Jerusalem Science Contest

Molecular Biology

Lecture 5: Genetic Engineering and Gene Therapy

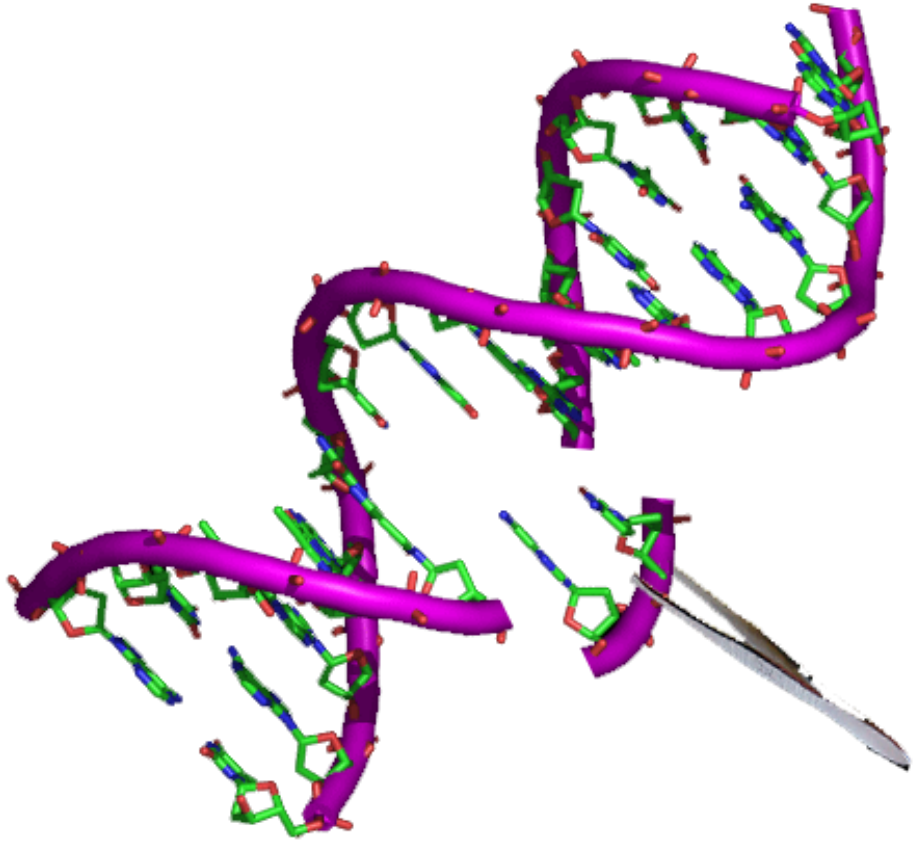


Image from Ciencias EspañolasKoS, CC BY-SA 3.0 <https://creativecommons.org/licenses/by-sa/3.0>, via Wikimedia Commons

Outside reading:
https://education.nationalgeographic.org/resource/genetic-technology/

Genetic Engineering

- Genetic engineering is the process of making specific, intentional changes to the genome of an organism
- Done to change the inherited traits of the organism
- Since the 1970s, scientists have developed more and more sophisticated techniques to do this
- Important to distinguish from selective breeding in agriculture (which aims to enhance traits using existing genetic variation) or random mutagenesis

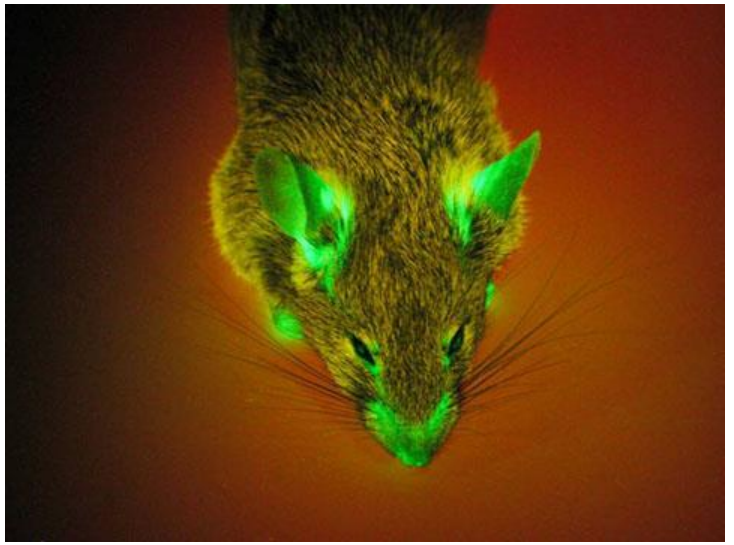


Image from Кнут Хаугланд, CC BY-SA 3.0 <http://creativecommons.org/licenses/by-sa/3.0/>, via Wikimedia Commons

