Gene Therapy for Rare Diseases: Development and Preclinical Research

Preclinical research is the earliest stage of developing a new therapy. The therapy itself is designed and optimized during this period, and many laboratory studies are done. One of the important goals of preclinical research is to make sure that the therapy is unlikely to cause serious harm (also called toxicity) before it is tested in people.

Preclinical research programs are scientifically complex and must be carefully planned and executed. Some preclinical research (such as certain types of studies done in animals) also must be carried out in compliance with strict regulatory guidelines.[1] Gene therapies are a relatively new—and quickly evolving—type of medical treatment.[1,2] As scientific advancements are made, regulatory requirements for these innovative treatments are being updated just as quickly to help ensure their quality and safety.[1]

Whether or not a potential new gene therapy can be tested in **clinical trials** (studies done in human beings) is determined by the strength and quality of its preclinical research. This brochure provides an overview of some of the steps, processes, and rules involved in getting a potential new gene therapy ready for clinical trials. Each step may take months, or even years. However, the work done during this period is essential to ensuring the safety and efficacy of potential new gene therapies, and to setting them on the course most likely to lead to successful clinical trials—and, by extension, to eventual availability to patients.[1]

Note: some words that may be unfamiliar are highlighted and are defined in the glossary at the end of this brochure.

Amicus Therapeutics®

Amicus Therapeutics has developed this educational resource in collaboration with the rare disease community and thought leaders.

Overview: How gene therapy works to treat rare genetic diseases[2,3]

Everyone has genetic information called **DNA** in his or her **cells**. DNA is inherited through **genes** that are passed down from the person's father and mother.

Genes provide instructions for the body to make proteins that cells need to work properly.

Genetic diseases are caused by **variants** (also called mutations) in the DNA that change the way the affected gene functions. These changes in function are what causes the **signs** and **symptoms** of the diseases.

Gene therapy is designed to add, remove, or change **genetic material** in a person in order to treat a disease. Gene therapies can be designed to target and "fix" genes in several different ways. These include:

- Adding healthy genetic material that functions normally. In diseases in which a **gene variant** causes a *loss* of protein function, a healthy gene can be added to restore function. Doctors and researchers sometimes use the term "**augmentation**" to describe this kind of gene therapy.
- Stopping a gene's disease-causing effects. In some diseases, a gene variant causes a *gain* of function for a protein that may have harmful results. In these cases, the goal of a gene therapy may be to turn off (or reduce the effects of) the gene variant so that it no longer causes disease. Doctors and researchers sometimes use the term "**suppression**" to describe this type of gene therapy. Conversely, healthy genes already present in the body can be turned on to help control the disease.

To learn more

Amicus offers additional resources that provide families with information about gene therapies; please contact us at patientadvocacy@amicusrx.com to request copies.

1. First steps

Before any new gene therapy can be designed, the gene (or genes) that cause the disease have to be identified. This process often involves extensive research and may take many years. A majority of rare diseases—about 80%—are caused by a problem in a single gene.[4]

Researchers also must decide which cells in the body the therapy should target. Which specific cells are targeted depends on the disease the therapy is being designed to treat.[1]

These important factors about the disease and how it affects the body help determine how the components of the gene therapy will be designed.[5]

4. Submitting results for regulatory approval[1]

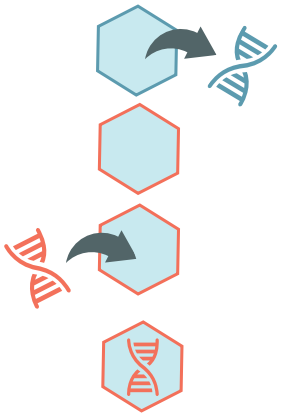
If preclinical research results demonstrate that the potential new gene therapy meets strict safety criteria and has potential therapeutic value, the data may be submitted to the region's or country's regulatory authority for official review.

Clinical research evaluating the safety and efficacy of the potential new gene therapy in human beings cannot begin until—and unless—the preclinical research data pass this regulatory milestone. And because standards for the rigor of preclinical research and the quality of its data are very high, only a small fraction of potential new therapies may go on to be tested in clinical trials!

2. Designing a gene therapy

Gene therapies can be made in many different ways. Processes that may be involved in creating one specific type of gene therapy (ie, an augmentation therapy that uses a viral vector) are described below.

