Gene Therapy from AA to V: Developing Gene Therapy for CNS Disorders

Voyager Therapeutics, Cambridge, MA

Gene therapy: Medicine gaining momentum

- Gene therapy is a rapidly growing area of drug development research with FDA approved drugs now available.[1]
- Gene therapy works by providing therapeutic genetic material to cells that may treat disease. [1,2]
- A broad range of treatments may be designed depending on the genes used.[1]
- For central nervous system diseases, long-term benefits may be possible. [1]

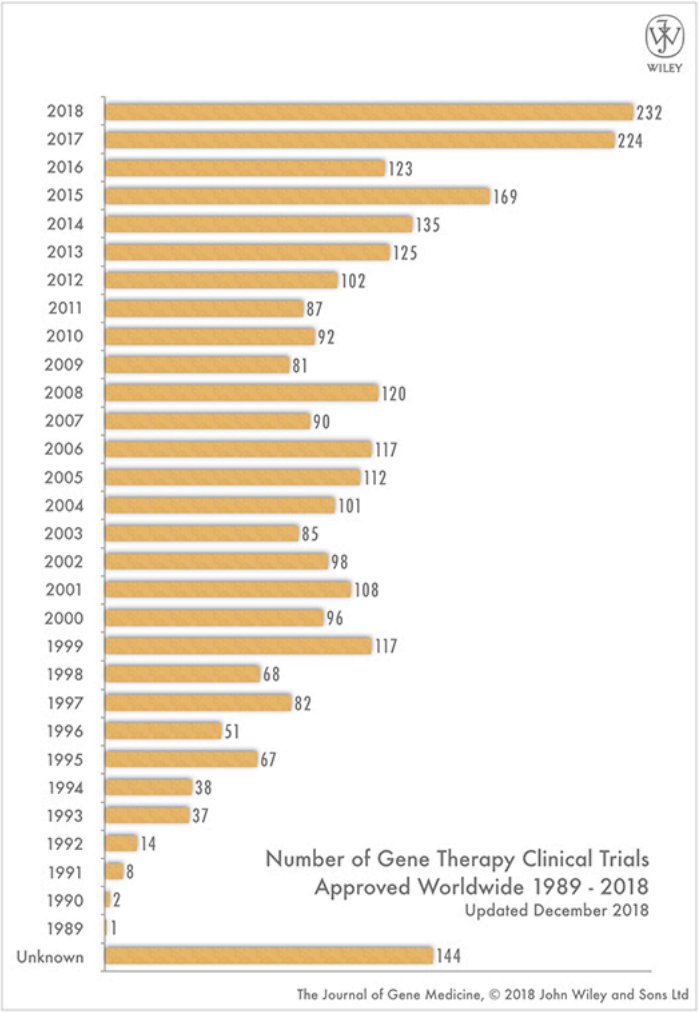
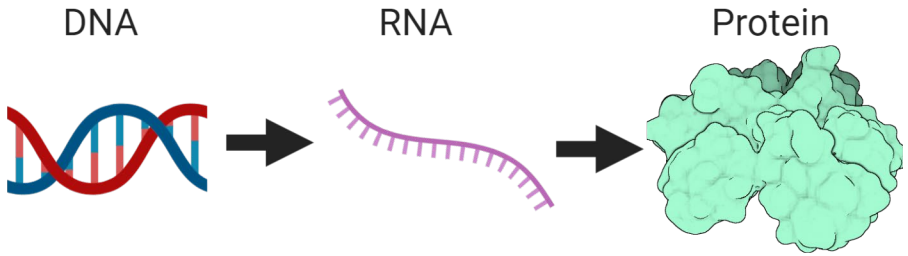


Image courtesy *J. of Gene* Medicine. http://abedia.com/wiley/years.php Accessed 27 May, 2020. 1. Anguela XM. Et al. *Annu. Rev. Med.* 2019. 2. United States Food and Drug Administration. *Human Gene Therapy for Rare Diseases: Draft Guidance for Industry*. https://www.fda.gov/media/113807/download. Accessed 27 May, 2020.

How genes work: DNA to protein

- DNA in cells contains genes, the blueprints for making proteins that the cell uses to function.[1]
- DNA is transcribed into RNA, a template that the cell translates into a protein.[1]



- Gene therapy can provide cells with additional DNA blueprints, which allows them to make new RNA.[1,2]
  - Some of these new RNAs make protein
  - Some of these new RNAs stop proteins from being made

1. National Institutes of Health. Help Me Understand Genetics. Available at: https://ghr.nlm.nih.gov/primer/therapy/procedures. Accessed 27 May, 2020. 2. Anguela XM. Et al. *Annu. Rev. Med.* 2019.

Think of gene therapy as a message that is coded into DNA and uses the cell's machinery to produce the drug

Message

Dear John,

We noticed an error. This is the tool you need to fix it.

Management

Envelope

Delivery

John Patient
23 CNS St
Human Body, 01234

Received

DNA sequence containing gene

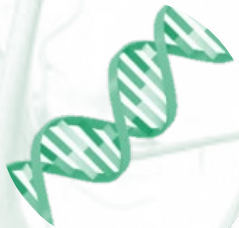
Gene packaged into vector

Targeted delivery of vector (eg, brain)

Cell uses gene to produce treatment

Think of gene therapy as a message that is coded into DNA and uses the cell's machinery to produce the drug

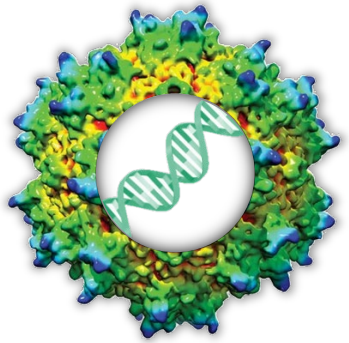
Message



DNA sequence containing gene

DNA acts like a blueprint the cell can use to make RNA and protein.

