About FivePlus
Beijing FivePlus Molecular Medicine Institute was established in 2005. The company has been dedicating itself to continuous innovation of viral vectors. The meaning of FivePlus is based on the five point scoring system and even better than five point, which perfectly interpret the culture of the company “the pursuit of excellence" and the mission of the company "committed to make a outstanding contribution in the development of viral vector and gene therapy".

A Novel Recombinant Virus Reagent Products for Efficient Preparation Of Hepatitis B Animal Models

About rAAV8-1.3HBV
2010 XiaoYan Dong, et al. \cite{1} reported for the first time that rAAV8-1.3HBV can be used for high performance of preparation of persistent HBV infected mouse model. rAAV8-1.3HBV, launched in 2010, is one of the self-developed product of Beijing fiveplus Molecular Medicine Research Institute Co., Ltd.. It is a kind of recombinant AAV8 virus carrying 1.3 copies of HBV genome, intravenous injection of which through to the tail of mice could make the mice persistently infected by HBV.

Application
Stable and efficient animal model plays a very important role in the development and evaluation of HBV pathogenic mechanism and antiviral medicines. At present, this model preparation has been widely accepted and used in the research of HBV candidate medicine evaluation and screening.

Principle
The product cleverly utilizes and combines the characteristics of that the genome containing the repeat area of the HBV genome could be duplicated in the liver of mice, as well as the tropism of hepatic cell of the AAV8 virus, which can well simulate the persistent state of HBV infection in mice.

Electron Microscopy of rAAV8-1.3HBV
Different Animal Model Preparations
Due to the species specificity HBV, its host range is very narrow. Which limited the preparation of HBV infected animal model. Chimpanzee is still the most effective animal model of HBV infection, but its endangered animal with limited resources and limited animal ethics. Chimpanzee model for HBV infection is also not efficient to prepare. As for small animal model, tree shrews can be naturally infected with HBV, but when mentioning the infection rate, infection duration, as well as stability and repeatability of test results, tree shrews model is not ideal. Therefore, the genetic background of the mouse is still make it a best uniform object of HBV study, however mouse could not naturally infected with HBV. After years of research, it develops many different method of preparation of the HBV mice model, such as transgenic mice, human mouse chimeric liver, rAAV8-1.3HBV mouse model and etc. Table 1 which shows the comparison of different HBV mice model.

<table>
<thead>
<tr>
<th>Mice Model</th>
<th>HBV Infection</th>
<th>HBV DNA Duplication</th>
<th>cccDNA</th>
<th>Advantage</th>
<th>Disadvantage</th>
<th>Generate Antibody</th>
<th>Price</th>
</tr>
</thead>
<tbody>
<tr>
<td>Transgenic Mice</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td>Stable Expression</td>
<td>Non infection model; HBV gene expression level is not uniform, individual expression level difference is huge; HBV genome integration on the cell chromosome, cannot be removed; easy to cause the mutation of the host gene</td>
<td>No</td>
<td>High</td>
</tr>
<tr>
<td>Hydrodynamic Injected Mice</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td>The model can be used to simulate the acute and persistent infection status</td>
<td>No repeated infection; Easy to cause injury; The success rate of the model is not high</td>
<td>No</td>
<td>Low</td>
</tr>
<tr>
<td>Human Mouse Chimeric Liver</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>HBV infection in human hepatocytes; replication; can be used to block the infection or inhibit cccDNA production.</td>
<td>difficult to prepare, cannot study the interaction between virus and host</td>
<td>Yes</td>
<td>High</td>
</tr>
<tr>
<td>Human Derived Mouse with Human Liver and Immune System</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>HBV infection in human liver cells, replication; can be used to study the immune response of the human body after HBV infection; can be used for the evaluation of immune therapy</td>
<td>Very difficult to establish</td>
<td>Yes</td>
<td>Very High</td>
</tr>
<tr>
<td>rAAV8-1.3HBV Intravenous Injected Mice</td>
<td>No</td>
<td>Yes</td>
<td>unknown</td>
<td>Good consistency; effective to simulate the persistent infection and immune tolerance state of HBV infection. It is the best model of HBV candidate medicine screen.</td>
<td>Not found yet</td>
<td>Yes</td>
<td>Low</td>
</tr>
</tbody>
</table>

Value
rAAV8-1.3HBV injection in mice and other animal to prepare animal model is of simple and high efficient and effective preparation, uniform stability, good dose-response relationship, as well as wide range of application, and has been widely used in evaluation and screening of hepatitis B therapy and candidate medicine development. It has brought a huge boost in terms of time, cost and effect in the preparatory work and pre clinical animal model stage of the of hepatitis B therapy and candidate medicine development. The application of this novel product is to accelerate the pace of research and development of hepatitis B therapy and candidate medicine. After years of experiment and research, it has been widely recognized by the global research institutions such as Shanghai Fudan University, Shanghai & France Pasteur Institute and etc., and gradually accepted by the global advanced pharmaceutical companies during their hepatitis B therapy and candidate medicine development.
## Feature and Advantages

<table>
<thead>
<tr>
<th>High Molding Rate</th>
<th>Dose per mouse as low as 4E+9 v.g. of rAAV8-1.3HBV can generate the high quality mouse model. (two strains option of C57BL/6 and BALB/c).</th>
</tr>
</thead>
<tbody>
<tr>
<td>Uniformity &amp; Stability</td>
<td>When rAAV8-1.3HBV was injected with the same dose, the HBsAg, HBeAg and HBV DNA were detected in the serum of mice, with similar index level. The model was uniform and stable.</td>
</tr>
<tr>
<td>Good Dose Effect Relationship</td>
<td>The level of HBsAg, HBeAg and HBV DNA in serum is positively related to the rAAV8-1.3HBV injection dose (1E+9 ~ 12 v.g. per mouse), which enables the level of HBsAg, HBeAg and HBV DNA in serum can be controlled during the preparation of HBV animal model in accordance to the requirements of the therapy and candidate medicine evaluation experiment;</td>
</tr>
<tr>
<td>Wide range of application</td>
<td>Can be applied on different animal including non-human primates</td>
</tr>
</tbody>
</table>

