# HBV-TAG

## 2021 CONFERENCE

# HBV (sAg) Targets that affect HDV

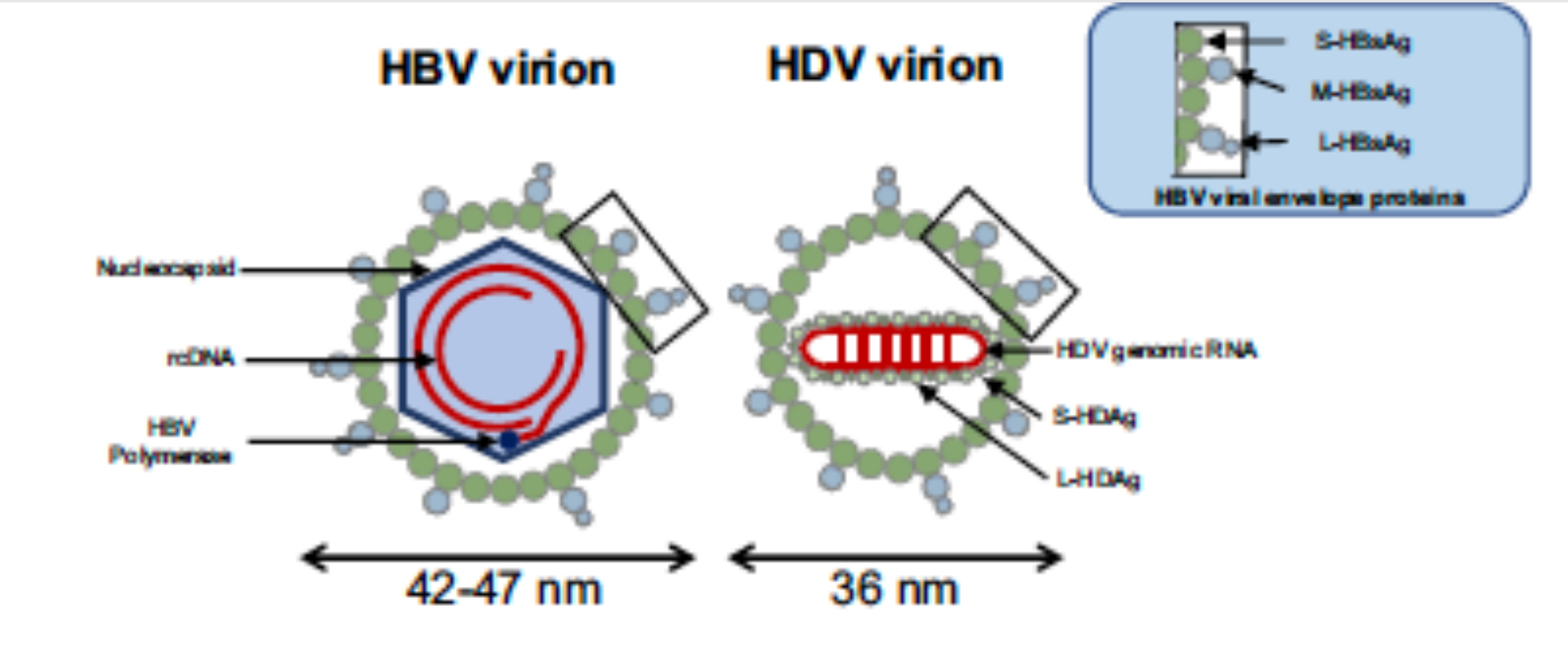
- Briefly review the most advanced experimental HDV antivirals
- Bring your attention to a new therapeutic approach  in development

**Conflicts:**

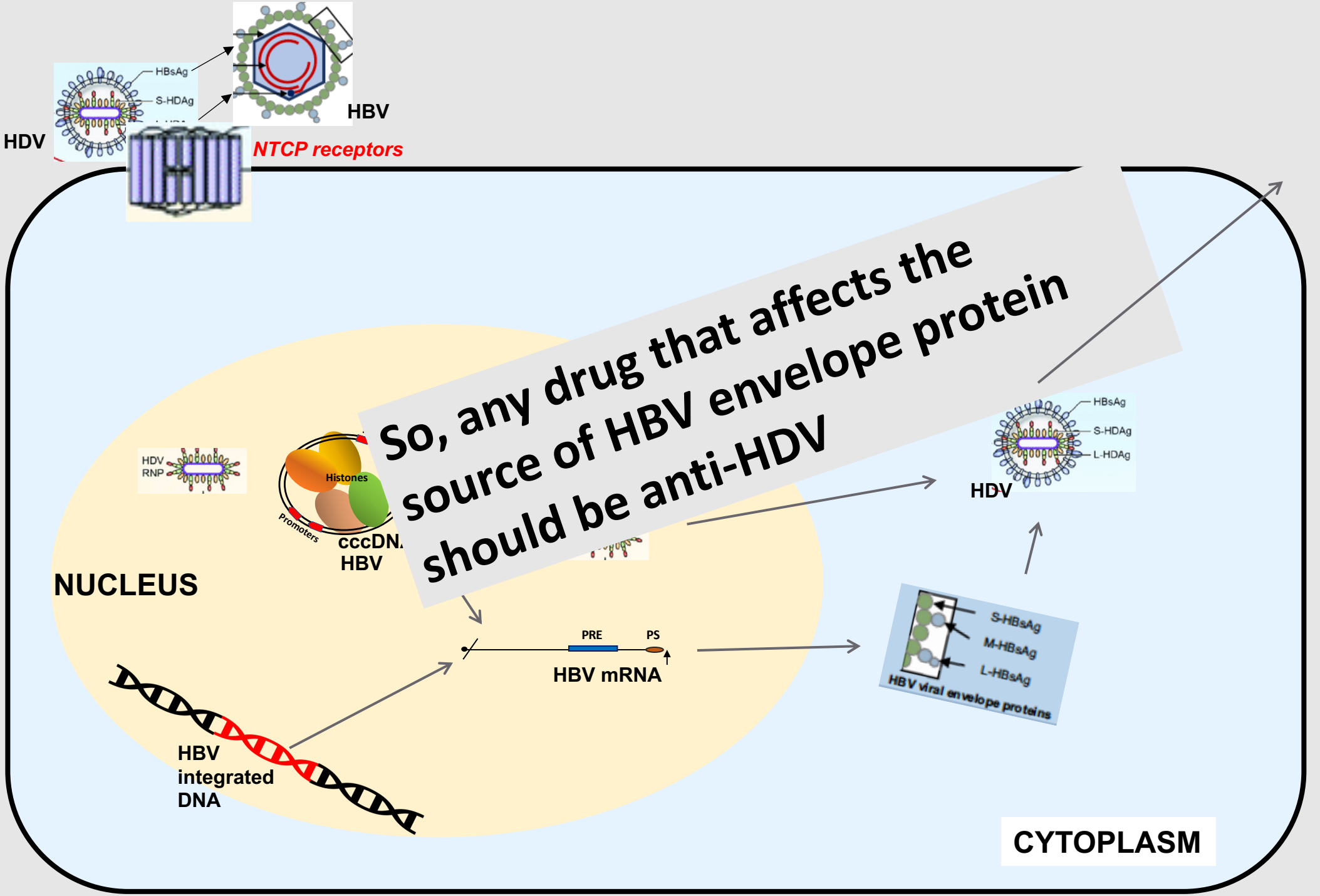
- Hepion (Board Member)
- Antegene (SAB Member)
- Co-Inventor (DHQ-E, several other experimental HBV drugs)

