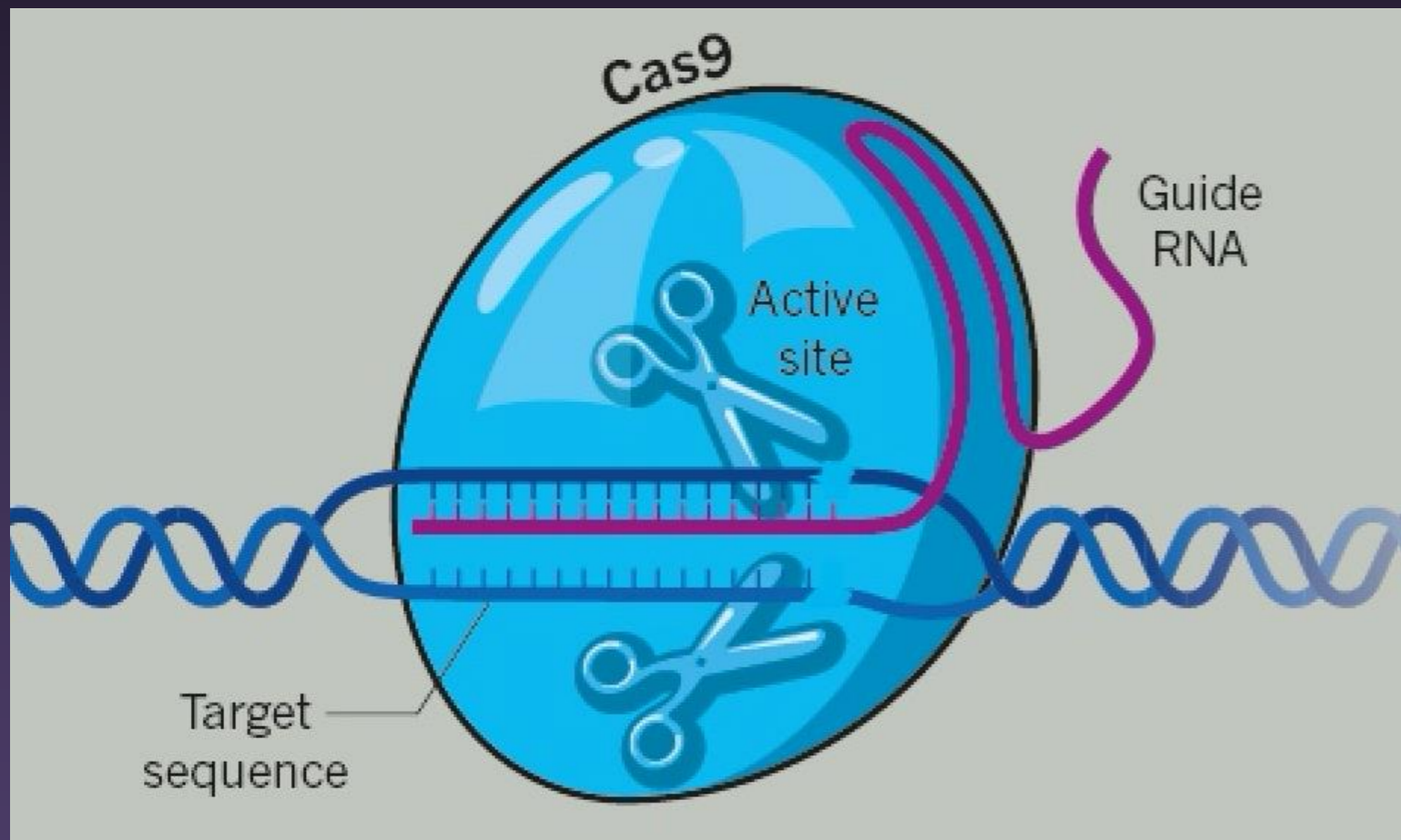# Living Wisely in an Era of Gene Editing at Will
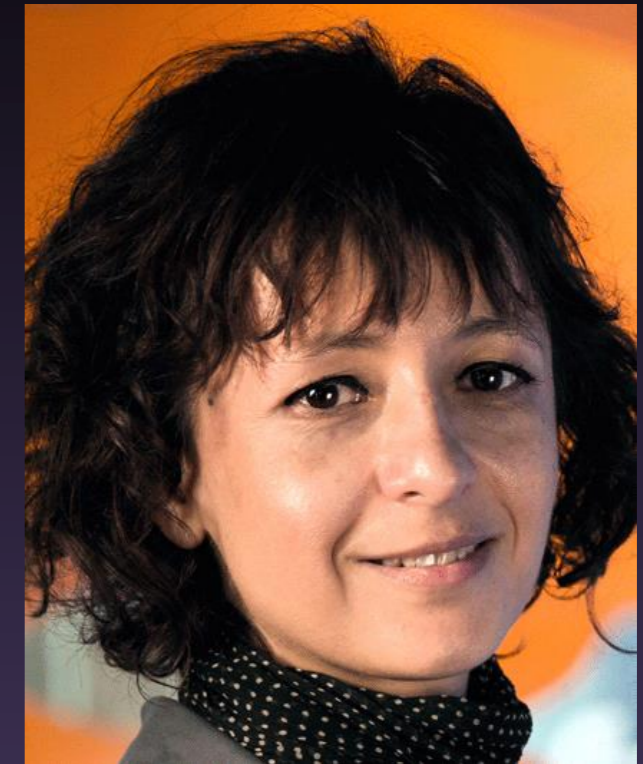
Anjeanette "AJ" Roberts, MACA, PhD
Research Scholar at Reasons to Believe

