CRISPR: Not Just a Drawer in Your Refrigerator

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Objectives

1. Discuss gene editing technology.
2. List potential uses for gene editing.
3. Discuss implications of genetic manipulation.

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Do these sound familiar?

- Genetic engineering
- GMOs – genetically modified organisms
- Human Genome Project
- GINA Act of 2008
- Gene therapy
- **CRISPR**

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Let's review…

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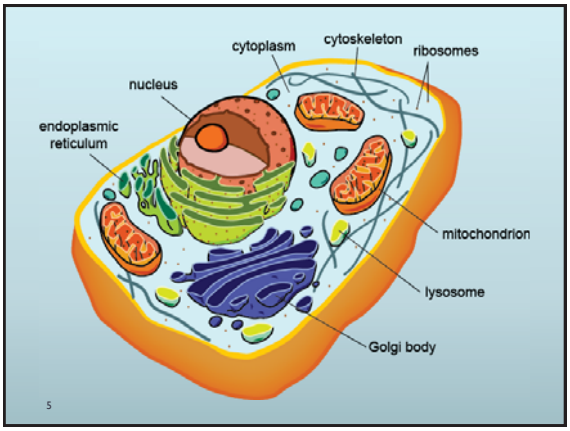

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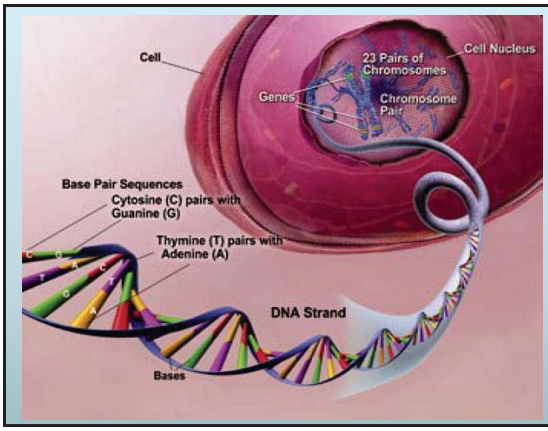
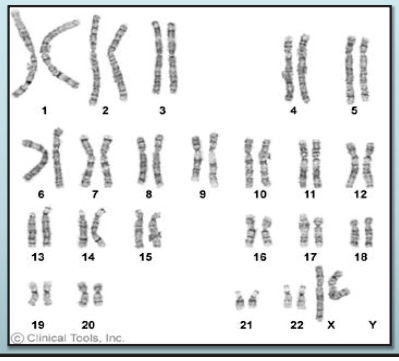


Human karyotype (female)



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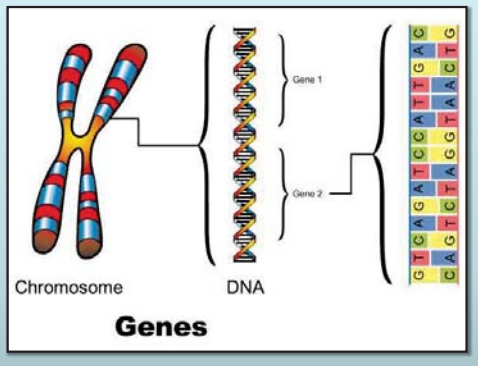

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How does DNA control everything?

- Specific **genes (DNA)** code for:
  - Structural proteins
  - Hormonal proteins
  - Respiratory proteins
  - Enzymes
- Certain genes get decoded/expressed, depending on cell type
- Only genes for a particular cell's function are expressed, others remain compressed.
- "Cellular instruction booklet"

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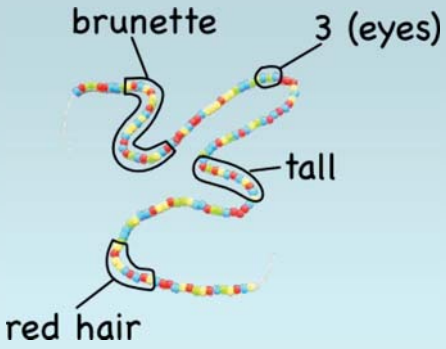

