Editor's Note – Transgenic technology allows genes from one organism to be placed into another to produce Genetically Modified Organisms (GMOs). One potential use of this technology is breeding forest trees to help them fight the exotic insects and diseases. However, as many are aware, the use of GMOs is highly controversial and there is worldwide concern over their safety to humans and the environment. The issue of GMO forest trees is particularly relevant to Kentucky as the newly established Forest Health Research and Education Center housed at the University of Kentucky was developed to work directly on forest health issues with a focus on genetics. As a concerned woodland owner you will be called upon to voice your opinion on the issue of using transgenic technology for forest protection and restoration. Ellen Crocker, post-doctoral scholar with the Forest Health Center and UK Forestry Extension, was requested by the Kentucky Woodland Magazine editors to provide all of us a sound background on this technology, enabling us to develop an informed opinion on it and its place in forest protection and restoration.

Photo courtesy: Paul Williams

The health of our forests is under attack. Invasive insects and diseases are increasingly prevalent in North American forests and can cause devastation, as seen below. How can we better defend our forests?

Decline of our Forests and Trees -Can Modern Genetics Provide a Solution?

s a society, it is our responsibility to decide what to do about the problems we have caused in our woodlands. If we want to protect native tree species from exotic insects and diseases, we will require solutions above and beyond traditional approaches. Modern scientific methods, including the use of transgenic technologies to create genetically modified (GM) trees, are being explored to deal with these problems. GM techniques have been met with resistance in agriculture and, in thinking about their potential use in forests and woodlands, there are many factors to consider. Should we allow GM trees at all? What if transgenic technology can help save some of our ecologically and financially important tree species or aid forest restoration? How about studying GM trees in the lab to breed better non-GM trees?

Science can provide solutions, but whether we use them is up to us. To make good decisions about which GM trees we should and shouldn't use, we first need to sort through the blind claims and profit-driven arguments to have a clearer picture of the risks and benefits associated with using genetic technologies in forestry. This series explores these topics and invites you to think for yourself about the future of our forests.

Threats on our doorstep

Our forests and woodlands are changing rapidly. But then again they have never been static, particularly when people get involved. We have been actively changing eastern

North American forests for thousands of years, removing or adding tree species and determining where forests and woodlands occur. These changes are often positive. For example, by focusing on sustainable wood-



People have been managing forests for many years, including these workers in the 1930s harvesting trees using donkeys. Photo courtesy: USDA Forest Service Southern Research Station Archive, Bugwood.org

land management we can make our woods more healthy and productive.

However, our forests and woodlands are increasingly facing threats that they have never encountered before and

that they might not be able to cope with. Human introductions of destructive invasive diseases and insects have decimated several tree species considered central to eastern forests. For example, American chestnut once dominated our forests, driving our local lumber economies and providing habitat and food for many animals. However, since the introduction of Asian chestnut blight in the early 1900s, American chestnut trees have been nearly eradicated.

Through the unintentional transport of contaminated



Increased worldwide transport (cargo ships, above, filled with shipping containers, right, and wooden packaging material) has facilitated this.

Photos courtesy: Larry R. Barber, USDA Forest Service, Bugwood.org

plants and woody material, we have released a Pandora's box of enemies attacking our trees. American chestnut is not the only giant to fall victim to our mistakes. American elms have mostly disappeared due to Dutch elm disease. On the west coast, an invasive disease is causing the epidemic sudden oak death, resulting in millions of dead oak and tanoak trees. Meanwhile, emerald ash borers, native to Asia, have killed ash trees throughout the region and



Thousand cankers diseases (above) and sudden oak death (right) are two of the diseases that could be devastating if they appear in Kentucky.

are currently decimating Kentucky's ash trees. European gypsy moth, Asian longhorned beetle, oak wilt, thousand cankers disease ... the list goes on and on, and the rate of new threats reaching our forests is only increasing as the world becomes more and more globally connected.