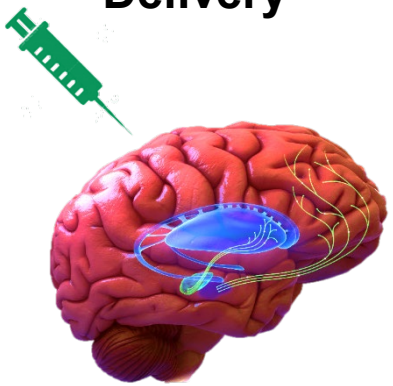
Envelope



Gene packaged into vector

A vector is a combination of the DNA being delivered and the capsid envelope that helps it enter cells. ***AAV is one type of capsid.***

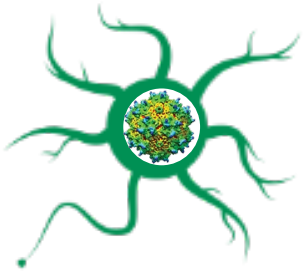
Delivery



Targeted delivery of vector (eg, brain)

The vector must be delivered so it can reach the target cells.

Received

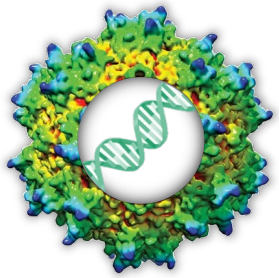


Cell uses gene to produce treatment

Cells that receive the gene can use the DNA blueprint to make the treatment.

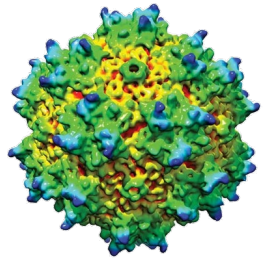
AAV gene therapy: Multiple factors shape development

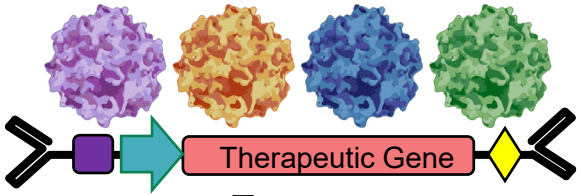
Gene & Cell targets



Capsid Selection and Gene Design

Capsid: The protein shell of AAV



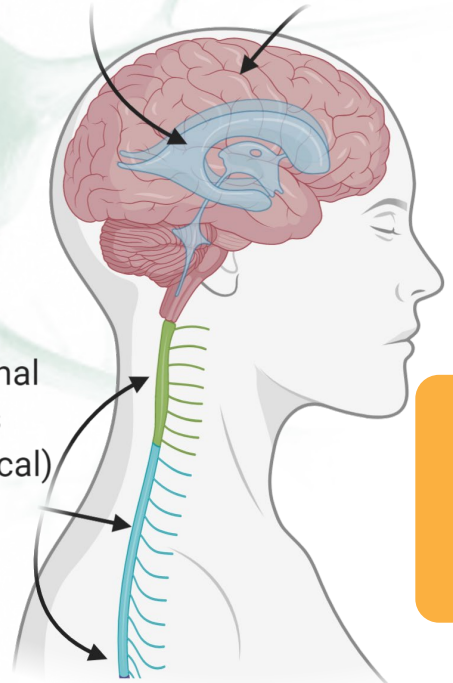


Route of Administration

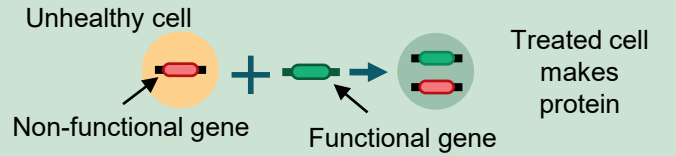
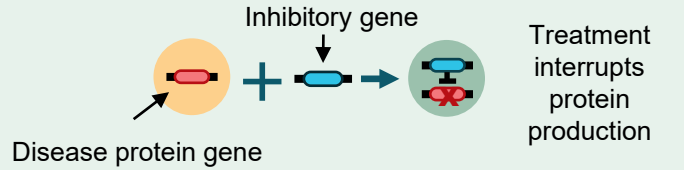
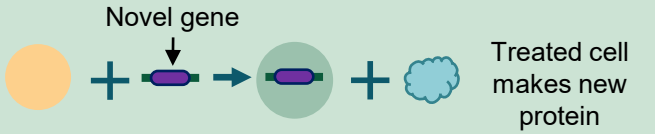
Into brain fluids (intracerebroventricular)

Direct to tissue (intraparenchymal)

Into spinal fluids (intrathecal)

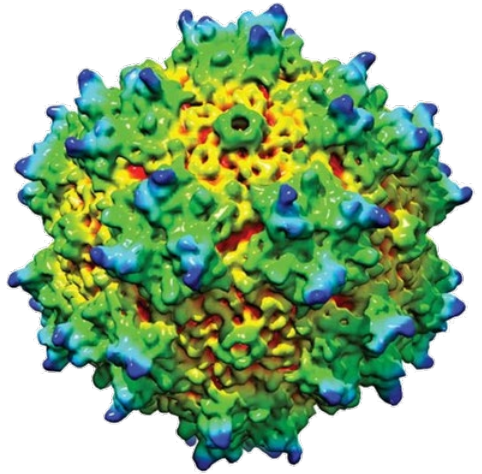


Medicinal genes can work in multiple ways

Treatment	Mechanism	Potential Result	Schematic
Gene Replacement	Adds a gene to cells where the ability to make a protein was lost	Missing protein produced	
Gene Reduction	Adds a gene that lowers the amount of protein made	Disease-related proteins reduced	
Gene Supplementation	Adds a gene that lets a cell make a new protein	New therapeutic proteins produced	

Adeno-associated viruses (AAVs) are promising packages to deliver gene therapy[1]