Restriction Enzymes: The Doorway to Genetic Engineering

- In 1970, Hamilton Smith and his lab isolated an enzyme from bacteria that could cut DNA at a specific sequence
- HindII enzyme cuts in middle of sequence 5’ GT(T/C)(A/G)AC 3’
- This was the first example of a “restriction enzyme”, allowing researchers to cut and paste different DNA molecules together in precise ways – “recombinant DNA”
- There are now over 3500 known restriction enzymes, able to cut at over 350 different DNA sequences

Restriction Enzymes

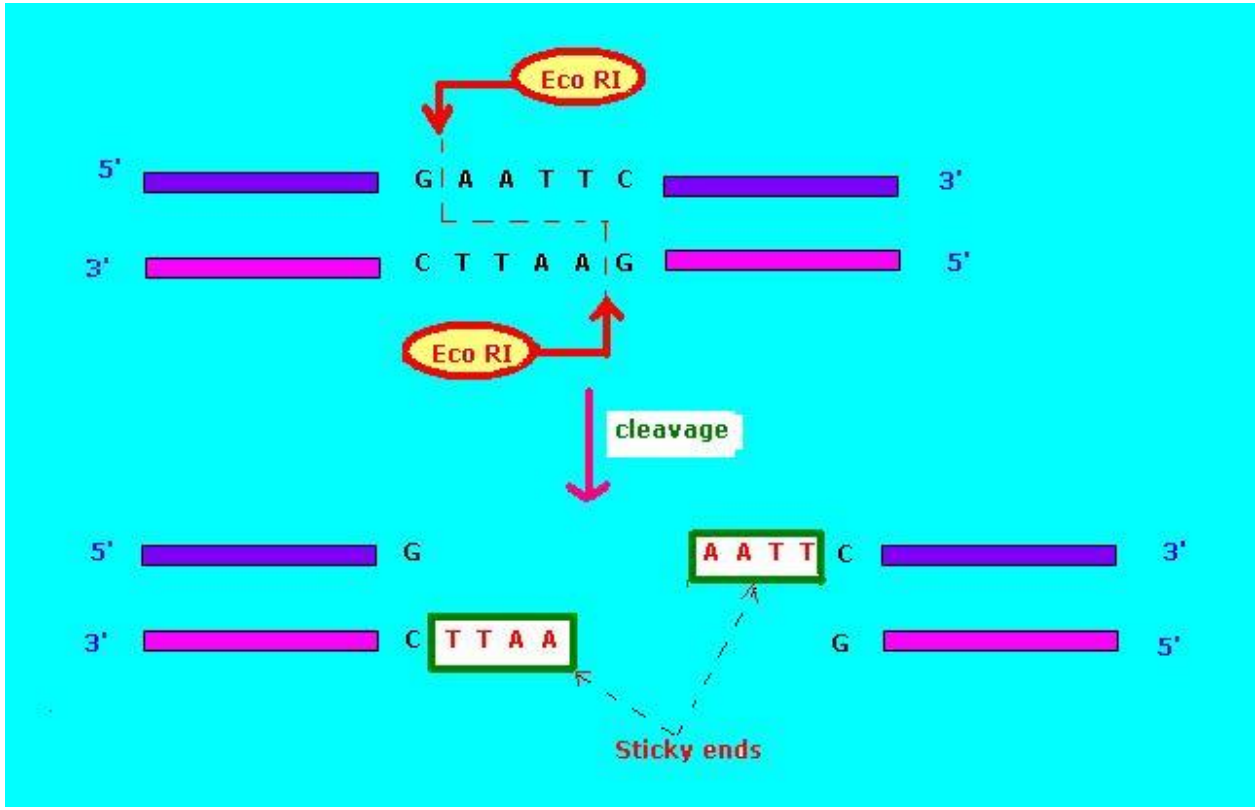


Image from Tinastella, Public domain, via Wikimedia Commons

Cloning DNA and GMOs

- Almost immediately after the discovery of restriction enzymes, they were used to "clone" DNA from one organism into another
- This resulted in genetically modified organisms, or GMOs.
- Many uses, from basic biological research to medicine to improved agricultural crops
- For example, essentially all human insulin today for diabetic patients is produced by recombinant *E. coli* bacteria
- Most corn, soybeans, and cotton grown in the U.S. also contain recombinant DNA

