Sun Grant/DOE Regional Feedstock Partnership

Final Technical Report

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Executive Summary

The U.S. Department of Energy (DOE) and the Sun Grant Initiative initiated the Regional Feedstock Partnership (referred to as the Partnership) in 2007 primarily to address information gaps associated with the likelihood for realization of the sustainable and reliable production of a billion-tons of biomass annually to support the U.S. bioenergy industry by the year 2030. Publication of *The Technical Feasibility of a Billion-Ton Annual Supply* (Perlack et al., 2005) in 2005, with its associated yield assumptions, led to the realization that the yield goals and assumptions utilized in that analysis required careful validation. To achieve the overall goal of validating key yield assumptions, the following objectives were developed:

1. Establishment of replicated field trials across regions to determine the impact of crop residue removal (primarily corn and small grains) on future grain yields and soil health.
2. Establishment of replicated field trials of some of the most promising dedicated energy crops (herbaceous annuals and perennials, as well as woody perennials) to demonstrate the potential performance of these feedstocks across the U.S.
3. Assessment of feedstock resources to be used to estimate a sustainable national supply potential.

Teams composed of representatives from Land Grant Universities, USDA ARS, DOE National Laboratories, industry, and other federal agencies were identified and assembled to address each of these objectives.

For Objective 1, members of the USDA ARS' Resilient Economic Agricultural Practices (REAP) team combined with university personnel to leverage on-going work and established new trials to determine sustainable corn residue removal rates. In addition, universities across the continental U.S. conducted surveys of published past research to define parameters for the use of cereal grain residue (primarily wheat) as a biomass feedstock.

For Objective 2, and based on input from DOE, USDA, Land Grant university scientists, and others, several herbaceous and woody energy crop species were selected for new or further evaluation across wide geographic areas as cellulosic feedstock sources. Species teams were organized to assess the yield potential of candidate herbaceous feedstocks including sorghum, energycane, *Miscanthus* x *giganteus* (hereafter referred to as miscanthus), switchgrass, and mixed perennial species (primarily grasses and some legumes) on Conservation Reserve Program (CRP) land. Woody species will be critical components of the bioenergy sector; therefore, species teams were also organized for shrub willow and hybrid poplar (*Populus spp.*). Field trials of each species were utilized to determine yield potential across the U.S.

For Objective 3, a GIS resource team was organized with representatives from each of the five Sun Grant regions. While some regional assessments were made, the multi-institutional team decided that the group from Oregon State University would utilize their PRISM ELM model to estimate national yield potential of the various species evaluated.

Key Outcomes and Impacts

- The Partnership demonstrated the production potential of diverse herbaceous and woody feedstocks across much of the U.S. for 5-7 consecutive growing seasons. Long-term studies such as this are not commonly possible due to limitations in funding cycles.
- Long-term field trials were valuable in demonstrating the importance of agronomic management practices and genetic resources to yield of sorghum, switchgrass, miscanthus, energycane, CRP mixed perennial grasses, poplar, and willow.
- National yield potential maps were developed for all species evaluated in the Partnership.
- Sustainable corn stover removal rates were quantified for use by producers, harvesters, and commercial lignocellulosic biorefineries.
- Data from the Partnership field trials not only validated initial *Billion Ton* estimates, but were instrumental in developing both the *Billion Ton Update in 2011*, and especially the *2016 Billion Ton Report*.
- The Partnership provided the opportunity to assemble and conserve a poplar germplasm collection that contains more than 20,000 clones for use in breeding programs. New clones resulting from crosses made using this germplasm collection have resulted in significantly improved cultivars that could be scaled up and deployed.
- Further validation of improved yields of new varieties/cultivars of biomass sorghum, energycane, hybrid poplars, and shrub willows was produced by the Partnership.
- The Partnership demonstrated the potential of energycane in northern parts of the southeastern U.S., particularly in relation to sugarcane varieties.
- For certain feedstocks (e.g., CRP mixed grasses, switchgrass, willow, and poplar), production economics were ascertained through Partnership field trials.
- Biomass samples collected as part of the Partnership are also being used to populate Idaho National Laboratory's Bioenergy Feedstock Library.
- Because of the size and scope of the Partnership, workforce development was a key component and outcome. Numerous undergraduate students (25), graduate students (15 MS and 8 PhD), and postdoctoral researchers (7) were trained by Partnership participants. This will help to prepare individuals for careers in the emerging fields of plant breeding, biomass production, feedstock supply logistics, and biomass conversion processes.
- More than 130 scientific publications have been authored by Partnership team members. Numerous other presentations, outreach publications, book chapters, and websites have also been developed to further the continued development of the bioeconomy.

Details for each species evaluated for the development of national yield potential maps are found in the following chapters as submitted by each team.

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Sun Grant/DOE Regional Feedstock Partnership

Final Technical Report

Introduction

The Sun Grant Initiative began in 2001 and was first authorized under USDA in 2002 through the Farm Bill. The mission of the Sun Grant Program is to:

(1) enhance national energy security through the development, distribution, and implementation of biobased energy technologies;
(2) promote diversification in, and the environmental sustainability of, agricultural production in the United States through biobased energy and product technologies;
(3) promote economic diversification in rural areas of the United States through biobased energy and product technologies; and
(4) enhance the efficiency of bioenergy and biomass research and development programs through improved coordination and collaboration between the Department of Agriculture, the Department of Energy, and the land-grant colleges and universities.

The program is led by five Regional Centers located at the following land-grant universities: the Pennsylvania State University (Northeast Regional Center), University of Tennessee (Southeast Regional Center), Oklahoma State University (South Central Regional Center), South Dakota State University (North Central Regional Center), and Oregon State University (Western Regional Center).

Publication of *The Technical Feasibility of a Billion-Ton Annual Supply* (Perlack et al., 2005; subsequently referred to as The Billion-Ton Study) by DOE and USDA in 2005, with its associated yield assumptions, led to the realization that these yield goals and assumptions required careful validation. The U.S. Department of Energy (DOE) and the Sun Grant Initiative initiated the Regional Feedstock Partnership (referred to as the Partnership) in 2007 primarily to address information gaps associated with the likelihood for realization of the sustainable and reliable annual production of a billion-tons of biomass to support the U.S. bioenergy industry by the year 2030.

The Billion-Ton Study identified the technical potential to expand domestic biomass production to offset up to 30% of U.S. petroleum consumption, while continuing to meet demands for food, feed, fiber, and export. However, after its release, a U.S. Department of Agriculture (USDA) Agricultural Research Service (ARS) assessment questioned the agriculture industry’s ability to increase yields and remove agricultural residues (such as corn stover) without reducing the productive capacity of agricultural lands.

Other questions arose about the likelihood that new practices (e.g., no-till) would be adopted to support such high annual biomass production, particularly in light of the traditional uses for crop residues (e.g., erosion control, feed, bedding, and as a soil amendment). Further, excessive removal of corn stover, which comprised a significant portion of the biomass resource in the Billion-Ton Study estimates, would deplete soil carbon content, an important indicator of soil productivity.

Recognizing an opportunity to leverage efforts, DOE's Bioenergy Technologies Office (BETO) collaborated with the Sun Grant Initiative (7 CFR §3430.1001) to form the Partnership to perform the fieldwork necessary to validate or modify the biomass feedstock yield assumptions described in the Billion-Ton Study.

In 2006 and 2007, workshops were held in each of the five Sun Grant regions to address the unique regional capacity to contribute to the vision of sustainably producing one billion tons of biomass feedstock annually by the year 2030. The workshops brought together research, government, industry, and other interest groups to work on a common framework for advancing biomass production and use in their respective regions. The potential for regional production of a diversity of biomass feedstocks was evaluated, obstacles and knowledge gaps were considered, research needs were identified, and key activities of the Partnership were outlined.

To generate the needed information, the Partnership was organized around primary biomass sources: 1) agricultural crop residues, 2) herbaceous energy crops including both annual and perennial grasses and CRP mixed grasses, and 3) short-rotation woody crops. Teams for these species included experts from Sun Grant Center and other Land Grant institutions and USDA-ARS. Other experts from national labs and industry were also participants in these teams where appropriate. To help translate new data into knowledge and transfer that knowledge to interested users, experts in Geographic Information Systems and outreach specialists were assembled as two separate teams to capture the program's progress and effectively disseminate it to the public.

One key recommendation from the 2006/2007 regional workshops was about the importance of conducting long-term field trials with corn stover and herbaceous and woody energy crops to validate yield potential assumptions. This required evaluation of conventional agricultural methods and best management practices, as well as consideration of crop development to increase yields of new energy crops.

Partnership field trials have provided yield and other information for key biomass feedstocks (i.e., corn stover, biomass sorghum, energycane, miscanthus, switchgrass, CRP mixed perennial grasses, willow, and poplar) across multiple years and in diverse environments across the United States (Figure 1).

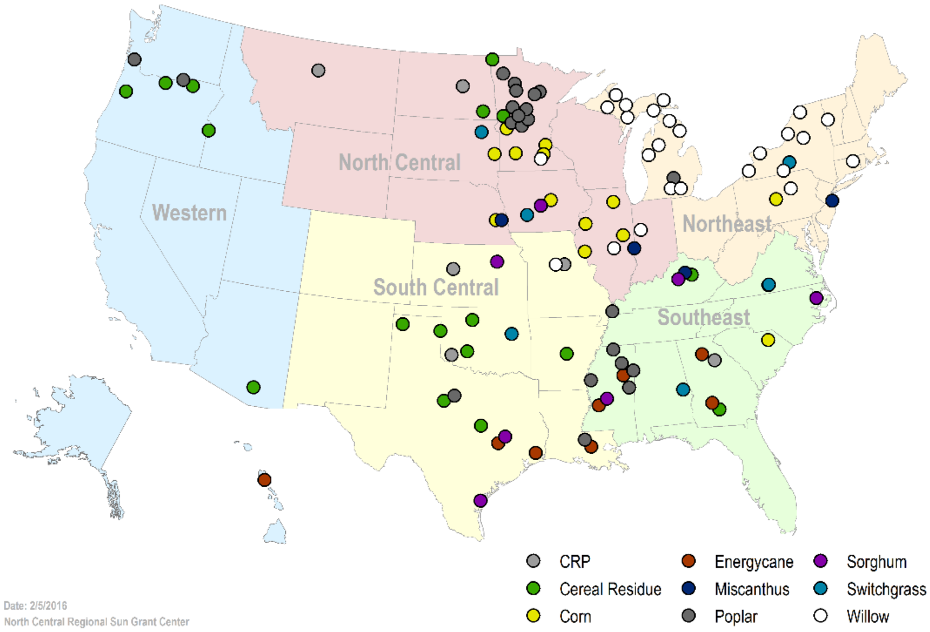

Figure 1. Location of Regional Feedstock Partnership multi-year field trials for diverse biomass feedstocks across the five Sun Grant regions.

Results from the Partnership have in fact helped validate the potential for sustainably producing one billion tons of cellulosic biomass annually. A better understanding of yield potential of various feedstock species across wide geographic gradients will be critical in biorefinery siting evaluations and in helping producers determine the economic potential of adopting dedicated bioenergy feedstocks into their business strategy. Farmers (including energy crop producers) and landowners will benefit from information on the different feedstock crops tested and from the management practices evaluated. Companies such as POET-DSM have used this information to create new jobs in rural areas as new supply chains for biomass feedstocks, biofuels, and bioproducts have been developed. Policymakers are using information from the Partnership to project future sustainable production of biomass across the country and to develop guidelines and policies related to development of the bioenergy, biofuels, and bioproducts industries.

In addition to yield potential, the Partnership addressed sustainability concerns that limit the use of corn stover as a feedstock, and convincingly documented the role of corn stover as a viable and significant bioenergy resource option for the United States. There is continued interest in the sustainable and renewable production of biofuels and chemicals amid concerns related to climate change, carbon sequestration in soils, and reliance on imported petroleum production for energy. The long-term nature of this research requires a concerted and continuous national effort (such as the Partnership) because yield stability and genetic effects under field conditions only become evident and relevant over multiple growing seasons.