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Human genome

- An individual's complete genetic makeup, including both genes and the "junk" DNA between the genes
- Present in every cell in the body
- Very large: >6 billion bp, >6.5 feet of DNA, ~38,000 genes
- DNA wound tightly around histone proteins into chromosomes to save room in the nucleus
- Individual genomes are unique but overall "map" of human genes is the same

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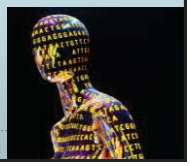
Human Genome Project (1990-2003)

- What is it?
  - Sequence of the entire human genome discovered
  - 23andMe, Ancestry.com, etc.
  - **G**enetic **I**nformation **N**on-discrimination **A**ct of 2008 (GINA Act)



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Genetic Engineering

- What is it?
  - Process that alters the genetic make-up of an organism by either removing or introducing DNA, often **from another organism – "cloning"**
  - Other organisms also have DNA

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Genetic similarities…

- Humans are:
  - 96% identical to chimpanzees
  - 60% identical to chickens
  - 60% identical to fruit flies
  - >60% identical to bananas

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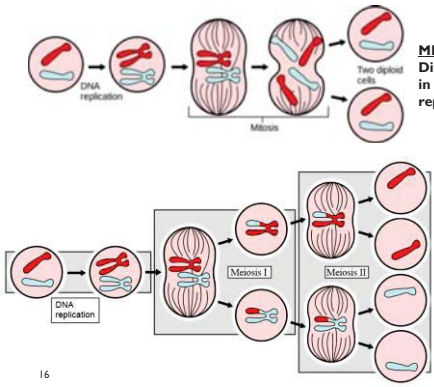

MITOSIS
Division of all cells in the body, except reproductive cells

MEIOSIS
Division of reproductive cells (sperm and egg)

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Genetically modified organisms (GMOs)

- What are they?
  - Any organism generated through genetic engineering
    - 1st bacteria - 1973
    - 1st animals (mice) – 1974
    - Insulin-producing bacteria commercialized in 1982
    - GM food since 1994
    - 2016 safety study

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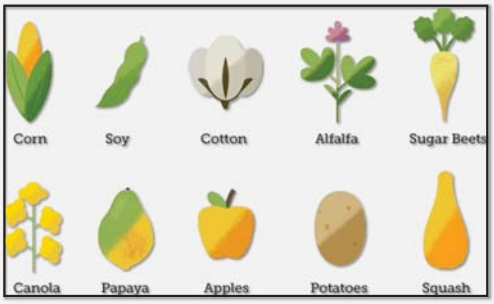
Commercially available GMO crops



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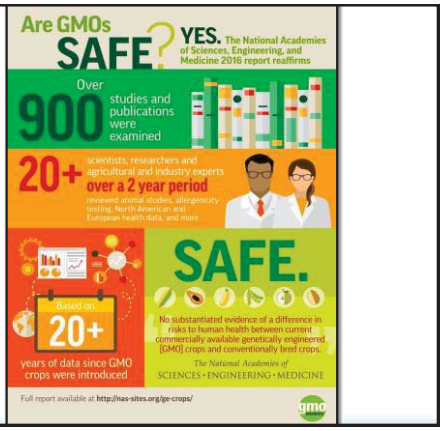

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A little history of genetic manipulation

- For thousands of years, humans have been engineering life through selective breeding – cross breeding organisms with favorable/desired traits to produce superior offspring
- Was not well understood until DNA and the genetic code were discovered
- 1960s – Scientists bombarded plants with radiation, to cause random variations in DNA, in hope of a superior variation by chance

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History, cont.

