LESSON PLAN & TEACHER'S GUIDE

Engineering the World Around Us:

Genome editing and the environment

Aim

How might genome editing be used to address the environmental issues we are facing?

Time

This lesson can be adjusted to fill 1-2 class periods.

Guiding questions

- What genetic technologies are being considered to address issues around food, animals, and the environment?
- What are the major hopes and concerns to weigh when thinking about genetics and preserving endangered animals?
- How might genome editing be an approach to solve problems related to food access, safety, and disease reduction?
- How can we as a society consider using genetic technologies when the risks and benefits are not yet fully understood?

Learning objectives

By the end of the lesson, students will be able to:

- Understand that scientific advances may allow scientists to genetically engineer the living world around us.
- Know that genome editing may hold promise for solving health and environmental problems, but presents uncertainty from ethical and ecological perspectives.
- Be able to debate how we allocate scientific resources.
- Explain the major points of excitement and hesitation related to genome editing and the environment.

Materials

Projector or Smartboard, laptop, handouts.

Standards alignment

Common Core Standards

CCSS.ELA-LITERACY.RST.11-12.1 Cite specific textual evidence to support analysis of science and technical texts, attending to important distinctions the author makes and to any gaps or inconsistencies in the account.

CCSS.ELA-LITERACY.RST.11-12.2. Determine the central ideas or conclusions of a text; summarize complex concepts, processes, or information presented in a text by paraphrasing them in simpler but still accurate terms.

CCSS.ELA-LITERACY.RST.11-12.4 Determine the meaning of symbols, key terms, and other domain-specific words and phrases as they are used in a specific scientific or technical context relevant to grades 11-12 texts and topics.

CCSS.ELA-LITERACY.RST.11-12.7. Integrate and evaluate multiple sources of information presented in diverse formats and media (e.g., quantitative data, video, multimedia) in order to address a question or solve a problem.

Next Generation Science Standards

This pgEd lesson integrates some of the NGSS practices and cross cutting concepts associated with the following disciplinary core ideas. The relevant portion of each disciplinary core idea is written out below.

HS-LS3: Inheritance and Variation of Traits

LS3.A:

- The instructions for forming species' characteristics are carried in DNA.

HS-LS4: Biological Evolution: Unity and Diversity

LS4.C:

- Changes in the physical environment, whether naturally occurring or human induced, have thus contributed to the expansion of some species, the emergence of new distinct species as populations diverge under different conditions, and the decline–and sometimes the extinction–of some species.

LS4.D:

- Humans depend on the living world for the resources and other benefits provided by biodiversity. But human activity is also having adverse impacts on biodiversity through overpopulation, overexploitation, habitat destruction, pollution, introduction of invasive species, and climate change. Thus sustaining biodiversity so that ecosystem functioning and productivity are maintained is essential to supporting and enhancing life on Earth. Sustaining biodiversity also aids humanity by preserving landscapes of recreational or inspirational value.

Background information

Advances in genetic technologies are accompanied by questions for society about if, where, and how to apply these tools. A new generation of genetic tools has opened a pathway to engineer the world around us to potentially improve human health, agriculture, and the environment. However, not everyone agrees these applications would necessarily be a benefit, while others worry about unintended consequences of these ecosystem-changing actions. In this lesson plan, we focus on three potential uses of these tools with the goal of (1) reducing toxicity of cassava, (2) modifying disease-carrying insects to preserve Hawaiian honeycreepers, an endangered species of bird, and (3) reviving an extinct animal - the woolly mammoth - to possibly aid in preventing permafrost thawing. These examples draw from current scientific research and environmental issues at the forefront of genetics and society. Each scenario raises a variety of complex ethical, environmental, and scientific conundrums for students to consider.

This interdisciplinary lesson requires students to consider perspectives from several fields (e.g., ecology, history, agriculture and genetics) to the issues presented. Students are encouraged to take a seat at the table and dive into debates about genetics, ecology, land use, and how to allocate scientific and environmental resources. This lesson is structured to allow discussion and debate at the start and end of each of the three case studies, requiring students to put themselves in the position of various stakeholders in each scenario.

A new generation of genetic tools

In the past decades, scientists have developed a range of genetic technologies that can change the characteristics of an organism by deliberately altering its genetic material. These approaches are collectively known as "genetic engineering". This lesson plan focuses on a type of genetic engineering called "genome editing", which can be used to make specific and targeted changes to an organism's DNA. One of the genome editing techniques that has generated the most excitement, due to its efficiency and ease of use, is called "CRISPR" ("clustered regularly interspaced short palindromic repeats"). Originally discovered in bacteria (see slide 3 sidebar), CRISPR is now being used as a tool with applications in many areas, including medicine, agriculture, and the environment. For more on the use of CRISPR and related genome editing tools for the purposes of improving human health, please see pgEd's lesson plan on "Genome editing and CRISPR" and this article on recent developments in research and clinical trials. In this lesson, students will explore three applications of genome editing to address environmental issues.

Case studies

Reducing toxicity of cassava

The first case study examines the possibility of genetically modifying cassava, an important food crop that is able to grow in poor soil conditions and can tolerate drought. Already a staple crop for many people around the world, cassava is anticipated to play an increasingly important role as a global food source as temperatures increase worldwide. When cassava grows under drought conditions, however, the levels of a toxin that the plant naturally produces increase. In high concentration, this toxin can cause permanent paralysis in humans. These toxic effects can be avoided by soaking the cassava in water for a couple of days and by eating the cassava as part of a protein-rich diet. However, in certain regions of the world, protein and water are scarce resources. Scientists, in consultation with other stakeholders, are considering the possibility of altering the DNA of cassava plants, so the plants produce less toxin. The goal is to create a plant that is safer for the millions of people who rely on cassava as a food source. Students are asked to weigh the risks and benefits of this case study as a narrow lens on the broader discussion about genetic modification of food crops. Ahead of this lesson we recommend teachers read: "Poverty Plus a Poisonous Plant Blamed for Paralysis in Rural Africa" by Amy Maxmen, *National Public Radio.*

