AASLD The Liver Meeting®

Dual-Targeting siRNA (HBx and PD-L1) Achieved Very Rapid and Durable Clearance of HBsAg and HBV DNA in Mice

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INTRODUCTION

- Chronic hepatitis B (CHB) is a global public health burden affecting approximately 257.5 million people worldwide [1]. In recent years, tremendous progress has be made for CHB drug development. However, low functional cure rate and high ratio of viral rebound after treatments stop are still two major challenges in clinical studies [2].
- We reported siRNA targeting PD-L1 in hepatocytes can effectively clear HBsAg and HBV DNA in mice. It also showed strong synergistic effects when combined with siRNA and ASO which directly inhibit HBV mRNA [3].
- We developed a dual-targeting siRNA (HBx and PD-L1 gene). In one molecule, it very effectively and rapidly cleared HBsAg and HBV DNA in mice without viral rebound.

AIM

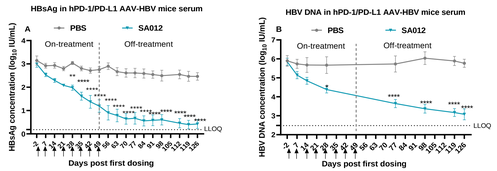
- To develop a platform technology for dual-targeting siRNA drugs;
- To achieve functional cure of CHB using a dual-targeting siRNA which simultaneously clears viral infection and enhances acquired immunity.

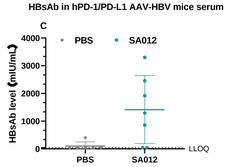
METHOD

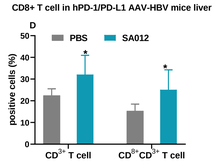
- hPD-1/PD-L1 AAV-HBV mice have been used in the SA012 alone and SA1211 efficacy evaluation studies.
- hPD-L1 HDI and Tg-HBV mice have been used in SA1211 maintaining respective activity evaluation experiments.
- Serum HBsAg, HBV DNA and/or HBsAb were detected at different time points during each study.
- Additionally, the safety profile, such as biochemistry, routine blood test and histopathology were assessed in rat and cynomolgus monkey toxicity studies with SA1211 Q2W for 3 doses.

RESULTS

1. SA012 (targeting PD-L1) can effectively clear HBsAg and HBV DNA by enhancing acquired immunity

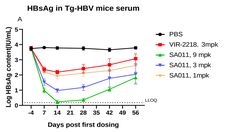


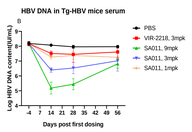




- 5/7 and 4/7 mice achieved the low limit of quantification (LLOQ) for HBsAg and HBV DNA, respectively. No obvious viral rebound was observed 11 weeks after SA012 discontinuation.
- SA012 induced high levels of HBsAb and 5/7 mice reached seroconversion after treatment.
- The number and percentage of $CD3^+$ and $CD3^+CD8^+$ T cells increased significantly in the liver, indicating the restoration of acquired immunity towards HBV infection.

2. SA011 (targeting HBx) is potent to rapidly reduce HBsAg and HBV DNA



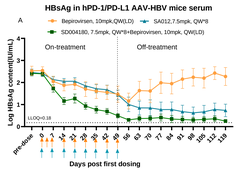


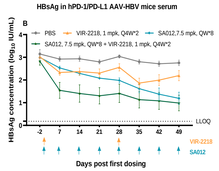
Scheme 1. Structure of Dual Targeting siRNA SA1211

SA012：anti-PD-L1
Enhance acquired immunity
Inhibit viral rebound

SA011：anti-HBx
Clear HBsAg and HBV DNA

3. SA012 shows strong synergy when combined with Bepirovirsen/ VIR-2218

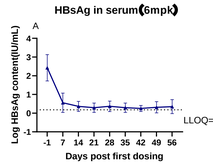


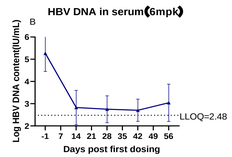


- (Up) Bepirovisen is an anti-HBV ASO. VIR-2218 is an anti-HBV siRNA. They had shown good results in HBV clinical trials. A: Serum HBsAg change after SA012 combined with Bepirovisen. B: Serum HBsAg change after SA012 combined with VIR-2218.
- When SA012 was combined with VIR-2218 or Bepirovirsen, synergistic effect in HBsAg reduction was observed.

- (Left) SA011 is a SiranBio developed GalNAc-conjugated siRNA targeting HBx. By single treatment, it showed rapid, potent and dose-dependent reduction of HBsAg and HBV DNA.

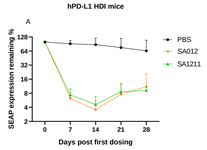
4. SA1211 single dose can clear HBsAg and HBV DNA in hPD-1/PD-L1 AAV-HBV mice

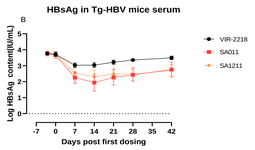




- Single dose treatment of SA1211 achieved rapid reduction of HBsAg and HBV DNA. 4/5 and 4/5 mice achieved the low limit of quantification (LLOQ) for HBsAg and HBV DNA **without viral rebound.**

5. SA1211 well maintained potency of each siRNA





- The potency of SA1211 in reducing PD-L1 mRNA in hPD-L1 HDI mice model was evaluated to be equivalent to that of SA012 alone.
- SA1211 has the same anti hepatitis B activity as SA011 molecule in Tg-HBV mice.
- **SA1211 effectively preserved the activity of each siRNA component.**

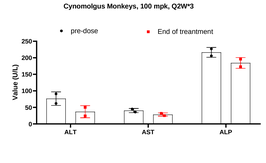
6. SA1211 shows excellent safety profile

A. In the 4-Week Subcutaneous Repeat-Dose Toxicity Study in Sprague Dawley Rats, the MTD was 200 mg/kg.

- No averse effect was observed in Tox studies, including body weight, food consumptions , clinical hematology and biochemistry.
- Pathologic dissection revealed mild to moderate changes in liver (vacuolar and basophilic granules in some animals), which is common for GalNAc-conjugated siRNAs.

B. The 4-Week Subcutaneous Repeat-Dose Toxicity Study in Cynomolgus Monkeys is on going.

- The experiment has ended and pathological testing is in progress.
- No test sample-related adverse effect was observed, including food intake, body weight, hematology and coagulation, serum biochemistry, immune functions and histopathological examinations.



- SA1211 showed excellent preclinical safety profiles in cynomolgus monkeys and in rats.

CONCLUSIONS

- We have developed two siRNA molecules which target PD-L1 and HBV X mRNA respectively, and combined them into a dual-targeting siRNA drug SA1211.
- SA1211 can reduce viral replication and enhance acquired immunity. It achieved rapid and sustainable clearance of HBsAg and HBV DNA in humanized PD-1/PD-L1 AAV-HBV mice.
- SA1211 showed excellent safety profiles in rats and NHP, which supports its further clinical development.

REFERENCES

1. Polaris Observatory Collaborators. Global prevalence, cascade of care, and prophylaxis coverage of hepatitis B in 2022: a modelling study[J]. Lancet Gastroenterol Hepatol. 2023.
2. Sequential Peg-IFN after bepirovirsen may reduce post-treatment relapse in chronic hepatitis B, Buti, Maria et al. Journal of Hepatology, Articles in Press .
3. Potent and sustainable HBsAg clearance and HBV DNA negative by a liver-targeting PD-L1 siRNA in mice. EASL 2024, poster top 354.

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