- 1970s – scientists inserted DNA fragments into bacteria, plants and animals to study and modify them
- Earliest GMO was a mouse (1974), making mice a standard tool used in research that saved millions of lives
- 1980s – Commercialized – first patent given for a microbe engineered to absorb oil
- First food modified in the lab went on sale in 1994 (Flavr Savr tomato)
- 1990s – Brief foray into human engineering to treat infertility

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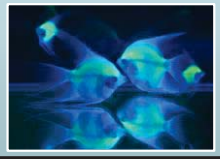
Present-day

- Today, super-muscled pigs, featherless chickens, fast-growing salmon, see-through frogs, luminescent fish
- We also produce many chemicals by means of engineered life: clotting factors, growth hormones and insulin


Trends in genetic engineering

- Until recently, gene editing was expensive, complicated, and took a long time to do.
- Nearly overnight, costs and time required have decreased dramatically

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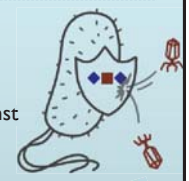
CRISPR

- Clustered regularly interspaced short palindromic repeats
- Modification of bacterial defense against viruses
- "molecular scissors"
- Man-made molecule that can be programmed to find mutated or diseased DNA

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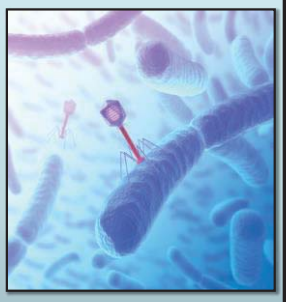
Infection by bacteriophages

- Viruses that target bacteria
- Need bacteria to be able to reproduce
- Most, but not all, bacteria do not survive infection
- Survivors "catalog" short DNA sequences from viral invaders, use to recognize and defend against subsequent infections



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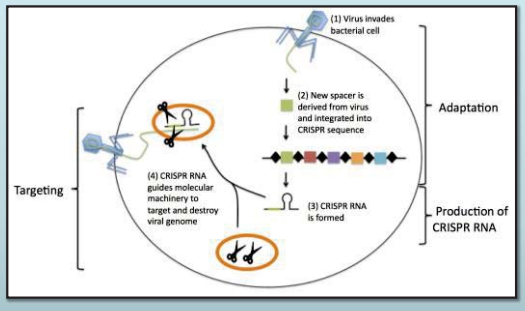

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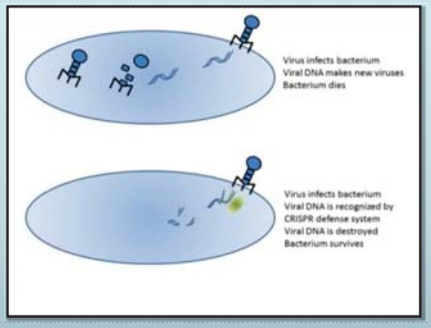
CRISPR in bacteria



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CRISPR-Cas9 System

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CRISPR in humans

- Since human genome has been sequenced, disease-causing genes/targets for CRISPR are continually being discovered
- CRISPR-Cas9 can be modified in the lab to recognize targets and remove or modify them
- Target-specific CRISPR introduced into human cells with DNA "flaw" of interest

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CRISPR in humans, cont.

- Human gene of interest can then be modified or eliminated by CRISPR-Cas9 system
- Cell's natural DNA repair machinery will replace eliminated gene with healthy DNA **OR**
- New, normal DNA may be incorporated
- Disease-specific CRISPR-modified cells then infused into patient **OR**
- CRISPR may be introduced directly into circulation or to a specific organ