Preserving endangered bird species – Hawaiian honeycreepers

The second case study focuses on a genetic intervention being considered to protect a species of Hawaiian birds, known as honeycreepers, that are on the verge of extinction, in part due to avian malaria. Hawaiian honeycreepers have no natural defenses against avian malaria, which is transmitted via mosquito bites. To escape the threat of disease, honeycreepers are forced to live at higher altitudes where the temperatures are too low for the mosquitoes to survive. However, to gather food, the birds have to travel into the valleys, where they are at risk of malaria infection. As average annual temperatures increase in Hawaii, the mosquitoes are able to survive at higher and higher altitudes. This is shrinking the honeycreepers habitat even further and bringing these birds to the brink of extinction. Since the early 2000s, the Hawaiian honeycreeper population has fallen an average of 68% (in their core habitat) to 94% (on the periphery of their living area). One idea to prevent the extinction of these birds is to reduce or eliminate avian malaria by editing the mosquito DNA. The idea is to introduce a genetic change into the DNA of the mosquitoes that would make them less able or unable to transmit avian malaria to the birds. This genetic change would be rapidly spread through the Hawaiian mosquito population by using a specialized technique called a "gene drive", which increases the likelihood that the genetic change is passed from one generation to the next. Students are asked to consider the risks and benefits of this case study, which presents a focus in the broader discussion about the various possible applications of gene drives.

> Students are more likely to be familiar with the human form of malaria, which causes fever, chills, vomiting, and, in some cases, death. The human malaria parasite, which is transmitted via mosquito bites, infects 210 million people annually and over 440,000 people die from malaria each year. Like humans, birds and other animals are vulnerable to related parasites.

Reviving extinct species: Could de-extinction of the woolly mammoth help to preserve the permafrost?

The final case study looks at the intersection of genetic technology, environmental issues, and de-extinction (the process of reviving an extinct species or creating an organism that resembles an extinct species). Students are asked to consider de-extinction of the woolly mammoth, often cited as one of the most compelling candidates for de-extinction. Woolly mammoth displays in natural history museums, with their unique features that symbolize the Ice Age (Pleistocene), have long sparked the imagination. One of the reasons woolly mammoths are being considered for de-extinction is the possible role they could play in slowing the thawing of permafrost – a frozen layer of soil. As average global temperatures rise, the thawing permafrost releases greenhouse gases into the environment, which further accelerate the increase in temperatures. To examine a strategy for preserving the permafrost, researchers in the Siberian region of Russia have gathered a number of large grazing animals (e.g., elk, musk oxen, and reindeer) in an effort to recreate the grasslands that covered this region thousands of years ago. The idea is that this grassland biome would reflect more sunlight during the summer and allow the winter cold to penetrate the ground further. Early reports indicate that this approach is lowering the temperature of the ground, which in turn can slow thawing of the permafrost and keep more of the greenhouse gases trapped in the frozen soil. To expand this idea of combatting permafrost thawing to a global scale, large herds of big grazers would be needed. One possibility that has been suggested is using genetic technologies for "de-extinction" of the woolly mammoth, which was a key part of this landscape before going extinct ~10,000 years ago. This project would require a mix of high-tech and low-tech solutions and highlights some of the moral and ecological dilemmas in the field. As students examine this case study, they will explore these multiple angles. Additional reading for teachers on this includes: "Born to rewild" by Eli Kintisch, *Science*. "De-extinction debate: Should extinct species be revived?" by Liza Gross, *WQED Science.* Also "Can Bringing Back Mammoths Help Stop Climate Change?" by Paul Mann, *The Smithsonian.*

In the classroom

Through in-class discussions, students will consider (1) how to weigh the risks and benefits of applying genome editing to environmental issues and (2) how to make sure all the stakeholders have a seat at the table. Class discussions will be informed by the advance homework reading and the slideshow. In addition, students should all receive a copy of the vocabulary list (page 26) ahead of the lesson presentation. As the slides are shown, students will pause at the start and end of each of the three case studies to discuss the major technical, ethical and ecological questions, giving special attention to the possible impacts on communities and how best to engage those most likely to be affected. By working through the questions, students will see the need for interdisciplinary approaches to the complex ecological questions posed in this lesson. Many of these issues – all part of the broader conversation about the safe and equitable use of biotechnology – require a multitude of voices, perspectives and expertise. This lesson, like all of pgEd's materials, emphasizes dialogue based on information, and asks students to draw their own conclusions. We recognize these topics can be highly controversial; we are not seeking to persuade students, but instead to enable them to form their own viewpoints, based on information from reliable sources.

Outline of resources and activities in this lesson

Activities

Three "Do Now" exercises (15 minutes total, integrated into slide show), slideshow (30 minutes), classroom discussion (15 minutes).

Engineering the World Around Us: Genome editing and the environment

Part 1: OVERVIEW FOR STUDENTS

Reading for students:

In advance of the lesson, ask students to read the following article that explores some of the major scientific and social issues at the intersection of genetic technologies and the environment. It covers only the third case study, but will provide some foundation for all of the examples.

"5 reasons to bring back extinct animals, and 5 reasons not to," by Breanna Draxler, *Discover Magazine*.

Vocabulary List:

Students may be unfamiliar with some terms in this lesson. The vocabulary list on page 26 could be assigned as homework ahead of the lesson, used as a guide during the presentation, or modified as needed.

Engineering the World Around Us: Genome editing and the environment

Part 2: DO NOW & SLIDESHOW (45 minutes)

The slideshow is located on the pgEd website along with this lesson, and accompanying explanatory notes for the slideshow are provided below.

Slide 2

This lesson plan explores the use of genome editing in tackling a variety of ecological problems. Students will be given the chance to consider the application of genetic technologies in three different situations: (1) agriculture, (2) insect-borne disease, and (3) de-extinction and permafrost preservation. The lesson plan is divided into three sections, one for each case study. Each case study includes its own "Do Now" and discussion questions.