Bacterial Type II CRISPR-Cas9 System

Reported in 2012 as an RNA mediated gene-editing system. Programmable gRNA targets "specific" DNA sites for cleavage

Cheap, simple, fast, widely accessible; over 30,000 researchers modifying cells, plants, other organisms, and human embryos.
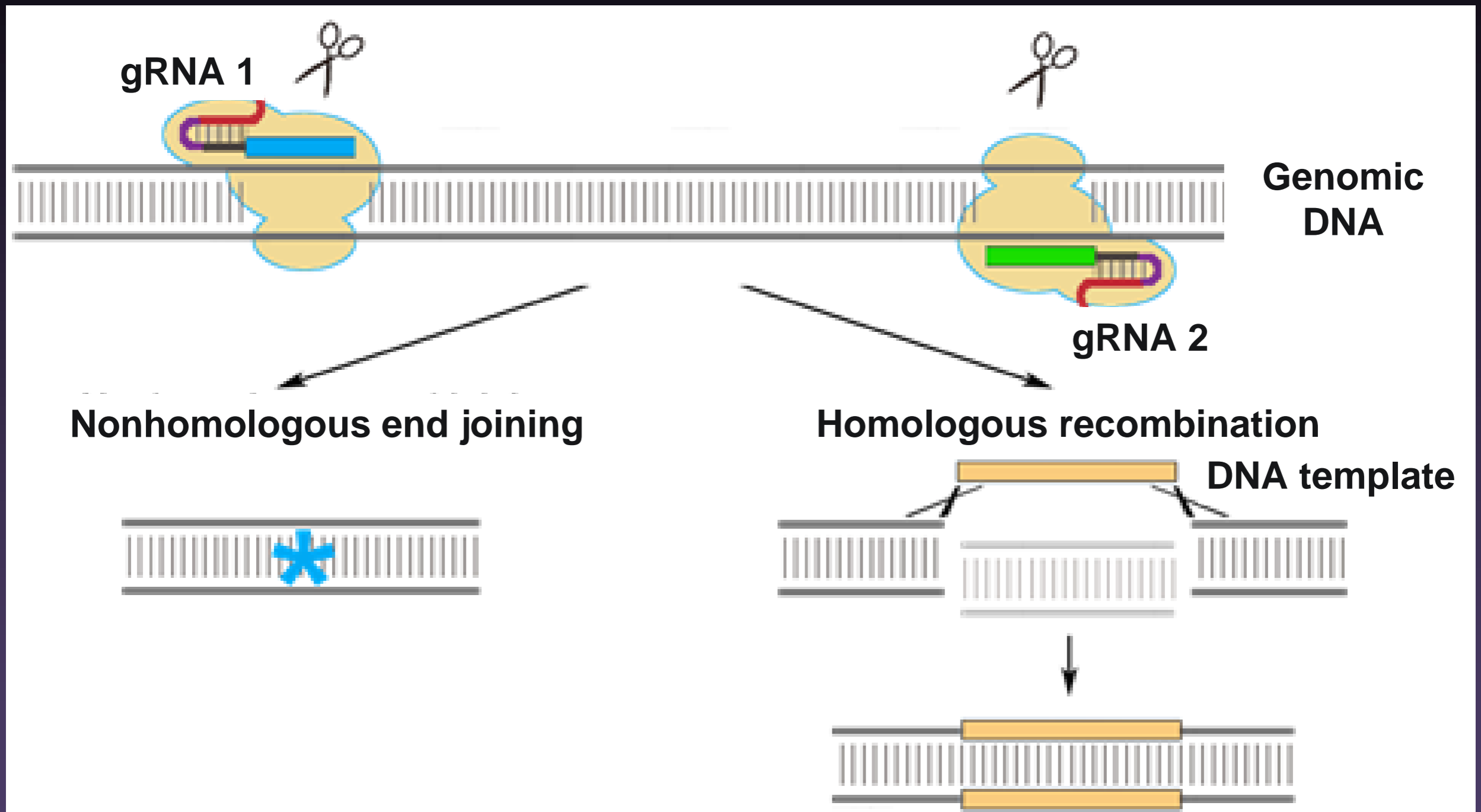