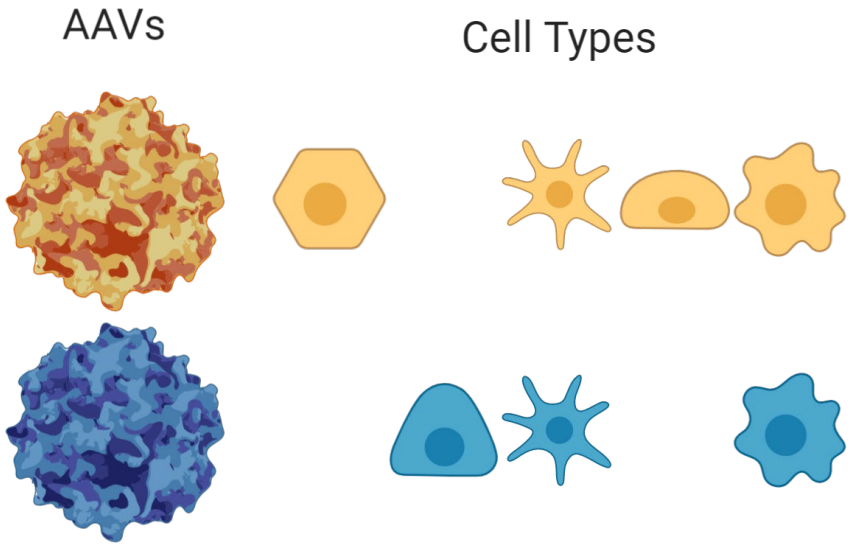
- They are not known to cause illness (non-infectious).
- AAVs do not multiply (non-replicating).
- They do not merge with or change the DNA of cells (non-integrating).
- In cells that don’t divide, like neurons, they may work for a long time.
- The immune system tends to have a mild response.
- Many types occur naturally with different properties.



1. Lee E. et al..Curr. Op. in Biomed. Eng. 2018.

Some differences in AAV serotypes

- The types of AAV are called serotypes.
- Serotypes differ from one another in several important ways.[1]
  - Different serotypes are able to enter different types of cells (tropism).



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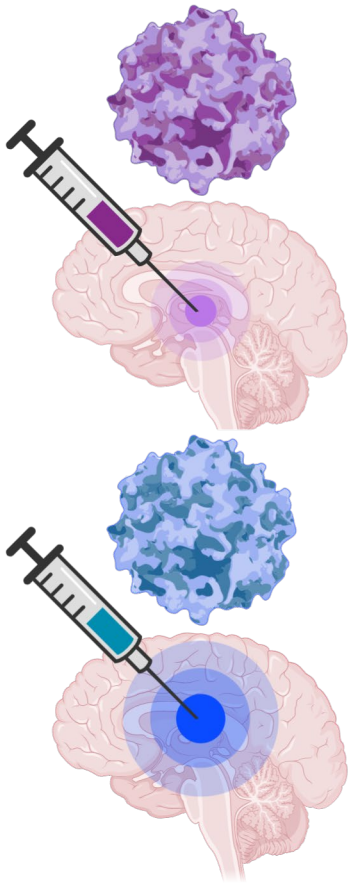

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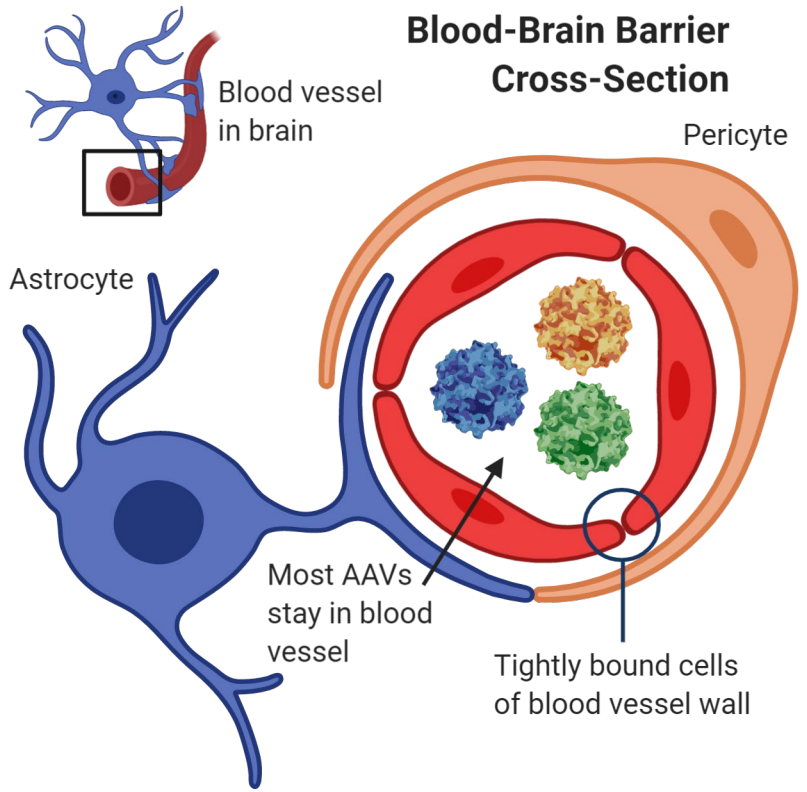

Tightly bound blood vessel walls along with supporting pericytes and astrocytes make up the blood-brain barrier

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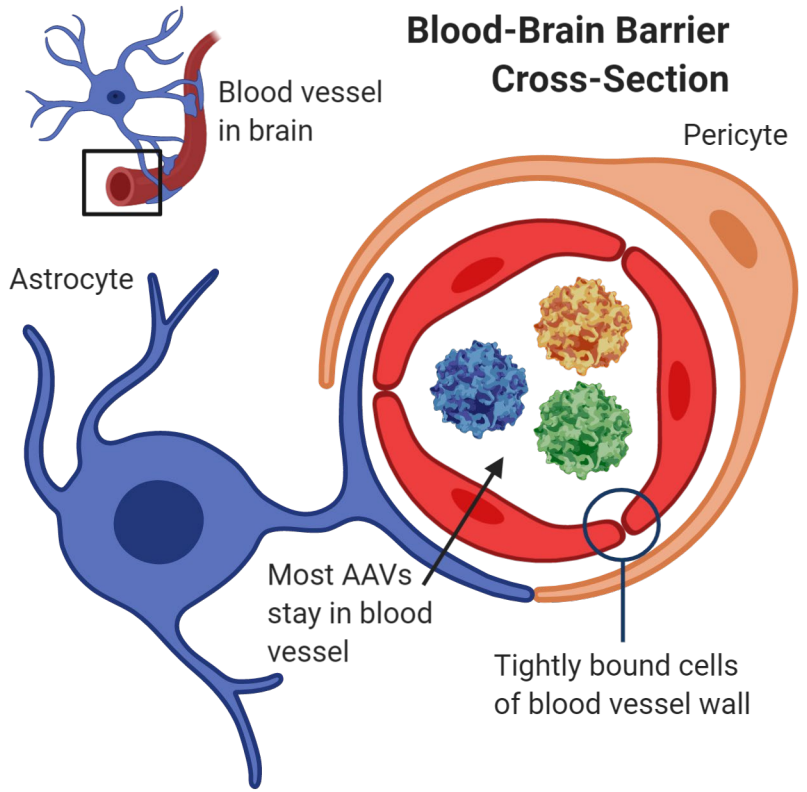
- New serotypes can be engineered if needed.[3]