Slide 3

What is genome editing? In the past decades, scientists have developed a range of genetic technologies that can change the characteristics of an organism by deliberately modifying its genetic material. These approaches are collectively known as "genetic engineering". One type of genetic engineering is the introduction of a gene into an organism to give it a new trait - for example, making bacteria that produce human insulin that can then be used as medicine.

This lesson plan focuses on "genome editing", a type of genetic engineering, used for making specific, targeted changes to an organism's own DNA. One of the genome editing techniques that has generated the most excitement, due to its efficiency and ease of use, is called "CRISPR" ("clustered regularly interspaced short palindromic repeats"). Originally discovered in bacteria (see sidebar), CRISPR is now being used as a tool with applications in many areas, including medicine, agriculture, and the environment. For example, scientists are investigating the use of

CRISPR - a tool found in nature. Scientists originally discovered CRISPR in bacteria, which use CRISPR as an "immune system" that protects the bacteria against viruses. After a bacterial cell is infected by a virus for the first time, it stores a little piece of the viral DNA as a molecular "mug shot". CRISPR uses this "mug shot" to recognize the virus in future infection events and then marks the viral DNA for destruction. This allows the bacteria to fight off the infection. Recognizing CRISPR's specificity and efficiency in making changes to DNA, some scientists decided to try and use the CRISPR system as a genome editing tool. Over the years, researchers have been able to use CRISPR in a huge variety of living things to make specific changes to their DNA.

CRISPR to "correct" genetic variants that cause diseases, with the goal of curing genetic diseases such as hemophilia, cystic fibrosis, and sickle cell disease. (For more on the use of CRISPR and related genome editing tools, please see pgEd's lesson plan on "Genome editing and CRISPR" and this article on recent developments in research and clinical trials.) CRISPR might also be used in other organisms (not humans) to address ecological issues impacting human health and safety.

Case Study 1: How might genome editing improve health and nutrition for people who rely on the cassava plant?

Slide 4

Do Now. Students will discuss the following scenario, which asks them to consider the application of genome editing to an urgent health issue. The aim of the discussion is to highlight some of the complex environmental and economic issues, as well as the role that scientists might/should play in collaborating with the people that are impacted by their work.

> You live in a rural village and your relatives are suffering from Konzo, a disease that causes paralysis. You rely on a plant called cassava as your main source of food. Cassava naturally produces a toxin. At high concentration, this toxin can make people sick with Konzo. However, soaking the cassava in water for a couple of days before eating it prevents this problem.
>
> Scientists have proposed to genetically alter the cassava plant to make it less dangerous. You wonder whether providing a clean source of water, such as a well, to your village could be a better solution. What are the questions you have for the scientists about their plan?

Student answers will vary. For example, students may ask: "What if the genetic alteration makes cassava less safe?", "Will the alteration of the cassava plant truly fix all the Konzo-related problems that the villagers are facing?", and "Will people who are already poor be asked to pay for the altered cassava plant seeds?".

Slide 5

Cassava is an important crop for over 800 million people worldwide. Cassava is a high-calorie food that can grow in nutrient-poor soil and tolerate drought. It is invaluable for populations in Asia and South America, as well as Africa, where it is eaten regularly by up to 40% of the population. Increasingly, the use of cassava is gaining popularity across the world - for example to make the "bubbles" in bubble tea.

Slide 6

Depending on how it is grown and prepared, cassava can cause a disease called Konzo. Cassava plants naturally produce a toxin that can cause Konzo, a disease that leads to paralysis and can potentially be deadly (see sidebar). This is particularly problematic when cassava is grown in arid conditions. Still, there are different approaches for avoiding Konzo. First, soaking cassava in water for several days reduces the plant's toxicity. Second, eating a protein-rich diet can help the body to break down the toxin more effectively. While these solutions may appear to be relatively simple, their implementation can be difficult due to systemic and historical barriers. In parts of Africa where Konzo is prevalent, European colonialism has left behind extreme poverty. Access to water and protein-rich foods is scarce, and people who are affected by paralysis are often not able to make the trip to the nearest river or well to collect the water needed for soaking the cassava. Moreover, waiting several days to soak cassava is not possible for people who are urgently hungry. This is why Konzo is considered a disease of poverty. Furthermore, Konzo maintains the cycle of poverty, as people with the disease lose the ability to work and collect water. We recommend teachers read: "Poverty Plus a Poisonous Plant Blamed for Paralysis in Rural Africa" by Amy Maxmen, *National Public Radio.*

Konzo is ravaging communities that rely on cassava as their main food source. When cassava grows in parched soil, it produces higher levels of a chemical called cyanogenic glucoside. This chemical can be dangerous to people because, when eaten, it is converted to a toxin called cyanide. Cyanide causes Konzo, which can lead to irreversible paralysis of the legs, organ damage, and potentially death. Cases of Konzo have officially been reported in Angola, Cameroon, the Central African Republic, the Democratic Republic of Congo (DRC), Mozambique, Tanzania, and more recently Zambia. The true number of people suffering from Konzo is difficult to obtain, as most people have no access to health care, and public health data collection is difficult. However, estimates suggest there are hundreds of thousands Konzo patients worldwide.

How might genome editing help improve health and reduce disease for the people who rely on cassava?

Slide 7

Editing the cassava's DNA could lower the plant's toxicity. Two genes are responsible for the toxicity of cassava. Scientists have proposed using a genome editing technique called CRISPR to make specific changes to these genes with the goal of reducing the plant's toxicity. One major advantage of using CRISPR to introduce genetic changes is that it is much faster than traditional breeding methods. Furthermore, CRISPR technologies can be applied to local varieties of the cassava plant, thus maintaining characteristics of the plant that make it well-suited to the region where it will be grown. The researchers are hoping to improve on current CRISPR methods, which introduce some foreign DNA into the target organism, so they are developing a new approach called 'DNA-free CRISPR'. Using the DNA-free CRISPR method, the cassava would only carry its own DNA, which would sidestep regulatory hurdles surrounding GMOs in certain countries (see sidebar) and allow the product to become available for use much faster. This project is in the early stages in 2020.

