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The Application of Genetic Engineering in Forestry: Factors That Prevent Its Commercialization

John Evers[1]*

Abstract
Forestry is an important industry that provides wood, paper, and many other commodities. Plant biotechnology has begun to play an important role in forestry. Despite its importance, forestry is behind in biotechnology when compared to agriculture. A question of particular interest is the lack of genetic engineering use in forestry, and what factors have caused this. Several factors are likely behind this phenomenon, including genetic engineering limitations, forestry economics, regulation, and public opinion. One major limitation in forestry is the lengthy generation span of trees. Another issue is the lack of forest tree domestication. Genetic engineering is still limited in ability, and some trees are difficult to genetically engineer. Investment in forestry, such as tree plantations, can be risky. Also, the economics are complicated. Environmental concerns exist, and public opinion can be negative towards genetic engineering, which can in turn influence politics. Clearly, there are many factors with several connections. This study looks at the lack of genetic engineering use in forestry, focusing on the challenges faced in the genetic engineering process, and how this process can be improved. The experiment will analyze genetic engineering limitations by genetically engineering economically important forestry trees and analyzing genetic transformation success rates and transgene stability. The impact of transgene introduction on tree physiology and areas in protocols that may benefit from more attention will also be examined. Expected results are provided and discussed. Future research directions to examine other factors impacting genetic engineering in forestry are outlined.

Keywords
Forestry — Transgenic — Genetic Modification

[1] *Department of Biological Sciences, University of North Texas*
***Faculty Mentor**: Dr. Tom Miles

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Introduction

The latter half of the twentieth century saw the production of an ever growing array of transgenic plants. These new and unusual organisms signaled a new dawn in biology: the genetic engineering age had begun. Even the pioneers of genetic engineering likely could not have foreseen just how large an impact their work would soon have. Today, genetically engineered crops are widely used in many parts of the world. Experiments in genetic engineering of forest trees are being carried out. The industry is rapidly growing and advancing technologically. Other technologies, such as plant tissue culture, have also become well established commercially. Many scientists hope that genetic engineering, modern cloning techniques and other technologies will help solve some major problems, such as feeding an ever increasing human population. However, genetic engineering and other biotechnologies are not without concerns, and a recent controversy has developed worldwide over genetic engineering, particularly in the political realm. Much like the story of GM (genetic modification) in agriculture, genetic engineering in forestry is also a controversial idea. While some improvements have been made over the years, much less progress has been achieved with forestry biotechnology than in agriculture. GM in forestry has yet to become fully commercialized.

1. Overview of Forestry Economics and Politics

Forestry is a very important part of the world economy. Humans rely heavily on wood products for a variety of uses, from paper to firewood. According to Fenning and Gershenzon [1], only about 12 percent of the world's wood harvest comes from tree plantations. The rest is harvested from natural forests. In some cases, destructive harvesting methods such as clear cutting are used. Wood and other forestry product demands are projected to increase in the near future. Timber demand will increase due to higher population levels and a growing global middle class, and the increased use of forestry products for biofuels and biomaterials.

Figure 1. Poplar Tree (photo by author)

Forestry uses either naturally occurring forests, which governments often protect and manage, and tree plantations as its main wood sources. At present, forestry uses traditional breeding techniques such as hybridization to produce trees with favorable genotypes, and cloning technology is already widely used by the forestry industry for rapidly replicating trees of a favorable genotype that have desirable characteristics, such as a fast growth rate [1]. Genetic engineering has yet to fully become commercialized in the forestry industry.

A few select tree groups are rather important in forestry: conifers, poplar trees, and eucalyptus trees. The genetics of these trees are better known when compared to the genetics of other tree species [2]. Of major note is that tree crops usually take many years to grow large enough to be harvestable before rotations can occur.

Problems with forestry often impede the development of tree plantations, the conditions under which genetically engineered trees would most likely be used. One problem is simply investment. Lots of investment is needed, and the investment is risky [1]. Trees need lots of space, yet another problem. Sometimes governments will provide subsidies or tax breaks to encourage people to invest in tree plantations, but investment is still limited.