How can we fight these threats and

defend our forests and woodlands? In some cases, we need to protect the trees we have from oncoming threats. In others we need to make the hard decision of whether to develop and reintroduce diseaseresistant versions of eradicated trees or

Photo courtesy: Bruce Moltzan, USDA Forest Service, Bugwood.org accept their loss, allowing them to live on only as a shadow of their former glory in selected preserves, managed gardens and our memories.

Building a stronger forest

While immediate threats can be met with pesticides, they also are expensive and can have non-target effects. Pesticides are useful for a variety of forest and woodland health issues, but only offer a short-term fix and then must be continually reapplied.

Regulatory programs aimed at preventing the spread of potential diseases and pests, both on our shores and within the United States, may reduce or slow down their arrival. However, the current programs clearly have been unsuccessful in a number of cases. In the long run these approaches are unlikely to provide complete protection as just one introduction is needed to result in new epidemic level devastation.

Breeding resistance is a better, more long-term, and sustainable means of giving trees the leg up they need to maintain or regain their natural role in our forests and woodlands. Breeding can defend against particular insects and diseases when traditional control techniques are ineffective. Conventional tree-breeding programs take time,

Vocabulary:

- Cisgenic plants: GM plants that have genes inserted into them from a different individual of the same (or a closely related) species.
- Conventional (classic) plant breeding: Intentional and repeated crossing of different plants (of the same or closely related species) followed by careful selection for desired traits.
- Genes: Regions of DNA that carry the information for inherited traits. They provide the recipe for proteins that make organisms (plants or animals) work.
- Genetic modification/genetic engineering/GM: Any intentional changes to an organism's genetic material using molecular biology. These changes include the mutation, insertion, deletion, or alteration of genes.
- Hybrid tree: A tree that is the offspring of two different tree species.
- Molecular biology: A scientific field focused on understanding the molecular basis of biology, genetics and biochemistry, especially involving the interactions of DNA, RNA, and proteins.
- Rapid cycle breeding: Using plants that develop more rapidly to accelerate the pace of plant breeding, especially useful for slow-developing trees. Can be developed using GM technology or conventional breeding.
- Subgenic plants: GM plants that have had genes deleted from their genome.
- Transgenic plants: GM plants that have had genes from another species inserted into their genome.



In some cases, chemical treatments are available for invasive pathogens and insects (such as the soil drench, above, and injections, right). However, these can be costly and time consuming and are typically not feasible on a landscape level. much longer than breeding programs for agriculture, but there are new techniques using GM technologies that can speed up the process. Regardless, the development of resistant trees may be the best long-term solution for several of our trees species under attack.

Photo courtesy: GRSM Resource Mgmt. Archive, USDI National Park Service, Bugwood.org



Photo courtesy: David Cappaert, Michigan State University, Bugwood.org

Searching for resistance

Any breeding approach starts by looking for particular traits, for example, trees naturally resistant to a particular disease or insect. However, in the case of American chestnut and others, researchers have found little resistance in our native populations. Because of this, several organizations have worked to breed hybrid chestnuts between the American species and a Chinese species resistant to the disease. This process is called conventional tree breeding, where a resistant tree (in this case Chinese chestnut) is bred with a susceptible tree (in this case American chestnut). The goal is that, after many generations of breeding, you will get a tree that has the resistance from one parent but all the other characteristics of the other parent. In this case, trees that look and grow just like an American chestnut but have the resistance of the Chinese chestnut.

While this seems straightforward, the reality is that conventional breeding in trees takes many, many years. First, you have to see which hybrids are resistant, which can take a long time as trees develop slowly and in some cases do not express symptoms of disease until they are more mature. Then this slow breeding and selection process must be repeated again and again so that the resulting tree has the characteristics of the susceptible native without tag-along traits.