Cloning DNA

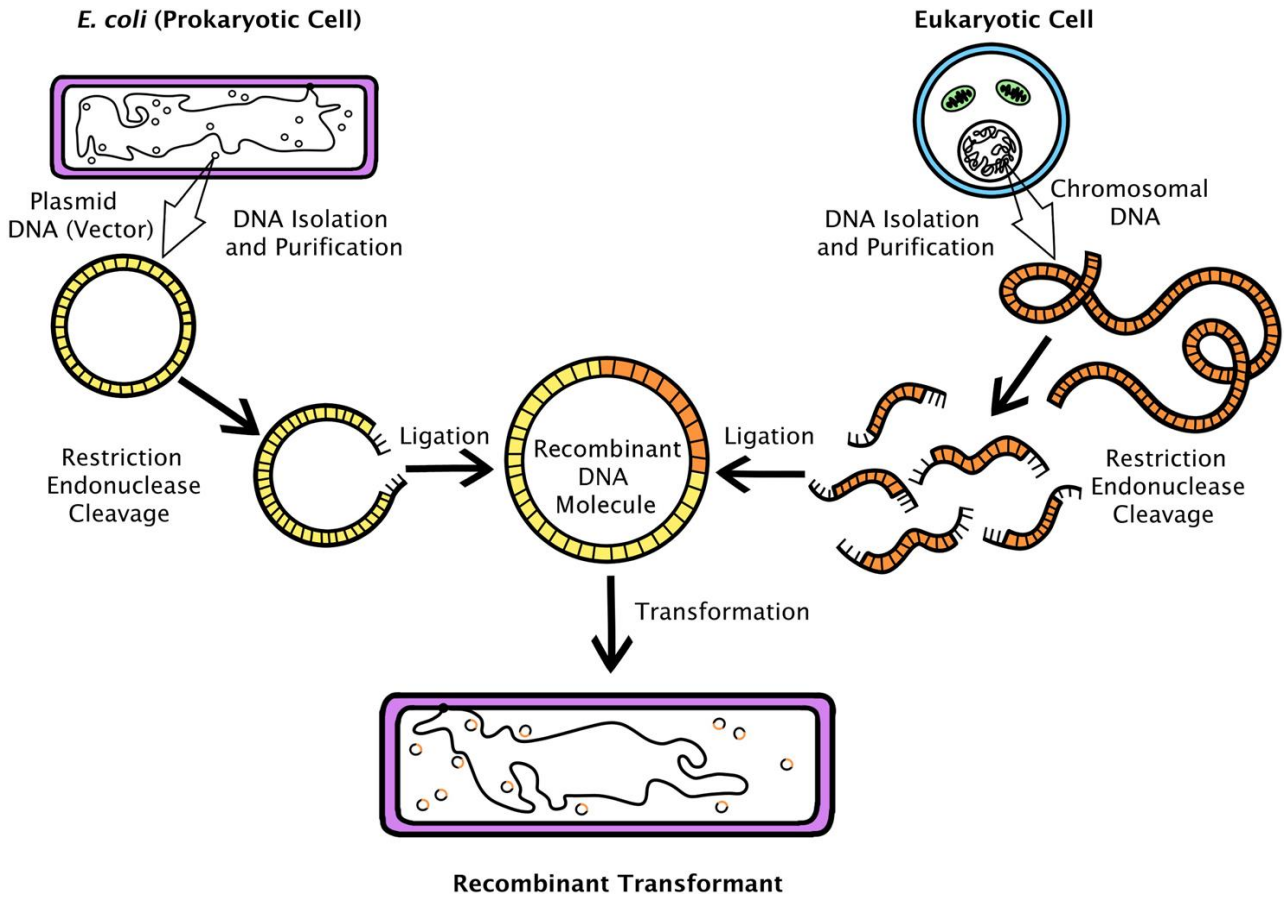