Fenning and Gershenzon [1] have an interesting theory regarding plantation forestry and ecocerticication schemes. Many environmental groups have become involved with the wood market, the main one being the Forest Stewardship Council, or FSC, and one of their main ideas is to cut back consumption of forestry products. These groups tend to favor sustainable harvesting plans and in more rare cases some types of ecologically friendly plantations. Transgenic trees are forbidden in ecocertification. In ecocertification, a governmental agency or private group, such as a non-governmental organization, certifies products that meet a set of guidelines that define whether a product is considered "sustainable" or not. Ecocertification schemes could contribute to a continued reliance on natural forests for forestry products, and the use of potentially harmful substitutes to wood like plastics that could contaminate the environment. Wood supply needs to be increased quickly to satisfy demand. Tree farms have been argued to be a great way to do so, especially when combined with biotechnology. The attempt to impose a green forestry economy by environmental groups stifles the growth of many tree plantations and forestry biotechnology.

Other groups are opposed to genetic engineering attempts, and in some cases have substantial political influence. Several governments are uncertain about genetic engineering, in some cases against it. Many regulation systems have been put in place, ones that unfortunately can only be overcome by wealthy, influential businesses and organizations most of the time [3]. Scientists will need to be ready to confront those with opposite views to convince the public of the benefits of genetic engineering.

1.1 Tree Genetic Engineering: Biological Factors

Two techniques are widely used in genetic engineering of plants: the Agrobacterium technique and the biolistics technique. In the Agrobacterium technique, a bacterium from the genus Agrobacterium (mainly *Agrobacterium tumefaciens*) is used as a vector to insert foreign genes into plant cells. Biolistics uses small DNA guns that shoot DNA-coated bullets into target plant cells, which uptake the foreign DNA and then regenerate. Walter notes that protocols, or guidelines, are being increasingly developed for these genetic engineering techniques in trees. Genetic engineering is already well established with trees, with many successes already had [4]. For example, many transgenic *Pinus radiata* trees have been made.

There is a common consensus among many scientists that trees are proving more difficult to genetically engineer than traditional crop plants. The primary factors are a lack of knowledge of the genetics of trees used in forestry, the long generation times trees have, and a lack of domestication of forestry tree species. In addition, there are some challenges involved with genetic modification of conifer trees, one of the most important groups of trees used in forestry. Many transgenic conifers have been genetically unstable when regenerated from transformed cells, meaning the introduced gene stops getting expressed [2]. This issue can also occur in other tree species. Many conifers are also more resistant to Agrobacterium infection, making this technique harder to use when applied to this group. Also, the efficiency demonstrated by biolistics is not as high as the Agrobacterium technique in most cases. Finally, the sheer diversity of trees is problematic. Many species exist, and the species themselves tend to be diverse in form, proving a problematic obstacle given the relative lack of knowledge of the genetics of most tree species.

Various experiments have been done to examine transgenic stability in forest tree species. A study done on transgenic stability in poplars has shown promise. In a study conducted by Li, Brunner, Meilan, and Strauss, two transgenes (green fluorescent protein, or GFP, and bialaphos resistance gene, or BAR) were used to examine transgene stability in poplars. During a span of 3 years in both a greenhouse and then a field trial, no trees experienced total loss of gene expression. This is still a relatively short time span however, especially when considering tree lifespan. Gene silencing can result in transgene instability and ultimately loss of function; usually several dynamics are involved in the process of gene silencing. Improvements in getting consistent transgene stability that is long term in forestry trees will be needed to ensure sterility of transgenic trees, which will be important for getting government and public acceptance.

Sometimes there are challenges in the introduction of transgenes. Introducing transgenes into an organism is a precise process, but there can be unforeseen impacts on other life processes of the organism. During experimentation, it was found that the poplar trees being tested using a LEAFY promoter did not perform as well as hoped. The attenuation system devised as a method to offset the negative impacts of the transgenic cytotoxin and floral promoter genes did not overcome the stunting impact on vegetative growth as well as hoped. The transgenic trees were similar in growth habit to the controls during the growing period in the greenhouse. Differences emerged in the field trial stage. The transgenic trees exhibited stunted growth and leaf yellowing during the field trial when compared to the non-transgenic controls, a problem that continued for two years. These results emphasize how critical early field trials can be.