- President and Board Member:
  Hepatitis B Foundation and its Baruch S Blumberg Institute/Pennsylvania Biotechnology Center

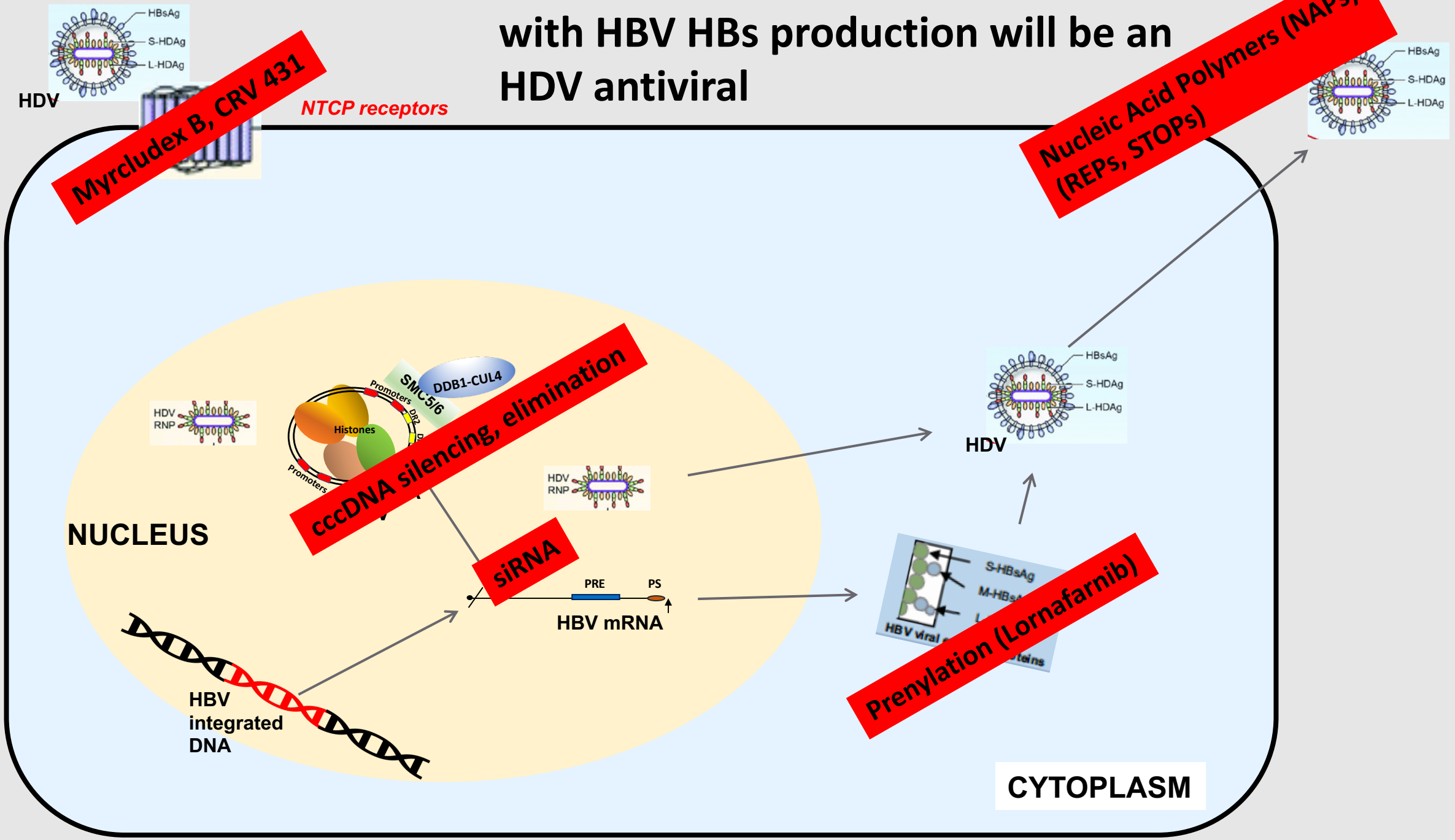
# HDV depends on HBV envelope polypeptides (SHBs, LHBs) to complete its replication cycle