More broadly, DOE investment in the Partnership has significantly advanced the knowledge base and public access to the data needed to begin answering many questions regarding sustainability and availability of biomass feedstock supplies and the production of bioenergy or other bioproducts. Data from the Partnership was incorporated into the Oak Ridge National Lab's Knowledge Discovery Framework (KDF). Other data related to biomass physical samples and their chemical characterization have been incorporated into Idaho National Lab's Bioenergy Feedstock Library.

Specifically, the Billion-Ton Study raised many scientific, technical, and economic concerns that were addressed through the sustained efforts of the Partnership. A well supported response to DOE concerns about soil organic matter raised by Partnership team members provided solid support for the practice of harvesting corn residues in some specific situations. This, in addition to populating the Bioenergy Knowledge Discovery Framework with field validation data, were direct, high-impact outcomes of the Partnership.

The Partnership has developed into a national resource for scientific, agronomic, and genetic information. Work completed in the Partnership trials have helped move each of these bioenergy crops down the commercialization path, thus supporting the Billion-Ton Vision. Further, Partnership data were instrumental in developing both the *Billion Ton Update* in 2011 and the *2016 Billion Ton Report*, which makes them significantly more credible resources for the community.

The following sections provide detail regarding each species evaluated in the Partnership as well as the development of yield potential maps for these feedstocks.

Cereal Crop Residues

Cereal Crop Residues Team: Russell Karrow, Oregon State University; Jing Dai, Wayne State University; Brent Bean and Gaylon Morgan, Texas A&M University; Bradford Brown, University of Idaho; William Bruening and Chad Lee, University of Kentucky; Jeff Edwards, Oklahoma State University, Michael Flowers, Oregon State University; Michael Ottman, University of Arizona; Joel Ransom, North Dakota State University; Jochum Wiersma, University of Minnesota.

Summary

Cereal residues could play an important role in terms of sustainably producing lignocellulosic feedstocks for the bioeconomy. This project evaluated the potential of cereal residues to be used as feedstocks, reevaluated the harvest index used in wheat specifically, and developed criteria for evaluation of the level of residue that may be removed without harming soil health.

Introduction

For the purpose of this project, cereals included wheat, barley, oats, triticale, and rye, as well as sorghum and millet grown for grain. In the last decade (i.e., 2004-2013), these cereals have been annually grown on more than 22 million ha (55 million acres) in the United States. In the previous decade, the combined acreage was more than 32 million ha (80 million acres) in some years. Initially, cereals might appear to be an obvious source of large quantities of biomass. Large areas of land are dedicated to cereal crops and these production areas are spread unevenly across the nation. These crops are typically harvested for their grain while plant stems and leaves (residues) are left in the field. There may be a general sense that these “waste” residues could be easily collected and used as a source of biomass; however, cereal growers, researchers, and extension and agricultural professionals recognize that this may not be the case depending on yield, soils, economics, etc. The vast majority of cereal acreage is grown under dryland conditions in areas with low to moderate rainfall and in environments where harsh winter or summer drought conditions could dramatically affect crop yields. In addition, many cereal growers participated in government crop support programs that require them to leave crop residues in place for soil conservation purposes. Without support program changes, these growers would not be able to harvest residues for biomass purposes.

Objectives

1. Establish a cereal project group among extension and research scientists across the US.
2. Query this group as to existing long-term cereal production plots across the US. The group will, in turn, query the managers of these plots as to their experiences with residue removal or addition in these plots. The question to be addressed is whether significant changes are expected in soil organic matter (SOM) or other soil characteristics with the removal of 50 or 100% of the straw in production areas where annual grain yields typically exceed 5,000 kg ha^{-1} (4,500 lb acre^{-1}).
3. Obtain available grain yield data (5-years) using National Agricultural Statistics Service county average data. Using the axiom that 45 kg (100 lb) of straw is produced for every 27 kg (60 lb) of grain (harvest index of 38% - typical for wheat and barley), areas with

less than 5,000 kg ha^{-1} (4,500 lb acre^{-1}; yields were excluded based on the assumption that approximately 680 kg ha^{-1} (1,500 lb acre^{-1}) and 1,360 kg ha^{-1} (3,000 lb acre^{-1}) is needed for soil conservation purposes and effective harvest, respectively.
4. Using geographic information system, plot areas of the United States where straw removal may be sustainably completed.
5. Collect a subset of straw samples from across the nation for analysis at the Idaho National Laboratory.

Methods

The cereals group took a different approach to the overall DOE program goals of assessing biomass production capability in each of the crop systems, assessing sustainability of biomass removal in each system, and determining biomass supply curves. A significant amount of information was already available on cereal residues, as were data from long-term plots that had been established to assess the effect of residue levels on soil quality. The group made the decision to utilize available information and trial work to address program goals rather than initiating new trials. Four primary task were completed to achieve these objectives including:

1. Mining long-term plot data for information on soil quality. A number of long-term (50+ years) cereal production plots exist in the United States and around the world. The current managers of these plots were asked to query their available data to address the effect of residue removal on soil quality. Could they document effects on SOM or other soil characteristics with the removal of residues?
2. Creating residue maps using existing data. Using NASS county data, grain yields for a 10-year period were obtained. Using this data and well accepted harvest index values (the amount of residue produced for each unit of grain), it was possible to create cereal residue maps over a 10-year period for all areas of the United States where NASS data were available.
3. Revising the harvest index value for wheat. At the beginning of this work the historic wheat harvest index value was 38% (Unkovich et al. 2010). Thus, for every bushel of wheat (27 kg [60 lb] of grain) produced, it was predicted that 45 kg (100 lb) of residue should also be produced [27/(45+27) = 38%]. Cereal agronomists in the group determined the harvest index for their plots over a 2-year period. The intent was to determine if the harvest index for newer commercial varieties of wheat was similar to or different than the historic and generally accepted 38% value.
4. Collecting sets of straw samples from across the nation for biomass quality assessment. Agronomists who conducted harvest index trials were asked to gather variety by site-specified straw samples after grain harvest, and to send these biomass samples for storage and possible chemical analysis at Idaho National Laboratory.

Results and Outcomes

Mining long-term plot data for information on soil quality. The Partnership sponsored a symposium on the topic of long-term plots and residue levels on soil quality, which was held at the American Society of Agronomy meeting in Pittsburgh, Pennsylvania, in 2009. Seven papers were given as part of this symposium, and their findings were later published as a symposium series in Agronomy Journal (Huggins et al. 2011).

From these papers and from other work that addresses the amount of cereal residue that must be left in place for soil maintenance purposes, one can draw the general conclusion that, in most situations, at least 3,370 kg ha^{-1} (3,000 lb $acre^{-1}$) of residue should be left on the ground to maintain soil organic matter content (Tarkalson et al. 2011). If mechanical harvest is then considered, agricultural engineers have estimated that at least 3,370 kg ha^{-1} (3,000 lb $acre^{-1}$) of residue must be available for collection for efficient harvest of the residue (R. Karrow, personal communication), resulting in a minimum residue yield of approximately 6,740 kg ha^{-1} (6,000 lb $acre^{-1}$) to sustainably and efficiently harvest the residue. Using the 38% harvest index value, this amount of straw corresponds to about 2,600 kg/ha of grain, which is significantly higher than most dryland acres are capable of producing in the average year.

Revising the harvest index value for wheat. To assess the accuracy of the historic wheat harvest index value (0.38), a survey was conducted on the aboveground biomass, grain yield, and straw yield of 12 cultivars of durum wheat, 40 cultivars of hard red spring wheat, 14 cultivars of hard red winter wheat, 174 cultivars of soft red winter wheat, 3 cultivars of soft white spring wheat, and 12 cultivars of soft white winter wheat in eight states (Arizona, Idaho, Kentucky, Minnesota, North Dakota, Oklahoma, Oregon, and Texas) from 2008 to 2010 (Wiersma et al. 2016). Across all wheat classes and locations, the harvest index ranged from 0.33 to 0.61, with an average of 0.45, which means that there is less residue per unit of grain harvest than was initially thought. By combining the minimum residue yield required for harvest (6,740 kg ha^{-1}; 6,000 lb $acre^{-1}$) with the updated, more realistic harvest index (0.45) for wheat, a net available residue map should only include those areas of the United States where grain yields exceed 5,526 kg ha^{-1} (4,920 lb $acre^{-1}$ or 82 bushels $acre^{-1}$). If a higher harvest index is used, then an even higher grain yield is needed to reach the minimum residue level). If a minimum 6,740 kg ha^{-1} (6,000 lb $acre^{-1}$) of residue value is used, this narrows areas available for wheat residue harvest to only several dozen across the entire country, which means that the contribution to annual sustainable cellulosic biomass feedstocks is negligible.

Creating residue maps using existing data. NASS county data was used to create grain yield maps and predicted straw yield maps using harvest index values. Maps for the 1999–2008 time period for barley, oats, rice, sorghum, wheat, and a combined straw total can be found on the Oregon State University website (http://sungrant.oregonstate.edu/projects/cereal-residue). Maps have been generated that show areas of the United States in which either a 5- or 10-year period predicted straw yields that exceeded 6,739 kg ha^{-1} (6,000 lb $acre^{-1}$), our suggested minimum harvest level. Reliable straw supplies would be essential for establishment of biomass processing plants. When looking at these maps and based on the assumptions made in these assessments, it is evident that despite the vast acreages and widespread production of cereal crops, there are few predicted locations for reliable biofuel production if cereal residues are used as the sole source of biomass. However, these resources could potentially be used to augment supplies of other feedstock materials. Two sample maps for wheat are shown in Figure 1.

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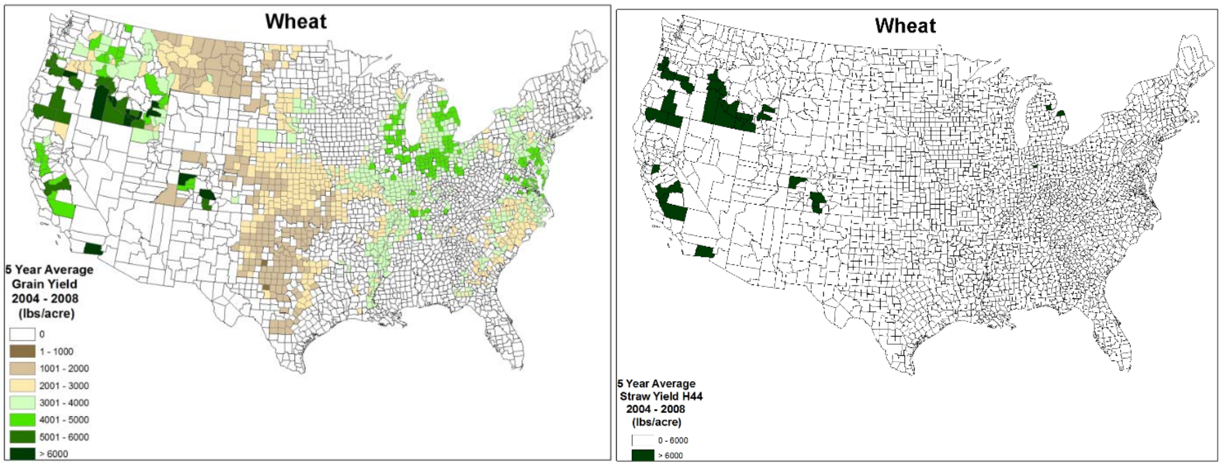

Figure. 1. Five-year average wheat grain yields (left panel) for the period 2004–2008, and potential wheat straw yield (right panel) using a harvest index value of 45%. The dark colored areas in the map on the right identify counties where straw yield would exceed 6,739 kg ha^{-1} (6,000 lb acre^{-1}) and, therefore, where it may be possible to consistently use wheat straw as a feedstock source for a biorefinery.

