Significant *in vitro* and *in vivo* inhibition of HBsAg and HBV pgRNA with ASC42, a novel non-steroidal FXR agonist

Xiaodong Li[1], Handan He[1], Qiong Zhou[2], Jinzi J.Wu[1]
1. Ascletis Bioscience Co., Ltd; 2. WuXi AppTec (Shanghai) Co., Ltd

INTRODUCTION

Hepatitis B virus (HBV) infection remains a major public health problem, and a better understanding of the mechanisms that control viral persistence and reactivation is essential for proposing new therapeutic targets to increase the rate of functional cure. Previous studies showed that activating Farnesoid X Receptor (FXR) can inhibit the transcriptional activity of cccDNA, and reduce the level of HBsAg [1].

ASC42 is an in-house developed, novel non-steroidal, selective and potent FXR agonist. The oral tablet formulation of ASC42 has been developed with the in-house proprietary technology and is stable at room temperature. As a candidate drug for NASH, ASC42 has recently received U.S. FDA Fast Track Designation.

AIM

The aim of this study was to evaluate the anti-HBV efficacy of an FXR agonist ASC42 via *in vivo and in vitro* studies.

METHOD

In vitro study: On Day 1, Primary human hepatocyte (PHH) cells (1600 HBV Ge / cells) were infected with type D HBV virus. On the second day, the cells were treated with ASC42, Entecavir(ETV), and vehicle (as negative control) respectively for 8 days, respectively. On the tenth day, the supernatant was collected for detection of HBV DNA, HBV RNA and HBsAg.

In vivo study: Male C57BL/6 mice were injected with AAV/HBV (type D) via tail vein to establish chronic HBV replication mode. From day 0 to day 27, all groups of mice were given intragastric administration once daily. The dosage of ETV was 0.1 mpk, and 10, 30 and 60 mpk for ASC42. Plasma was collected at - 1, 7, 14, 21 and 28 days post-dosing. The anti HBV efficacy of the compounds was evaluated by quantitative detection of HBV pgRNA, HBV DNA and HBsAg in plasma samples.

RESULTS

***In vitro* study (PHH model):**

- ASC42 showed a significant inhibition on HBV RNA with EC50 of 0.09 μM, while ETV showed no inhibition on supernatant HBV RNA(Figure 1. A, B).
- ASC42 significantly reduced HBsAg with EC50 of 0.79 μM, while ETV showed no inhibition on supernatant HBsAg(Figure 1. C, D).
- ASC42 and ETV showed a dose-dependent inhibition on supernatant HBV DNA, with EC50 of 0.62 μM and 0.009 nM, respectively (Figure 1. E, F).

***In vivo* study (AAV/HBV mouse model):**

- ASC42 dose-dependently inhibited HBV pgRNA, HBsAg, HBV DNA in mouse plasma. High-dose group of ASC42 (60 mg/kg) inhibited HBV pgRNA, HBsAg, and HBV DNA by 0.60 log10 copy/μl ($p<0.01$), 0.38 log10 IU/μl ($p=0.002$), and 0.77 log10 copy/μl ($p<0.05$), respectively, in relative to vehicle control group (Figure 2).
- After ETV (0.1 mg/kg) treatment, HBV DNA in mouse plasma decreased significantly, while HBV pgRNA and HBsAg showed no obvious reduction.

