⁺ individuals following viral reactivation with LRAs.¹³ Using this method, we evaluated HDACi **10** in reversing HIV latency from aviremic patient CD4⁺ T cells and inducing HIV *gag* p24 proteins (Figure 3). CD4⁺ T cells isolated from the blood of HIV⁺ individuals were treated with vehicle (DMSO), HDACi **10** (50 nM, about 1xEC₅₀ free), and vorinostat (400 nM) for 72 h. CD4⁺ T cells were lysed, and *gag* p24 protein was quantified. HDACi **10** significantly induces HIV *gag* p24 protein at 0.06 pg/mL with significant difference ($n = 3$, $p < 0.05$) from vehicle (DMSO) treatment (Figure 3A). This induction is to a greater extent than observed following treatment with 400 nM vorinostat, which is a much weaker HDAC inhibitor with EC₅₀ of 1600 nM in the 2C4 cell activation assay. The higher concentration of vorinostat (2000 nM) caused significant cell death due to its cytotoxicity. Furthermore, dose titration of HDACi **10** from 15 nM to 500 nM resulted in increased levels

Table 3. HDAC Inhibitors on SAR for Imidazole Replacements



Struct & Comp #	HDAC1 IC ₅₀ (nM) ^a	HDAC2 IC ₅₀ (nM)	HDAC3 IC ₅₀ (nM)	HDAC6 IC ₅₀ (nM)	HDAC8 IC ₅₀ (nM)	Cell Assay (5%NHS) EC ₅₀ (nM) ²² %Act @max dose
 9	1.4	8.5	1.2	>45000	280	178 132% @1.25 μM
 21	3.1	21	3.2	>900	603	1700 161% @5 μM
 22	25	81	20	>45000	445	3600 192% @10 μM
 23	21	86	15	>45000	212	1400 191% @10 μM
 24	16	61	13	>45000	466	4300 192 @10 μM
 25	0.20	0.82	0.17	>900	32	14 155% @ 39 nM
 26	51	101	23	>900	>900	2100 171% @ 10 μM

^aReported values are the average of ≥ 2 independent measurements with standard deviation less than 3-fold of the reported mean. ^bNT = not tested.

Table 4. HDACi 10 Selectivity over HDAC Enzymes

Comp #	HDAC1 IC ₅₀ (nM)	HDAC2 IC ₅₀ (nM)	HDAC3 IC ₅₀ (nM)	HDAC4 IC ₅₀ (μM)	HDAC5 IC ₅₀ (μM)	HDAC6 IC ₅₀ (μM)	HDAC7 IC ₅₀ (μM)	HDAC8 IC ₅₀ (nM)	HDAC11 IC ₅₀ (μM)
10	0.08	0.47	0.09	>50	>50	>45	>50	3.0	>50

Table 5. Pharmacokinetic Parameters of Compound 10

PK Parameters	Mouse		Rat		Dog	
	IV dose	PO dose	IV dose	PO dose	IV dose	PO dose
Dose (mg/kg)	2	10	0.5	5	1	2
Cl (mL/min/kg)	54		156		61	
V _{ss} (L/kg)	3.9		37		1.5	
AUCN (μM·h)	1.38		0.118	0.188	0.61	
T _{1/2} (h)	5.2		5.5		2.3	
C _{max} (μM)		0.48		0.050		0.071
T _{max} (h)		0.25		0.38		0.75
F (%)		18		16		7.2
Plasma protein binding %		96.2		88.6		91.2

Plasma protein binding % in human plasma is 91.5.

of viral protein (Figure 3B). For the dose increase to 150 nM and 500 nM, the HIV gag p24 protein expression decreased,

presumably due to cytotoxicity of HDACi 10 at high concentration for extended duration in culture. This class-I HDACi 10 was evaluated in the safety test in the rodent model which showed a limited safety margin; however, the report is out of the scope for this letter and would be published separately.