Collecting sets of straw samples from across the nation for biomass quality assessment. Several hundred straw samples were collected from existing variety testing plots for use in determining harvest index values. Collected samples were from common cereal check cultivars and from the most promising experimental lines. These samples have been cataloged and are in storage at Idaho National Laboratory for use in potential assessments of cereal residue-derived biofuel production.

Key Outputs

Peer Reviewed Publications

1. Gollany, H. T., R. W. Rickman, Y. Liang, S. L. Albrecht, S. Machado, and S. Kang. 2011. “Predicting Agricultural Management Influence on Long-Term Soil Organic Carbon Dynamics: Implications for Biofuel Production.” *Agronomy Journal* 103 (1): 234–46. doi:10.2134/agronj2010.0203s.
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Where Do We Go from Here

Site-specific information will be the key to using cereal residues as a biomass source for biofuel production. Site-specific management information and technologies, as opposed to "clear cut" strategies, may open residue harvest options. Differential harvest in fields on a real-time basis is now possible. Straw balers can be attached directly to combines, and grain yield sensing technologies could be used to allow baling of those areas of a field where straw loads are adequate to support both soil health and straw harvest. In areas where grain yields are high but not quite high enough to allow for every-year harvest of straw, differential harvest among fields over time may allow consistent biomass harvest in that area.

Cereal residues will be a part of the biomass and biofuels future of the United States but not on the scale that was originally envisioned by some. Site-specific, sustainable harvests are likely to be made in areas where cereal residues are a part of a mixed (or multiple) feedstock strategy for biomass processing plants.

References

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Corn Stover

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Summary

DOE investment in the Partnership has significantly advanced the scientific understanding and public access to data that are needed to assess the sustainability of biomass feedstock supplies and production of bioenergy or bioproducts. One of the findings crucial for securing private investments in the biofuel industry has been documentation that adequate quantities of corn stover can be collected to meet biofuel industry demands, while leaving enough residue on the field to protect and sustain the soil resource (Karlen et al. 2014; Johnson et al. 2014). This understanding has also led to development of sub-field, site-specific simulation models that are helping change perceptions within the agricultural community regarding this feedstock. This corn report summarizes 239 site-years of field research examining effects of corn stover removal rates at 36 sites in seven different states.

Introduction

Corn stover was selected as a focal point for one of the Sun Grant Regional Partnership teams because in the original Billion Ton Study (BTS) (Perlack et al., 2005), US Department of Energy (DOE) and Department of Agriculture (USDA) scientists had projected an annual supply of 446 million tons of crop residues (Figure 1), of which corn stover accounted for 75 million dry tons. The US Environmental Protection Agency (EPA) concurred that stover was indeed “the most economical agricultural feedstock … to meet the 16 billion gallon cellulosic biofuel requirement” (Schroeder 2011), and estimated that 7.8 billion gallons (29.5 billion liters) of ethanol would come from 82 million tons (74 million megagrams) of corn stover by 2022. Similarly, Lavigne and Powers (2007) evaluated ethanol production from corn grain and stover with respect to energy use, energy security, and resource conservation metrics, and concluded that using corn stover as a feedstock for advanced biofuels was more consistent with US national energy policy priorities than producing more biofuel from grain.

Since its hybridization in the 1930s, corn has become a highly-productive crop that is grown worldwide. Stover, the aboveground plant material left after grain harvest, was identified as a major feedstock for bioenergy production (Perlack et al., 2005) because of the vast area used for corn production in the US. From 2011 through 2013, the crop was planted on an average of 39.4 million hectares (97,272,000 ac) (USDA 2013) each year and produced an average of 419 billion liters (11.9 billion bushels) of grain. The total amount of stover can be estimated from grain yields using a harvest index (HI). The HI represents the ratio of harvestable biomass per unit of total biomass produced [*i.e.*, (stover mass) / (stover + grain mass)]. At plant physiological maturity, the HI value is approximately 0.5, though values can range from 0.50 to 0.55 depending upon plant population, genetics, location, and growing season characteristics. Assuming an HI of 0.50, the total stover quantity associated with 11.9 billion bushels of grain would be 255 million Megagrams (282 million tons).

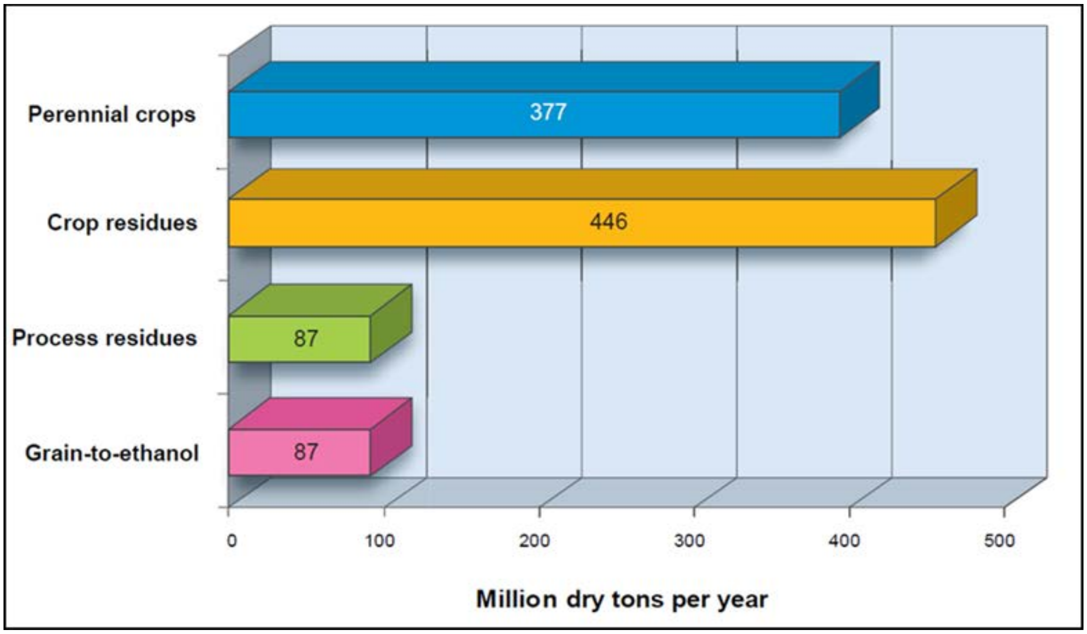

Figure 1. Bioenergy feedstock sources projected to be available by the mid-21st century in the original Billion Ton Study (BTS).

One advantage for using corn stover as a biofuel feedstock is the coproduction of food and fuel on the same land, thereby reducing concerns that biofuel production will result in significant and undesirable land use change (Searchinger et al., 2008). Another advantage is the well-developed nature of corn production and the industry that supports it throughout the US. The corn industry has excelled at discovering fundamental genetics, using that information to improve crop performance, and thus has the capacity to develop hybrids for coproduction of biomass and grain. Furthermore, the US Corn Belt has an extensive transportation infrastructure for moving grain and agricultural products from the field to storage and processing facilities, and ultimately to markets within and outside the region. Collectively, these reasons suggest that emerging cellulosic biofuel and bioproduct industries will be developed using corn stover in the Midwest (Moore et al., 2013). Other potential feedstocks (Figure 1) and their supporting biofuel and bioproduct industries will likely be more important in other regions of the country.

A closer examination of both the BTS and EPA estimates shows they assumed only 25 to 28% (rather than 100%) of the total US corn stover production would be available as a potential bioenergy feedstock. This assumption recognizes the most critical challenge and educational goal associated with production, collection, transport, storage, and conversion of biomass to energy: that ***corn stover is not a "waste" product associated with grain production***. Stover and all other crop residues are essential for protection against soil erosion, provision of soil carbon, and cycling of essential plant nutrients. Excessive biomass harvest for any use can easily disrupt these important functions, or ecosystem services. Figure 2 illustrates the inherent tension between economic drivers and ecosystem services associated with crop residue (Wilhelm et al., 2010).

Balancing the multiple ecosystem service needs for corn stover with the projected economic value of stover is critical to meet the nation's bioenergy goals. Early projections made in the BTS indicated that bioenergy production would be affordable when stover prices approximated \$35 ton (\$38.57 Mg^{-1}) at the farm gate. This projection was quickly challenged with data that showed macronutrient replacement costs alone (Hoskinson et al., 2007) could account for 30% of the projected stover price. Many farmers and researchers expressed concern that when all costs (equipment, labor, other management) and potential soil carbon declines were accounted for, there simply would be no incentive to harvest stover at the projected price. Others argued, however, that harvesting a portion of the stover could increase net farm income and simultaneously reduce crop residue management costs that currently range from \$45 to \$65 ha^{-1} (\$20 to \$30 ac^{-1}) (Duffy 2014).

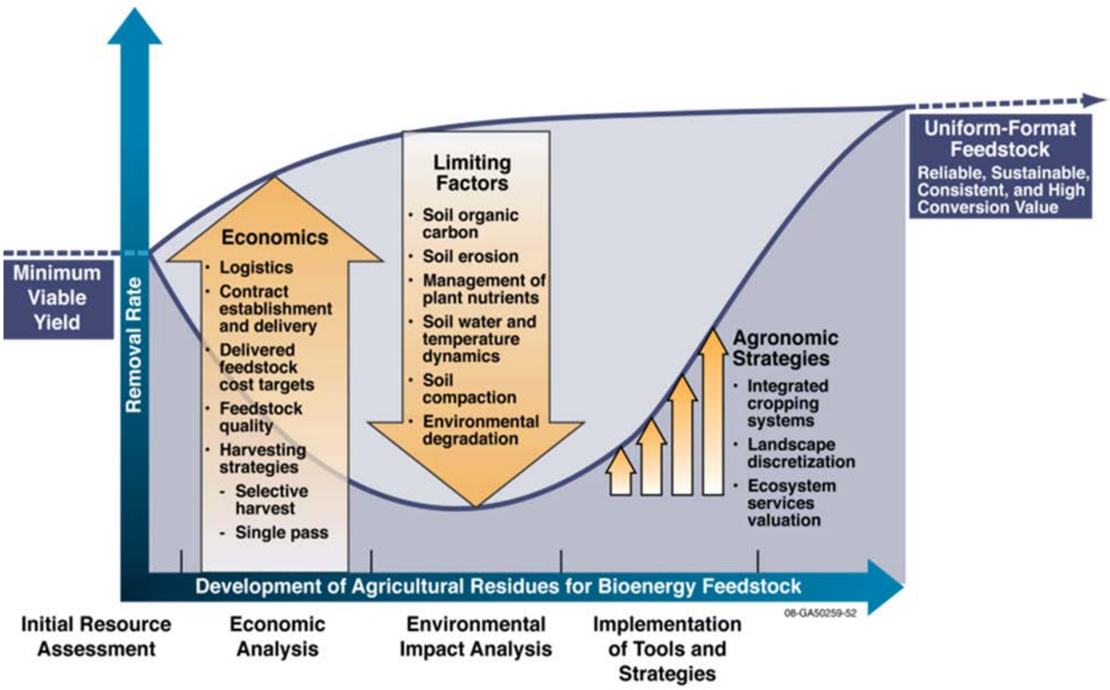

Figure 2. Economic drivers and limiting factors that ultimately determine sustainable quantities of biomass available for bioenergy or other bio-product uses (from Wilhelm et al. 2010).

Objectives

To help strive for a balance between the economic drivers and limiting factors (Figure 2), the USDA Agricultural Research Service (ARS) Resilient Economic Agricultural Practices (REAP) team (formerly the Renewable Energy Assessment Project team) linked with the Department of Energy Bioenergy Technologies Office's Sun Grant Regional Feedstock Partnership Corn Stover team. Specific objectives for this unique inter-agency, multi-institutional partnership were to: (1) implement field trials quantifying short- and long-term effects of harvesting corn stover on soil health and feedstock supplies, (2) develop methodologies and collect greenhouse gas (GHG) and water quality impact data associated with corn stover harvest strategies, and (3) develop tools to help guide implementation of sustainable agricultural residue harvest for bioenergy production.