Genetically Modified Organisms (GMOs) and DNA-free CRISPR. Different countries and organizations define GMOs slightly differently. In general, a GMO is an organism whose DNA has been altered using genetic technologies in a way that does not occur naturally through breeding or mutation. The CRISPR technique often involves introducing foreign DNA (i.e., from bacteria), and therefore, CRISPR-edited organisms are regulated as GMOs in some parts of the world (e.g., Europe). If successful, the DNA-free CRISPR method, described in the cassava case study, would make changes to the organism's own DNA without introducing foreign DNA. In other words, you would not be able to distinguish between (1) the genome of an organism carrying a genetic change brought about by DNA-free CRISPR and (2) the genome of an organism carrying that same genetic change through natural processes of mutation and breeding.

Slide 8

Scientists and communities are considering the risks and benefits of using genome editing to lower cassava toxicity. Using genome editing to lower the cassava's toxicity could have unintended consequences. For example, the cassava's toxicity appears to be correlated with its ability to tolerate drought, as the drier the circumstances are, the more toxin the plant produces. Could the edits negatively affect the plant's drought-tolerance, a beneficial trait for growing in many regions across the globe? Furthermore, cassava's toxicity is thought to provide a defense against insects. Could the edited cassava plants require farmers to use pesticides in order to grow their crop? Lastly, what will the economic implications be? Who will own the plants as well as the seeds of the edited cassava? Will farmers be able to afford this new crop?

As students consider the use of genome editing to make cassava safer and more sustainable, what are other possible approaches? The DNA sequence of the cassava plant, published in 2016, allows scientists to better understand the genes involved in toxicity. Could this provide a roadmap for using conventional breeding methods to choose cassava varieties with specific beneficial genetic traits? Stepping back further, Konzo is a disease of poverty with lack of access to water and protein-rich food being the main contributors. Should disease prevention efforts focus on a genetic solution? Or should the focus be on breaking the cycle of poverty that is at the root? Might a combination of approaches be the best way forward?

Case Study 2: Can genetic tools help protect a species of bird from becoming extinct?

Slide 9

Slide 9: Do Now. Students will discuss the following scenario in which they imagine themselves as a scientist consulting with a community to explore a controversial idea: using genetically modified mosquitoes to combat malaria infections.

> You are a scientist. You are visiting an island where a species of birds is in danger of extinction because they suffer from a disease that is given to them through mosquito bites. You think that the best way to rescue the birds is to use genome editing to wipe out the mosquitoes on the island.
>
> You know that the people who live on the island need to be partners in this project. How do you establish such a partnership? What is the information you need to gather from them, and what is the most important information for you to share? What are you looking to learn?

Student answers will vary. For example, ideas to establish a partnership may include: (1) setting up a lab in the affected area, (2) hiring staff from the island who can provide local expertise, and (3) hosting a series of meetings to have dialogue with the general public. Engaging local communities is important from an ethical point of view to build trust between scientists and the public and to ensure informed consent for projects going forward in their environment. Furthermore, community partnerships can bring to the table diverse types of knowledge [e.g., local entomology (study of insects) and epidemiology as well as local social structures and politics], which can lead to the development of better strategies for addressing the problem. Examples of on-going gene drive (see slide 12) projects - how they have interacted with and impacted local communities - can be found in this article.

Slide 10

Hawaiian honeycreepers - colorful birds that are culturally important to the Indigenous people of Hawaii and beloved by many people all over the world - are at risk of extinction. The people of Hawaii and others are seeking a solution to protect Hawaiian honeycreepers from going extinct. Honeycreepers are an important part of the ecosystem on the Hawaiian islands and hold historical and cultural importance as well. The Indigenous peoples of Hawaii have used feathers from honeycreepers to make ceremonial cloaks called *ahu'ula* and helmets known as *mahiole*.

However, the population of Hawaiian honeycreepers is dwindling, in part because their habitat is being destroyed by human activity. Another threat to the survival of these birds are mosquitoes that carry an avian form of malaria. Avian malaria is a bird disease caused by parasites that spread to the honeycreepers through mosquito bites. Hawaiian honeycreepers are specifically adapted for life in Hawaii, where mosquitoes did not exist until the 1800s. As such, these species of honeycreepers have no natural defense against avian malaria. Since the early 2000s, the population of Hawaiian honeycreepers has fallen an average of 68% (in their main living area) to 94% (at the outer edges of their habitat).

Slide 11

Environmental changes are increasing the threat of avian malaria to Hawaiian honeycreepers, driving these birds closer to extinction. Since the introduction of mosquitoes and avian malaria to the Hawaiian islands in the 1800s, honeycreepers have been forced to live at higher altitudes where the temperatures are too low for the mosquitoes to survive. However, to gather food, the birds have to travel into the valleys, where they are at risk of malaria infection. With average annual temperatures increasing in Hawaii, mosquitoes are now able to survive at higher and higher altitudes. This is shrinking the honeycreepers' habitat even further and bringing them to the brink of extinction.

How can Hawaiian honeycreepers be preserved? Habitat restoration is part of the solution, but other approaches may be needed as well. One of the possibilities being considered is the use of genome editing. (For more on this as well as other ideas being explored, see this summary of a meeting between scientists, policymakers and community stakeholders: "To Restore a Mosquito Free Hawaii".) How might genome editing help prevent the Hawaiian honeycreepers from going extinct?

Slide 12

This slide is animated to introduce this concept to students in three steps.

a) Editing the DNA of mosquitoes could be used to prevent them from infecting Hawaiian honeycreepers with avian malaria. Two approaches have been suggested: (1) to introduce a genetic trait in the mosquitoes that would greatly reduce and possibly fully eliminate the mosquito population, and (2) to introduce a genetic trait in the mosquitoes that would make them unable to carry the avian malaria parasite, so they can no longer transmit the parasite to the honeycreepers.

b) The challenge of introducing these genetic traits into the population of mosquitoes living in the wild is that, under normal sexual reproduction, the trait will only be passed to ~50% of the next generation. This means that the trait would not spread very widely in the Hawaiian mosquito population.

c) To address this problem, a genetic technology known as a "gene drive" could be used to increase the likelihood that a genetic trait will be passed to the next generation. In this way, a gene drive allows for a specific trait to quickly spread in a population. With the advent of CRISPR, the gene drive approach has become a practical reality. For a more detailed explanation of gene drives, see slide 21.