One of the major challenges in tree breeding is the time it takes trees to reach maturity. In some cases this can be a few decades. Tree maturity times and flowering intervals limit breeding options. Genetic engineering to modify flowering can be used to open up new opportunities to cross tree species that previously could not be due to differences in flowering timing. Trees can also be engineered to flower earlier, allowing for shortening the time until maturity. A major limitation has been slower advances in gaining knowledge on the molecular mechanisms that play a regulatory role in the maturation process of trees. While knowledge about genes and processes that regulate flowering in trees is better understood, additional research is still needed to improve existing knowledge of tree flowering regulation and to improve upon the manipulation of flowering.

Other biotechnologies exist that are often used in conjunction with genetic engineering. Cloning techniques, such as tissue cultures, are commonly used to grow and regenerate transgenic plants, for example. There are issues that can be encountered in cloning techniques however. In a particular example, Nair and Vijayalakshmi [5] noted some challenges in callus survival rate they faced when trying to create transgenic *Eucalyptus tereticornis* trees. Many of the calli being prepared for transformation died or otherwise became unusable.

Other DNA-based technologies are used, such as DNA sequencing and DNA mapping, which can help identify novel genes and broaden understanding of tree genetics. Many plant adaptations are of interest to scientists, such as osmotic mechanisms and genes that regulate stress, which are given particular attention to by Altman [2]. These particular plant traits could be important in improving plant resistance to stressful environmental conditions, an area of major interest among plant genetic engineers.

Ultimately, two major biotechnology strategies will arise [2]. These are the non-transgenic and transgenic approaches. Each will involve different biotechnologies but will be linked in their goals to improve plants, primarily for human use but also environmental restoration.

1.2 Promise and Problems

Many benefits of genetic engineering are already evident in agriculture. It is widely agreed by many people that genetic engineering and other biotechnologies could provide significant benefits to forestry. Genetic engineering could improve tree genotypes, make trees grow faster, and shorten crop rotation periods. Wood yield could be increased [1]. Disease and pest resistance could be improved to minimize losses. The potential to increase plant resistance to environmental stress, such as dry conditions, could be quite useful [2]. Trees could even be engineered for functions such as phytoremediation.

Genetic engineering does not come without hazards. The possibly of fertile transgenic trees becoming feral and eventually pests is a hypothetical possibility. Many trees used in forestry have related species that occur in the same area, and cross pollination is of concern to many people, in particular environmentalists. Transgenes could contaminate wild population gene pools. The gene or genes being introduced into a tree species must be carefully considered so as to avoid unwanted impacts. A solution is to breed or genetically engineer sterility into transgenic trees to ensure they do not spread or contaminate wild tree stocks with transgenic pollen. Another issue is that tree plantations are likely to be used with genetic engineering, which tends to reduce biodiversity in the cultivated area. However, tree plantations are often more biologically diverse than agricultural cropland, and may serve as habitat for some organisms [1]. Some precautions should be taken to be sure biodiversity is not too reduced in tree plantations and some wildlife can persist. In fact, genetic engineering could actually help increase biodiversity, allowing for the creation of new genotypes [4].

Furthermore, genetic engineering is controversial. Many people believe genetic engineering of organisms is dangerous and poses environmental and health risks. It has become a major political topic. Scientists should do a better job with communicating the benefits of genetic engineering and how to minimize risk. They should be honest and willing to take criticism. A precautionary principle rule is emphasized that should be used when dealing with genetic engineering (Walter, 2004). Many groups will try to influence public opinion, but people are generally more open-minded when it comes to new techniques and technologies [4]. Politicians must stick to science if genetic engineering and other biotechnologies are to be fully successful in implementation. However, regulations tend to be difficult to change or replace once implemented [3].