Team members used new plot- and field-scale studies as well as several on-going experiments to address each specific objective. Research participants agreed upon a "core" experimental design consisting of: (1) replicated plots on highly productive soils representative for each location, (2) the use of no-tillage or the least amount of tillage needed to produce a successful corn crop, and (3) planting either continuous corn or both phases of a corn-soybean

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rotation at one or more field sites associated with each location. Three stover harvest strategies were implemented: (1) no removal, (2) moderate removal (nominally referred to as 50% harvest), and (3) maximum feasible collection which was dependent upon the location and available equipment, and nominally referred to as >90% harvest of the above-ground biomass. Baseline soil samples were collected to a depth of approximately one meter before and after the five-year study period. Additional soil and plant samples were collected when feasible.

Methods

New and existing SGRP Corn Stover Team studies provided 239 site-years of data from 36 replicated field experiments (Table 1), of which 31 were within the US Corn/Soybean Belt (Figure 3). Four Pennsylvania sites were selected because of increasing regional corn and soybean production and ongoing complimentary GHG research at those sites. Thirty-five studies were conducted on loam, silt loam, clay loam, or silty clay loam soils. The 36th site, located on a loamy sand in the Southeastern Coastal Plain near Florence, SC, was included because a similar multi-location stover harvest research was conducted at this location (Karlen et al., 1984) following the 1970s energy crisis. Furthermore, it was hypothesized that if adverse effects of stover harvest were going to be detected quickly, it would most likely occur on highly weathered loamy sand rather than on structured, heavy-textured soils in the Midwest.

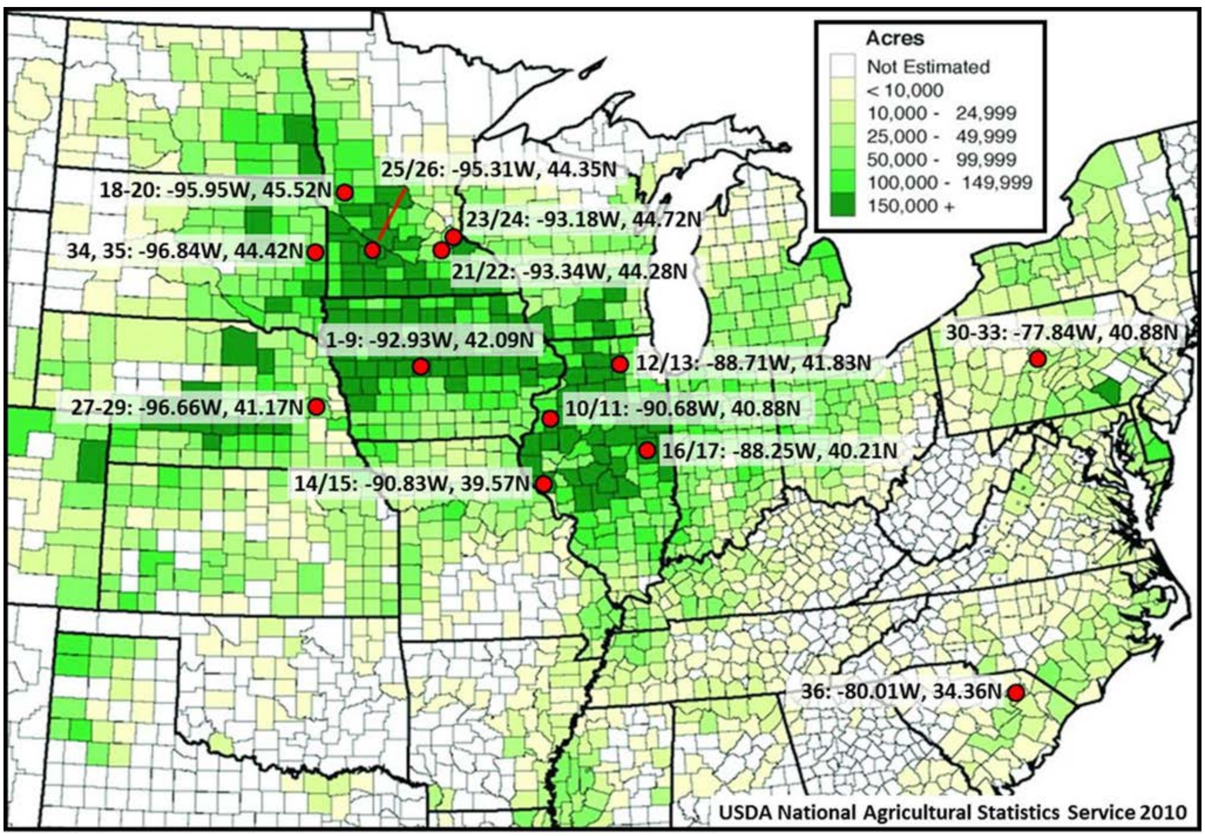

Figure 3. Location of Sun Grant Regional Partnership Corn Stover team research sites.

The study sites represented nine longitudinal bands (−96°, −95°, −94°, −93°, -91°, −89°, −88°, −79°, and −78° W) and six latitudes (45°, 44°, 42°, 41°, 40°, and 34° N) within the USA.

Primary tillage practices at the various sites included chisel plowing, moldboard plowing, disking, or strip tillage in addition to the core no-tillage treatment. Stover was harvested at the various sites for 5 to 12 years (Table 1). Commercial corn hybrids adapted and recommended for each location were used. Grain yields were determined either by hand harvesting a known area within each treatment or by using various commercial combines to harvest the entire plot. Yields were adjusted to a constant water content of 155 g kg^{-1} for comparison among studies. Stover yields were also determined in several different ways including (1) collecting samples from a 2-m length of row and fractionating the biomass into (a) plant parts above the ear shank, (b) below the ear shank, (c) cobs, and (d) grain; (2) attaching a canvas tarp to a commercial-scale combine to collect all residue from each plot, weighing all of the residue, and then dividing and returning a portion to each plot to create removal rates of 0, 50, or 100 % of the biomass; (3) harvesting with a single-pass combine designed to simultaneously collect grain and stover (Karlen et al., 2011); (4) collecting stover with a Hesston Stackhand[1] after combining the grain; or (5) collecting stover with a commercial flail chopper after combining the grain. Stover mass was calculated using dry weights from the hand-harvested samples, or for the mechanically collected material, it was determined by weighing, subsampling, and drying a representative sample to a constant weight, so that the quantities of stover removed could be compared among sites at a water content of 0 g kg^{-1}.

The range and overall mean values for grain and stover yields for all 36 studies, as well as mean N, P, and K removal for the 28 studies where those parameters were measured, were compiled and subjected to a meta-analysis using a SAS general linear model (GLM). Longitude, latitude, tillage, rotation, and stover harvest methods were evaluated for significant effects ($P \leq 0.1$). No-tillage treatments were compared to all other primary tillage methods as a group (i.e., conventional tillage). Stover harvest methods were also compared using two groups: "machine harvest" for locations measuring stover removal mechanically versus "calculated" for locations using a harvest index, summation of plant fractions, or the difference between the measured amount of residue after removal compared to the amount calculated to be present from the hand-harvested samples.

Results and Outcomes

Specific Objective 1 was fully met as documented by grain yields for no, moderate, and high stover removal treatments at the 36 SGRP sites (Table 2). Overall, grain yields ranged from 5.0 to 14.2 Mg ha^{-1} (80 to 227 bu ac^{-1}). Averaged across all sites, years, management practices, and hybrids, grain yields for the no-, moderate-, and high-stover harvest rates were 9.8, 10.1, and 10.1 Mg ha^{-1} (156, 160, and 160 bu $acre^{-1}$), respectively, with average stover yields of 0, 3.9 and 7.2 Mg ha^{-1} (0, 1.7 or 3.2 tons ac^{-1}), respectively (Karlen et al. 2014). Comparisons between the no-removal and moderate- or high-removal treatments showed a slight increase in grain yield of about 0.3 Mg ha^{-1} (4 bu $acre^{-1}$) across all sites, although results varied by location. When the no-removal treatment yields were lower, we assumed this was primarily due to residue management problems such as nitrogen immobilization and slower early-season plant growth and development.

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[1] Mention of a trademark or proprietary product is for information only and does not represent an endorsement by the USDA Agricultural Research Service, Department of Energy, or any university partner associated with this project.

The quantity of stover harvested at all sites showed substantial seasonal variability due to differences in growing conditions (i.e., planting dates, rainfall, temperature patterns, etc.), field-specific lodging caused by severe wind storms, and/or yield loss due to drought or hail. This variability confirms that corn stover harvest decisions must be site specific and even subfield specific to ensure that they are sustainable (Muth et al., 2012).

Stover harvest increased mean annual N, P, and K removal by an average of 24, 2.7, and 31 kg ha^{-1} (22, 2.4, and 28 lb ac^{-1}), respectively, for the moderate-removal treatment and by 47, 5.5, and 62 kg ha^{-1} (42, 4.9, and 55 lb ac^{-1}), respectively, for the high-removal treatment. Among locations, there was substantially more variation in nutrient removal than might be implied by the overall means. This presumably reflected hybrid differences in nutrient uptake and removal patterns controlled by corn genetics, biotic (e.g. soil pH, soil type, etc.) and abiotic (precipitation, temperature, etc.) factors, and their interactions. Another factor contributing to the site-specific variation was the time of sample collection. Stover nutrient concentrations at physiological maturity were generally higher than just before harvest (data not presented). Therefore, calculated nutrient removal rates for sites using nutrient concentration data at physiological maturity were also greater. Previous studies (Karlen et al., 2011) also showed stover nutrient removal to be lower when based on material collected during machine harvest than on hand-harvested samples collected near physiological maturity. This was especially notable for the moderate removal rate which consisted primarily of cob and upper plant parts, for which the upper parts were typically present at physiological maturity but tended to desiccate and break off by combine harvest. For the high-removal treatments, nutrient removal estimates were generally higher, presumably because higher amounts of soluble elements such as K, Cl, and NO_3^--N were still present in the lower stalk fraction (Hoskinson et al., 2007). Nutrient removal measurements were not made for Illinois sites because those studies were designed and implemented before the SGRP was formed and criteria for the core experiment were established.

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Table 1. USDA-ARS Resilient Economic Agricultural Practices (REAP) and Sun Grant Regional Partnership research sites (from Karlen et al., 2014).