Slide 13

Multiple viewpoints and issues need to be considered as scientists and communities weigh the risks and benefits of using genetically-engineered mosquitoes to prevent the extinction of the Hawaiian honeycreeper. Using genome editing could help to halt the rapid decline of the Hawaiian honeycreeper population; however, there are several aspects of this approach that need further consideration.

1. Which genetic trait would be the most beneficial to introduce into the wild mosquito population: (1) one that would greatly reduce and possibly eliminate the mosquito populations, or (2) one that would prevent the mosquitoes from carrying the avian malaria parasite? Given that the mosquitoes are not native to Hawaii, wiping them out could be considered as a "reset button". However, what if the mosquitoes have become an integral part of the ecosystem during the ~200 years that they have been present in Hawaii? What if other species are now dependent on the mosquitoes for their survival? In this light, preventing mosquitoes from carrying the avian malaria parasite instead of possibly wiping out the mosquito population might be preferable. However, these mosquitoes are known to be able to carry a number of diseases that can affect animals as well as humans (e.g., Western equine encephalitis and West Nile virus). Preventing

the mosquitoes from carrying avian malaria parasites will not prevent them from transmitting these other diseases.

2. Could the use of a gene drive to rapidly spread the genetic trait of interest through the Hawaiian mosquito population have far-reaching effects (e.g., spreading the genetic trait beyond the target population)? For example, a gene drive targeted to wipe out the local mosquito population on Hawaii could end up driving this species to extinction across the globe. Scientists are trying to design built-in "safety mechanisms" to limit the effects of the gene drive to the target species or even to reverse the gene drive if unintended consequences arise.

3. As students consider the use of genome editing to combat avian malaria in Hawaii, what are other possible approaches? In urban areas, methods such as (a) limiting the sources of standing water where mosquitoes breed (called "source reduction"); (b) targeted application of insecticides; and (c) using devices that attract and kill female mosquitoes (called "lethal ovitraps") have proven to be successful in controlling mosquito populations. However, these approaches are challenging and costly to apply in the rural and forested areas of Hawaii where the honeycreepers live. Another possible method is the Sterile Insect Technique (SIT), which involves releasing male mosquitoes into the wild that are either infertile or produce offspring that do not survive long enough to reproduce. This method can be successful in eliminating a local mosquito population, but may not work if the released males do not compete effectively with the wild male mosquitoes for breeding with wild females. SIT is considered "self-limiting" as the trait that causes the mosquitoes to be infertile (or produce offspring with limited survival) dies with them.

As described above, each approach brings risks for unintended consequences as well as potential benefits. For example, eliminating the mosquitoes could save Hawaiian honeycreepers from going extinct, but, at the same time, another species that has come to depend on these mosquitoes for food could be pushed to the brink of extinction. What are reasonable risks and worthwhile benefits? These are questions that require strong partnerships with communities to gather diverse expertise (such as knowledge of local entomology and epidemiology as well as local social structures and politics) for developing strategies to save the Hawaiian honeycreepers.

Case Study 3: Can preservation of the permafrost be helped by using genetic tools to create a cold-resistant "woolly" elephant?

Slide 14

Do Now. Students are asked to discuss the following scenario about the use of genome editing to bring back extinct species.

> You are a farmer in Siberia, tending animals and vegetables on your land. You heard a team of scientists are hoping to bring the woolly mammoth back from extinction. These animals once roamed exactly where you live. How do you feel about this plan? What are your questions about it?

When students are asked to imagine this project from the perspective of people living on the land that the revived animals would occupy, students are likely to express skepticism or excitement. Some might express concern about the possibility that Indigenous people living in areas of the world where woolly mammoths used to roam (e.g., Siberia, Alaska, Canada, and Greenland) are not part of the decision-making process and stand to lose more and more of their land.

Slide 15

What is de-extinction? De-extinction is the process of reviving an extinct species or creating an organism that resembles an extinct species. Genome editing tools have made this a possibility that some people are interested to consider. The woolly mammoth is often cited as one of the most compelling candidates for de-extinction. In natural history museums, displays of woolly mammoths, with their unique features that symbolize the Ice Age (Pleistocene), have long sparked the imagination. One of the reasons mammoths are being considered for de-extinction is the potential role they could play in slowing the thawing of permafrost. The project to bring them back would require a mix of high-tech and low-tech solutions, and highlights some of the moral and ecological dilemmas in the field. We will explore this idea in the next couple of slides.

Slide 16

The thawing of permafrost has a big impact on our environment. The permafrost is a layer in the ground that remains below 32ºF (0ºC) for at least two years in a row. Permafrost is found in areas where the average temperature rarely gets above freezing - on land as well as below the ocean floor. Approximately 20% of the earth's land surface is permafrost, which is mainly found in tundra and taiga landscapes of the Northern Hemisphere (e.g., in Siberia, Alaska, Canada, and Greenland). As average global temperatures rise, the total area of permafrost is shrinking. Thawing of the permafrost is a particular concern because the frozen soil stores significant

Tundra and Taiga landscapes. Tundra landscapes are defined by the presence of permafrost and are often bare, as the frozen soil makes it difficult for roots to penetrate the ground. Only moss, lichen, and low-growing shrubs, are able to live there. The more southern taiga landscapes have a milder climate than the tundra and the permafrost is not continuous. Due to an increased growing season and a thicker layer of active soil on top of the permafrost, taiga landscapes are able to support the growth of trees in large forests.

amounts of carbon that, if released into the Earth's atmosphere, could have significant global impact. It is estimated that the permafrost contains twice as much carbon as is found in the Earth's atmosphere today. According to current models, as much as 5-15% of the carbon stored in permafrost soils will be released into the atmosphere by the end of this century. This carbon is released in the environment in the form of carbon dioxide and methane, two greenhouse gases that trap infrared radiation near the Earth's surface and therefore could accelerate the rise of average temperatures around the globe. In regions such as Alaska (where 85% of the land has permafrost), the infrastructure is increasingly affected as the solid frozen foundations disappear. Furthermore, permafrost is thought to contain frozen and preserved bacteria and viruses. The concern is that these pathogens might revive when the permafrost melts and could (re-)introduce diseases into the world for which we have no natural defenses or treatments.