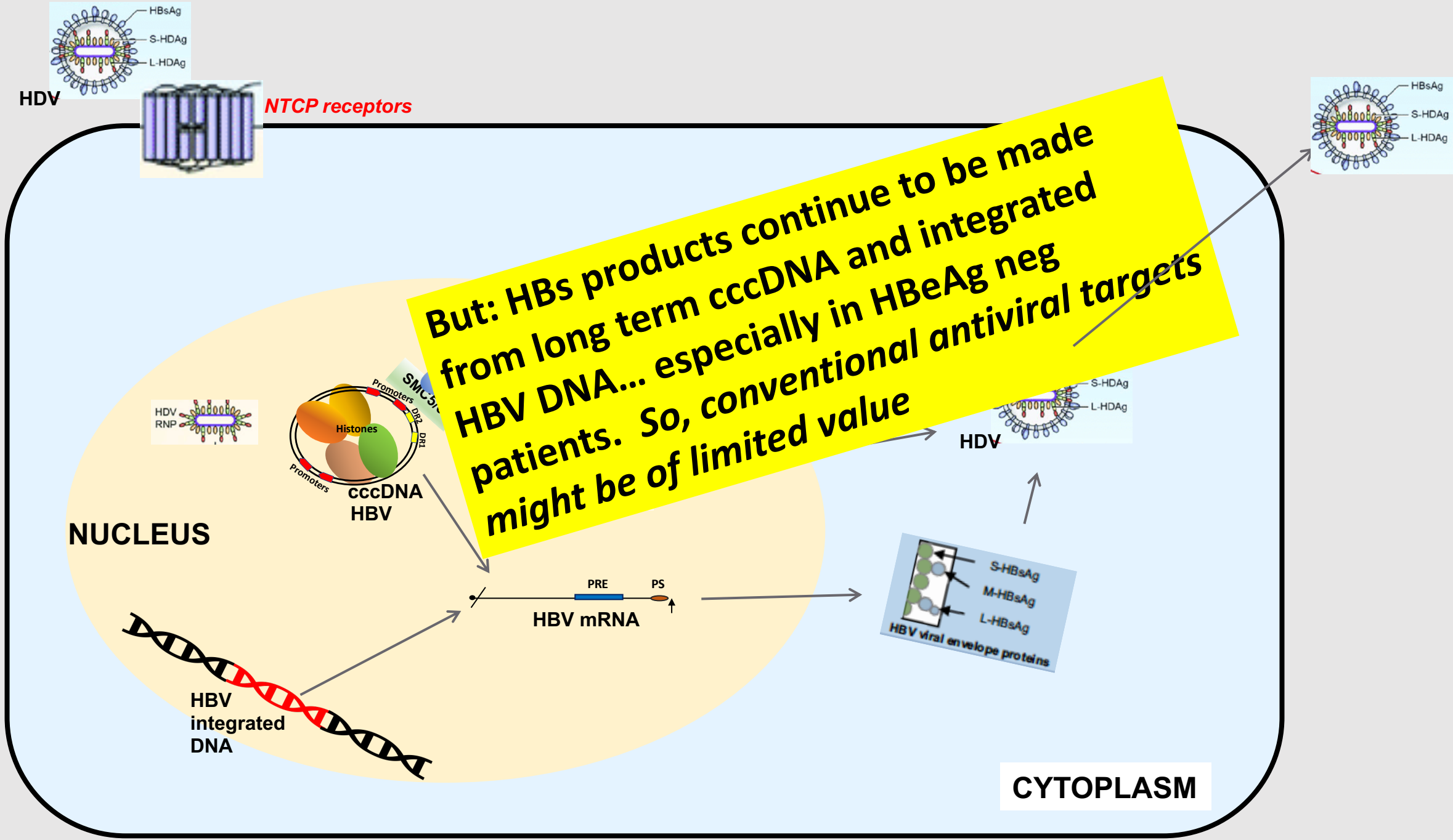



Asselah, T., Loureiro, D., Tout, I., Castelnau, C., Boyer, N., Marcellin, P. and Mansouri, A., 2020. Future treatments for hepatitis delta virus infection. *Liver International*, *40*, pp.54-60.


HBsAg
S-HDAg
HDV
HBV
NTCP receptors
So, any drug that affects the source of HBV envelope protein should be anti-HDV
HDV RNP
Histones
Promoters
cccDNA
HBV
HBsAg
S-HDAg
L-HDAg
HDV
NUCLEUS
PRE
PS
HBV mRNA
S-HBsAg
M-HBsAg
L-HBsAg
HBV viral envelope proteins
HBV integrated DNA
CYTOPLASM