Site	State	County	Dominant soil or soil association†	Texture‡	Elev§	Lat§	Long§	MAP§	MAT§	Management practices¶	Age
					m	°N	-°W	cm	°C		yr
1/2	IA	Boone	Clarion-Nicolet-Webster	SiCL, L	340	42	94	92	10	CP & NT / MM (single row)	5
3/4	IA	Boone	Clarion-Nicolet-Webster	SiCL, L	340	42	94	92	10	CP & NT / MM (twin row)	5
5/6	IA	Boone	Clarion-Nicolet-Webster	SiCL, L	340	42	94	92	10	CP / MM + 8 or 18 Mg ha^{-1} biochar	5
7	IA	Boone	Clarion-Nicolet-Webster	SiCL, L	340	42	94	92	10	NT / MM + CC	5
8	IA	Boone	Clarion-Nicolet-Webster	SiCL	293	42	94	92	10	CP / MM	8
9	IA	Boone	Clarion-Nicolet-Webster	L	350	42	94	92	10	CP / MS	8
10/11	IL	Warren	Muscatine	SiL	221	41	91	98	11.0	CP & NT / MM	6
12/13	IL	DeKalb	Flanagan	SiL	263	42	89	93	9.3	CP & NT / MM	7
14/15	IL	Pike	Clarksdale	SiL	198	40	91	92	11.6	CP & NT / MM	7
16/17	IL	Champaign	Drummer	SiCL	225	40	88	103	11.2	CP & NT / MM	7
18	MN	Stevens	Barnes-Aastad	CL, L	350	45	96	65	5.8	NT/95 / MS	8
19	MN	Stevens	Barnes-Aastad	CL, L	350	45	96	65	5.8	NT/05 / MS	8
20	MN	Stevens	Barnes-Aastad	CL, L	350	45	96	65	5.8	CP/05 / MS	7
21/22	MN	Rice	Garwin	SiC, SiL,	290	44	93	71	6.1	MbP & ST / MM	5
23/24	MN	Dakota	Waukegan	SiL	289	45	93	89	6.9	CP & ST / MM	5
25/26	MN	Redwood	Normania-Ves-Webster	L	344	44	95	71	6.9	CP & ST / MM	5
27	NE	Saunders	Yutan & Tomek	SiCL, SiL	366	41	96	76	10.5	NT/NI# / MM	12
28	NE	Saunders	Tomek & Filbert	SiL, SiL	366	41	96	76	10.5	NT/Irr# / MM	12
29	NE	Saunders	Tomek & Filbert	SiL, SiL	366	41	96	76	10.5	D/Irr# / MM	12

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Site	State	County	Dominant soil or soil association[†]	Texture[‡]	Elev[§]	Lat[§]	Long[§]	MAP[§]	MAT[§]	Management practices[¶]	Age
30	PA	Centre	Opequon-Hagerstown complex	SiCL, SiL	352	41	78	97	9.7	NT / MM	5
31	PA	Centre	Opequon-Hagerstown complex	SiCL, SiL	352	41	78	97	9.7	NT / MM +CC	5
32	PA	Centre	Opequon-Hagerstown complex	SiCL, SiL	352	41	78	97	9.7	NT / MS + CC	5
33	PA	Centre	Opequon-Hagerstown complex	SiCL, SiL	352	41	78	97	9.7	NT / MS + CC (twin-row)	5
34	SD	Brookings	Kranzburg-Brookings	SiCL	490	44	96	58	6.1	NT / MS	12
35	SD	Brookings	Kranzburg-Brookings	SiCL	490	44	96	58	6.1	NT / MS+CC	8
36	SC	Darlington	Goldsboro-Lynchburg-Coxville	LS	140	34	79	130	17	NT/IRSS / MM	5

[†] Soil classification information for each site is provided in Appendix Table A1.

[‡] Texture – L = loam; CL = clay loam; SiC = silty clay; SiL = silt loam; SiCl = silty clay loam; LS = loamy sand

[§] Elev = elevation above mean sea level; Lat = latitude; long = longitude; MAP = mean annual precipitation; MAT = mean annual temperature

[¶] Crop management practices –

Tillage – CP = chisel plow; NT – no-tillage; ST – strip tillage; MbP – moldboard plow; D – disking; IRSS – in-row subsoiling (done beneath each row at planting); Note – the /95 and /05 indicate the studies were initiated in 1995 and 2005, respectively

Crop -- MM – continuous corn (maize); MS – corn (maize) /soybean; CC – cover crop;

[#] Water management (imposed only at sites 27 – 29) – NI = non-irrigated; Irr = irrigated

Table 2. Mean corn grain and stover yields for rotation and stover harvest method associated with a meta-analysis of data representing 239 site-years of replicated field studies from 36 research sites associated with the USDA-ARS Resilient Economic Agricultural Practices (REAP) and Sun Grant Regional Partnership (from Karlen et al., 2014).

Factor		Grain†		Stover‡	
Removal Rate	None	Moderate	High	Moderate	High
Rotation		Mg ha^{-1}		Mg ha^{-1}	
Corn/Corn	9.84	10.19	10.24	4.01	6.97
Corn/Soybean	9.08	9.24	9.00	3.45	7.89
$LSD_{(0.10)}$	NS	NS	NS	NS	NS
Harvest Method					
Machine	9.19	9.81	9.62	3.72	6.62
Calculated	10.16	10.15	10.34	4.06	7.79
$LSD_{(0.10)}$	NS	NS	NS	NS	0.98

† Grain yields at a water content of 155 g kg^{-1}
‡ Stover yield at 0 g kg^{-1} water content

In an ancillary experiment conducted at University Park, PA, two years of stover data (2009–2010 and 2010–2011) showed significant yield and nutrient removal differences when autumn versus spring harvest dates were compared. They reported mean stover yields of 8.25 and 5.98 Mg ha^{-1} ($LSD_{(0.05)}$ = 0.74) for the autumn and spring harvests, respectively. The associated N, P, and K removal rates were 60, 4.7, and 60 kg ha^{-1} when harvested in autumn compared to 40, 3.25, and 18 kg ha^{-1} when harvested the following spring ($LSD_{(0.05)}$ = 11, 0.78, and 12 kg ha^{-1}, respectively) (Karlen et al., 2014). This comparison helps illustrate the degree of change associated with measuring nutrient composition in corn stover harvested at different times after physiological maturity.

Comparisons between sites with continuous corn versus a corn/soybean rotation showed no significant differences (Table 2) even though mean rotated corn yields were lower for all three stover harvest treatments. Since corn grain yields are generally 5 to 20% higher when grown in rotation (Karlen 2004), this unexpected response is presumably because most crop rotation sites were located in the northern and western portions of the study area (Table 1).

The Corn Stover Team fulfilled Specific Objective 2 by conducting several ancillary studies on a broad range of environmental issues (e.g., soil organic carbon (SOC), GHG emissions, microbial communities, cover crops, etc.). Data were collected from both new and existing field studies that were established prior to the 2005 BTS (Perlack et al., 2005) In fact, the ARS-REAP team (http://www.ars.usda.gov/research/programs/programs.htm?np_code=212&docid=21224) was organized to provide a coordinated response to that report. Although the BTS had made a reasonable attempt to balance the need for crop residues to protect soil resources from wind and water erosion, several ARS scientists were concerned that other crop and soil productively issues, especially the soil organic carbon (SOC) balance (Wilhelm et al., 2004) was not given enough emphasis to ensure the sustainable development of the bioenergy industry. In 2005, it was not known if returning enough residue for erosion control was sufficient to prevent a loss in

SOC. A study published by Wilhelm et al. (2007) demonstrated that indeed it could take considerable more residue to sustain SOC compared the amount needed for erosion control. Fortunately, enhancing the ARS-REAP efforts through the ARS-REAP/Sun Grant Regional Partnership Corn Stover team has provided sufficient technology to help ensure the critical feedstock supply needs can be met in an agronomically, environmentally and economically sustainable manner. An outcome of the Corn Stover Team partnership was the inclusion of SOC constraints in the *Billion-Ton Update* (BT2; US DOE, 2011) and Biomass Research and Development Board (BRDB) (BRDB 2011) reports. This optimism was confirmed by the 2014 launch of three full-scale corn stover bioenergy conversion facilities in the US (i.e., POET's Project Liberty in Emmetsburg, IA; Abengoa's Hugoton, KS biorefinery; Dow/DuPont's Nevada, IA biorefinery). Indeed, this is a strong market signal, but it also indicates that cellulosic feedstock supplies must increase dramatically to meet bioenergy production demand in a sustainable manner.

The results from SGRP/REAP studies emphasize the extreme variability associated with different soils, weather patterns, and crop growth conditions (Karlen et al., 2014). For 35 studies, the estimated average <u>minimum</u> residue return rate to the soil was 6.4 ± 2.2 Mg stover ha^{-1} yr^{-1} (2.85 ± 0.98 tons ac^{-1} yr^{-1}), below which there is increasing probability for significant SOC reductions and a subsequent loss of ecosystem services the soil provides (Johnson et al., 2014). A conservative estimate of minimum average grain yield required before any stover could be removed is based on the upper residue return value (3.83 tons ac^{-1} yr^{-1}), correcting for water content in a standard bushel of grain (15.5% or 155 g kg^{-1}), and assuming a HI of 0.5. This estimate leads to the conclusion that fields with average corn grain yields less than 157 bu ac^{-1} yr^{-1} (8.8 Mg ha^{-1}) should leave all stover in the field (e.g. none should be harvested). Although this value illustrates the academic calculation that many will make, in reality, ***the most important point of this comprehensive SOC and crop residue review is that the extreme variability refutes any notion that there is a universal minimum residue requirement.*** Instead, the Corn Stover Team unanimously recommends that crop residue harvest decisions must be made at the local level if not the individual field, or better yet, subfield management level (Johnson et al., 2014).

Corn Stover Team studies were also conducted to quantify SOC effects of stover harvest on soil aggregation and particulate organic matter. For three Regional Partnership sites in the western Corn Belt (Brookings, SD; Morris, MN; and Ithaca, NE), soil aggregate distribution was negatively impacted by stover harvest unless another carbon source, such as cover crop residue, was added to replace the harvested stover carbon (Osborne et al., 2014). Quantitatively, this soil physical property change was confirmed by lower amounts of soil organic matter (SOM), fine particulate organic matter (fPOM), and total particulate organic matter (tPOM) when crop residues were harvested. Studies quantifying crop residue harvest effects on the least limiting water range (LLWR), an indicator of soil compaction effects, showed that soil physical properties were degraded by loss of SOM due to tillage and possibly erosion. These results again supported the conclusion that crop residue removal rates should be limited to levels that maintain or even increase SOM levels.

Another targeted, multi-location study across Corn Stover team partnership sites included an examination of crop residue harvest impacts on the soil microbial community. Soil microbial fatty acid and DNA analyses from four Partnership locations (Brookings, SD; Florence, SC; Ithaca, NE; and Morris, MN) with contrasting soils, climatic conditions, and substantial differences in both SOM and pH showed that high (~7.2 Mg ha^{-1} yr^{-1}) stover harvest rates tended to reduce the fungal-to-bacterial ratios in all soils (Lehman et al., 2014). This is an important metric because soil fungi are known to secret mucilage capable of binding particle together into aggregates. This response was also consistent with the previously reported decrease in aggregate stability and increase in erodible soil fraction.

Greenhouse gas emissions were estimated using static chamber data from nine corn production systems that were being managed using various crop residue and tillage management practices across the Corn Belt. Although GHG emission were highly variable, moderate and high stover harvest rates generally decreased total soil CO_2 and N_2O emissions by 4 and 7%, respectively, when compared to no stover removal. Decreased GHG emissions were attributed to lower stover C and N inputs and possible microclimate differences due to changes in soil cover (Jin et al., 2014). The GHG knowledge base was further increased by summarizing automated continuous chamber CO_2 and N_2O flux data collected between spring 2010 and spring 2012 for three levels of stover harvest (none, moderate, and maximum collectable). The results show no significant difference in N_2O emission as a function of stover harvest, but CO_2 loss from the high removal plots was slightly lower than from the non-removal plots. The CO_2 emission difference between the two treatments was much smaller than the amount of C removed with the stover, implying that C was being lost from the high-removal plots; indeed, this phenomenon was confirmed by rigorous soil sampling.

Economic assessments were recognized as being crucial for fledgling cellulosic bioenergy industries to succeed and also for encouraging producers to adopt more sustainable soil and crop management practices, including the use of no-tillage and cover crops – two practices that will also enhance opportunities for stover harvest. To ensure that such assessments are valid and representative, economic analysis could not begin until a substantial portion of the five-year Regional Partnership measurements and management information were collected and compiled in a common database. To date, the team's initial evaluations show that breakeven field-edge biomass prices range from \$26 to \$42 Mg^{-1} in Iowa and from \$54 to \$73 Mg^{-1} in North Dakota (Archer et al., 2014). Additional economic assessments are planned as the team seeks resources to continue the multi-location effort stimulated by Sun Grant Regional Partnership investments.