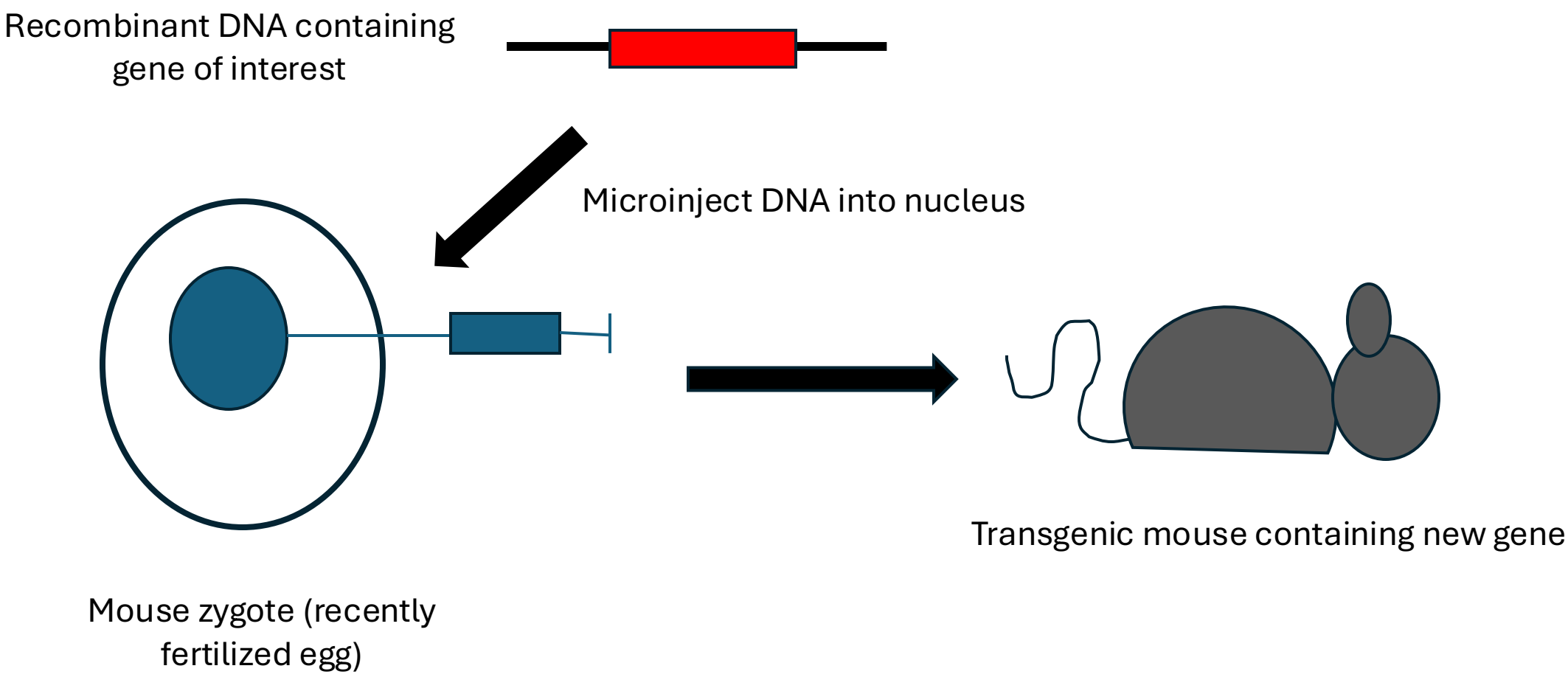