Slide 17

How might thawing permafrost be slowed or even reversed? One idea to protect the thawing permafrost is to undertake a massive effort to return the current tundra and taiga landscapes to grassland biome that covered this region thousands of years ago. This would entail large herds of grazers (such as deer, horses, and bison) roaming the landscape to help establish and maintain the grasslands. During the short summer, grasslands keep the ground cooler as their light color reflects sunlight more effectively than the dark-colored shrubs and trees of the tundra and taiga, respectively. During winters, when snow covers the ground, the grazers in the grassland biome keep the ground cooler. In tundra and taiga biomes, the snow acts as a "blanket" that protects the ground below from the bitterly cold winter temperatures. By contrast, in grassy landscapes, grazing herds of herbivores disrupt this snow blanket by tamping down the snow and disturbing the snow cover as they look for food underneath. This compaction and removal of snow exposes the ground below to the cold winter air, which prevents the permafrost from thawing and may even expand this frozen layer, allowing for more carbon to be contained.

Slide 18

Pleistocene Park: a test case for restoring grasslands in Siberia. To test the idea that recreation of a grassland biome could prevent thawing of permafrost, researchers have gathered a number of grazing animals (such as elk, musk oxen, and reindeer) in a 5,000-acre reserve called the Pleistocene Park in Siberia. Preliminary data suggest that this effort lowers the ground's temperature and slows the thawing of the permafrost, keeping more of the greenhouse gases trapped in the frozen soil (more on these data at "Born to rewild" by Eli Kintisch, *Science*). While encouraging, this effect would need to be translated on a larger scale to combat the thawing of permafrost globally. Large herds of big grazers would be needed to disrupt the mossy tundra and forested taiga landscapes and recreate the grassland biome of the past. A key animal in this landscape was the woolly mammoth, which was largely extinct by the end of the Pleistocene epoch (roughly 10,000 years ago).

Could de-extinction of large woolly mammoth herds recreate the vast grassland biome and help to preserve the permafrost?

Slide 19

A leading strategy is using CRISPR to introduce some genetic traits from woolly mammoths into the DNA of its close relative, the Asian Elephant. In other words, the goal of the woolly mammoth de-extinction project is to create a cold-resistant "woolly" elephant, sometimes called a "mammophant". Though a number of technical hurdles remain, this project was able to get underway because of (1) the discovery of preserved mammoth DNA; (2) the existence of a close living relative of the mammoth, the Asian elephant; and (3) the availability of genome editing tools, such as CRISPR. By analyzing preserved (though highly degraded) mammoth DNA, scientists have been able to identify genes responsible for key traits that helped the mammoth to survive in cold climates. These traits include longer hair, more fat under the skin, and a circulatory system adapted to cold temperatures. Using CRISPR, researchers are introducing these traits into Asian elephant cells in a laboratory setting. This is possible because Asian elephants and woolly mammoths are closely related, sharing 99.96% of their DNA. To make a mammophant, hundreds or maybe thousands of genome edits are likely needed. As of 2020, CRISPR has been used to introduce some woolly mammoth traits in the DNA of Asian elephant cells, and this work is on-going.

> **If scientists are trying to bring back a woolly mammoth, why not a dinosaur?** De-extinction is not being applied to dinosaurs for technical reasons as well as many ethical and ecological concerns. This is a great opportunity to remind students that without preserved DNA and a currently living close relative that could serve as a surrogate, dreams of a T-Rex will continue to live on only in movies such as *Jurassic Park*. Students are often interested to hear that, from a genetic perspective, the closest living relative to ancient dinosaurs are modern birds – including such backyard favorites as the chicken. See the *Smithsonian's* "Dinosaurs' Living Descendants" for more.

Still, if this project is to go forward, several steps and challenges lie ahead. Once the Asian elephant cells have been edited (thus creating mammophant cells), these cells could then be used to generate a mammophant embryo. That embryo could then be transferred into the uterus of an Asian elephant, with the goal of starting a pregnancy that would give rise to a baby mammophant.

Slide 20

Multiple viewpoints and issues need to be considered as scientists and communities weigh the risks and benefits of using genome editing to make a cold-resistant Asian elephant. Beyond the technical challenges of creating a mammophant, described in slide 19, there are a number of ecological and ethical questions as well.

1. Is it acceptable to use the Asian elephant, an endangered species, in this project? There could be risks to an animal used as a surrogate to carry a mammophant pregnancy. Researchers working on this project have suggested the use of an artificial womb to eliminate the surrogacy issue; however, this technology is still in its infancy and could not yet support the development of a mammophant to term. Further concerns exist around the use of Asian elephants in raising the mammophants after they are born. Elephants are very social animals, and the social effects of this project on both the Asian elephants and the mammophants present many unknowns. For example, how would we ensure appropriate rearing and socialization of the mammophants that will provide them with the necessary skills to live in mammophant herds and survive cold climates, when the Asian elephants cannot continuously live there? How would the Asian elephant herd be affected when baby mammophants are introduced? Or when those mammophants are subsequently removed from the herd to inhabit tundra and taiga landscapes?

However, this project may also yield unexpected benefits for preserving the Asian elephant. When scientists working on the mammoth de-extinction project heard about a strain of herpes virus that is deadly for Asian elephant calves, both in captivity and in the wild, they began an effort that may lead to a cure for this disease.