Specific Objective 3 was fulfilled through excellent collaborations between ARS, university agricultural scientists, and DOE engineers from INL. Throughout this five-year study, field data were used to develop and validate a first-generation "corn stover tool" that ultimately gave rise to the Landscape Environmental Assessment Framework (LEAF) (Muth et al., 2012). The LEAF tool has been made available to private sector investors in the bioenergy industry through an application for smart phones/tablets that enable in-field determination of the general site suitability for stover harvest. A version of LEAF is also being used to guide on-the-go, site-specific single-pass corn grain and stover harvest as well as subsequent tillage operations. The development of LEAF illustrates the need for more subfield-scale research to understand how interactions between location, soil resources, weather, and management (e.g. tillage, hybrid selection, cover crops) affect the sustainability of corn stover management.

Synergistic Activities:

With the exception of the Illinois sites, which were incorporated into the project in the fourth year, core data from the original Corn Stover team sites have been contributed to the DOE Knowledge Discovery Framework (KDF) database.

Dried and ground biomass samples from participating sites were also submitted to the INL to develop a new near-infrared (NIR) interpretation algorithm to quantify plant constituents needed to estimate their value for conversion to bioenergy. This effort was led by Dr. Gary Gresham and his colleagues. The samples also provided an opportunity to bridge this Sun Grant – ARS activity with the NIFA-funded CENUSA collaborative agricultural project (CAP), which supplied student intern labor to assist with analysis. To date, two manuscripts reporting on the chemical characterization of the various corn stover fractions have been prepared and submitted for publication. Data have also been contributed to the INL biomass characterization library (https://bioenergylibrary.inl.gov/Home/Home.aspx) which can also be accessed through the KDF (website under development).

Key Outputs

As of July 2014, at least 36 peer-reviewed manuscripts, 8 outreach publications, 20 proceedings papers, and 39 presentations were delivered. The Corn Stover Team funds also helped support 8 graduate students and another 10 undergraduate students.

This multi-location, multi-agency project has produced a tremendous amount of publicly available information to support cellulosic bioenergy and bio-products industries. We consider the 239 site-year dataset to be one of the most comprehensive research efforts ever conducted to provide replicated field validation for projections such as those in the revised Billion Ton Report (US DOE 2011). However, those reading this report are cautioned not to use these multi-location mean values for site-specific planning or developing corn stover harvest strategies. As pointed out for grain and stover yields, nutrient removal, SOC changes, soil structure changes, microbial communities, GHG emissions and several other indicators, spatial and temporal variability are tremendously wide ranging and must be accounted for in developing local stover management plans.

Numerous peer-reviewed technical journal publications, outreach pamphlets, proceedings papers, and presentations have been prepared by every Corn Stover team member. Several of the outputs identifiable through the ARS information system are listed below. Another major contribution summarizing Corn Stover team research is in a special issue of BioEnergy Research that includes 14 papers providing substantially more detail than has been covered in this SGRP species report. Undoubtedly, a significant portion of this information will also be used to continue developing and validating simulation models such as DAYCENT and LEAF. The data is also being used to strengthen interactions among the ARS and other DOE labs and with the NRCS.

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Where do we go from here?

Overall, the REAP/Regional Partnership Corn Stover team has been highly successful in providing information needed to help establish the fledgling bioenergy industry in the USA. Fiscal resources provided by the DOE have been highly leveraged by a very dedicated group of ARS scientists and their university partners. As stated by a 2013 DOE review team, “this project provided a broad assessment of stover yield potential, feedstock characteristics and sustainability metrics. LEAF provides an analytical framework to explore the balance between economic drivers and sustainability constraints. Development of the database for further meta-analyses by team members is noteworthy. Additional research is needed to quantify effects of cover crops, perennial segments within extended rotations, as well as proper utilization of animal manures.

Future work should specifically address development of best management practices for biomass harvest, while also improving soil and water quality relative to conventional production practices." These recommendations are being pursued by developing investment opportunities for industry, nonprofit, government agency, and other partners through the Agricultural Technology Innovation Partnership (ATIP) Foundation. Public support for the ATIP and a continuation of the Sun Grant Association were both encouraged in the US Farm Bill: http://docs.house.gov/billsthisweek/20140127/CRPT-113hrpt-HR2642-SOM.pdf

This statement directly affects future activities associated with the Corn Stover team because one of the two initial public-private-partnership (PPP) projects, endorsed by the Farm Bill language, is designed to fulfill the recommendations made by the 2013 DOE review team. To date, seven founding partners (bioenergy and other industries benefiting from the Corn Stover Team technology) have agreed to contribute funds for continuing several site-specific studies and cross-location assessments to address additional critical needs (i.e., cover crops, animal manures, extended crop rotations, soil health, reduced tillage intensities, SOC assessments, etc.). Nine initial Statements of Work (SOW) from core and ancillary Regional Partnership sites have been submitted to the ATIP Foundation. A USDA Liaison Committee working with the ARS National Program Officers and the PPP participants are currently reviewing these proposals. Other components of the SOWs may be of greater interest to other potential partners, including other government agencies such as the DOE and non-profit organizations. These developments are extremely encouraging to Corn Stover Regional Partnership team members and are a critical continuation of the initial DOE investment in cross-location research activities coordinated by the Sun Grant Association for this important bioenergy feedstock species.

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Sorghum

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Summary

Sorghum [*Sorghum bicolor* (L.) Moench] is one of four herbaceous dedicated bioenergy crops identified in the Regional Feedstock Partnership as critical to producing 1 billion tons of biomass annually (Gill et al., 2014). Of these four crops, sorghum is unique as it is a drought tolerant, annual crop established from seed that is readily tractable to genetic improvement. The purpose of this study was to assess the yield potential and stability of sorghums grown across diverse production environments in the USA. For this study, six sorghum genotypes (one cultivar, five hybrids) were grown in yield trials in seven locations in six states for five years (2008-2012). Variation in dry and fresh yield was attributable to not only genotypes, but also to the effects of year, location, and year x location. Even with the highest yielding genotype, environmental conditions were a major factor in determining the yield in a given year. This variability affects the consistency of the biomass supply for ethanol production. In general, the southeastern U.S. had the highest mean yields for fresh weight and dry weight, indicating that this area may be the most reliable for biomass production. Significant variation was detected among genotypes for fresh weight, dry weight, moisture content, and brix, revealing that sufficient variation within sorghum exists for continued improvement and that certain hybrids are more tractable for biomass/bioenergy production. With dedicated bioenergy sorghum germplasm and proper production environments, sorghum will be a valuable tool in the goal of the sustainable production of one billion tons of dry biomass each year in the USA.

Introduction

Sorghum is an important crop species in the U.S. and around the world. Because of its substantial heat and drought tolerance, sorghum production is traditional in semi-arid, sub-tropical and tropical regions of the world. In addition to abiotic stress tolerance, sorghum is very responsive (in terms of productivity) to more favorable conditions. Sorghum is grown as a grain forage, syrup and more recently, energy crop. There has been renewed interest in sorghum as a bioenergy crop for several reasons. First, sorghum has an established production history as a crop which eliminates the time required for crop domestication, production and market development and reduces concerns regarding producer acceptance and adoption. Second, there is a well-established seed industry that is proficient in genetic improvement and seed production. Third, the annual nature of the crop, while a detraction to some, increases the speed and efficiency at which sorghum can be genetically improved and deployed in a production environment. Finally, sorghum has evolved as a standard genetic model for the improvement of bioenergy crops. Combined, these factors confirm that sorghum will play an important role in the development and evolution of dedicated energy crops.

Sorghum is a genetic diploid with base chromosome number of n=10 and 2n=2x=20 and a genome size is that is larger than rice but substantially smaller than corn or other grass species. Botanically, sorghum is a typical grass with a deep and fibrous root system, primary culm and the capacity for both basal and axillary tillering. Reproductively, sorghum has a complete flower that is exerted through the top leaf sheath prior to anthesis. Sorghums are predominantly self-pollinated but outcrossing does occur at rates between 2-30% although the precise frequency is a function of both genotype and environment. While it is self-pollinated, the crop is highly amenable to commercial hybrid seed production through the use of cytoplasmic male sterility systems.

As an energy crop, sorghum is unique in that there are several different types of sorghum that are and can be used for biofuel or bioproduct production. Grain sorghum is now a significant contributor to biofuel production in the US. Sweet sorghum is being deployed in conjunction with sugarcane in sugar to ethanol conversion facilities and biomass sorghums produce significant quantities of structural carbohydrates. Different types of forage sorghums, are being integrated into the biomass sorghums which are photoperiod sensitive meaning that, when grown in long day environments, they do not flower and can accumulate large quantities of biomass.

Regardless of type, sorghum is a seed-propagated annual crop which is grown primarily as a hybrid crop. Unlike several other bioenergy crops, stand establishment is typically not a major problem and the costs associated with planting sorghum are significantly less than other crops. Stand establishment of sorghum requires a well- prepared seedbed and adequate moisture to initiate the germination process. Therefore, the planting and stand establishment process in energy sorghums is similar if not identical to that of grain sorghums. From a productivity standpoint, plant population and row spacing are probably the most critical management factors and the optimum density and spacing depends on the type of production system. For example, sweet sorghum processors prefer thick stalks that mimic sugarcane, however higher yields are typically associated with higher plant densities. Furthermore, row spacing modifications are limited to those that fit with existing harvesting equipment. Optimizing population and distribution to fit production programs is of significant importance.

Sorghum is an efficient user of soil nutrients because its fibrous root system is very efficient at taking up nutrients within the root profile of the crop. Relative nutrient requirements depend on the type of sorghum and the yield potential of the crop. Nitrogen requirements in sorghum depend greatly on the type of sorghum being produced. For example, biomass and sweet sorghums required less nitrogen per ton of biomass produced compared to grain crops (Grain sorghum and corn). However, overall tonnage of these types was higher and the optimum fertilization for grain and biomass sorghums were similar for the growing season. As with any biomass crop, removal of all biomass from the production environment is not sustainable long term due to removal of soil nutrients and carbon. In addition, the long duration of growth for biomass sorghums is mitigated by the recycling of nitrogen from stems to leaves such that harvested stems contain relatively low N concentrations. Further research in this area will be of primary importance in optimizing productivity and nutrient utilization in sorghums.

Sorghum is known and grown for its ability to use water efficiently and to maintain productivity through periods of drought, which is important because biomass production from energy crops is expected to be rainfed. Within sorghum there is significant variation for tolerance to drought among both sorghum types and genotypes within each group. There are differences in the level of drought tolerance between sweet sorghums and biomass sorghum. Since biomass sorghums do not initiate reproductive growth until very late in the season (and in some cases, not at all), they have an inherently greater level of tolerance to water stress during the growing season than either grain or sweet sorghums.

As a crop species domesticated thousands of years ago and distributed throughout the world, sorghum is a host plant for an array of plant pests and pathogens but only a few have real and significant economic importance. For bioenergy sorghum, several diseases and insects will be or already are of substantial economic importance. Anthracnose is probably the most significant but root and stalk rots, ergot, head smut, and downy mildew may also be problematic. For most of these diseases, breeding for host-plant resistance remains the most cost-effective method of disease control. Insect pests that affect the stalk will be important; stalk borers of all types (mostly *Lepidoptora* sp.) will be of significant importance.

Within energy sorghums, delayed maturity is consistently associated with increased yield so long as moisture and nutrients are not limited and initiation of harvest should coincide with near optimum yields. Another consideration in harvest of the biomass sorghum is that there will be some form of continuous harvest because storage of large stockpiles of biomass is not economically or logistically feasible. Given that there was a diversity of sorghum types, ranging from single cut to multiple cut hybrid, it is critical to identify when it is best to harvest and how to manage that production. Multiple cut sorghum hybrids can be harvested up to four times in a single year and these types produce higher yields under multiple harvest than single harvest. However, this production system has higher nitrogen requirements and the composition is typically leafier and therefore has higher nutritive value. Single cut hybrids accumulate higher biomass yields on a per cut basis but they are not selected for regrowth and typically do not produce ratoon crops. For sweet sorghum, ratoon crops have been minimally successful. In the US, up to a three month harvest window with maximum yields in the middle of this season can be expected for sweet sorghum.