Emmanuelle Charpentier

Jennifer Doudna

http://embomolmed.embopress.org/content/7/4/363

http://alchetron.com/Jennifer-Doudna-509475-W

Editting Mechanisms

gRNA 1

Genomic DNA

gRNA 2

Nonhomologous end joining

Homologous recombination

DNA template

https://www.thebestgene.com/CRISPRInfoPage.do

- Deletion (indel): loss of function
- Deletion: gain of function

- Repair of alleles associated w/disease
- Introduction of new genes

Applications

In organisms (in the lab and into nature)

- Assess developmental & tissue specific regulation

- Build new designer organisms: bacteria, insects, plants/crops, animals

  - Expressing new or improved proteins

  - Disease resistant organisms

  - Produce disease models for research of enigmatic diseases

Applications
In humans (somatic and germline)

- Somatic cell modifications: useful in treating some diseases and disorders

- Genetic disorders/congenital diseases: e.g. Duchenne Muscular Dystrophy, hemophilia, sickle cell anemia, cystic fibrosis

- Cancers (sarcoma, myeloma, melanoma)

- Infectious diseases: HIV, Herpes, Zoster, Hepatitis B and C

Applications
In humans (somatic and germline)

- Non-viable and viable embryos

- Understand (ab)normal human development

- Molecular eugenics or designer babies

  - Expensive, not highly efficient (IVF)

  - Ethically questionable

  - No regulation in some countries

Concerns and Cautions
Is it wise to release genetically modified species into nature?

- Could be implemented to eradicate or modify disease vectors: mosquitos, ticks, etc.

- This leads to one of the biggest fears surrounding CRISPR: the creation of gene drives designed to spread through a population of organisms at an unnaturally fast rate.

Concerns and Cautions II

Is it wise to release genetically modified species into nature?

- Could result in creating a more vulnerable species

- One could relatively easily and intentionally (or unintentionally) engineer a species into extinction.

- DNA editing in the wild may have irreversible, unforeseen ecological outcomes.

  o Mosquitos

General Concerns

- High off-target hits disrupting unintended sites (0.1 to more than 60%)

- Challenge of efficient delivery to specific tissues/cells

- Modifications to Cas9 (3-4 amino acid substitutions) can increase specificity while maintaining efficiency -> claims of undetectable hits on off-site targets.