1.3 Summary

The field of genetic engineering and related biotechnologies in forestry is an ever evolving one. Several biotechnologies exist with a wide range of applications. There are many possibilities, but they don't come without some risk. Many advances have been made, but many more remain to be done. Knowledge of tree genetics still has far to go, and tree plantations are yet to reach their true potential. Genetic engineering techniques and methodology also need to be refined and fine-tuned for particular tree species. Current genetic engineering experimentation in forestry occurs, but hasn't yet become the commercial reality some people envision.

While forestry economics and the GM controversy in politics play a role, limits in biotechnology and knowledge gaps concerning trees remain significant limiting factors. The challenges posed to scientists hoping to bioengineer trees need to be addressed before other barriers can be overcome. This is an area that needs to see several more experiments done to improve existing techniques and find new ones. Meanwhile, the GM controversy will certainly continue. Scientists must be transparent and clear with the public if they wish to highlight the benefits of genetic engineering technology. With improvements in biotechnology and a successful outreach effort, the scientific community can make widespread commercialization of transgenic trees a reality. Given growing environmental challenges and resource demands, now efforts are more pertinent than ever.

2. Methodology

Genetic engineering in agriculture is widely used today, but is much less prevalent in the forestry industry. Many possible factors are likely to blame for this, but a clear picture of why genetic engineering has not taken the same path as it has in agriculture is yet to fully emerge. The intent of this research is to identify the factors involved that have stifled the application of genetic engineering in forestry, with the focus of this experiment on biological challenges involved in creating transgenic trees.

2.1 Genetic Engineering Experiment

One of the challenges with getting transgenic trees commercialized is that many tree species are difficult to genetically engineer, particularly conifers. Experimental replication of these claims will be very useful in seeing what challenges are faced in tree genetic engineering technology. The primary purpose of this experiment is to examine how easily certain tree types can be genetically engineered, and if these traits are stable and have no short or long term negative impacts on the trees. A secondary purpose is to examine differences in how the trees react to transformation attempts and how their culture conditions vary, as not all of the trees will be able to be grown under the exact same conditions due to differences in their evolutionary histories. A third purpose is to see if the Agrobacterium or biolistics technique is consistently more effective at creating transgenic trees.

Given the enormous diversity of different tree species and the difficulty of obtaining specimens of certain species, only a few select tree types will actually be used in the experiment out of practical reasons. The tree groups that will be used will be poplar, pine, and eucalyptus species. Each tree species will have its own designated "species group" with a non-transgenic control grouping and two transgenic groupings (for the two genetic engineering techniques being investigated). Two fairly commonly used, preferably fast-growing species in forestry will be selected for in these 3 groupings. Black cottonwood (*Populus trichocarpa*) and eastern cottonwood (*Populus deltoides*) are the chosen poplar species, loblolly pine (*Pinus taeda*) and Monterey pine (*Pinus radiata*) are the chosen pine species, and blue gum (*Eucalyptus globulus*) and forest red gum (*Eucalyptus tereticornis*) are the chosen eucalyptus species. Teak (*Tectona grandis*), being relatively important in forestry and sometimes grown in plantations, will also be studied in its own group. Preferably, one individual specimen tree of each species will be used for cloning use, that way the genetics of all the individuals of each species grouping will be identical. Therefore, if there are any observed differences in growth rate, health, and other factors within a tree species group, the possibility of these observed differences being due to differing genetic compositions can be ruled out. There will be differences between the tree species groups due to different genetics, which will be taken into account. If tissue culture propagation is best done from seeds, than seeds will be obtained for those trees where this is necessary. In the case of trees cultured from seed, differences in genetics between individuals will be taken into account.

Two commonly used genetic engineering techniques will be tested for their effectiveness in this experiment: the biolistics technique and the Agrobacterium technique. The two will be compared to each other to see which one is more effective, and with which types of trees. Though there are other genetic engineering techniques and leaving them out will be a limitation, the biolistics and Agrobacterium techniques are among the most commonly used and well known, and therefore of the greatest interest.

