A Highly Potent and Safe Allosteric HIV-1 Integrase Inhibitor, STP0404

Seohyun Ahn[1], U. Kim[1], W. Y. Seo[1], S. Choi[1], T. Maehigashi[2], J. Lindenberger[3], B. Mahboubi[2], C. Gavegnano[2], J. Xu[2], M. Kvaratskhelia[3], Baek Kim[2], Kyungjin Kim[1]
[1]ST Pharm Co. Ltd., Seoul, South Korea, [2]Center for Drug Discovery, Emory University, Atlanta, GA, USA, [3]University of Colorado Anschutz Medical Campus, Aurora, CO, USA

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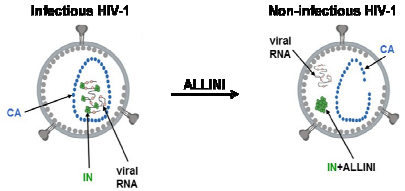
BACKGROUND and SUMMARY

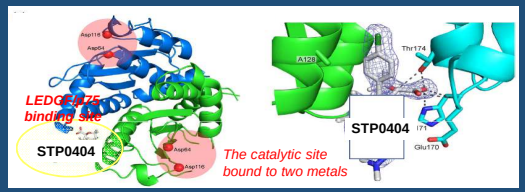
Allosteric Integrase Inhibitors (ALLINIs) are a new class of anti-HIV agents that target the host LEDGF/p75 binding site of the viral integrase (IN) and also interfere with the IN-viral RNA interaction which is essential for viral maturation process. While a number of ALLINI candidates have been developed, no ALLINIs have been advanced to clinical applications, potentially due to their toxicity and efficacy issues. Here, we report discovery and development of a highly potent and safe ALLINI STP0404 with outstanding antiviral efficacy and preclinical properties.

MODE OF ACTION/ALLINIs



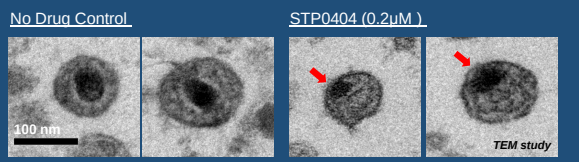
Aberrant IN oligomerization and loss of IN binding to viral genomic RNAs are induced by ALLINI binding to LEDGF/p75 binding site of IN, leading to mislocalization of viral RNA during viral maturation and formation of non-infectious viral particles[1].

Figure 1. Proof-of-Concept by X-ray crystal structure



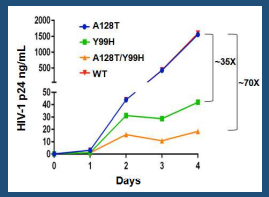
- Binding and interaction mode of STP0404 with HIV-1 IN was determined by solving the X-ray crystal structure of HIV-1 IN catalytic core domain (CCD)-STP0404 complex.
- STP0404, as an ALLINI, binds to the LEDGF/p75 binding site formed between two IN monomers.

Figure 2. Novel Mode of Action of STP0404



- vRNA can be seen inside of viral capsid with high electron density in TEM (No Drug Control).
- STP0404 interferes with IN-RNA binding with high electron density at the outside of viral capsid cores[2].
- New MOA for HIV-cure as "maturation inhibitor"

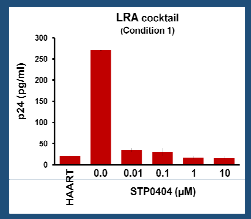
Figure 3. Replication kinetics of resistant viruses



- Y99H/A128T are STP0404 resistant mutations that significantly reduced HIV-1 replication.
- Y99H is responsible for replication delay of Y99H/A128T mutant virus.

- *CEMx174 cells were infected with HIV-1 89.6 to analyze replication kinetics.*
- *In comparison with WT HIV-1 replication: Y99H mt: 35-fold reduction and A128T/Y99H mt: 70-fold reduction*

Figure 4. Anti-HIV-1 reactivation activity from latently infected primary resting T cells



- Anti-HIV-1 rebound activity from latently infected T cells were determined with three reactivation agents (LRA, PMA, and IL-15)[2,3].
- STP0404 effectively suppresses HIV-1 rebound in all three reactivation conditions.

- *Condition 1: Pre-treated with STP0404 before infection, but removed during infection and thereafter.*

CONCLUSION

- **First therapeutic candidate targeting ALLINI.**
- **Significant antiviral activity against Raltegravir-resistant strains.**
- **Y99H (35-fold) and A128T/Y99H (70-fold), STP0404-resistant mutations, significantly reduce HIV-1 replication kinetics.**
- **As a maturation inhibitor, proof-of-concept of ALLINI & novel MOA of vRNA mislocalization were confirmed.**
- **STP0404 significantly suppresses HIV-1 rebound from latently infected primary T cell reservoir.**
- **HIV-1 natural variants, A124N and T125A, display wild type level sensitivity to STP0404 in CEMx174 cells.**
- **No significant clinical signs in MTD & DRF toxicity studies.**
- **No toxicity issues after 4-week GLP repeated toxicology study in rat & dog.**
- **Exploring the therapeutic potential of long-acting ARV is on-going: i) weekly *po* regimen, ii) monthly *im* or *sc* regimen.**
- **STP0404 will be moving to phase 1 clinical development in Q1 2020.**