# In principle: anything that interferes with HBV HBs production will be an HDV antiviral



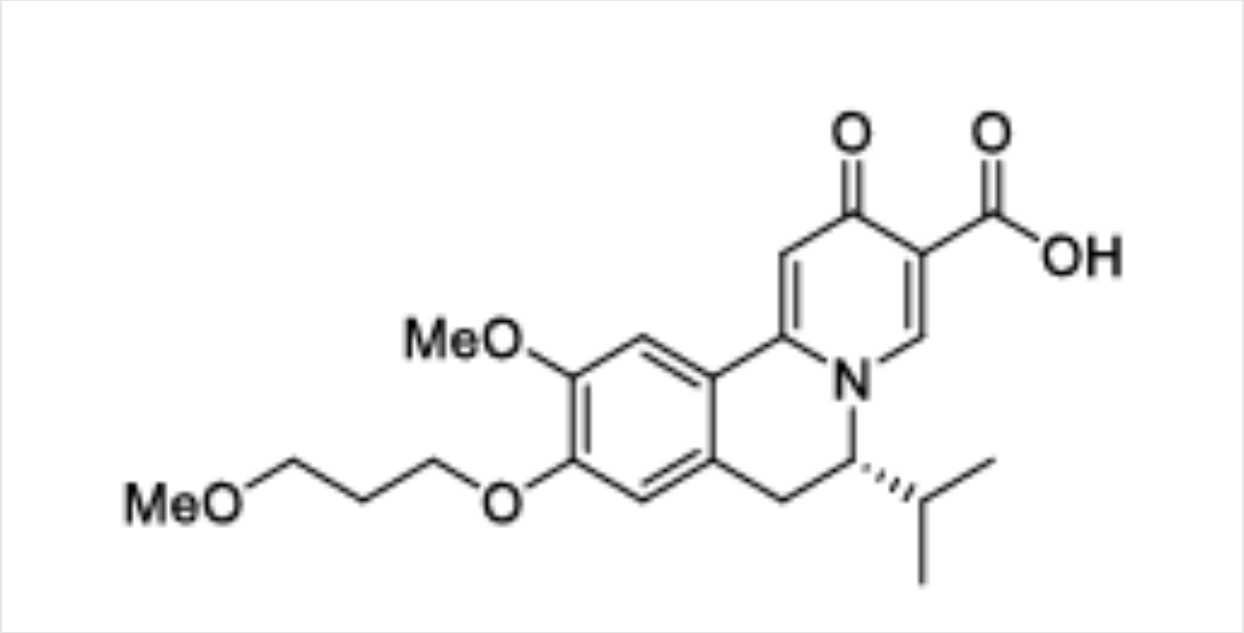

HBsAg
S-HDAg
L-HDAg
HDV
NTCP receptors
But: HBs products continue to be made from long term cccDNA and integrated HBV DNA… especially in HBeAg neg patients. So, conventional antiviral targets might be of limited value
HDV
RNP
Histones
Promoters
DR2
DR1
cccDNA
HBV
NUCLEUS
PRE
PS
HBV mRNA
S-HBsAg
M-HBsAg
L-HBsAg
HBV viral envelope proteins
HBV
integrated
DNA
CYTOPLASM

# Attention turning to drugs that

- Kill or eliminate HBV infected cells by immuno-stimulation or other means (i.e. Therapeutic vaccine combinations)
- Target HBs directly (i.e. NAPs, siRNA)