2.2 Species Group and Analysis

Each species group will contain three groups: a control group of the tree species that has been cloned but not genetically transformed, and two groups that have been cloned and genetically transformed, using the biolistics for one transgenic group and the Agrobacterium technique for the other transgenic group. For the Agrobacterium and biolistics techniques, one grouping of each will be done per tree species tested, with a total of 14 transgenic tree groups overall. There will be 7 different species groups overall. Each tree will be labeled with its own individual number for identification, and also labeled by species and what group. All of the trees will be cloned using tissue culture method. Given that cloning success will most likely vary from one tree species to the next, it is best to make several cloning attempts so as to maximize the number of individual trees. Not all clones may survive, so taking several clones for each species control and transgenic groups increases the chance that a decent number of trees will survive and regenerate into full plants. Of note, tissue culture methods will vary for each tree species, but this is an aspect that is a goal of the experiment to understand what the differences are in tissue culture and other procedures between tree species and how to improve existing procedures. The differences in tissue culture technique for each tree will be accounted for in the study.

As mentioned in discussing tissue culture, since different tree species are being examined, different protocols will be used for each tree species since not all trees are cultured or engineered the same exact way. Any differences in techniques of creating and culturing transgenic trees will be documented in detail using journals. Details will be important in journal entries regarding procedures, so care must be taken to properly document all important aspects of the tree genetic engineering process for each species.

Once the tissue cultures have regenerated into new young trees, the regenerated plants will be transferred onto a gel medium or directly into soil depending upon what the accepted protocols are for each species. The gel media will only serve as a temporary growth medium for the young trees, and acts a soil substitute that contains important nutrients. The trees will be grown under lab conditions that remain consistent, with unwanted variables such as drafts in the room minimized so as to not impact growth and health. Trees will all be subjected to the same uniform lighting and temperature conditions. Watering may vary between tree species due to their natural history, a fact that will be taken into account when comparing between tree species results.

As the trees grow, they will eventually be taken out of the gel media (for those trees that were started in gel media) and potted into larger pots. As the trees grow larger, they will be moved to a designated experiment greenhouse where all the trees will be together and easily observable. This may have to be done at varying times for each tree species because they will not all grow at the exact same rate. At all transferring stages, the species groups will not be separated, nor will their control and transgenic groups be separated from each other. Keeping track of the groupings is vital so individual trees are not misplaced in the wrong group.

The trees will be allowed to grow for two years in the greenhouse, which has several advantages. Growing the trees to full maturity would take too long, up to several years, would be too costly, time consuming, and could generate unwanted opposition from the public due to concerns about GM trees. In addition, keeping the transgenic trees in a greenhouse very significantly reduces environmental risks, and there will be less concern from governmental agencies since the trees are enclosed. The trees would also be protected from weather and would be under consistent growing conditions. Finally, the eucalyptus and teak trees would not survive the winter outside, so they must be kept in a greenhouse to ensure their survival.

Tree growth rate and observations on tree health, such as healthy or yellow leaves, or normal or stunted growth, will be considered and documented during the experiment. Growth rate, genetic engineering success rates for the biolostics and Agrobacterium techniques, and similar data will be collected on tables that make note of what trees are being measured, and out of what group. This data will be compiled at the end of the experiment. Plant health observations, notes on protocol differences, morphology observations, and observations on transgene expression will be kept in a plant journal that keeps track of the date of observations, tree that was observed, and out of what group.

The transgenic trees will be engineered with the GFP (green fluorescent protein) gene that will cause the tree tissue to fluoresce under ultraviolet light. This will provide an easy visual test to see if a tree was successfully transformed, and if a tree stops expressing the trait, this can give an idea of how stable the trait is. It also allows for seeing if expression of the introduced gene is in all plant tissues or only some. This genetic engineering experiment, which examines the issues involved in the genetic engineering process of a set of commercially important forestry trees will help contribute to a better understanding of where gaps in knowledge of tree genetic engineering remain, and where improvements can be made in protocols. By researching the impact of the introduction of the GFP gene into the selected tree species, areas that need refinement in the genetic engineering process can be identified, as well as patterns that might be of interest to plant scientists, such as how the trees react to the introduction of foreign genetic material. This will ultimately help refine and improve the genetic engineering process of trees. It is critical that gaps in knowledge of tree genetics and the genetic engineering process be addressed if genetically engineered forestry trees are to ever become widely commercialized and realize their full potential.