Image from Angela N.H. Creager, CC BY 4.0 <https://creativecommons.org/licenses/by/4.0>, via Wikimedia Commons

Genetically Modified Mammals

- Bacteria and plants are relatively easy to modify genetically.
- Mammals pose more of a challenge, but more recent developments have allowed genetic engineering of mice, turning them into one of the most important medical research tools.
- Introducing a new gene into a mouse produces a ***transgenic*** mouse, while deleting a gene can produce a ***knockout*** mouse
- Tissue-specific and “conditional” knockouts are also possible
- These techniques all involve manipulating zygotes (recently fertilized eggs) or early embryos

How To Make a Transgenic Mouse



Gene Therapy

- The only way to cure genetic disorders in humans is to “fix” the mutation that causes the disease.
- This is a form of genetic engineering called “gene therapy”
- Diseases such as Tay-Sachs disease, Huntington’s disease, cystic fibrosis, and sickle cell disease are attractive targets for gene therapy

Somatic Cell Gene Therapy

- The current laws regulating gene therapy forbid making genetic changes that can be passed down to future generations.
- This means that gene therapy approaches can only be used to alter somatic cells (differentiated cells of the body) and not germ cells (sperm and eggs) or human embryos.
- This requires approaches that can target specific tissues or organs in the body, or genetically engineering cells in the lab and reintroducing them into the patient.
- This also generally limits treatment to diseases where the patient can survive past infancy

Gene Therapy Approaches

- One gene therapy approach uses ***retroviruses*.**
- These viruses use RNA as their genetic material instead of DNA.
- During infection, they make a DNA copy of their genome and insert it into a chromosome of the infected cell
- Recombinant retroviruses can be used to insert a “good” copy of gene into cells of a patient

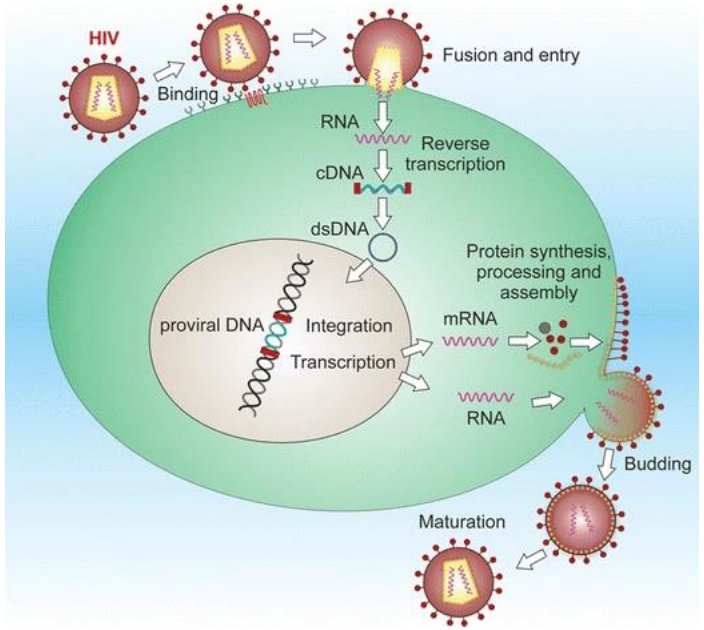


Image from Giovanna De Chiara, Maria Elena Marcocci, Rossella Sgarbanti, Livia Civitelli, Cristian Ripoli, Roberto Piacentini, Enrico Garaci, Claudio Grassi, Anna Teresa Palamara, CC BY 2.5 <https://creativecommons.org/licenses/by/2.5>, via Wikimedia Commons

Gene Therapy Approaches

- Retroviruses have some limitations as gene therapy “delivery” systems.
  - Rapid mutation rates, limited cell types that can be infected, sometimes even cause cancer

- Other viruses such as adenoviruses are also being used to get around some of these limitations

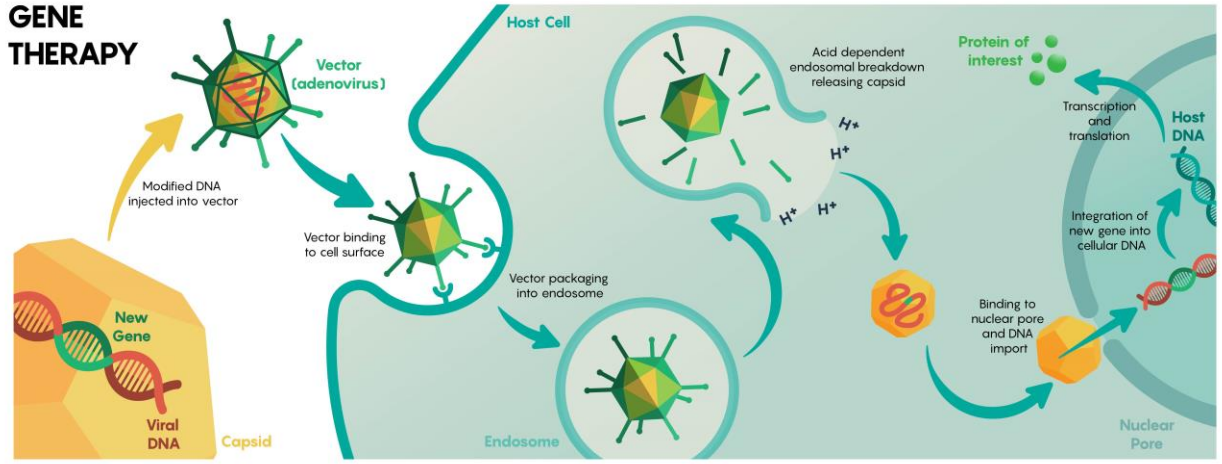


Image from University of Toronto, CC BY 4.0 <https://creativecommons.org/licenses/by/4.0>, via Wikimedia Commons