In summary, we described the identification of a series of class-I HDAC inhibitors based on an aryl ketone. SAR of the aryl ketone led to the discovery of the 3-isoxazolyl ketone as the best zinc binding group for the HDAC inhibitors, which have excellent HDAC1, 2, 3 inhibitory activity and are highly selective against other subtype HDACs. The X-ray crystal structure of HDACi 10 bound to the HDAC2 enzyme demonstrates that favorable interactions of the isoxazole group in the foot pocket of HDAC2 lead to the high potency. The lead HDACi 10 can activate HIV latency in a Jurkat model using 2C4 cells, with exceptional single digit nanomolar potency. HDACi 10 has an acceptable pharmacokinetic profile

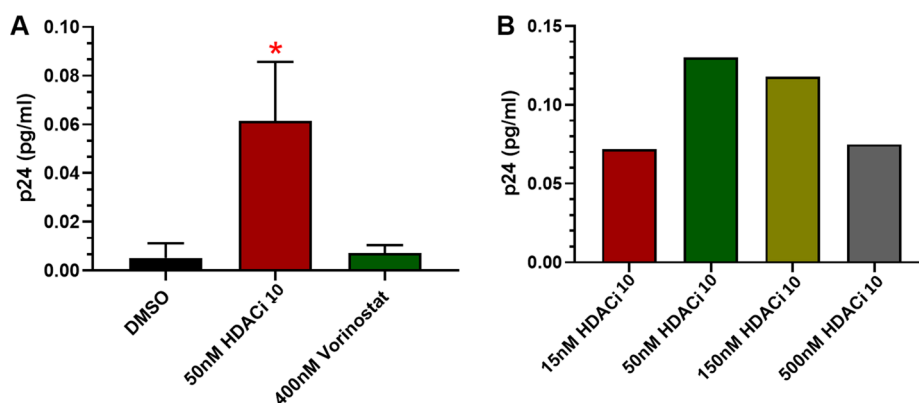


Figure 3. HDACi 10 activates cART-suppressed HIV patient CD4⁺ T cells with detectable HIV gag p24 proteins. (a) HDACi 10 activates aviremic patient's CD4⁺ T cell HIV latency and induces HIV gag p24 proteins significantly higher than the vehicle, vorinostat. ($n = 3, p < 0.05$). (b) Dose titration of HDACi 10 on activation of aviremic patient's CD4⁺ T cell HIV latency and induces HIV gag p24 proteins.

in preclinical species with moderate oral exposure. The binding inhibitory kinetic assays demonstrated that the 3-isoxazolyl ketone HDACi 10 has a very slow off rate from the HDAC3 enzyme. Furthermore, HDACi 10 activates ART-suppressed HIV⁺ patient latent CD4⁺ T cells and induces the expression of HIV gag p24 proteins. Overall, we have identified a series of class-I HDAC inhibitors with excellent inhibitory potency and selectivity, in which the lead HDACi 10 demonstrates efficacy to activate HIV latency and induces protein p24 for potential cellular kill of HIV latent CD4⁺ T cells when combined with other killing methods.

■ ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge at <https://pubs.acs.org/doi/10.1021/acsmchemlett.0c00302>.

Synthetic methods, characterization of key compounds, biology assay protocols, and data (PDF)

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Notes

The authors declare no competing financial interest.

■ ABBREVIATIONS

HDAC, histone deacetylase; HDACi, histone deacetylase inhibitor; HIV, human immunodeficiency virus; LRA, latency reversing agency; AIDS, acquired immune deficiency syndrome; cART, combined antiretroviral therapy; AEs, adverse effects; SAR, structure activity relation; DMF, dimethylformaldehyde; THF, tetrahydrofuran; DCM, dichloromethane; DMAP, 4-dimethylaminopyridine; EDCI, 1-(3-dimethylaminopropyl)-3-ethylcarbodiimide hydrochloride; HOBT, 1-hydroxybenzotriazole hydrate; MMP, 4-methylmorpholine; TFA, trifluoroacetic acid; MeOH, methanol; COMU, (1-cyano-2-ethoxy-2-oxoethylideneaminoxy)dimethylamino-morpholinocarbenium hexafluorophosphate; GPCA, global progress curve analysis; PK, pharmacokinetic.

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