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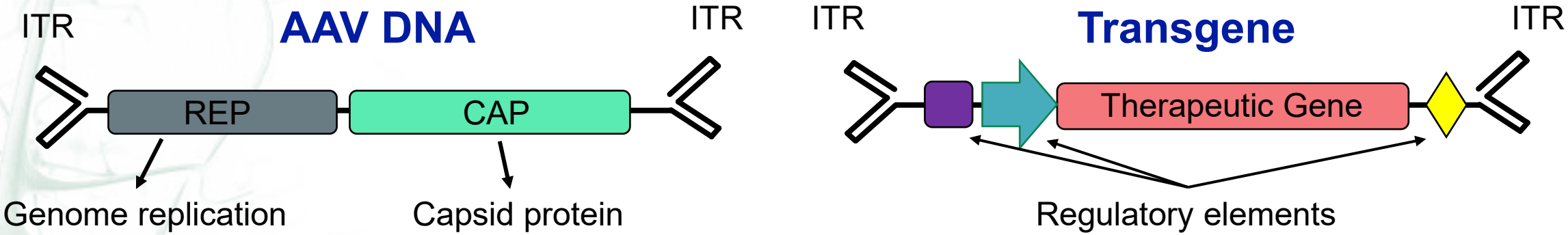
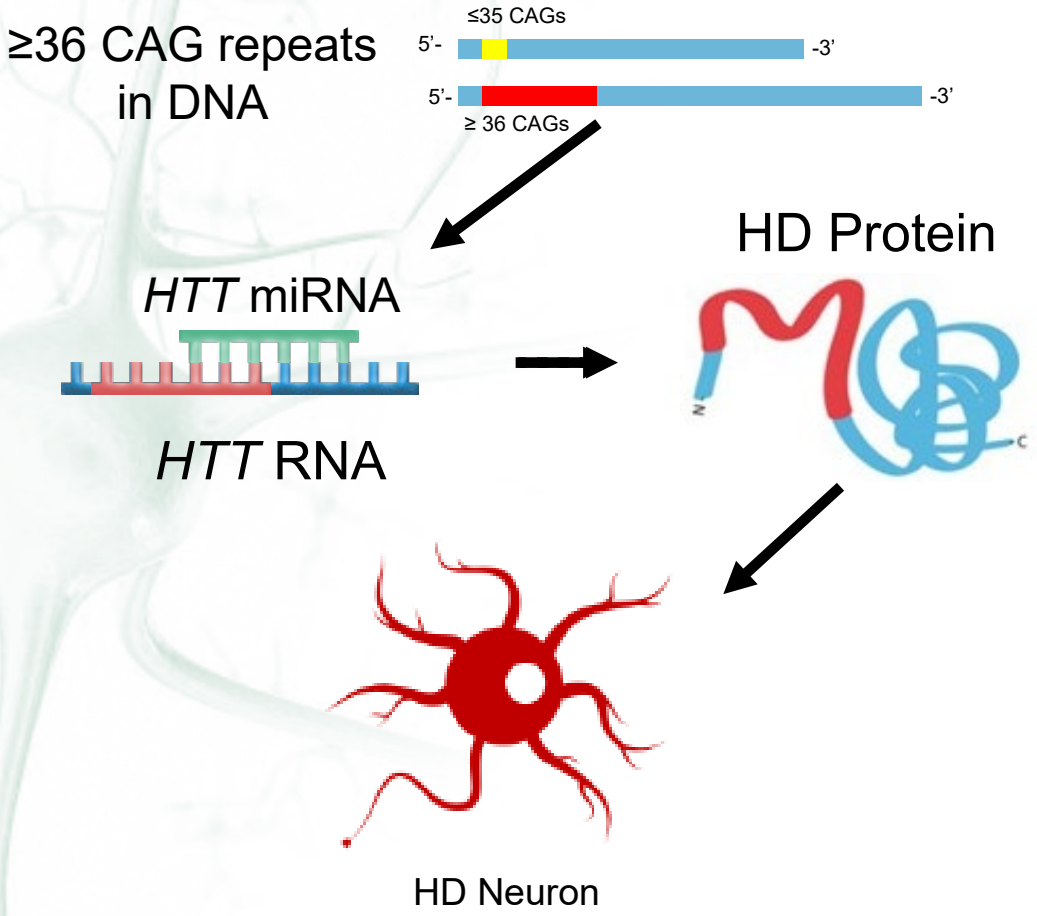
Designing the therapeutic transgene to put in the AAV package



- AAV contains genes for replication (REP) and the capsid (CAP), along with inverted terminal repeats (ITRs) at both ends.[1]
- The ITRs help to assemble the AAV vector and keep the therapeutic gene stable inside cells.[1]
- REP and CAP genes are replaced with the therapeutic transgene (the therapeutic gene and regulatory elements).
  - Regulatory elements help control which cells will use the gene and how much of the treatment those cells will make.[1]

1. Wang D. et al. Nat. Rev. Drug Disc. 2019

An RNA interference transgene may reduce protein in HD



RNA-interference (RNAi) reduces protein production

- Micro RNAs (miRNA) are naturally made by cells to help them break down RNA before it makes a protein.[1]
- A gene therapy can deliver miRNA that targets *HTT* RNA so that diseased HTT proteins are not produced.
- Studies have shown reducing both normal and abnormal *HTT* RNA is an effective treatment strategy in animal models of HD[2,3]

1. National Center for Biotechnology Information, Probe Database Glossary: RNAi. Available at: https://www.ncbi.nlm.nih.gov/probe/docs/techrnai/. Accessed May 30, 2020. 2. Boudreau RL et al. Mol Therapy. 2009. 3. Drouet V et al. Ann Neurol. 2009.