2. Where would the newly de-extincted animals live? There are people currently living in the tundra and taiga landscapes where the mammophants are suggested to be introduced. How will their voices be weighed in this discussion – specifically, the voices of Indigenous peoples in these regions who, throughout history, have seen their claims to land being stripped away?

3. If mammophants are introduced, how would they impact the local ecosystems? There are some examples of success when an animal is reintroduced to its habitat (see National Science Foundation's Understanding the ecological role of wolves in Yellowstone National Park). However, given that mammoths have been extinct for several thousands of years, it is hard to know what impact they might have. Should we proceed when the consequences of introducing newly revived species into wild ecosystems might have outcomes that are hard to predict?

4. Do we have an obligation to try any and all efforts to prevent thawing of the permafrost even if those efforts are expensive, have a high risk of failure, could disrupt existing ecosystems and biomes, or require the use of endangered animals? How do we balance the risks of proceeding with this project with the risks of not proceeding, if there is a chance, however small, that the mammophants could play a role in preventing the release of carbon from the permafrost?

Slide 21

Science supplement - Gene Drives. Mosquitoes carry two sets of chromosomes, one inherited from each of their biological parents. This means that, for each gene, there are two copies (or alleles) present. These alleles do not necessarily have the same sequence, though it is possible that they do. When a mosquito reproduces, it will pass one copy of each of its genes to its offspring. In other words, there is a 50% chance that a certain allele will be passed on to the next generation. Under these conditions (left panel), spreading a specific genetic trait of interest (left panel: red) through the entire mosquito population is inefficient.

Gene drive mechanisms increase the likelihood that a specific genetic trait will be passed to the next generation (right panel). Rather than a 50% chance, the trait (right panel: red) could be inherited by most, if not all, of the offspring, thus allowing it to spread through a population at a much faster rate. The way this is achieved is by introducing the genetic sequence that codes for a DNA-cutting enzyme (right panel: blue) together with the genetic trait of interest (right panel: red). When the DNA-cutting enzyme is produced (right panel: scissors), it will cut the chromosome that does not carry the trait of interest. The cell's repair mechanism will fix this break by using the corresponding sequence from the intact chromosome (which includes the genetic trait of interest and the sequence of the DNA-cutting enzyme; right panel: red + blue) as a template. In other words, the DNA-cutting enzyme (right panel: blue) "drives" the inheritance of the genetic trait of interest (right panel: red) as well as itself (right panel: blue) so that, in the ideal case, 100% of the offspring receive the genetic trait of interest.

In the honeycreeper case study, two genome editing approaches were proposed to combat avian malaria: (1) one that would prevent the mosquitoes from carrying the avian malaria parasite and (2) one that would greatly reduce or possibly eliminate the mosquito populations. The first approach can be achieved by introducing genes that encode antibodies that attack the malaria parasite. By introducing these genes (represented by red in right panel) together with a DNA-cutting enzyme (represented by blue in right panel), this genetic trait can efficiently be spread through the wild mosquito population in Hawaii. This would result in mosquitoes that, when infected with malaria, have an immune system that attacks the parasite so they can no longer transmit the parasite to the honeycreepers.

The second approach involves using a gene drive (represented by blue in right panel) to spread an edited version of a gene called *doublesex* through the mosquito population (represented by red in right panel). Female mosquitoes that receive two copies of the altered *doublesex* gene from their biological parents are unable to reproduce. When the altered *doublesex* gene drive is initially introduced in the population, most mosquitoes will have at least one parent that does not carry the altered *doublesex* gene. As the gene drive spreads further through the population, the number of mosquitoes that receive an altered *doublesex* gene from both biological parents increases. This also means an increase in the number of females that cannot reproduce, causing the mosquito population to collapse and possibly be wiped out.

With advances in genome editing researchers have been working on generating gene drive tools that can have widespread application beyond the mosquito example discussed here. In particular, CRISPR provides a DNA-cutting enzyme called Cas9 that is very specific and efficient and can be applied in a wide variety of organisms.

Part 3: CLASSROOM ACTIVITY (15 minutes)

Slides 8, 13, and 20 all provide discussion questions. We encourage teachers to return to these slides at the end of the presentation and ask students to further delve into the questions. Teachers may also ask students to read an additional article and continue the conversation in the next class. Finally, this time could also be used for a short, written in-class response to one of the questions or case studies that most captures the student's attention.

Part 4: ASSESSMENTS & HANDOUTS

Homework assignment:

In addition to participation, assess students through a persuasive essay that addresses one of the case studies from this lesson, with students choosing an argument and supporting it with evidence. Teachers may want to use articles embedded within this lesson plan, or some of the articles and videos below in the "Additional resources" section.

"Engineering the World Around Us: Genome editing and the environment" quiz answer key

(see page 28 for quiz)

1. Genome editing is making a change to an organism's DNA at a specific site.

2. Konzo is a paralytic condition caused by the consumption of a toxin in cassava that our bodies convert to cyanide. Konzo is a disease of poverty, with lack of access to water and protein-rich food being the main contributors.

3. Scientists are considering using a genome editing technique called CRISPR to make changes to the two cassava genes linked to the plant's toxicity, without losing any beneficial traits that these genes might provide, such as drought-tolerance and resistance to pests.

4. Hawaiian honeycreeper birds are at risk of extinction because of (a) habitat destruction by humans and (b) avian malaria, worsened by increasing average annual temperatures that allow the mosquitoes that carry the malaria parasite to survive at higher altitudes where the honeycreepers live.

5. The main goal of using a gene drive is to quickly spread a genetic trait of interest through the wild population of mosquitoes. To protect the Hawaiian honeycreepers, the desired genetic trait would either eliminate the mosquito population or make them mosquitoes unable to carry avian malaria.

6. Key questions include: (a) which genetic trait would be most suitable to introduce into the wild mosquito population in Hawaii? (b) what are the possible effects on the existing ecosystem? (c) how can spreading of the genetic trait

beyond the target population (i.e. the mosquitoes in Hawaii) be prevented? (d) what are reasonable risks and worthwhile benefits with regards to this approach? (e) what does a partnership look like between scientists and community?