Sweet sorghum and biomass sorghum breeding efforts focus on hybrid development. Parental lines are being developed and studies of developed hybrids indicate improvement in productivity. In addition a range of maturity hybrids will be necessary to ensure optimized hybrids for a continual harvest season. Breeding for biomass production uses similar approaches currently used to produce hybrid forage sorghums except that they are bred for the single harvest management scheme. Most of the breeding effort will be focused on the pollinator parent because existing seed parental lines possess most of the traits needed in a seed parent for biomass hybrids. Equally important to the development of the yield and quality potential of energy sorghums is the inherent protection of that yield potential. Thus, breeding for stress tolerances, both abiotic and biotic is critical for both adaptation and productivity.

Objectives

The objective of this project was to establish and perform replicated field trials of energy sorghums to gather biomass production, compositional quality and sustainability data that documents biomass yield at different regional locations for assessing potential expansion of sorghum as a bioenergy feedstock resource.

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Methods

Six sorghum genotypes were evaluated in seven environments over five years. The six genotypes evaluated were Graze All, a PI sorghum-sudan forage hybrid; Graze N Bale, a PS sorghum-sudan forage hybrid; TX08001, a PS bioenergy hybrid; M81-E, a moderately PS sweet sorghum variety; Sugar T, a moderately PS sweet sorghum silage hybrid; and 22053, a moderately PS brown midrib (bmr) silage hybrid. TX08001 was developed by Texas A&M Agrilife Research, M81-E was developed in Mississippi by the USDA-ARS [3], and the remaining four hybrids are produced and marketed by Advanta, Inc. primarily as forage sorghums for silage, green chop, grazing and hay. Given this background, it must be noted that they were not developed specifically for bioenergy. However, at the initiation of this project, numerous groups were utilizing these types for bioenergy uses due to the paucity of energy sorghum types and that is why they were included in this study.

The seven locations used for testing were: Manhattan, Kansas (KS); College Station, Texas (CS); Corpus Christi, Texas (CC); Ames, Iowa (IA); Lexington, Kentucky (KY); Raymond, Mississippi (MS); and Roper, North Carolina (NC) (Figure 1). All yield trials were rain fed; no supplemental irrigation was used in any location. In all locations and years, trials were planted in a randomized complete block design but plot size and number of replications varied across locations due to space availability and management capacity. Standard production practices specific to each location were observed for fertilizer, tillage, and herbicide application. Target plant densities were 125,000 plants per hectare for the sweet sorghums (Sugar T and M81-E), 150,000 plants per hectare for the bioenergy types (22053 and TX08001), and 200,000 plants per hectare for the forage sorghums (Graze All and Graze N Bale). Agronomic traits evaluated at each location were fresh weight, moisture concentration of the biomass, dry weight, and brix. Biomass samples were collected at harvest and dried in a forced air oven at 60°C for a minimum of 72 hours, to obtain the moisture concentration and dry weights. Several environments in various years were lost due to insufficient rainfall or inconsistency in the data quality.

Figure 1. Map showing Sun Grant Regional Feedstock Partnership energy sorghum evaluation sites.

Dried biomass samples were collected at harvest, and ground in a Wiley Mill (Arthur H. Thomas Co., Philadelphia, PA.) to a particle size small enough to pass through a 2 mm screen. Samples were analyzed for ash, protein, lignin, glucan (cellulose), and xylan (hemicellulose) composition on a FOSS near-infrared XDS Rapid Content Analyzer (FOSS NIRSystems, Inc., Laurel, MD.). Spectrum data were converted into composition data using a calibration curve developed by the Texas A&M University sorghum quality lab and the National Renewable Energy Laboratory (Wolfrum et al., 2013). Several environments in various years were lost due to insufficient rainfall or inconsistency in the data quality. All analyses of variance were conducted using the mixed models procedure of SAS version 9.3 (SAS Institute, 2011). Replications, nested within year and location, were considered a random effect; all other sources of variation were considered fixed effects. Multiple comparison procedure tests were conducted using the general linear model procedure of SAS version 9.3. GGE biplot software was used in an effort to subdivide the locations into mega environments in order to make inferences about the feasibility of producing sorghum as a dedicated bioenergy crop in different areas of the USA (Yan and Tinker, 2006). Rainfall is reported as the total amount received at each location for the complete calendar year (January-December).

Repeatability values were calculated using the equation

$$x = \frac{\sigma_g^2}{\frac{\sigma_e^2}{rt} + \frac{\sigma_{ge}^2}{t} + \sigma_g^2}$$

where χ is repeatability, σ_g^2 is genetic variance, σ_e^2 is the error variance, σ_{ge}^2 is the genotype x envrionment variance, *r* is the number of replications, and *t* is the number of locations.

In conjunction with modelers at Oregon State University, national yield potential maps were developed using PRISM-ELM (Daly et al., 2018).

Results and Outcomes

Relative Effects of Genotypes and Environments

During the years of evaluation the seven locations used for this study varied widely in terms of annual rainfall, seasonal temperature, and length of growing season and represented different adaptation zones. Furthermore, within the years tested, rainfall at individual sites varied widely from year to year.

In the combined analysis, the main effects of year, location, and genotype, and many of the interaction terms were significant. The majority of the variation observed in the data from this experiment was attributed to the effects of year, location, and year x location (Table 1). The large variability due to environment may require that breeding efforts be conducted on a more regionalized basis instead of breeding germplasm adapted across the USA. This conclusion is confirmed by the significant variation observed for year × genotype, location × genotype, and year × location × genotype for each trait (Table 1). The significant effect due to genotype for each trait indicates that there is considerable variation in sorghum that can be used to breed improved varieties and hybrids for ethanol production. However, the large amount of variability caused by environmental conditions must be taken into consideration when evaluating the minimum land area required for an ethanol production facility.

Table 1. Mean squares from the combined analysis of variance and the percent variation attributable to each source of variation (from Gill et al., 2014).

	Fresh Weight	Var %[a]	Moisture	Var %	Brix	Var %
year	4.65×10^{-9}***	4.5	1076.1***	8.9	20.7***	1.1
loc[b]	1.17×10^{-10}***	15.9	544.2***	6.2	193.0***	16.7
year*loc	6.29×10^{-9}***	42.5	763.4***	44.0	32.3***	12.8
rep(year*loc)	1.09×10^{-8}***	1.3	9.9ns[c]	0.5	3.2ns	0.0
gen	8.48×10^{-9}***	9.9	434.6***	4.3	31.4***	2.1
year*gen	3.70×10^{-8}***	1.8	56.6***	2.5	7.2***	1.4
loc*gen	5.42×10^{-8}***	4.0	101.1***	6.6	15.1***	6.1
year*loc*gen	3.04×10^{-8}***	10.0	54.6***	16.5	12.0***	22.2
error	6.16×10^{-7}	10.2	7.6	10.6	3.6	37.8
R^2	0.93		0.93		0.79	
CV	15.8		3.9		17.3	

[a] Percent of the total variation due to each effect

[b] Loc, location; rep, replication; gen, genotype

[c] ns, not significant

* Significant at the 0.05 probability level; ***Significant at the 0.001 probability level

Across all environments, significant differences were detected among locations for each agronomic trait evaluated (Table 2). The locations with the lowest average yields were in the regions traditionally associated with grain sorghum production (CC, CS and KS). Grain and forage sorghum are common in these regions because they are more drought tolerant; but the results clearly indicate that these same regions will not produce the highest yields and may not be well suited for biomass and bioenergy production due to persistent seasonal droughts unless supplemented with irrigation. The locations in the southeastern USA had greater yields for fresh weight and dry weight due to longer growing seasons, greater rainfall, and the adaptation of sorghum genotypes to warmer climates.

Significant variation was observed among genotypes for all traits (Table 3). Of the entries, Graze All had the lowest mean fresh and dry weight, which is partially because it is an early flowering PI sorghum sudangrass hybrid. These hybrids are designed for multiple harvests, which was not practiced in this study. The greatest average yields were produced by TX08001, which is a hybrid bred specifically for biomass production. M81-E had a similar fresh weight as TX08001, but the mean dry weight of this cultivar was significantly less because it is a sweet sorghum and has a greater moisture content than TX08001. As expected, the highest brix values were in M81-E and Sugar T, which were the two entries known to have greater soluble sugar concentrations. Sweet sorghums have value in systems that produce both a wet and dry processing stream as fermentable sugars are found in both the juice and the biomass.

Table 2. Means and ranges for fresh weight biomass, moisture concentration, and brix averaged over all genotypes and years for each location (from Gill et al., 2014).

Site	Fresh Weight (MT/ha)[a]		Moisture (%)		Brix (%)	
	Mean[b]	Range	Mean	Range	Mean	Range
Corpus Christi, TX (CC)	30.5e	4.1-84.4	70.3b	39.8-90.7	9.1e	6.0-13.7
College Station, TX (CS)	40.1d	5.7-89.0	73.9a	52.9-83.8	11.5cd	6.2-18.2
Ames, IA	58.4b	29.3-105.5	73.0a	66.4-79.8	13.5a	7.5-19.3
Manhattan, KS	41.5d	13.9-79.8	67.3c	51.0-80.5	13.2ab	8.4-16.3
Lexington, KY	52.0c	28.4-91.9	69.6b	51.7-89.6	12.2bc	6.0-17.2
Raymond, MS	63.8a	17.5-117.8	74.0a	53.3-85.0	8.2e	4.1-15.4
Roper, NC	61.2ab	15.4-127.8	69.3b	54.4-80.8	10.7d	5.0-18.7
HSD (P<0.05)	3.7		1.3		1.3	

[a] Metric tons per hectare

[b] Means within a column followed by the same letter were not significantly different at the 0.05 probability level based on Tukey's honestly significant difference [21]

Table 3. Means and ranges for fresh weight biomass, moisture concentration, and brix averaged over all environments for each genotype (from Gill et al., 2014).

Genotype	Fresh Weight (MT/ha)[a]		Moisture (%)		Brix (%)	
	Mean[b]	Range	Mean	Range	Mean	Range
22053	41.9d	4.2-79.9	70.7b	45.8-88.2	10.7bc	5.0-17.7
Graze All	35.1e	4.1-113.3	68.4c	39.8-86.1	10.0c	4.1-16.8
Graze N Bale	55.3b	7.9-116.0	73.1a	44.0-89.6	10.1bc	4.6-18.7
M81-E	58.2ab	5.4-118.7	72.6a	52.6-87.8	12.0a	5.2-18.2
Sugar T	51.3c	12.3-108.5	73.2a	51.7-90.7	11.9a	4.3-19.3
TX08001	58.6a	9.1-127.8	69.5c	55.0-86.8	10.9b	6.4-16.0
HSD (P<0.05)	3.2		1.1		0.9	

[a] Metric tons per hectare

[b] Means within a column followed by the same letter were not significantly different at the 0.05 probability level based on Tukey's honestly significant difference.

Of the entries in the test, the hybrid 22053 was the only brown midrib genotype, a mutation that results in lower lignin. Lignin can interfere with extraction of cellulose and hemicellulose for fermentation. While this trait has value in forage sorghum, there are potential limitations. The yield of 22053 was low relative to the top yielding entries, and the hybrid consistently lodged late in the season in most locations.