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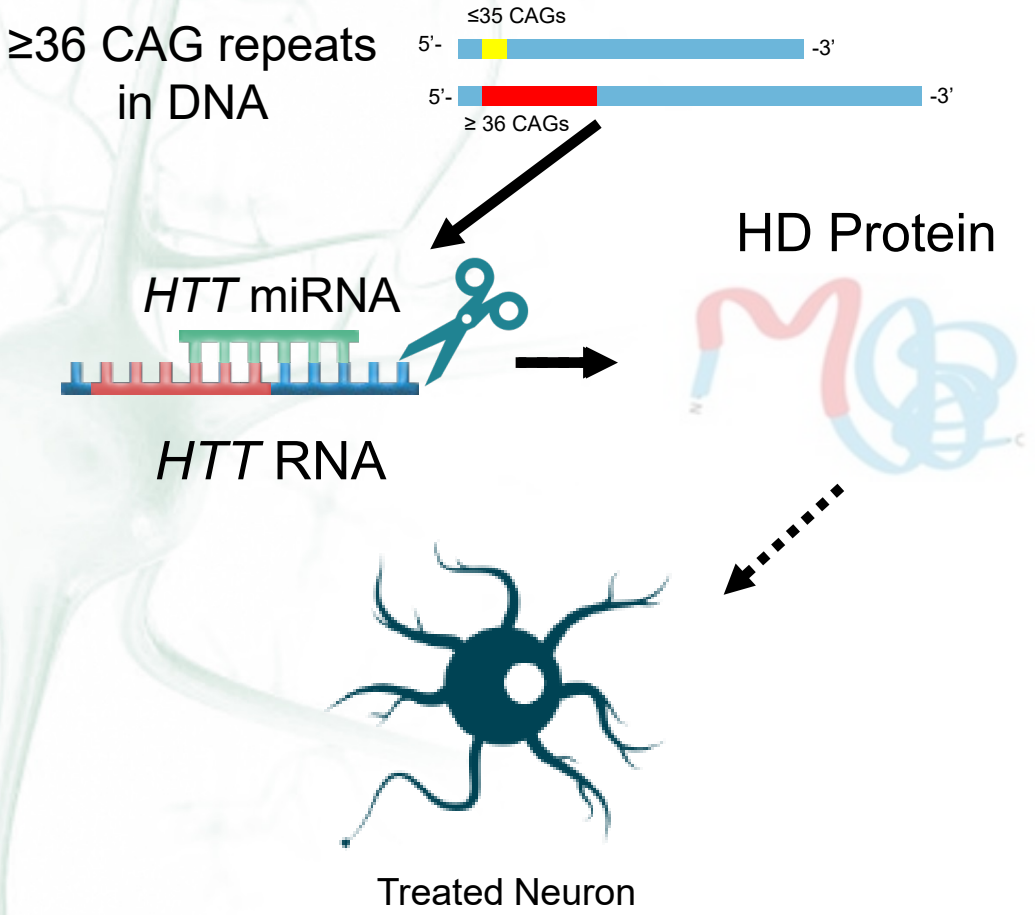

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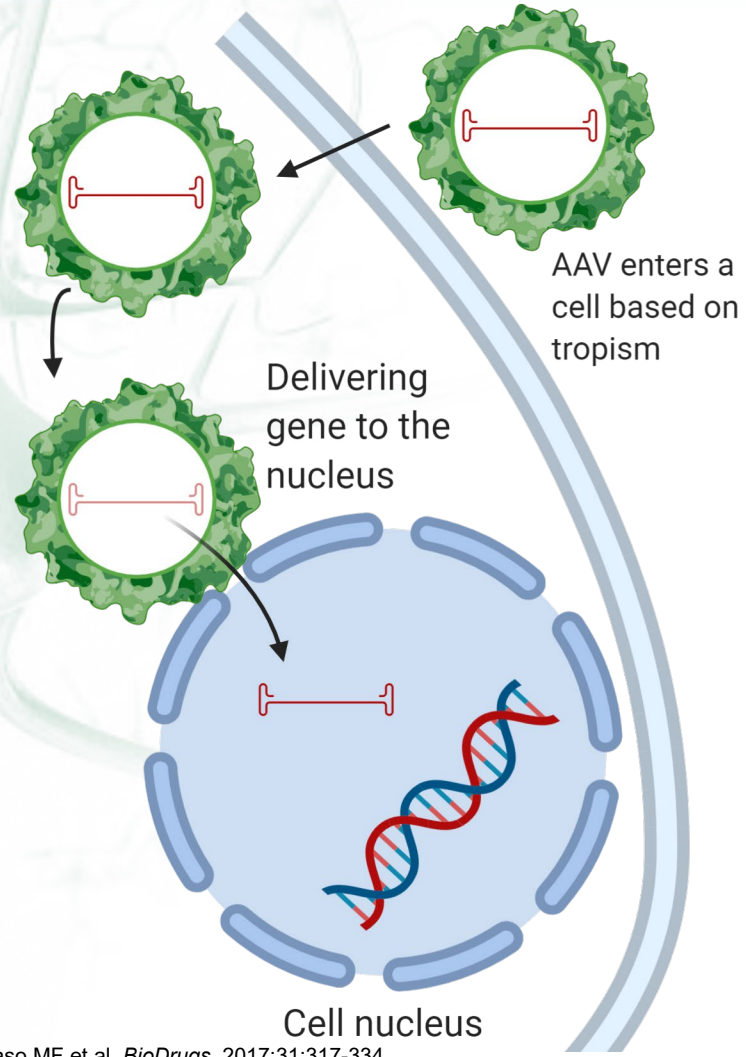
Delivering the transgenes to the cells



- Once AAV enters a cell, it delivers the therapeutic transgene to the nucleus.[1]