Genetic material and vector are combined to form the gene therapy[5,8]



The genetic material of the virus that will be the vector is removed (or disabled)

The virus's capsid is modified as needed

The new genetic material designed to treat the disease is inserted into the viral vector (ie, the empty, modified viral capsid)

The gene therapy—consisting of the vector with the new genetic material inside—is now complete

A gene therapy has two main components—genetic material and a vector

The genetic material in a gene therapy provides information that helps the targeted cells function correctly. For example, it might provide instructions to the cells for making a missing or faulty protein.[5]

The **vector** of a gene therapy is the delivery system that brings the genetic material to the targeted cells.[5] Many different types of vectors exist. Researchers decide which type to use based on the disease and the cells being targeted.[7]

The genetic material of a gene therapy typically includes DNA designed to correct the disease-causing gene variant as well as a section of DNA called a **promoter**.[6] The function of a promoter is to help turn genes on or off within the targeted cells. There are two main types of promoters[6]:

- those that activate genes only in specific cell types (called tissue-specific promoters)
- those that allow for gene expression in a wider variety of cell types (called ubiquitous promoters)

What type of promoter is used (as well as other aspects of how the promoter is designed) is determined by several factors. For example, a gene therapy that acts on many different cell types may be more desirable in some cases, whereas targeting a more narrow range of cells may be safer and more effective in others.[6]

Viruses are often used as vectors because they are good at targeting and entering cells.[3] Several different species of virus can be used as vectors. Researchers choose the species they believe will work best for the specific disease being treated.

When a virus is used as a vector, it is modified in several ways to make it able to safely and effectively deliver the desired genetic information to cells. The virus's own genetic material is removed (or disabled) so that it cannot cause disease. The virus's outside shell, or **capsid**, also may be modified to help it[5,8]:

- target the correct cells within the body
- successfully insert the genetic material into the cells
- avoid unwanted side effects

3. Planning and conducting preclinical research

The main phases of preclinical research are described below. Some phases may overlap, and their overall order may vary (depending on multiple factors). Preclinical research programs usually require a total of about 4-5 years to complete.[9]

Planning the research program

- Data about the disease to be treated (as well as any existing data about therapy components, if available) are carefully reviewed
- A program of laboratory research—including ***in vitro*** and ***in vivo*** studies—is mapped out to create a scientific rationale for the potential new therapy by[1]:
  - defining a possible dose range and dose schedule
  - confirming that the therapy targets the correct cells
  - identifying potential toxicities or other risks
  - helping to determine patient eligibility criteria for future clinical trials

Choosing animal species/ developing animal models

- Animal species used in testing are chosen based on[1]:
  - relevant similarities to humans
  - susceptibility to the vector
  - immune tolerance of human genetic material
- Healthy animals are used for some studies; some animals also may be modified to create **animal models of disease**[1]

Proof-of-concept (POC) studies

- POC studies are small *in vitro* or *in vivo* studies designed to test the scientific rationale developed during the planning process
  - data from these studies are used to refine the design of definitive preclinical studies (see below)[1]
  - early POC studies also may influence the choice of animal models to be used in later studies[1]

Proof-of-concept studies *are generally done with small batches of the gene therapy that were created in a laboratory.*

Definitive preclinical studies

- Definitive preclinical studies are larger studies that are designed similarly to clinical trials[1]
- Definitive preclinical studies include[1]:
  - *Efficacy studies* designed to evaluate whether the therapy may effectively treat the disease (these studies often compare three groups of animals: animals with disease who receive the therapy, animals with disease but are not treated, and healthy animals)[1]
  - *Safety and* ***toxicology*** *studies* designed to test for adverse reactions to the therapy (these studies are usually done in healthy animals of two different species, often a rodent and a non-human primate)
- Safety and toxicology studies must be conducted in compliance with extensive regulations known as Good Laboratory Practices (GLPs), which cover equipment use, laboratory processes, humane animal care, and other important research elements[1,10]
- Regulatory agencies use data from these important studies to determine whether a potential new therapy may move forward into clinical trials

Some types of ***definitive preclinical studies*** *must use the same gene therapy product that will be used in clinical trials. This means that a large-scale manufacturing process must be developed before preclinical research can be completed*[1,4]