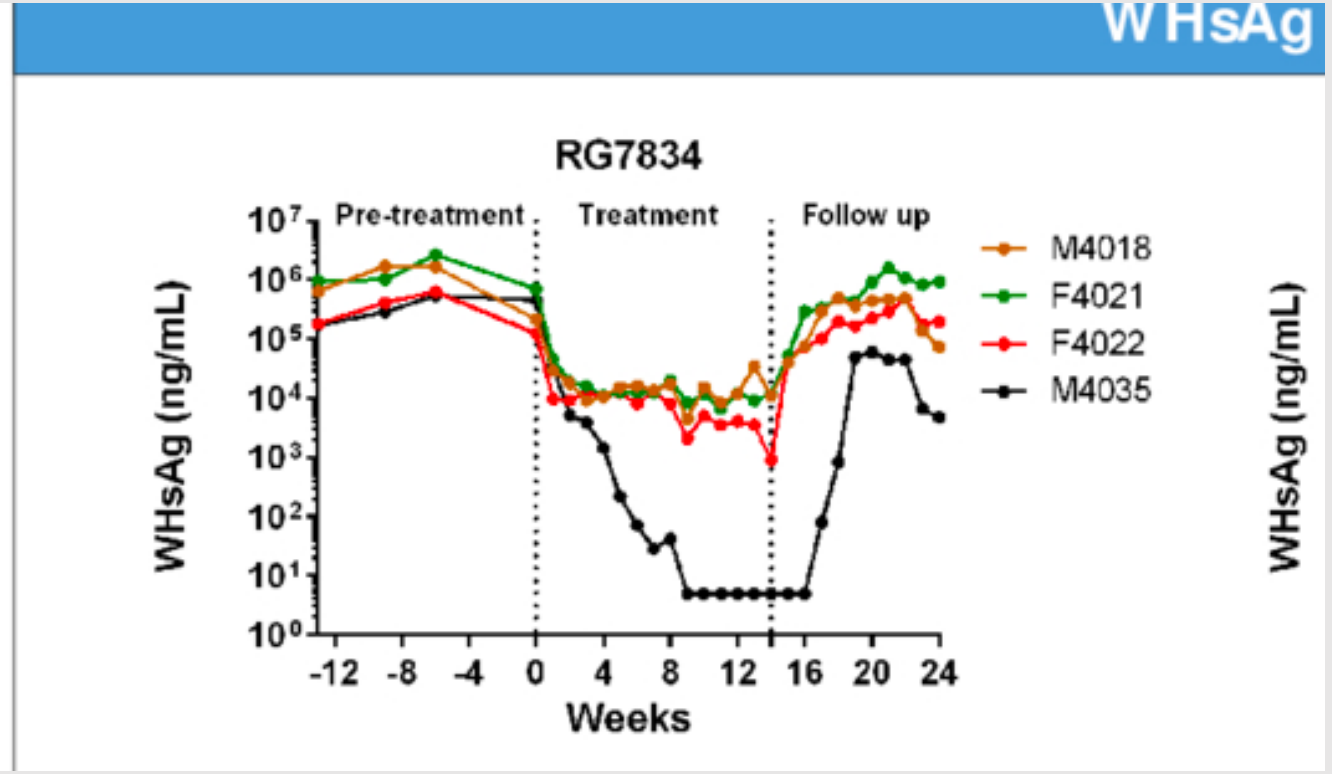
- But, there is another possibility…

# 2017: Mueller et al (Roche) report a small molecule that selectively reduced HBs




**Dihydroxyquinoline DHQ (RG7834)**

# In animals (mice, woodchucks) dramatic HBs reductions

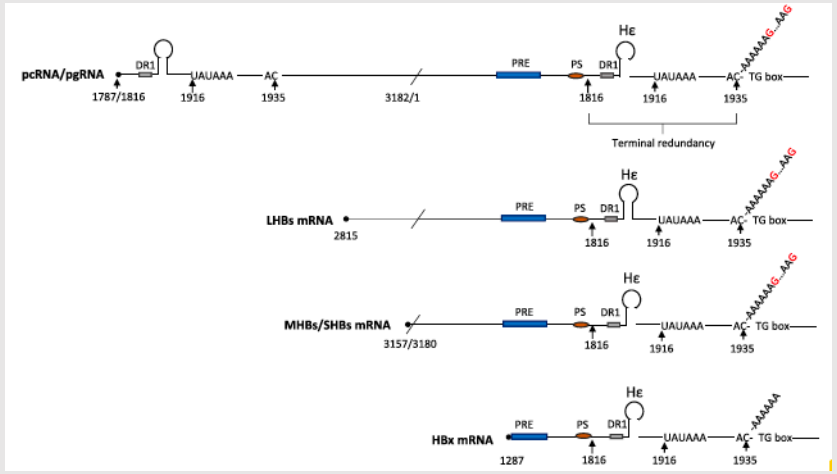


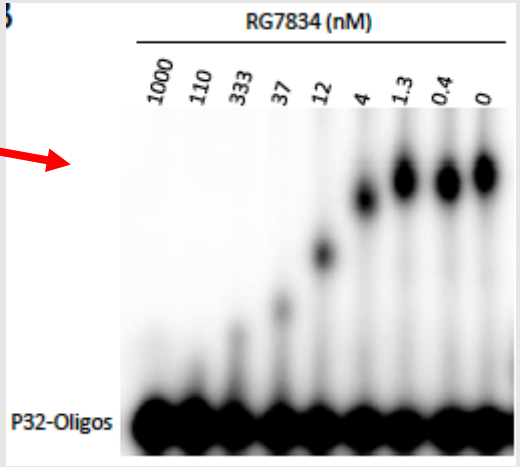

Mueller H, Wildum S, Luangsay S, Walther J, Lopez A, Tropberger P, Ottaviani G, Lu W, Parrott NJ, Zhang JD, Schmucki R. A novel orally available small molecule that inhibits hepatitis B virus expression. Journal of hepatology. 2018 Mar 1;68(3):412-20.

# DHQ works by:

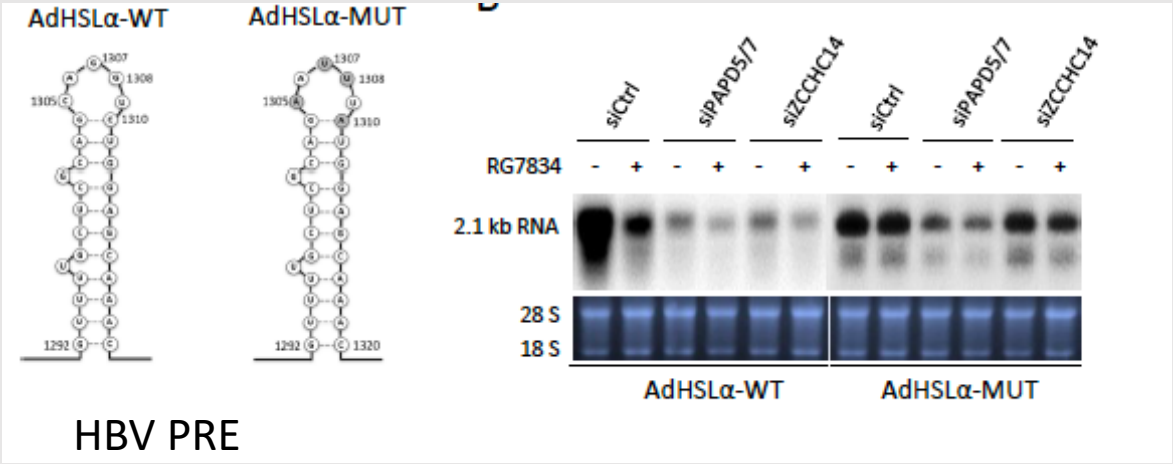
***(an RNA De-stabilizer")***

- **Roche: Targeting a host enzyme, PAPD5 / 7 (Mueller et al 2018)**
- **Blumberg: Works through "De-stabilizing" HBV mRNA "PRE" sequence (Sun et al, et al 2018, 20)**

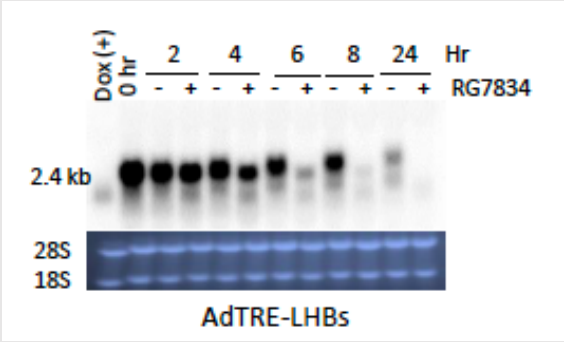



PAPD5 enzyme assay (Sun et al, 2020)