2.3 Study Parameters and Assessment

How exactly the trees will react to the introduction of the GFP transgene could take several paths, but some reasonable predictions can be made. It is expected that some of the tree species will accept the introduced transgene, making the initial genetic engineering attempt successful, while other tree species will not be successfully transformed. Of the trees that are successfully genetically transformed, the GFP gene is expected to be stable and expressed throughout the approximately 2 year period of the experiment, at least on some level. Some of the trees, such as the poplars, are expected to be transformed with the GFP gene relatively easily, while for other tree species (such as the eucalyptus) expected results are less certain.

Transgene expression should be full or partial, at least in some tree tissues. It was mentioned by Altman [2] that conifers tend to have less stable transgene expression. Hence, it is possible that the pine trees, which represent conifers in the experiment, will have a lower rate of GFP transgene stability. Other than the pines, transgene stability rates are expected to be similar among the other tree species, which are all eudicots. No significant differences are expected in how the trees express the GFP gene, nor should any negative impacts on plant physiology or health occur due to the introduction of the GFP gene.

Of the two genetic engineering techniques used, the Agrobacterium technique is expected to be higher in its success rate than the biolistics technique at transforming the trees. This is because the Agrobacterium technique uses a bacterium as a selective vector that is more precise than the biolistics technique, which involves the less precise method of shooting DNA-coated pellets with a gene gun into target cells. As was noted earlier, Altman mentioned that the Agrobacterium technique is not as effective at transforming conifers [2]. Therefore, the rate of successful genetic modification in the pine tree species using the Agrobacterium technique may be lower when compared to the other tree species.

The accepted protocols used for each tree species in this experiment should not radically vary. However, some differences are expected in how the protocols will need to be carried out, because the trees have different growth and development requirements. For example, the hormone concentrations needed for a successful tissue culture of pines are unlikely to work for growing eucalyptus tissue cultures. Tree growth rate is not expected to be changed and should be normal for each tree species. The growth habit of each tree (tall, bushy, etc.) should be normal as well. Root morphology is also expected to be normal for all of the trees. Growth rates and patterns will vary between tree species, but this will be a function of naturally occurring differences in the genetics between the tree species being studied and not the GFP transgene. The transformed trees should be healthy with no negative side-effects from the introduction of the GFP gene.

3. Discussion

Testing the impacts of introducing a transgene into the genome of different tree groups is a good place to start for a better understanding how to improve the genetic engineering process for trees. GFP gene loss of function in engineered trees would signal a need to invest more effort in improving the long-term functioning of genes introduced into the genomes of trees. Similarly, partial, but not full, GFP gene expression may imply a need for more research into the issue of transgene loss of function and improving gene delivery systems. Loss of function of the GFP at a higher rate in the selected pine species than the species in the other groups of trees would support the idea mentioned by Altman [2] that conifers tend to have transgene stability issues. If challenges are encountered in the genetic engineering of one tree group significantly more so than the others, such as eucalyptus, then more studies should be conducted to better understand why that particular group of trees is more challenging than others to successfully genetically transform. Also, while not expected in the proposed experiment, if negative impacts to tree health were to occur due to the introduction of the GFP gene, this would warrant a strong need for deeper investigation to understand how the introduced gene may be causing a negative impact on tree health or morphology.