1. Naso MF et al..*BioDrugs*. 2017;31:317-334

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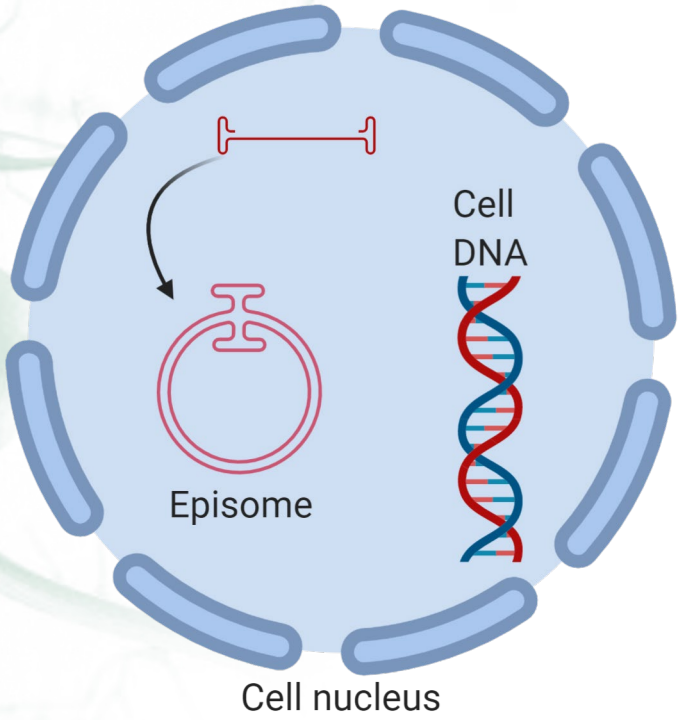

- Once AAV enters a cell, it delivers the therapeutic transgene to the nucleus.[1]
- The two ITRs can attach to each other to make a closed loop called an episome.[1]
- This episome is separate from the genes of the cell.[1]

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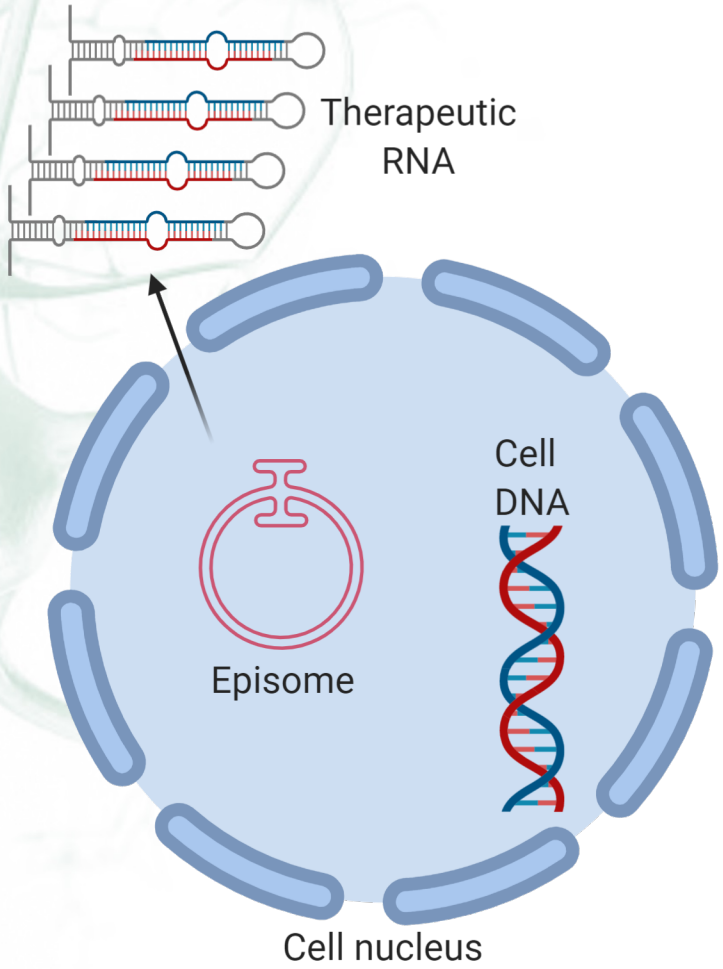
Delivering the transgenes to the cells



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- The two ITRs can attach to each other to make a closed loop called an episome.[1]
- This episome is separate from the genes of the cell.[1]
- The new episomal gene may be used by the cell to continually make the new treatment.[1]