### Important Warning

- By the beginning of the experiment, please browse our published paper [1].
- The rAAV8-1.3HBV viral products and related animal model products has the infectivity and pathogenicity to humans and a variety of mammals, therefore it is suggested to be classified into Biosafety Level 2 (BSL-2), and it should be strictly managed and controlled during the process of production, sale, transportation, use and other aspects. The related animal model is suggested to classified into animal laboratory Biosafety Level 2 (ABSL-2), and treated equivalently as HBV transgenic mice.
- The excretion of rAAV8-1.3HBV infected mice carry HBV virus, which may have infectivity and pathogenicity to primate.

**Reference [1] & [3]**

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![Fig 1. The Levels Of HBV DNA In Mice Serum At 32 Week After rAAV8-1.3HBV Injection At Different Doses](image1)

![Fig 2. The Levels Of HBcAg Expression (100x) at 32 Week After rAAV8-1.3HBV Injection At Different Doses](image2)

*The Black arrow indicates HBcAg positive cell. The Hollow arrow indicates inflammatory cell.

Note: the candidate medicine research can start after 1~2 week after injection of rAAV8-1.3HBV
animals, including human, and to tree shrews, ducks and other animals. Hence, protective measures is needed during transport and operation.

- The institutions utilizing and operating rAAV8-1.3HBV product are required to have a Laboratory of Biosafety Level 2 (BSL-2). The operator should be vaccinated with hepatitis B vaccine, and with hepatitis B immune ability. The operator should be trained by professional biological safety and obtain biological safety training certificate, and operate in strict accordance with the biosafety regulations and manuals.

- The packaging, transport, feeding, experimental operation of the mice model products infected by rAAV8-1.3HBV (adr/ayw) need special conditions and requirements, such as animal laboratory with Biosafety Level 2 (ABSL-2). The operator should be vaccinated with hepatitis B vaccine, and with hepatitis B immune ability. The operator should be trained by professional biological safety and obtain biological safety training certificate, and operate in strict accordance with the biosafety regulations and manuals, in order to avoid risk of contamination or infection to the operator, the environment and other unrelated subjects.

- The institutions utilizing and operating rAAV8-1.3HBV product and rAAV8-1.3HBV infected animal model should have the qualification of biosafety.

**Product Type**

The serum type of HBV: type ayw (GenBank accession number: KX470733)

type adr (GenBank accession number: KX449554).

**Product Specifications And Prices**

<table>
<thead>
<tr>
<th>Item Code</th>
<th>Item Name</th>
<th>Package</th>
<th>Price (¥ RMB)</th>
<th>Item Code</th>
<th>Item Name</th>
<th>Package</th>
<th>Price (¥ RMB)</th>
</tr>
</thead>
<tbody>
<tr>
<td>AMV-001</td>
<td>rAAV8-1.3HBV adr</td>
<td>1E+12v.g.</td>
<td>12000.00</td>
<td>AMV-002</td>
<td>rAAV8-1.3HBV ayw</td>
<td>1E+12v.g.</td>
<td>12000.00</td>
</tr>
<tr>
<td>AMV-001-1</td>
<td>rAAV8-1.3HBV adr</td>
<td>1E+11v.g.</td>
<td>2000.00</td>
<td>AMV-002-1</td>
<td>rAAV8-1.3HBV ayw</td>
<td>1E+11v.g.</td>
<td>2000.00</td>
</tr>
<tr>
<td>AMV-001-2</td>
<td>rAAV8-1.3HBV adr</td>
<td>5E+11v.g.</td>
<td>9000.00</td>
<td>AMV-002-2</td>
<td>rAAV8-1.3HBV ayw</td>
<td>5E+11v.g.</td>
<td>9000.00</td>
</tr>
<tr>
<td>AMV-001-3</td>
<td>rAAV8-1.3HBV adr</td>
<td>5E+12v.g.</td>
<td>53000.00</td>
<td>AMV-002-3</td>
<td>rAAV8-1.3HBV ayw</td>
<td>5E+12v.g.</td>
<td>53000.00</td>
</tr>
<tr>
<td>AMV-001-4</td>
<td>rAAV8-1.3HBV adr</td>
<td>1E+13v.g.</td>
<td>102000.00</td>
<td>AMV-002-4</td>
<td>rAAV8-1.3HBV ayw</td>
<td>1E+13v.g.</td>
<td>102000.00</td>
</tr>
</tbody>
</table>

**Lead Time**

If not in a large order quantity, delivery on the second working day of the confirmation of orders, and the arrival time of the goods is according to the courier lead time; if a large order quantity, please contact sales manager in advance for more specific detail.

**Product Application Example and related literature reference**


[2]. Guo Y et al, Immunosuppressant dexamethasone can significantly extend the expression of hepatitis B virus antigens in the HBV mouse model by hydrodynamic transfection method. 2010,26(01): 20-26


[6]. Zeng Y et al, Establishment of a tree shrew model of acute hepatitis B virus infection by transduction with a
recombinant adeno-associated virus 8 carrying 1.3 copies of HBV genome. acta laboratorium animalis scientia sinica, 2013, 21(03): 36-41.