  - This is great for increasing specificity in lab experiments.

  - Concern over limits of detection

  - Are they really screening the entire human genome?

  - Not true for all gRNAs

Concerns

Hubris and the Human Genome

Despite 2004 reports of completion of the human (euchromatic) genome sequence (~3.1/6.2 Bbp)

- ~ 10% of the >3.1 Bbp human genome is absent from the human reference genome, either (un)sequenced/assembled

- Person-to-person variability ranges 10 to 50%

- ENCODE and continued research indicates ncRNAs are associated with >90% of Hu genome

- Many diseases are not monogenic and may be due to differences in regulatory ncRNAs.

Hubris and the Human Genome

Genome Reference Consortium – screenshot 24 July 2016

www.ncbi.nlm.nih.gov/projects/genome/assembly/grc/human/issues/

Hubris and the Human Genome

- Chromatin remodeling and differential expression of genes during development, in various tissues, and under varying physiological or environmental conditions makes the human organism extremely complex and dynamic on a genetic level.

- Germline modifications affect all future generations

- If evolutionary biologists are right and horizontal (lateral) gene transfer occurs in mammals/humans as in bacteria and simple eukaryotes, changes to somatic cells may have generational consequences

Concerns and Cautions

- In studies seeking to produce the most minimalistic cell, researchers have identified a bacterial cell that needs only 473 genes. But of these essential genes, 149 are of unknown function.

"We must be able to make informed choices in a climate of prevailing ignorance." ~ John Pollock

- Our confidence in what we know and what we can do needs to be tempered by the truth of how much more we still do not understand.

Overconfidence in our under-standing AND access to techniques that allow almost anyone to effect change is a little like allowing children to play with matches.

PLAYING WITH

It is not hard to imagine, nor farfetched to think, that someone could start some-thing that will not be able to be reversed and will not be able to be controlled. In this case, ignorance is not bliss.

Points for Ethical Consideration

- Naiveté, overconfidence (hubris), and accidents are not trivial issues.

- In some parts of Europe, genetic engineering is illegal outside of professional facilities. Extreme differences exist in government (public) verses private pursuits.

- Once we cross the line and modify the human genome in a germline cell it becomes part of the human gene pool affecting all future generations. Changes by a few affect generations w/o consent.

C. S. LEWIS

THE
ABOLITION
OF MAN

HOW EDUCATION DEVELOPS
MAN'S SENSE OF MORALITY
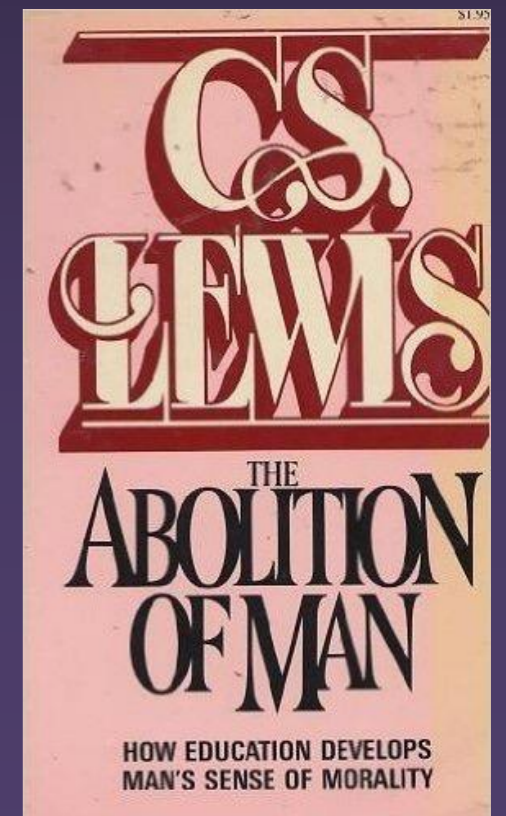
Faith Tempering Unrestrained Editing

- The Judeo-Christian Scriptures make it clear that as those made in the image of God we have been given dominion and stewardship of creation.