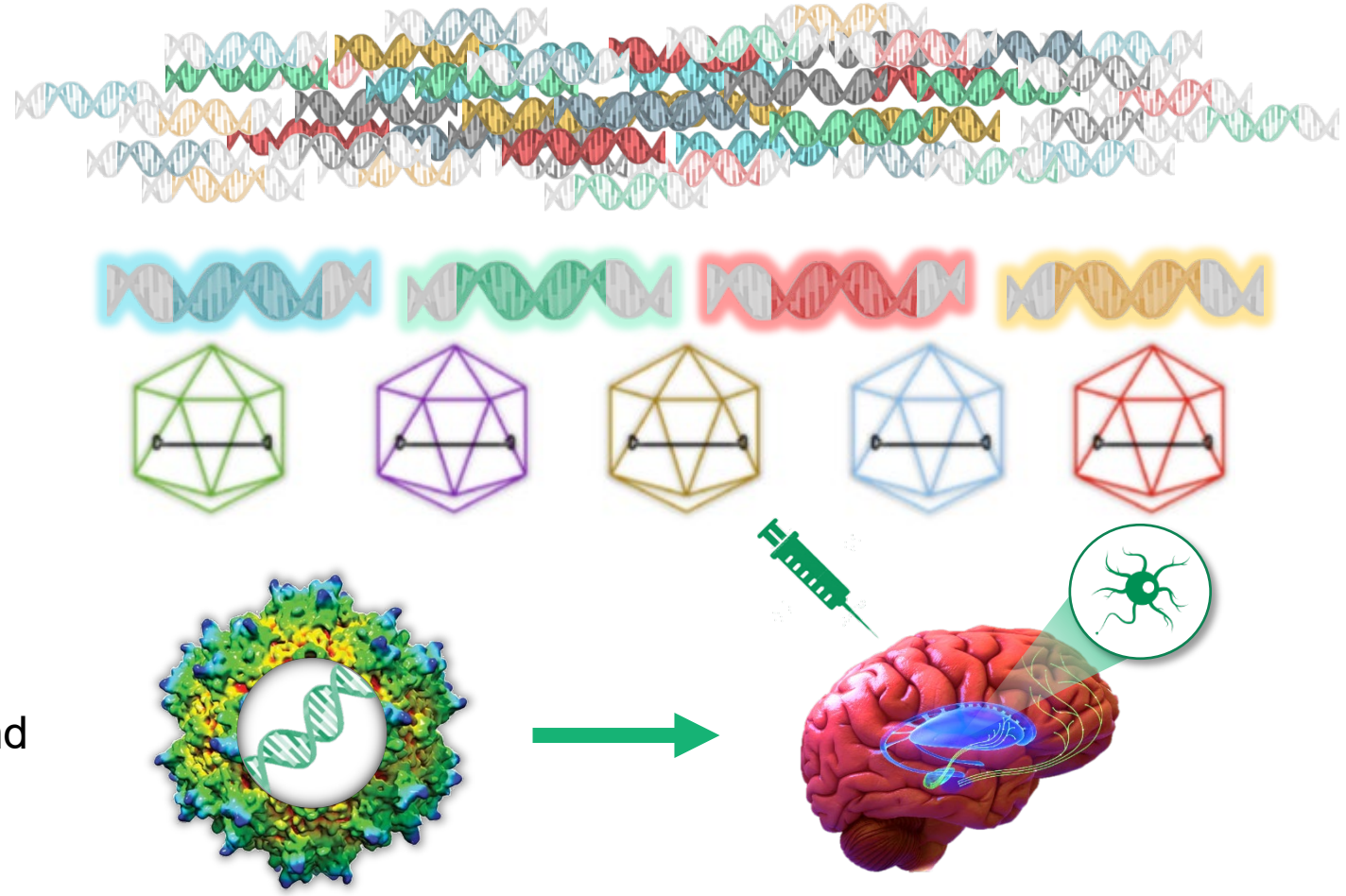
1. Naso MF et al..*BioDrugs*. 2017;31:317-334

Building an investigational gene therapy

Transgenes are designed and tested for accuracy, safety, and potency.

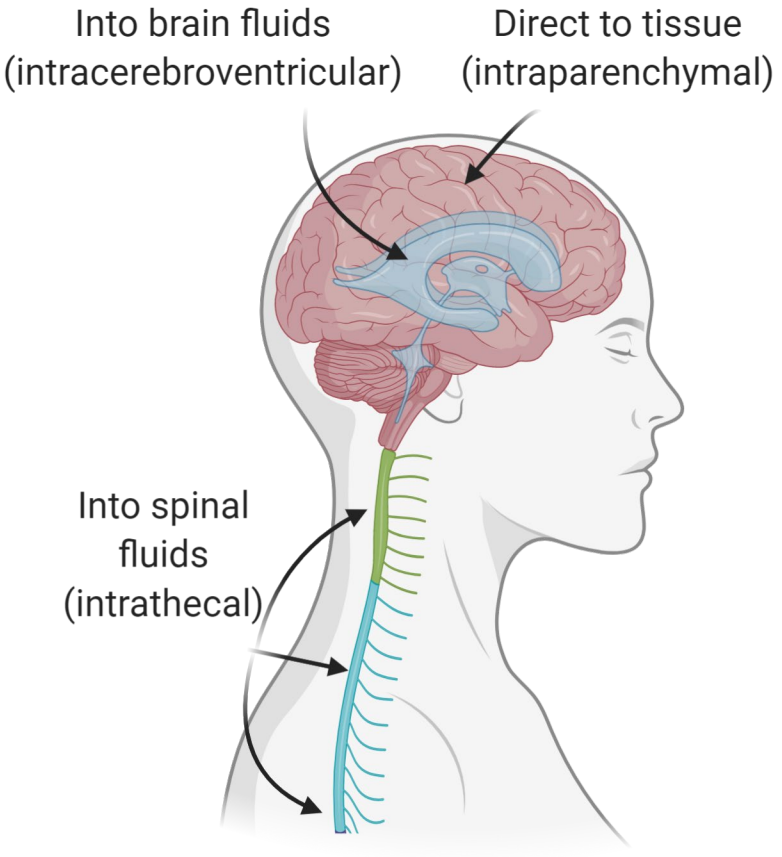
The leading transgenes are packaged into different AAVs and tested in cells.

The leading vector will help determine how the gene therapy will be delivered to target cells and tested in animals.



The blood-brain barrier makes treating cells in the brain a challenge

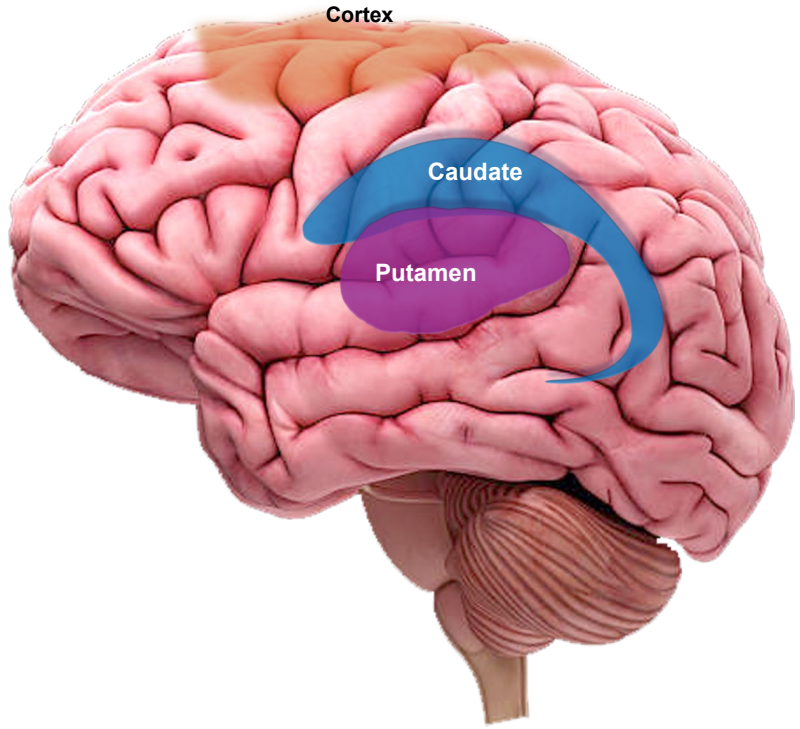
- Direct delivery into the brain (intraparenchymal) places the vector exactly where intended.[1]
- Delivery to the cerebrospinal fluid (CSF) in the brain or spinal cord may allow the vector to travel with the CSF to cells in many places.[1]
- If the AAV serotype can cross the blood-brain barrier, IV (intravenous) injection may be possible.[1]