7. Essential factors needed for de-extinction of a species, such as the woolly mammoth, include (a) preserved DNA and (b) a close living relative who could act as a surrogate. Additionally, one would need substantial financial investment, a habitat in which to re-introduce the animals, as well as a process for consulting with the public and securing agreements with governments and communities that might be impacted.

8. Concerns around de-extinction include: (a) will there be a space for the revived animals to live? (b) how are the voices of people impacted by the return of an extinct species weighed? (c) what might the impact be of the reintroduction of extinct animals into existing ecosystems? (d) what are reasonable risks and worthwhile benefits with regards to this approach? (e) in the case of the woolly mammoth, is the use of the endangered Asian elephant in this project justified?

STUDENT HANDOUT

Vocabulary List

Biome: A specific area of the planet that is defined by the animals and plants that can live in that area's physical climate (e.g., desert, tundra, tropical rainforest). A biome can contain a number of ecosystems.

Cassava: A root vegetable native to South America. It is a major staple food in countries around the world. After rice and maize, it is the largest source of carbohydrates for people living in the tropics. Cassava must be properly prepared before it is eaten or it can be toxic.

CRISPR: CRISPR stands for "clustered regularly interspaced short palindromic repeats". Found naturally in bacteria, scientists have adapted CRISPR as a tool to make targeted changes to the DNA of a variety of organisms.

Cyanide: A toxic chemical. The cassava plant contains a naturally-occurring chemical that is turned into cyanide when consumed by humans. If the cassava is prepared properly, this precursor to cyanide can be removed, so the cassava is safe to eat.

De-extinction: The process of reviving an extinct species or creating an organism that resembles an extinct species.

Ecosystem: A community of organisms and their environment that interact with each other through cycles of nutrients and energy.

Extinct: Describes a species for which there are no living specimens.

Gene Drive: A technique for increasing the likelihood of a specific segment of DNA to be passed from parents to their offspring (above the usual 50-50 chance). A gene drive enables a genetic trait to spread more widely through a target population.

Genome: An organism's complete set of DNA.

Genome editing: Making a change to an organism's DNA at a specific site.

Malaria: An infectious disease caused by parasites that invade red blood cells. The parasites are transmitted to humans and other animals via bites from mosquitoes that carry them.

Mammophant: Mammophant is a combination of the words "mammoth" and "elephant". It is the term used to describe an organism that carries DNA from both Asian elephants and woolly mammoths.

Paralysis: Losing the ability to move part(s) of your body.

Permafrost: A layer in the ground that remains below 32ºF (0ºC) for at least two consecutive years.

Surrogate: An animal or person who carries and bears offspring on behalf of another. In the context of woolly mammoth de-extinction, an Asian elephant will function as a replacement for the non-existent mammophant parent.

Taiga: The world's largest biome. Taiga is found at northern latitudes and is characterized by coniferous forests (e.g., pines and spruces). Also known as "boreal forest" or "snow forest".

Tundra: The world's coldest biome, which is found in the Arctic as well as high mountain regions. Tundra is characterized by the presence of a permanently frozen soil (permafrost) and a cold climate with little rainfall. These conditions result in a treeless landscape with moss, lichen, and low-growing shrubs.

Engineering the World Around Us: Genome editing and the environment

QUIZ

Name________________________________ Date_____________

1. What is genome editing?

2. What is Konzo, and what are the causes of this disease?

3. What genetic changes are researchers thinking of making to the cassava plant?

4. List two reasons why the Hawaiian honeycreeper birds are at risk of extinction.

5. Describe the main goal of using a gene-drive to alter mosquitoes that are impacting Hawaiian honeycreeper birds.

6. Name three questions that scientists and local community members should consider as they develop a genome editing strategy to introduce a genetic trait that prevents mosquitoes from infecting the Hawaiian honeycreepers with avian malaria.

7. Imagine you are a scientist planning to bring back an extinct species such as the woolly mammoth. List at least two essential factors needed for a project to be considered scientifically possible.

8. What are three points of concern about using genetic engineering to revive extinct animals?

Engineering the World Around Us: Genome editing and the environment

ADDITIONAL RESOURCES FOR TEACHERS

These articles and videos highlight the scientific and societal questions related to genome editing and the environment and provide additional background for the case studies in this lesson. This lesson will be updated regularly, so please visit www.pged.org for additional reading.

pgEd's lesson plan on "Genome editing and CRISPR".

Videos

CRISPR: A gene editing super power (video) by *SciShow.*

Cassava in Color (video) by *Crop Trust*

What's a gene drive? (video) by *Risk Bites.*

Hawaiian I'iwi bird (video) by *The Nature Conservancy*

Genetic Engineering and Diseases: Gene Drives and Malaria (video) by *Kurzgesagt.*

"Should we bring extinct animals back to life?" (video) by *LifeNoggin.*

Articles and Books

Bitter Harvest: Cassava and Konzo, the Crippling Disease by Amy Maxmen, *Global Health Now.*

"The Plan to Rescue Hawaii's Birds with Genetic Engineering" by Antonio Regalado, *Technology Review.*

"Huia-like bird could sing from the branches once again, but what are the limits?" by Jack Fletcher, *The Press/Stuff NZ.*

A recent public apology from a scientist working to do the type of community consultation and partnership development needed for a safe and successful project, by Kevin Esvelt, *Sculpting Evolution* (blog).

Scientists Release Controversial Genetically Modified Mosquitoes In High-Security Lab, by Rob Stein, *KPBS-National Public Radio*.

"How to Clone a Mammoth: The Science of De-extinction", a book by Beth Shapiro. Dr. Shapiro's "The Royal Institution" talk is also great for students.

"De-Extinction Debate", a point/counterpoint series from *Yale Environment 360.*

"These Are the Extinct Animals We Can, and Should, Resurrect" by Beth Shapiro, *Smithsonian Magazine.*

"Artificial womb could grow mammoth-elephant hybrid" by Claire Reilly, *CNET*

"Countries with the most endangered mammals" by Ellie Kincaid, *Business Insider*.