Another area of interest besides the GFP transgene genomic stability, expression, and impact of introduction into tree genomes, is the success rate of the Agrobacterium-mediated gene transfer technique versus the biolistics technique for genetic engineering. If one technique is consistently more effective at genetically transforming the different tree species than the other technique, then that technique might be more widely applicable to different trees for future genetic engineering projects. If, however, success rates vary between tree species or species groups, then more studies may be necessary to identify the most effective genetic transformation methods for use with a particular tree group, such as with poplars, eucalyptus, or pines. If the Agrobacterium technique has a lower success rate with pines compared to the other species groups, or biolistics works better than Agrobacterium for genetically transforming pines, then biolistics might be a more effective technique for transforming conifers, and research into developing or identifying bacterial or viral vectors that can better introduce transgenes into conifers than Agrobacterium should be examined more. This would also provide more evidence for verifying the issues encountered with transforming conifers with Agrobacterium that Altman [2] mentioned. Challenges encountered at any point during the genetic engineering process should be carefully noted. If an issue comes up, such as a problem getting the tissue cultures of a particular tree species established, then the issue can be noted and future experiments could be designed to identify the problem. This is a practice that should be more widely used, that way protocols can have problem areas more readily identified and resolved.

More studies like this one need to be done to identify gaps in knowledge in the genetic engineering process of trees so protocols can be improved. With the identification of problem areas, resources and funding can be better allocated to addressing challenges that are still faced in creating transgenic trees. This will lead to getting answers quicker on issues that may be holding back genetic engineering efforts in certain forestry tree species in a more efficient and effective manner, speeding up efforts to create transgenic trees that can be commercialized.

At first, experiments should focus on trees in which their biology is better known and genetic engineering techniques are sufficiently established with. However, there needs to be an expansion in experimentation to trees that are used in tree farming or may be in the near future that are not as well known. With dwindling natural forests that will no longer be able to sustain harvesting and increasing pressure from environmental threats such as disease, invasive insect pests, and climate change, tree farming will become more important as a source for forestry products, and the genetic and species diversity of trees used in managed forests and plantations will likely increase as growing pressures force the forestry industry to increase tree genetic stock. Also, studies should include tree species from different areas of the globe where forestry activity occurs, not just a few select regions. Both temperate and tropical species need to be accounted for.

While outreach attempts have been made by the scientific community and industry to educate the public about genetic engineering technology, current attempts are still not proving sufficient. The public is still wary of genetically modified crops even though the technology has been legal for years and proven safe, and campaigns are ongoing by environmentalists, consumer groups, and other groups to restrict or ban genetically modified organisms. As long as the public remains wary of genetic engineering technology, attempts to commercialize transgenic trees will be likely to fail.

More efforts need to be made by the scientific community and industries interested in transgenic trees to communicate what transgenic trees are, how the technology works, and what the benefits are. It needs to also be stressed that transgenic trees are not dangerous to human health and must undergo years of testing with regulatory oversight by government agencies. Scientists and industry could engage in more public talks, including presentations and internet streaming videos that are open to anyone interested in listening. Such events would provide great chances to answer questions people may have and directly engage the public. Scientists should also make more of an effort to make research papers and results available to the public for free through online journals, that way anyone interested in learning about genetic engineering technology can easily research the topic on their own. Groups could also be put together by scientists to fact-check claims made by anti-GMO activists so incorrect information can be corrected. The corrected information could then be given to the public in talks, video streams, and on websites that people can readily access.

While genetically engineered trees are likely years away from being commercialized on a global scale, improvements have been made in the tree engineering process. Identification of areas needing improvement will be vital the next several years to refine the genetic engineering process for trees and to make them marketable. With careful planning and reasonable regulation, genetically engineered trees could be used to supply the majority of the forestry products society needs while taking pressure off of natural forests, which should be brought under increased protection from logging and other commercial activities. Genetically engineered trees are no panacea to the current issues of deforestation and increasing demand for forestry products; however, genetic engineering could certainly alleviate these issues while helping build a more sustainable global economy.

4. Future Research

While the current experiment will focus on biological challenges involved in creating and commercializing transgenic trees, such challenges are far from the only obstacles to overcome if transgenic trees are to be widely used in forestry. There are many economic, legal, and political factors also involved that need to be examined to better understand how these factors impact tree genetic engineering research efforts and commercialization attempts. A case study would be useful in better understanding economic and regulatory factors involved, and a survey to get insight into how the general public perceives transgenic trees.