Depending on how well that works, you are still left with a hybrid tree, in this case a mix of American and Chinese chestnut. In the meantime, forests are not standing still, waiting for the return of the American chestnut. New species, including many invasives, are taking their place in forests and woodlands. The more time that passes the harder it will be for American chestnut to regain an ecologically significant role.

Using the genetic toolbox

In recent years, advances in molecular biology have opened up a whole new world of possibility in tree breeding. New technology is giving us a bigger and better toolbox to fight tree diseases and insect pests and keep pace with incoming threats.

Our increasing understanding of genetics presents many different possibilities when it comes to tree breeding. In the past we selected resistant trees somewhat blindly, waiting to see whether symptoms developed and hoping that their absence represented a genetic superiority over susceptible individuals that would be inherited by future generations. Now we can specifically look for the genetic components of resistance, enabling faster and more precise breeding.

Perhaps the most well-known—and most controversial—application of new molecular techniques is the direct changing of genetic information, either by adding, removing, or moving around genes. While everyone has heard of "genetically modified," or "GM," plants, most people envision them in the context of agricultural systems where they are used extensively. While tree plantations

may be somewhat similar to a farm, forests and woodlands are different and require different types of GM approaches. For example, the trees must be self-propagating, diverse, and provide a slew of other ecosystems services, not just fill a narrow niche for our uses alone.

As with any new technology, there



Scientists have used conventional breeding approaches to develop more resilient plants for many years. Now, new technologies are enabling researchers to even better understand tree genetics and defenses.

Photos courtesy: Rachel McCarthy, Cornell University - NEPDN, Bugwood.org

is plenty of confusion and misinformation regarding GM plants, especially with trees. On the one hand, unintended side-effects are possible from the use of GM trees. On the other hand, many tree species are in jeopardy because of human actions and we need to develop better solutions to maintain our strong native forests. GM forest trees are still years away from potential widespread use here, but before then, it's important to understand the science behind them and to develop informed views on their use.

What does genetically modified mean?

Any organism whose genetic material has been altered by modern genetic engineering techniques is considered a genetically modified organism (GMO). While this may sound simple, it actually includes a wide range of different motivations and approaches. Take a look at a few example GMOs:

- Bacteria modified to produce insulin, developed in 1978 by the biotech company Genentech
- Genetically modified mice for lab research to provide insight into human cancer and other diseases, first developed in 1984 by university researchers
- Corn and soybeans modified to be resistant to herbicides, first developed by the company Monsanto in 1995

• Vitamin A enriched rice (golden rice) to minimize a type of malnutrition that kills hundreds insulin, GMOs are increasingly a part of our modern world. Current estimates suggest that approximately 90% of all corn, soybean and cotton grown in the U.S. is genetically modified.

From corn to

of thousands of children each year, first reported in 2000 by Swiss researchers at federal and university institutes

- Eucalyptus trees modified to tolerate cold weather and for plantations in North America, currently under development by a mix of tree biotech, pulp and paper firms
- American chestnuts modified to be resistant to Chestnut blight, recently developed by university researchers and a non-profit organization (American Chestnut Foundation) collaboration

Reading this list, you might have different reactions to different GMOs. Are these reactions based on their use, the type of organism being modified, the reason for their development or who developed (and is benefiting from) them? Since GM refers to a technology that can be used in many different ways, you might approve of some and not others. To help break this down further, let's focus on several particular aspects of GM development and purpose.

Genetic information used in GM plants can come from a wide variety of different sources. On the one hand, we now

have the technology to move specific genes around between different individuals of the same tree species. For example, defense genes can be moved from resistant trees to susceptible trees, protecting them from infection. This type of GM plant, called "cisgenic," only has genetic information added from another of the same (or of a closely related) species. The same end effect could be reached with traditional breeding, but GM technology allows scientists to work much faster and more precisely.

On the other hand, genetic information might be brought in from a more distantly related species, referred to as "transgenic." This is the type of technology most commonly used to create herbicide- and insect-resistant agricultural crops by taking bacterial genes for those traits and putting them in plants. The use of transgenic technology to produce GMOs, of course, has raised public concern. But before this issue can be discussed rationally as it relates to forests and woodlands, it is important to know how scientists might utilize these technologies in tree-breeding programs.