HBV PRE

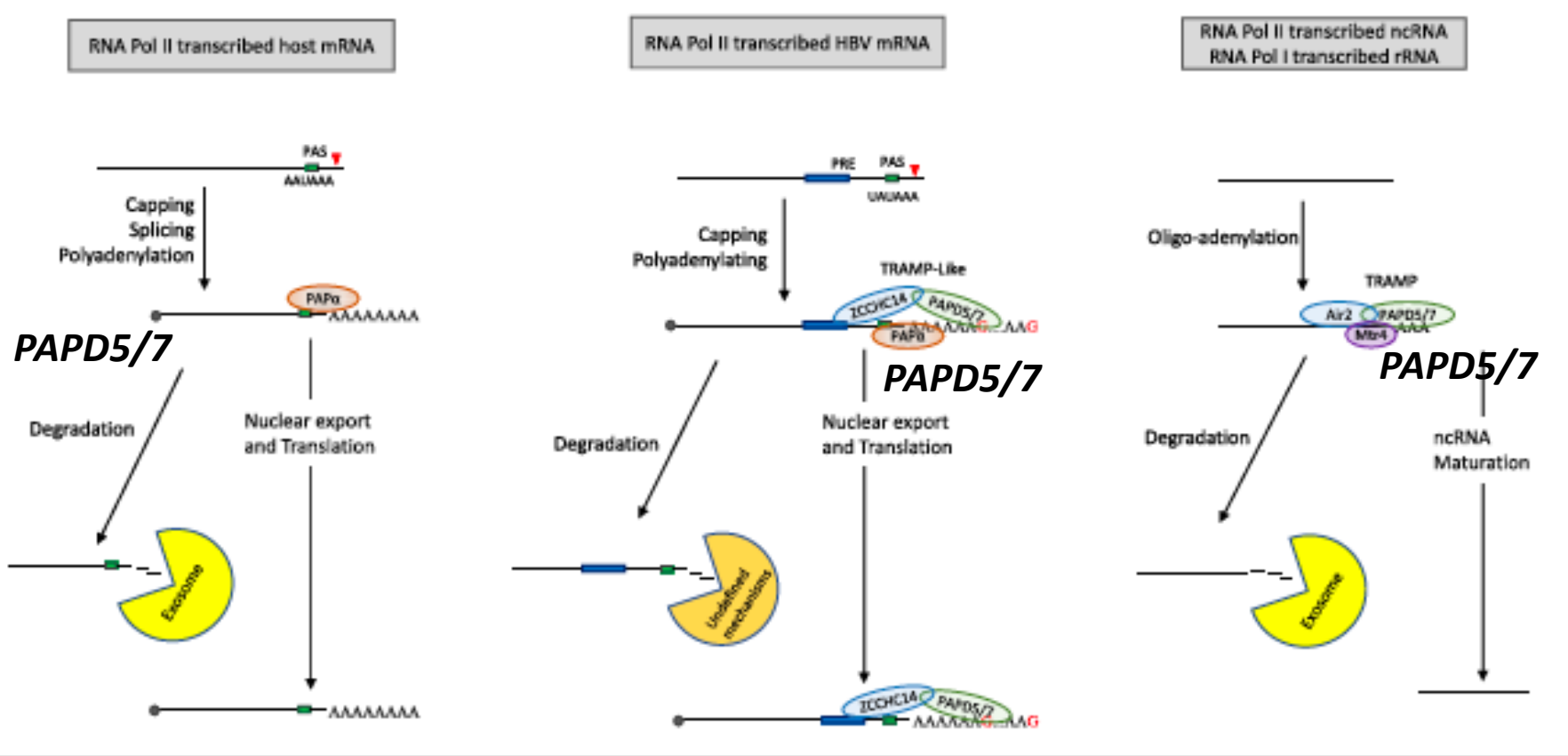


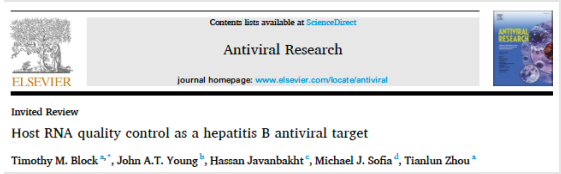

Northern blot: (Sun et al, 2018)

But: Almost all cell /defective host mRNA is degraded by the PAPD5/7 pathway:

*HBV does the opposite.*

So exciting: suggests HBV behaves more like a non coding RNA! All new strategy for antivirals

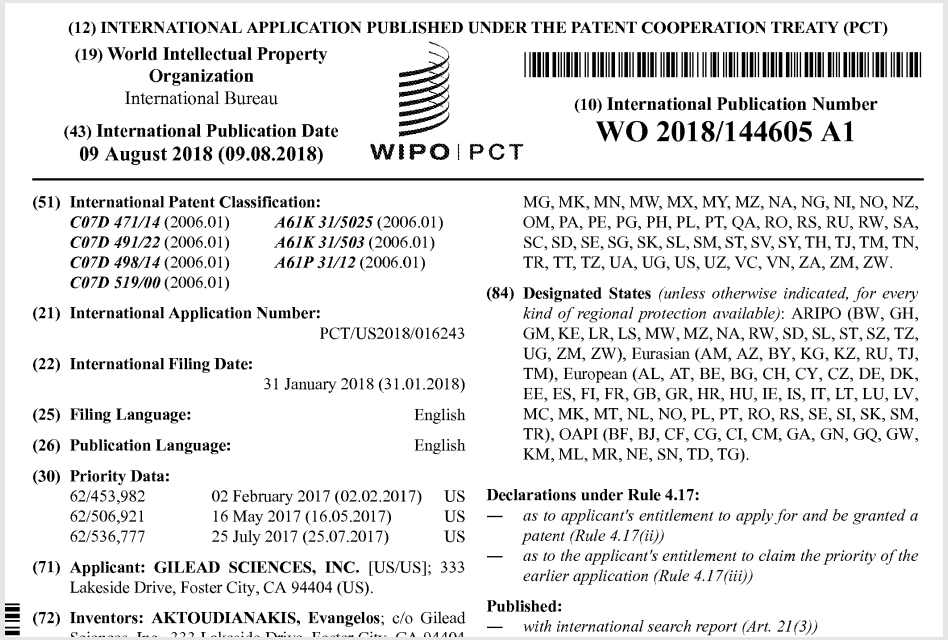




Contents lists available at ScienceDirect
Antiviral Research
journal homepage: www.elsevier.com/locate/antiviral
Invited Review
Host RNA quality control as a hepatitis B antiviral target
Timothy M. Block, John A.T. Young, Hassan Javanbakht, Michael J. Sofia, Tianlun Zhou

# DHQs/HBV RNA Destabilizers: Extremely promising, first in class, new approach

- But then, development abruptly stops…
- ?Neuro toxicity

# DHQ may have toxicity



(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(19) World Intellectual Property Organization
International Bureau

(43) International Publication Date
09 August 2018 (09.08.2018)

WIPO | PCT

(10) International Publication Number
WO 2018/144605 A1

(51) International Patent Classification:
*C07D 471/14* (2006.01) *A61K 31/5025* (2006.01)
*C07D 491/22* (2006.01) *A61K 31/503* (2006.01)
*C07D 498/14* (2006.01) *A61P 31/12* (2006.01)
*C07D 519/00* (2006.01)

(21) International Application Number:
PCT/US2018/016243

(22) International Filing Date:
31 January 2018 (31.01.2018)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:
62/453,982 02 February 2017 (02.02.2017) US
62/506,921 16 May 2017 (16.05.2017) US
62/536,777 25 July 2017 (25.07.2017) US

(71) Applicant: **GILEAD SCIENCES, INC.** [US/US]; 333 Lakeside Drive, Foster City, CA 94404 (US).