1. Hudrey, E. et al. *Neuron*. 2019.

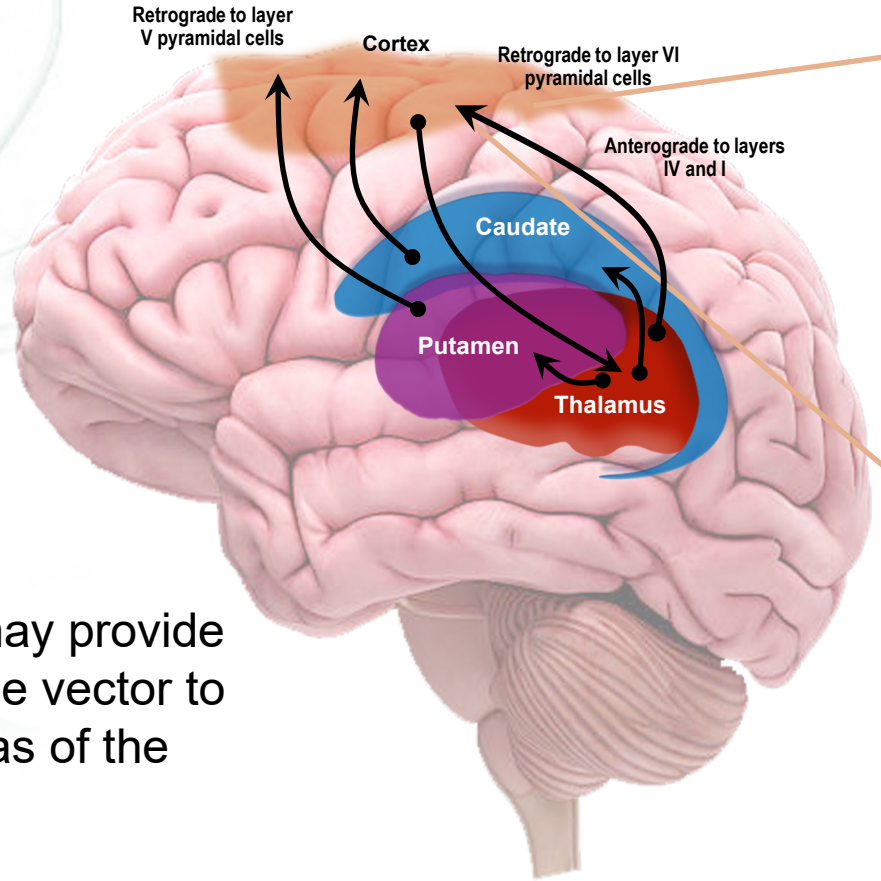
Multiple areas of the brain are important to target to treat HD[1]

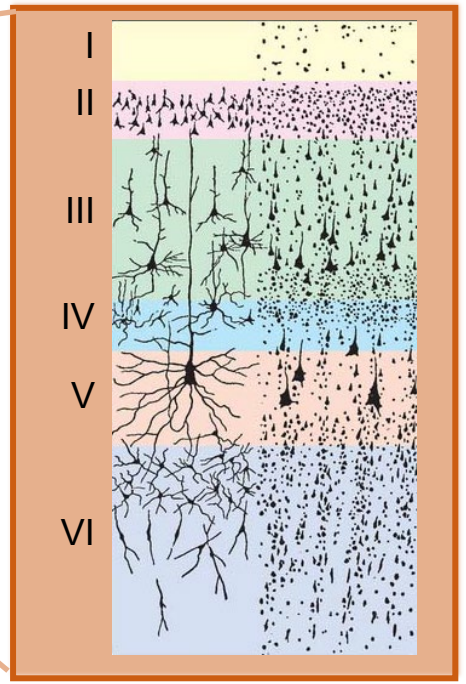
Structure	Related To:
Putamen	Motor coordination, involuntary movements
Caudate nucleus	Emotional and behavioral control, learning and memory
Cerebral cortex	Behavior, mood disturbance, motor impairment



1. Rangel-Barajas C. et. al. *J of Huntington's Disease*. 2016.

The AAV vector can travel using the connections between key structures in the brain[1,2]





Anterograde and retrograde to neurons in multiple layers of the cortex

The thalamus may provide a pathway for the vector to reach other areas of the brain.

Anterograde: moving forward along a pathway
Retrograde: moving backwards along a pathway

1. Rangel-Barajas C. et. al. *J of Huntington's Disease*. 2016. 2. Salegio, EA. et al. *Gene Therapy*. 2013..

Next steps: From lab to clinic

- The potential gene therapy must be thoroughly tested before it can be given to people to determine:[1]
  - If the therapy is likely to work as expected;
  - That the therapy is likely to have an impact on the disease;
  - If the therapy is likely to be safe and can be safely administered;
  - The possible side effects that could occur in people; and
  - That the therapy can be consistently produced at high quality.
- This information is presented to the FDA, which will decide if the potential therapy will be cleared for testing in clinical trials by granting Investigational New Drug status.[1]

1. US Food and Drug Administration. Investigational New Drug (IND) Application. https://www.fda.gov/drugs/types-applications/investigational-new-drug-ind-application Accessed 26 May, 2020

Thank you for your interest.

For more information, find us at voyagertherapeutics.com

For questions regarding VY-HTT01 or the VYTAL clinical trial, please contact clinicaltrials@vygr.com

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