- We have also been given the model and command to serve one another in mitigating aspects of evil and human suffering.

- But Scripture also shows us that we fail to acknowledge God, choosing to abandon our honored yet humble place as steward-creatures to rather be like gods and manipulate the creation for our own desires

Faith Tempering Unrestrained Editing

- So how do we balance our rightful place as stewards endowed with the image of God without over-reaching and presuming god-like privileges?

- By humbly acknowledging the Creator and realizing that we are never given dominion over one another, just Jesus' command to love and serve one another.

Now, while there are still many technical challenges to overcome, is the time to discuss policy and regulatory issues.

Needed: Good Stewards of Genome Editing

7/21/2016
Genes & DNA

A Bacterial Protein (CRISPR/Cas9) Will Change Life on Earth

7/14/2016
Science Technology

Pigs Thinking Like Humans: Reality or Hogwash?
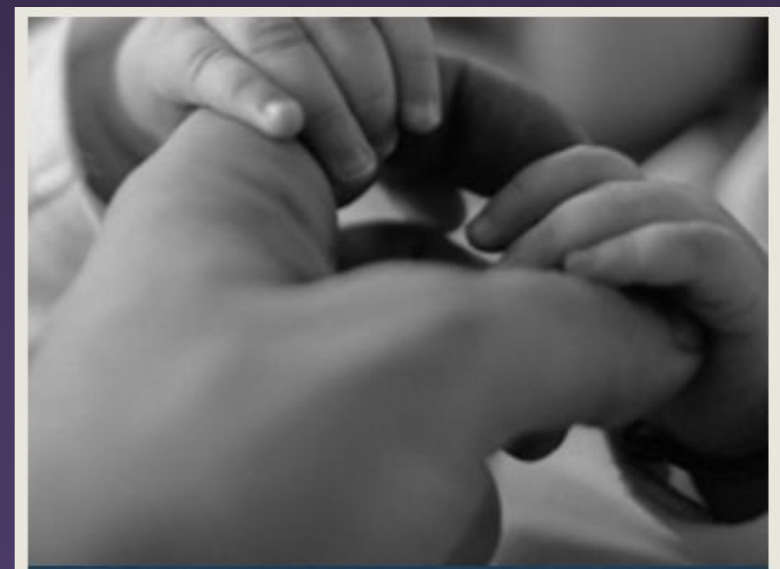
7/7/2016
Human Uniqueness

Imago Dei and the Ethics of Gene Modification

2/25/2016
Stem Cells/Cloning

More can be found at Reasons to Believe website: www.reasons.org

Exploring the Human Genome: Can We Make Predictions from Infancy?

9/14/2015

Not for showing… reference only.

Guidelines issued by an International Summit on Human Gene Editing

- 1-3 December 2015 - Recommendations or guidelines

    - Basic and preclinical research is needed and should proceed according to appropriate legal and ethical rules and oversight.

    - Any modifications of human germline cells or embryos should not be used to establish a pregnancy.

    - Clinical use of modified human *somatic* cells for treating disease should be appropriately and rigorously evaluated within existing and evolving regulatory frameworks for gene therapy.

    - Clinical use of modified germline cells: risks of inaccurate editing, incomplete editing (mosaicism), difficulty in predicting harmful effects, obligation to consider implications for the individual and future generations, once modifications are introduced could be difficult (impossible) to remove, genetic enhancement could exacerbate social inequalities or be used coercively, purposefully altering human evolution.

Guidelines on Human Gene Editing

- Recommendations or guidelines (continued)

  - Germline editing:

    - should not occur unless or until relevant safety and efficacy issues have been resolved (not until the science is known and shown to be safe)

    - must be based on balancing risk, benefits, and alternatives

    - should not be done until there is broad societal consensus on the appropriateness of the proposed application

    - preformed only under appropriate regulatory oversight

    - should be revisited on a regular basis as the views of society change and scientific understanding is advanced - need for an ongoing international forum of not just scientists and clinicians

  - Discourage unacceptable activities while advancing human health and welfare