Gene Editing

- Viral delivery of genes can only fix genetic disorders caused by a complete lack of a protein (i.e. gene is completely non-functional): “recessive” diseases

- Some diseases are caused by a malfunctioning protein that can’t be fixed by just adding a “good” copy of the gene: “dominant” diseases

- A new approach called ***gene editing*** allows us to just correct the mutation in the gene, hopefully curing the disease

Gene Editing

- Gene editing uses another enzyme discovered in bacteria: Cas9
- Using a “guide RNA” that is complementary to a target DNA sequence, Cas9 cuts the DNA at a specific site
- A corrected DNA sequence is then provided, allowing the cell to repair the cut DNA with the desired sequence

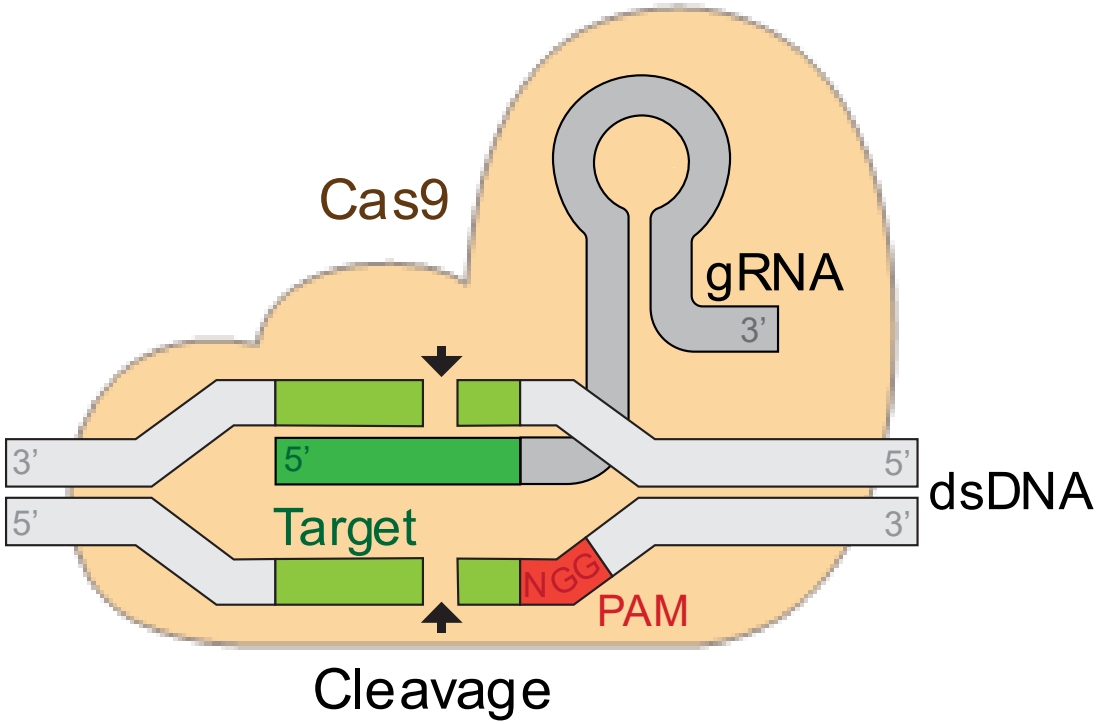


Image from Mariuswalter, CC BY-SA 4.0 <https://creativecommons.org/licenses/by-sa/4.0>, via Wikimedia Commons

The Future of Gene Therapy

- Gene therapy has the potential to allow us to cure all kinds of genetic diseases.
- Genetically engineered immune cells may also be a key to new cancer treatments.
- New genetic tools are very powerful, but also introduce ethical issues
- If any gene sequence can be changed, there is the potential to move away from just diseases toward “cosmetic” genetic engineering
- There is also the possibility of producing ”designer babies”

Next Lecture: Viruses and Vaccines