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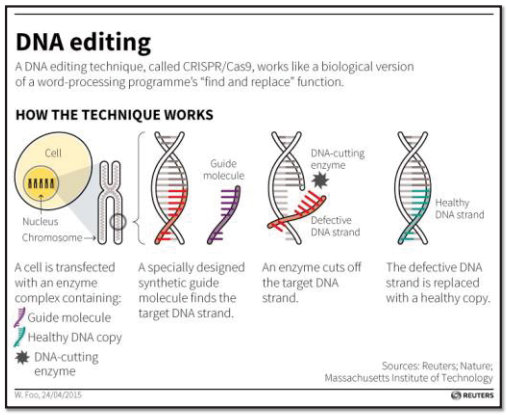
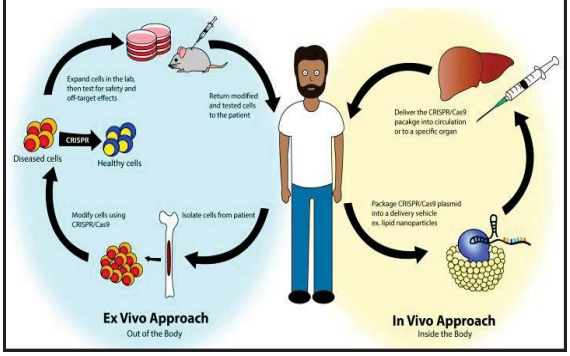


Two approaches to treatment



Human implications

- Genetic/inherited defects, some just annoying, some deadly
  - Color blindness
  - Hemophilia
  - Huntington's Disease
  - Sickle cell disease
- 3000+ genetic diseases are caused by just a single mutation in DNA, rather than a change in larger stretches of DNA
- Cas9 already being modified to fix just a single mutation in the cell

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Things to consider…

- Changes in somatic/non-sex cells will not be passed on to offspring
- Changes in sex cells (sperm and egg) may be heritable
  - Overall modification of the human race?
  - Disease eradication?
  - Greatly prolonged life expectancy?
  - Designer babies – comparison with current pre-natal testing

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More things to consider…

- CRISPR, while very accurate, is not perfect
- Some mistakes and mis-edits
- Don't yet know how certain genes may impact others
- Banning genetic research/editing will only cause scientists to go to other countries where the technology is accepted, possibly with less oversight and transparency

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Other possible uses

- In utero CRISPR treatment?
- Remove malaria from mosquitos
- Treat HIV and other viruses
- Make disease-resistant crops
- Make multiple gene edits at once – complex diseases
- The list goes on…

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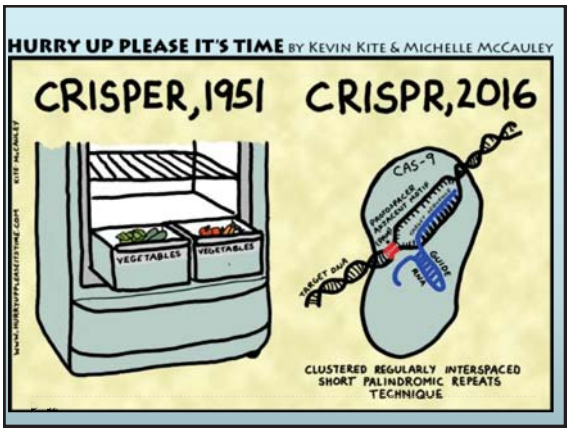
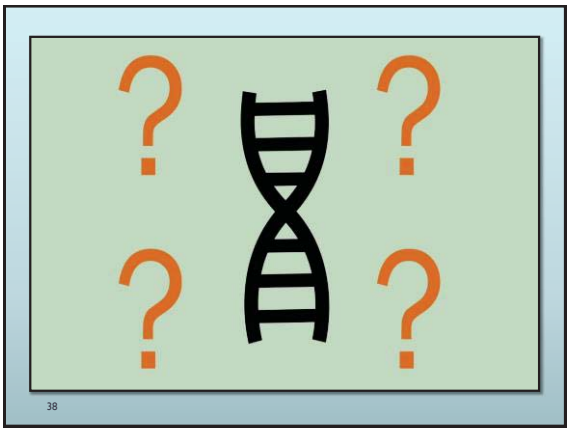

HURRY UP PLEASE IT'S TIME BY KEVIN KITE & MICHELLE MCCAULEY
CRISPER, 1951
CRISPR, 2016
VEGETABLES
VEGETABLES
CAS-9
TARGET DNA
GUIDE RNA
CLUSTERED REGULARLY INTERSPACED SHORT PALINDROMIC REPEATS TECHNIQUE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