RESULTS

Table 1. Physicochemical property and ADME

ADME				
Physicochemical property	MW	LogD	pKa	
	495.02	2.05	3.23	4.57
Equilibrium Solubility (μg/ml)	FaSSGF	FeSSIF	FaSSIF	Water
	131	320	292	49.8
Kinetic Solubility (μg/ml)	pH 2.0		pH 7.4	
	389		>500	
	Rat	Dog	Monkey	Human
Microsomal stability ($T_{1/2}$, min)	>145	>145	133.3	135.9
Hepatocytes ($T_{1/2}$, min)	44.4	24	>216.8	>216.8
Plasma stability (%, 60 min)	104	97.8	100	99
Plasma protein binding (%)	99.2	99.7	99.5	99.5
Fresh human serum binding (%)	99.6			
CYP Inhibition (IC_{50}, μM) (1A2/2B6/2C8/2C9/2C19/2D6/3A4-M/3A4-T)	NI/35.2/13.4/41.8/>50.0/NI/>50.0/>50.0			
CYP Induction (EC_{50}, μM)	CYP3A metabolism		PXR activation	
	17		>100	
Off-target				
Lead Profiling Screen	68 receptors & Ion Channels		No activity	
Kinase Profiling Screen	468 kinases		No off-target effect	

Table 2. Antiviral activity and cytotoxicity

*TI: Therapeutic Index

Cell lines	HIV-1 strains	EC_{50} (nM)	CC_{50} (μM)	TI*
PBMC	NL4-3	0.24	>10,000	>41,075
MT-4		3.65	>10,000	>2,740
CEMx174	89.6	1.23	>65,610	>53,341

Table 3. Antiviral activity in Raltegravir-resistant strains

Compounds	Average IC_{50} (range, nM)	
	PBMC	MT-4
STP0404	0.08 (0.02~0.22)	2.49 (0.95~3.48)
Zidovubine	7.96 (0.22~20.7)	37.94 (29.7~57.6)
Raltegravir	1,227.70 (12.5~3,036)	2525 (351~4,322)
Elvitegravir	-	2751.5 (276~10,000)
Dolutegravir	-	4.57 (3.07~8.54)

RAL-resistant strains: 4736_2, 4736_4, 8070_1, 8070_2, 1566_1

Table 4. Pharmacokinetic parameters

Parameters	Cyno-Monkey		Beagle Dog		SD Rat	
	1 mpk (p.o)	1 mpk (i.v)	2 mpk (p.o)	2 mpk (i.v)	10 mpk (p.o)	5 mpk (i.v)
$T_{1/2}$ (hr)	5.25	8.02	6.90	6.11	4.56	3.83
AUC (hr·nM)	950	3,601	4,683	9,260	78,047	42,676
C_{max} (nM)	193	-	3,983	-	21,380	-
F_t (%)	26.9	-	50.6	-	92.8	-

Table 5. GLP toxicology study

Genetic Toxicity	
Ames test	Not mutagenic
Chromosomal Aberration test	Not mutagenic
Micronucleus test	Not genotoxic
Safety Pharmacology	
Central Nervous System	No effects
Respiratory System	
Cardiovascular System	
Repeated-Dose Toxicity	
NOAEL	Males / Females
Rats (4w + R2w)	300 mpk / 600 mpk
Dogs (4w + R2w)	90 mpk

- In dog (4-wk GLP tox study), there were no STP0404-related clinical signs and toxic changes observed.
- More than 30 different organs were evaluated, but there were no STP0404-related gross lesions and histological findings.

Table 6. First in human dose calculation[4]

Species	Dose (mpk)	Species	Equivalent Dose (mpk)
Dog (Km 20.00)	90	Human (Km 37.00)	48.65

FIH calculation

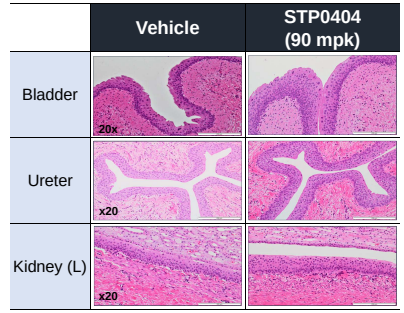
- 70 kg adult, qd (48.65 x 70) = 3405.5 mg
- First in human (FIH) 3405.5/10 = **340.5 mg**

Table 7. Phase 1 study design

Study	Cohorts (subjects)	Dose (mpk)
SAD	4 (32)	200, 400, 600, 800
MAD	3 (30)	200, 400, 600
FE	2 (12)	200

Clinical Site: Eurofins OPTIMED, France

Figure 5. Dog 4-week toxicology study



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2. J. Kessl et al., *Cell*, 2016, 166, 1257
3. L. Vranckx et al, *EbioMedicine*, 2016, 248.
4. ICH S9 Guideline.

ACKNOWLEDGEMENT

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