GM tree motivation: from research to restoration

People want to use GM technology to breed forest trees for many different reasons. For the most part, they fall under

How Plants are Genetically Modified

How are plants genetically modified? In most GMOs, genes are either changed (mutations), added (insertions), or removed (deletions).

Mutations: Many different substances and conditions can increase the number of mutations in genetic information. These mutations usually have a random effect, making them more useful for learning about the genetic basis of traits, than field application.

Insertions: Most GM plants have a gene added. This change can be done many different ways, for example:

- "Gene guns" can physically shoot particular genes into plant cells. Sometimes, the plant then will incorporate the genes into its DNA, however it is relatively inefficient and non-targeted (the gene could wind up anywhere in the genome).
- Some bacteria and viruses have natural equipment that let them insert genes into plants, and this ability can be used to transfer the desired genes.

Deletions: Newer technologies enable targeted gene removals and replacements.

• Genome editing is a broad type of genetic modification that uses artificially engineered nucleases ("molecular scissors" that snip DNA precisely) to create breaks at specified parts of the genome and cut or add new genes. the following major goals: research testing, improved harvested yield and forest restoration.

In highly controlled lab or greenhouse environments, GM trees can give us insight into key mechanisms that then might be applied in a broader context. One example is by developing test trees that mature more rapidly. Normally, trees take a long time (5-7 years) to mature, which makes finding and selecting particular traits a slow process. However, by modifying—for example—the genes that control flowering time, GM trees can mature much more rapidly. After, the modified DNA can be removed from the offspring trees, producing a native tree without the DNA alteration that led to early flowering in the parents. Referred to as "rapid cycle breeding," these fast-developing GM trees can be used to speed up our discoveries and let researchers know whether they are on the right track without introducing GM plants to the environment.

In contrast to this experimental use of GM trees, there's also great industry interest in developing GM trees to increase quality and quantity of harvested timber in forests and plantations. Two approved GM forest tree varieties are available internationally: poplars modified for insect resistance in China and eucalyptus designed for yield increases in Brazil. The companies behind their development argue that, as these GM trees are more efficient and can do more with less land, they could decrease the conversion of natural forests into plantations. In addition, companies are investigating a wide range of other financially beneficial uses of GM forest trees for use in the United States. Given that GM approaches have been used for several agricultural trees (apple, plum, papaya), it is likely that the same will apply to forest trees. However, the long-term ecological impacts of such trees in forested settings is less clear.

A third goal of GM technology for forest trees would be restoration. Restoration can be done by strengthening or reintroducing native species that have been decimated by invasive diseases and insects and is a fundamentally different objective from improving trees for increased economic production. In this case, the goal is restoring ecological balance, not financial gain. As with the American chestnut, an increasing number of important tree species are being jeopardized by human-introduced invasive threats. GM technology provides one pathway to addressing this problem by breeding resistance to handle the onslaught of exotic threats that are occurring at an increasing rate. However, given the long lives of trees and the rapidly changing nature of our climate and eastern forests, it will be a challenge to predict which traits are important and which are not in the long run.

"GM" is an umbrella term, covering the broad set of different techniques, origins, and goals that drive modern molecular plant breeding. You might be in favor of some GM plants while against others. Some of the concerns about GM technology may prove well founded, others overblown, but by looking at each proposed GM plant independently (the reason behind its development, the technique used to develop it) we can each develop a clearer picture of what resonates with each of us and why.

Stay tuned: Next time we will dive deeper, focusing on several case studies of potential GM trees under development.

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Kentucky's diverse woodlands provide many benefits and are ecological and economic assets. It will likely require a variety of approaches to ensure that future generations enjoy the same abundant forest resources that we do.

Photo courtesy: Tom Barnes