(72) Inventors: **AKTOUDIANAKIS, Evangelos**; c/o Gilead

MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PA, PE, PG, PH, PL, PT, QA, RO, RS, RU, RW, SA, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW.

(84) Designated States *(unless otherwise indicated, for every kind of regional protection available)*: ARIPO (BW, GH, GM, KE, LR, LS, MW, MZ, NA, RW, SD, SL, ST, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, RU, TJ, TM), European (AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, RS, SE, SI, SK, SM, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, KM, ML, MR, NE, SN, TD, TG).

Declarations under Rule 4.17:
— *as to applicant's entitlement to apply for and be granted a patent (Rule 4.17(ii))*
— *as to the applicant's entitlement to claim the priority of the earlier application (Rule 4.17(iii))*

Published:
— *with international search report (Art. 21(3))*

| Decreased Neurite Length SSMD Values | 0.001 µM | 0.01 µM | 0.1 µM | 1 µM | 10 µM | 100 µM |
|---|---|---|---|---|---|---|
| RG7834/ R07020322 24 hr | NS | NS | NS | **-3.4** | **-5.9** | **-4.5** |
| 4 hr | NS | NS | NS | NS | **-3.8** | **-4.0** |
| Vincristine 24 hr | NS | NS | NS | **-4.9** | **-5.4** | **-6.6** |
| 4 hr | NS | NS | NS | **-7.3** | **-5.7** | **-10** |

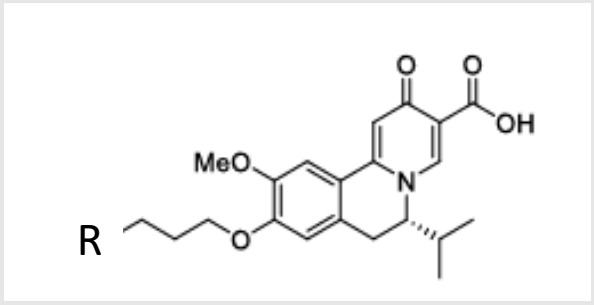
NS = no significant effect and > -3 SSMD Value. Values of ≤ -3 values are significant for decreased length

From: Aktoudiannakis, 2018, p.226

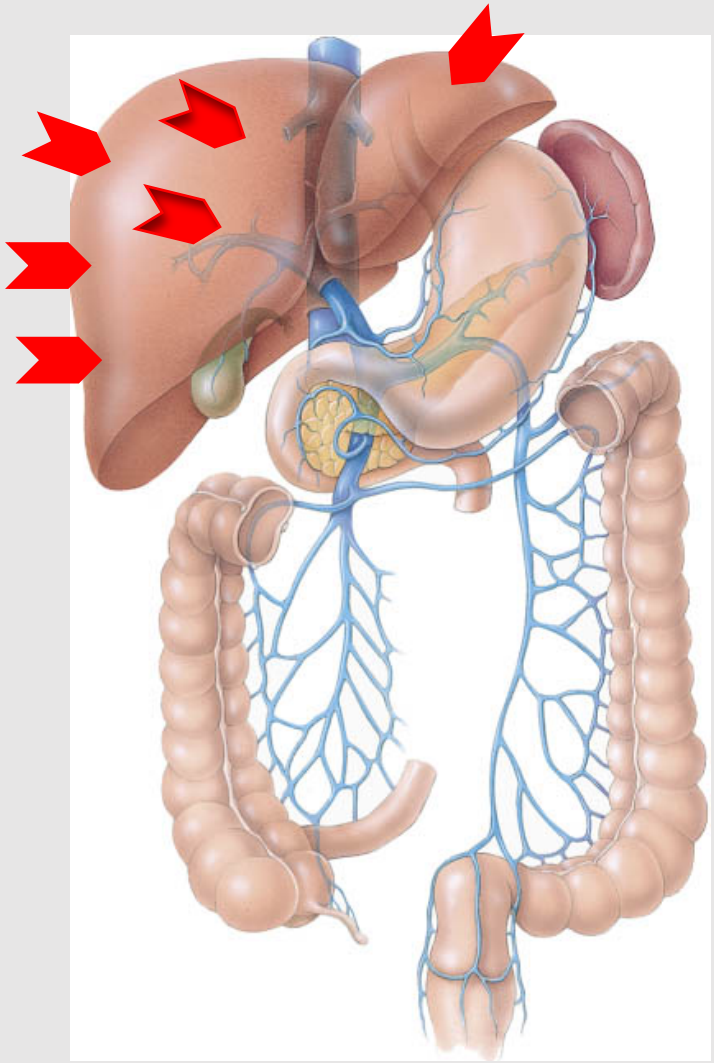
Aktoudianakis, E.; Canales, E.; Currie, K. S.; Kato, D.; Li, J.; Link, J. O.; Metobo, S. E.; Saito, R. D.; Schroeder, S. D.; Shapiro, N.; Tse, W. C.; Wu, Q.; Hu, Y. E. Patent: Compounds for the treatment of hepatitis B… WO2018144605 A1