Additional experiments on biological and protocol factors will not be ruled out. Examination of the impacts of the introduction of other genes or gene combinations other than GFP would be a useful addition to the insight gained from the original genetic engineering experiment. Making changes in protocols for experimentation could also prove useful in identifying areas in the genetic engineering process that need more attention. In addition, other tree types could be studied.

4.1 Case Study

For a look at forestry economics and regulation, a case study would be very useful. The states of Alabama and New York would be chosen as the two subjects of this case study because they both have different regulatory climates, with one being heavier with regulations than the other. This case study does not consider the full U.S. forestry economy and also leaves out world forestry economy dynamics, which would be a limitation. However, studies on the U.S. and world forestry economies would simply be far too costly and time consuming. A smaller case study of Alabama and New York would give a good idea of how the modern forestry economy works and provide a decent comparison of two different regulation systems. The time span of interest would be from 1990 to 2015, the time span in which genetic engineering of trees could have hypothetically become commercialized and genetic engineering became established with trees. Much of the data of interest will deal with production and profitability, such as yearly tree harvests, the number of tree plantations per state, and how profitable plantations and tree farms are. The other area of interest is regulation and related factors, such as what level of environmental protection exists, and political makeup. These factors influence how much forestry activity occurs in the state. States with less regulation are more likely to have more forestry industry activity than states with more regulation, hence making Alabama and New York a good comparison for these aspects.

The relevant information would be gathered by various means. Some data would come from government records, while other data would come from company records. Several industries and local and state governments would be asked to voluntarily participate in the case study. They would be asked to voluntarily give records and estimates for research purposes. While not all desired information can be gotten this way, it should gather a sufficient amount of information for study use. Current and past state laws, regulations, and court rulings regarding forestry would also be considered. U.S. federal laws and regulations regarding forestry apply to both Alabama and New York equally, but should still be considered in the study because these have a significant impact on forestry economics. Political data, such as Electoral College maps and the results of state government elections in Alabama and New York, is also of value to this case study. Once sufficient data has been gathered, it will be compiled for each state under one of three categories: economic, legal, and political. Economic data (graphs, tables, etc.) would be compiled on a timeline basis for each state. Laws, regulations, and court rulings would also be put on a timeline, note made of which ones are still in place, when they were enacted, and what laws and regulations are no longer in place. Political data would also be put into timeline format. This way, a change over time picture will emerge for each state, and correlations between the three categories can be visualized. This setup will also allow a good comparison between the Alabama and New York forestry economies and how they changed over time in relation to each other.

5. Conclusion

While the GFP gene is a widely used transgene in genetic engineering experiments, this is far from the only transgene that has been engineered into trees. Herbicide resistance, insect resistance, and other various traits have been engineered into trees. Thus, it would be good to test the success rates in transforming trees with other transgenes or transgene combinations and examine the impact the introduced genes have on tree physiology and growth dynamics.

Protocols are another good place for experimentation. Identifying problem points in the genetic engineering process for certain tree species allows for experimentation of current techniques. Based upon results, techniques in the process could be refined for existing protocols, or in the case of species where protocols are not well established, new protocols could be created. For example, the necessary concentrations of growth hormones for successful culturing of eucalyptus calli could be experimented with to see what the most optimum concentrations are.

At the end of the study, all the data and conclusions can be put together to create a full image of the factors involved, their links, and how they have interacted to create an environment that so far has not proven favorable to widespread use of genetic engineering in forestry. This information can be farther combined with the previous research on genetic engineering challenges in trees to generate an even fuller understanding.

Author Biography

John Evers graduated Summa Cum Laude in fall of 2015 from UNT with a Bachelor of Science in Biology with a concentration in plant science. He was a student of the Honors College and received the Board of Reagents Chancellor's Scholarship and the Honors College Distinguished Scholar Award, and is a member of Phi Kappa Phi. He is currently pursuing a Ph.D. at UNT under the mentorship of Dr. Brian Ayre.

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