Glossary

Animal model of disease: an animal bred or genetically engineered to have a disease process similar to a specific disease that occurs in human beings

Augmentation: gene therapy that adds a properly functioning gene into cells that have poorly functioning gene variants

Capsid: the outside shell of a virus that may be modified to target specific cell types

Cell: basic building block of all living things

Clinical trial: voluntary research studies conducted in people and designed to answer specific questions about the safety or effectiveness of drugs, vaccines, other therapies, or new ways of using existing treatments

DNA (deoxyribonucleic acid): substance within genes that contains instructions, or code, for making proteins

Gene: made up of DNA; some genes act as instructions to make molecules called proteins

Genetic material: DNA (or sometimes RNA [ribonucleic acid], a biochemical that helps DNA send its biological instructions) provided by a gene therapy to treat a disease

Gene variant (also known as mutation): a change to the structure of a gene that may alter the gene's function, sometimes resulting in diseases or conditions

***In vitro* study:** research done outside of a living organism (*in vitro* is Latin for "in glass")

***In vivo* study:** research done in living organisms (*in vivo* is Latin for "in life")

Preclinical research: laboratory studies done in animals or in equipment such as test tubes (see ***in vitro study*** and ***in vivo study***, above)

Promoter: a part of the genetic material in a gene therapy that helps turn genes on or off within the targeted cells

Sign: objective evidence of a disease or condition that may be recognized by the patient, as well as others

Suppression: gene therapy that turns off a gene variant that is not functioning properly

Symptom: subjective evidence of a disease or condition that may only be perceived by the person who has the disease

Toxicology: the study of potential adverse effects of chemical, physical, or biological agents on living organisms (including methods of preventing or reducing these adverse effects)

Variant: (see **gene variant**, above)

Vector: a biological agent (virus) or biochemical agent (liposome or polymer) used to carry and transfer genetic material into a cell

References:

1. Preclinical Assessment of Investigational Cellular and Gene Therapy Products–Guidance for Industry. Food and Drug Administration Center for Biologics Evaluation and Research. November 2013; reviewed 2019. https://www.fda.gov/regulatory-information/search-fda-guidance-documents/preclinical-assessment-investigational-cellular-and-gene-therapy-products. Accessed June 18, 2020. **2.** Anguela XM, High KA. Entering the modern era of gene therapy. *Annu Rev Med.* 2019;70:273-288. doi:10.1146/annurev-med-012017-043332 **3.** American Society of Cell and Gene Therapy. Gene Therapy Basics. https://www.asgct.org/education/gene-therapy-basics. Accessed June 22, 2020. **4.** Human Gene Therapy for Rare Diseases–Guidance for Industry. Food and Drug Administration Center for Biologics Evaluation and Research. January 2020. https://www.fda.gov/media/113807/download. Accessed June 18, 2020. **5.** Genetics Home Reference. National Institutes of Health U.S. National Library of Medicine. Help Me Understand Genetics. Gene Therapy. How does gene therapy work? June 2020. https://ghr.nlm.nih.gov/primer/therapy/procedures. Accessed June 18, 2020. **6.** Powell SK, Rivera-Soto R. Viral expression cassette elements to enhance transgene target specificity and expression in gene therapy. *Discov Med.* 2015;19(102):49-57. **7.** Nayerossadat N, Maedeh T, Ali PA. Viral and nonviral delivery systems for gene delivery. *Adv Biomed Res.* 2012;1:27. **8.** Nance ME, Duan D. Perspective on adeno-associated virus capsid modification for Duchenne muscular dystrophy gene therapy. *Hum Gene Ther.* 2015;26(12):786-800. doi:10.1089/hum.2015.107 **9.** Paul SM, Mytelka DS, Dunwiddie CT, et al. How to improve R&D productivity: the pharmaceutical industry's grand challenge. *Nature Reviews Drug Discovery.* 2010;9:203-214. **10.** The Drug Development Process. Step 2: Preclinical Research. U.S. Food and Drug Association. Reviewed 2018. https://www.fda.gov/patients/drug-development-process/step-2-preclinical-research. Accessed June 21, 2020.

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Please discuss any medical questions with a health-care professional (HCP).

If you would like to provide feedback on this educational resource or would like additional information please contact:
patientadvocacy@amicusrx.com.