# Avoid neuro, off liver toxicity
# Produce a liver targeted DHQ (DHQ-E)

OAT B receptor

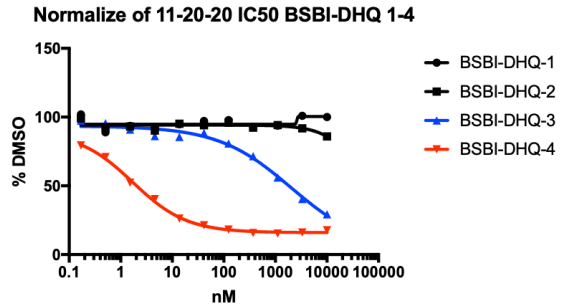



DHQ-E

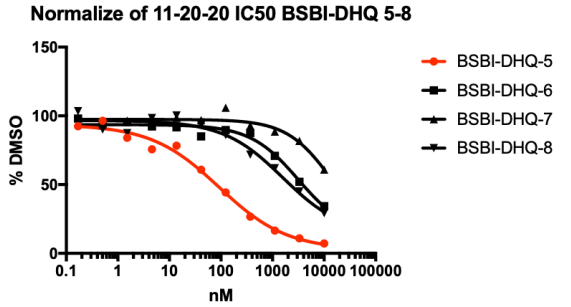


Blumberg: Lab of Yanming Du

# DHQ-E retains excellent nano molar anti-PAPD 5 activity




| | |
|---|---|
| 65-10-A | |
| 65-05-B-OH | |
| 65-10-B | 2008 +/- 1589 |
| 65025-A-OH | 1.713 +/- 0.4534 |




| | |
|---|---|
| 65-25-B-OH | 92.01 +/- 32.63 |
| 65-25-C-OH | |
| 65-42A-OH | |
| 65-42C-OH | 1587 +/- 1048 |

# DHQ-E uses the OAT P1 receptor

## OATP1B1 and 1B3 Transporter Substrate

Table 1. Substrate of OATP1B1 in OATP1B1 -HEK293 Cells

| Compound Number | Compound ID | Test Concentration (μM) | Uptake Ratio (-/+Inhibitor) | Uptake Ratio (Transporter (-Inhibitor) /Mock(-Inhibitor)) | Comment |
|---|---|---|---|---|---|
| Inhibitor | Rifamycin SV | 100 | - | - | - |
| Positive Control | Estradiol 17-β Glucuronide | 10 | 81.84 | 48.70 | - |
| 1 | BSBI-65042-E-OH | 1 | 11.17 | 18.99 | A potential in vitro substrate for OATP1B1 |
| 2 | DHQ-1 | 1 | 0.38 | 0.71 | Not an in vitro substrate for OATP1B1 |

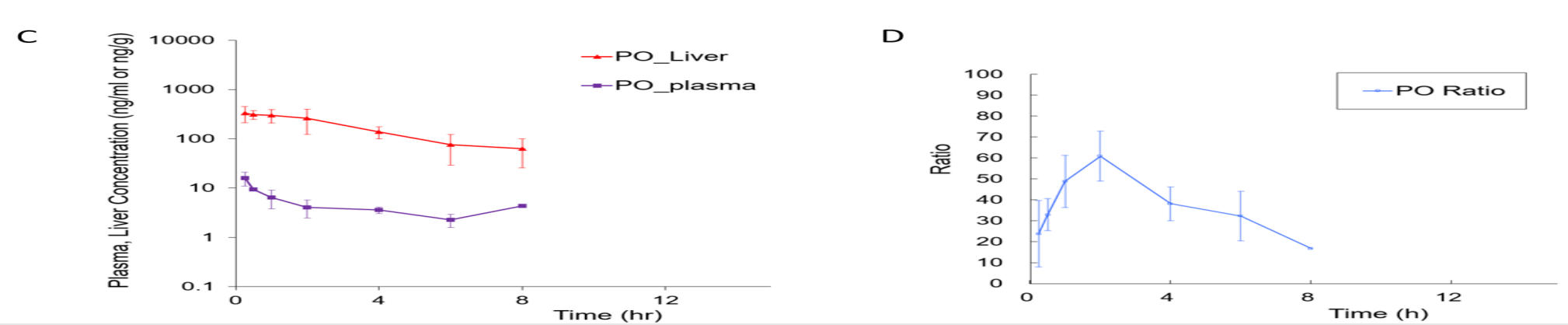
Table 1. Substrate of OATP1B3 in OATP1B3 -HEK293 Cells

| Compound Number | Compound ID | Test Concentration (μM) | Uptake Ratio (-/+Inhibitor) | Uptake Ratio (Transporter (-Inhibitor) /Mock(-Inhibitor)) | Comment |
|---|---|---|---|---|---|
| Inhibitor | Rifamycin SV | 100 | - | - | - |
| Positive Control | Estradiol 17-β Glucuronide | 10 | 86.15 | 98.86 | - |
| 1 | BSBI-65042-E-OH | 1 | 11.57 | 24.80 | A potential in vitro substrate for OATP1B3 |
| 2 | DHQ-1 | 1 | 0.42 | 0.56 | Not an in vitro substrate for OATP1B3 |

A compound is considered as a potential substrate of particular transporter when both the **Uptake ratio 1 and Uptake ratio 2 are greater than 2**

Hepatoselective Dihydroquinolizinone Bis-acids for HBsAg mRNA Degradation **(2021)**
N. Hwang[=], L. Sun[=], D. Noe, P. Lam, T Zhou, T Block*, Yanming Du*

# DHQ-E accumulates in the liver (*in vivo*), doesn't cross the blood brain barrier *(in vitro)*




Hepatoselective Dihydroquinolizinone Bis-acids for HBsAg mRNA Degradation **(2021)**
N. Hwang[=], L. Sun[=], D. Noe, P. Lam, T Zhou, T Block*, Yanming Du*

# Summary

- Any drug that interferes with HBV envelope (S,L) production or function should have anti-HDV activity

- Drugs that Kill or eliminate HBV infected cells by:

❖ *immuno-stimulation or other means (Therapeutic vaccine combinations)*

❖*Target HBs directly (NAPs) or HBs mRNA (siRNA, antisense)*

Are getting new attention

- A new strategy that targets HBV mRNA for degradation (HBV PRE, host quality control)
  - Liver targeting of this drug is hoped to reduce toxicity and make it viable