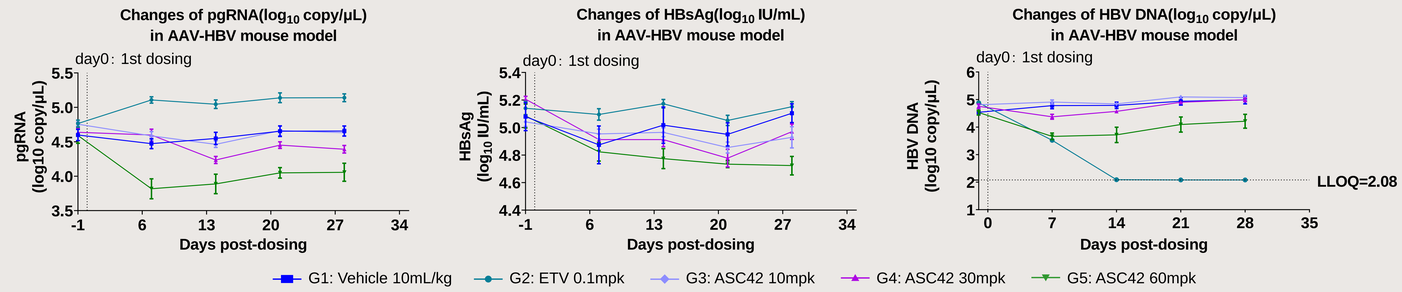

Figure 2. Effects of ASC22 and ETV on the reduction of pgRNA, HBsAg and HBV DNA in AAV/HBV mouse model

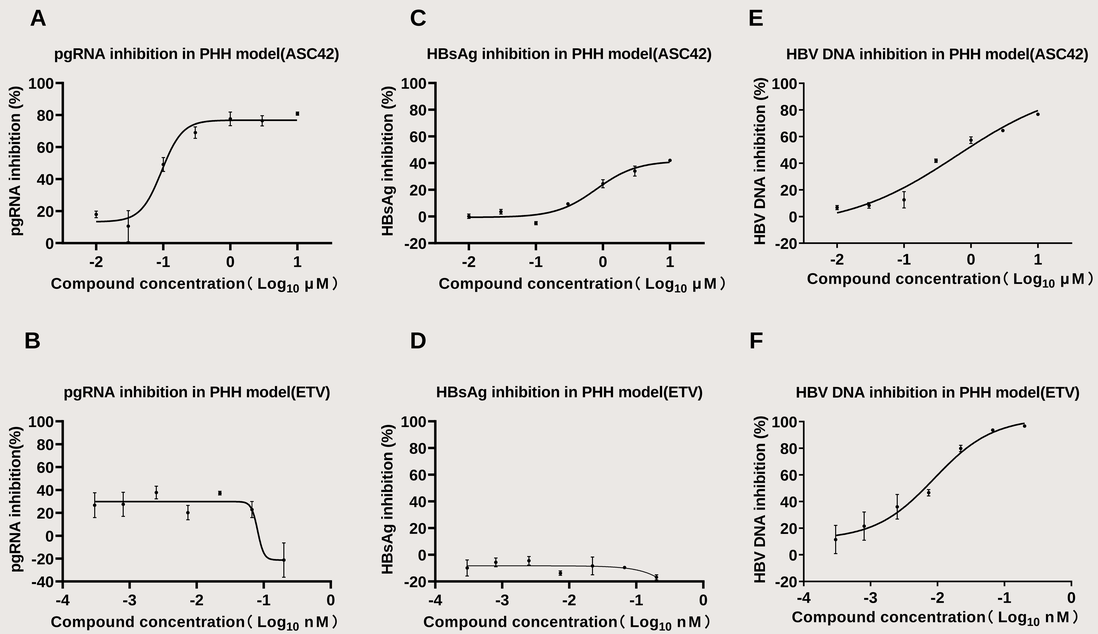

Figure 1. Effects of ASC22 and ETV on the antagonism of pgRNA, HBsAg and HBV DNA in PHH model

CONCLUSION

These *in vitro* and *in vivo* studies demonstrated that ASC42, a FXR agonist, significantly inhibited HBV pgRNA and HBsAg, indicating that ASC42 has therapeutic potential to functional cure of HBV infection. The results support the advancement of ASC42 into clinical trials in human.

REFERENCES

[1] Mouzannar K, Fusil F, Lacombe B, et al. Farnesoid X receptor-alpha is a proviral host factor for hepatitis B virus that is inhibited by ligands *in vitro* and *in vivo* [J]. FASEB journal 2019, 33(2): 2472-83. DOI: 10.1096/fj.201801181R.

CONTACT INFORMATION

Ascletis BioScience Co., Ltd.
website: https://www.ascletis.com
Address: 12F, Building D, 198 Qidi Road, HIPARK, Xiaoshan District, Hangzhou, Zhejiang Province, PRC, 311200
Tel: +86 571 85389730