Stability Analysis

The GGE biplot for fresh weight data divided the locations into three mega environments composed of KS, CC and CS in one, NC, MS, and KY in another, and IA by itself (Figure 2, left). Similar results were found for GGE analysis of the dry weight data. These groupings are logical in the context of environment and they imply that adaptation and hybrid type will vary between these environments. Thus, genotypes developed for one mega environment are not likely to perform similarly in the other mega environments. Consequently, breeding and improvement programs will be directed at those different environments. The adaptation of genotypes to mega environments is demonstrated in the genotype-centered biplot (Figure 2, right). For fresh weight, the cultivars M81-E and TX08001 performed best in NC, KY, and MS and their performance in some of the other environments was slightly lower relative to other genotypes (Figure 2, right).

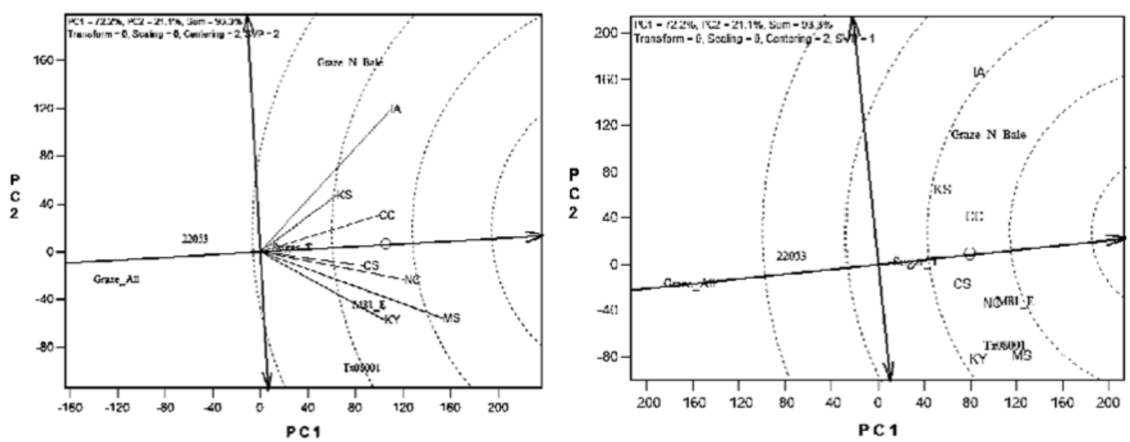

Figure 2. (Left) Location-centered biplot for fresh weight yield grouping the seven locations into mega environments. (Right) Genotype-centered biplot for fresh weight yield showing the performance of each genotype relative to the grand mean (from Gill et al., 2014).

Consistency of Yield in the Top Yielding Hybrid

Ultimately, biomass yield is best estimated by evaluation of the hybrid that consistently produces the greatest yield and it is this hybrid that should be used to measure productivity of the crop in a biomass production plan. Across these tests, the hybrid TX08001 produced the greatest mean fresh and dry weights (Table 3) but the consistency of this production varied from year to year within a location (Table 4). For example in CS between 2009 and 2012, the dry weight yield of TX08001 ranged from 4.3 to 20.9 MT ha^{-1}. In this situation, the low yields were likely due to dry weather, which is common in the western locations. In the southeast testing sites, the variation from year to year was reduced (Table 4).

Unlike grain, which can be easily transported for processing, biomass conversion facilities will require locally produced biomass because transportation is cost prohibitive. Consequently, when evaluating potential production locales, high yield is important but consistency of yield is equally, if not more important. Many of the potential conversion processes assume that the biomass provided to the conversion facility arrives dry. Based on sorghum phenology and the data collected from these trials, it is our conclusion that biomass sorghum is a crop that will be harvested at high moisture concentration because it is very difficult to dry in the field. In the current study, there were differences in moisture concentration among the entries, ranging from the low 40% range to nearly 90% (Table 3). For TX08001, which was in the lowest average moisture concentration grouping (Table 3), the moisture concentration ranged between 65 and 76% at harvest in four locations over four years (Table 4). Biomass sorghums have significantly thicker stems and are harvested later in the season and both of these factors are less conducive for dry down. Consequently, processors who use sorghum will likely have to adopt systems that handle wet biomass. While the additional moisture increases transportation costs, the water in the sorghum genotypes tested in this trial contains substantial amounts of fermentable sugars, so the extraction process yields both juice and bagasse. Thus, it is envisioned that sorghum high in juice sugar concentration and biomass will be processed much like sugarcane.

Table 4. Agronomic performance of the biomass sorghum hybrid TX08001 in College Station, TX, Ames, IA, Raymond, MS, and Roper, NC in four consecutive years (2009-2012) (from Gill et al., 2014).

Location	Year[a]	Fresh weight	Moisture concentration	Dry weight	Brix
		(MT/ha)[b]	(%)	(MT/ha)	(%)
College Station, TX	2009	65a[c]	71b	19ab	8c
	2010	57a	76a	14b	8c
	2011	16b	72b	4c	10b
	2012	65a	68c	21a	13a
LSD (P<0.05)		19	3	6	2
Ames, IA	2009	40b	71bc	12b	13a
	2010	57a	73ab	16ab	13a
	2011	58a	70c	17a	13a
	2012	59a	75a	15ab	11a
LSD (P<0.05)		15	2	5	3
Raymond, MS	2009	74b	72a	21b	nd[d]
	2010	69b	67bc	23ab	12a
	2011	78ab	66c	27ab	10ab
	2012	94a	70ab	29a	8b
LSD (P<0.05)		18	3	7	2
Roper, NC	2009	104a	67a	35a	10b
	2010	73b	66a	25b	14a
	2011	47c	65a	16c	10b
	2012	67b	68a	21b	12b
LSD (P<0.05)		16	3	4	2

[a] TX08001 was not included in 2008

[b] Metric tons per hectare

[c] Means within a column followed by the same letter were not significantly different at the 0.05 probability level based on Fisher's least significant difference test

[d] nd, no data

The harvest dates for the greatest yielding hybrids were typically late in the season. For northern locations, this was typically from mid-September to early October. In southern locations the harvest season was longer, ranging from late August through early November. In all locations, harvesting earlier than optimum lowered yields and harvesting later than optimum reduced yield and quality (because the crop begins to degrade). Consequently, given that sorghum is a crop with high moisture, storage systems must be developed or the crop must be harvested as needed. If the latter, then complementary crops are essential to maintain a harvest window sufficient to justify capital costs for processing and conversion. In the southeastern USA, there is an opportunity to combine sugarcane and sorghum, which would use much of the same processing equipment. Regardless of the method and conversion system, crop complementation will be essential to the productivity and economic efficiency of biomass conversion.

Although some composition traits show high repeatability in certain locations, the environment causes large fluctuations in values of ash, protein, lignin, glucan, and xylan across locations and years. Processors should expect variability in the composition of biomass delivered to the refinery due to differences in uncontrollable factors such as weather and soil type and controllable factors such as management practices. It may be possible to minimize a portion of the variability by standardizing management techniques and breeding for enhanced stability, but ultimately the environment will largely determine final composition of the biomass at harvest. More research is needed to determine how differences in rainfall and temperature within a given location may affect the composition of the biomass at harvest. Extensive genetic variation within sorghum and advanced molecular tools coupled with traditional breeding practices will allow researchers to manipulate the composition in response to demand from ethanol refineries. With superior genotypes and stable production environments, sorghum will be a valuable tool in the goal of the sustainable production of one billion tons of dry biomass each year in the USA.

Using data generated from the Feedstock Partnership trials as well as other yield data collected, and combined with basic growth parameters and weather data, the PRISM-ELM model for bioenergy sorghum indicates that sorghum has high yield potential across a wide range of the Central and Eastern U.S (Figure 3). Yields in the far northern U.S. (> 42° N) trend lower due to the cooler temperatures and short growing season. In the Southeast, while the productivity is high overall, the relative increases and reductions are associated with soil fertility and quality.

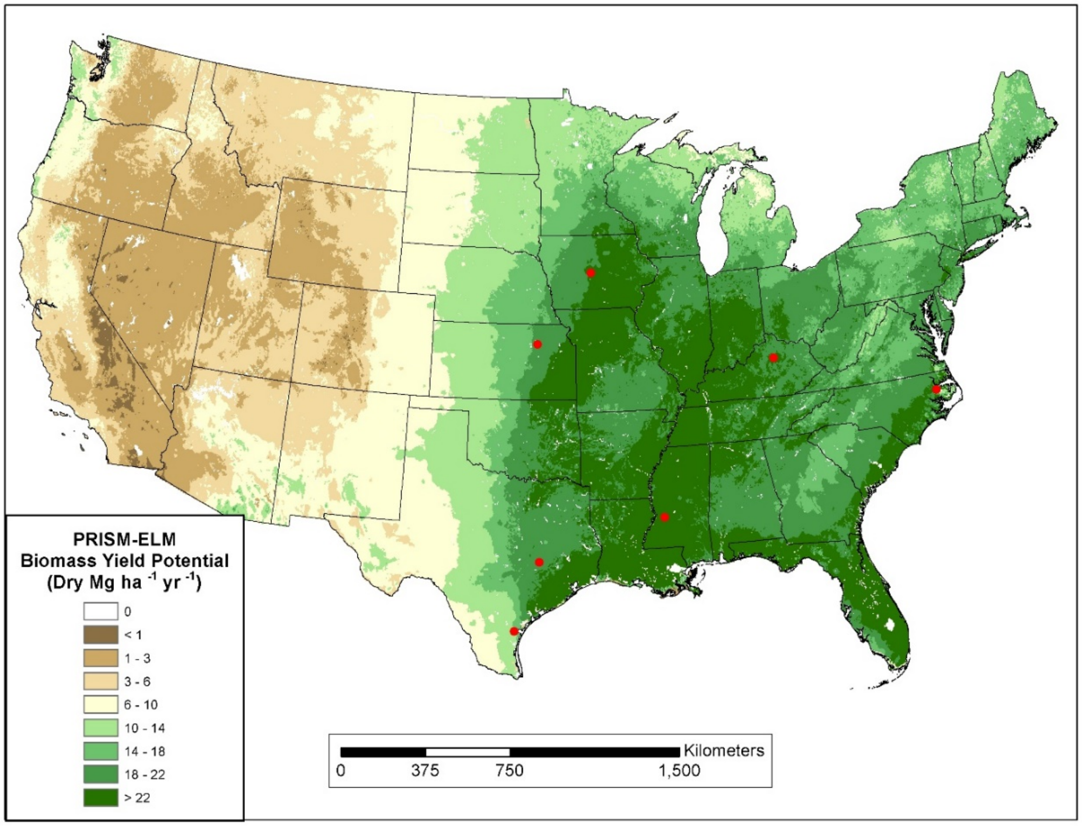

Figure 5. Biomass yield potential of sorghum for the U.S. generated using the PRISM-ELM model and based in part on in part on Regional Feedstock Partnership Field Trials (red dots) (from Lee et al., 2018).

Key Outputs

Peer Reviewed Publications

Burks, P. S., T. J. Felderhoff, H. P. Viator, and W. L. Rooney. 2013. "The Influence of Hybrid Maturity and Planting Date on Sweet Sorghum Productivity during a Harvest Season." *Agronomy Journal* 105: 263–7. doi:10.2134/agronj2012.0317.

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Hoffmann, L., and W. L. Rooney. 2016. "Accumulation of Biomass and Compositional Change over the Growth Season for Six Photoperiod Sensitive Sorghum Lines." *BioEnergy Research* 7 (3): 811–5.
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Sorghum Sustainability

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Summary

Modern bioenergy feedstocks, such as bioenergy sorghum (*Sorghum bicolor* L.), are being developed to supply future cellulosic biofuel demands. How these cropping systems impact greenhouse gas (GHG) emission of CO_2 and N_2O from the soil is unknown. We studied effects of N fertilization, residue management, crop sequence [corn (*Zea mays* L.)-sorghum vs. sorghum-sorghum], and their interactions on CO_2 and N_2O emissions from bioenergy production scenarios in central Texas. Overall, CO_2 and N_2O fluxes were higher than those observed by others in the United States despite drought conditions throughout much of 2010 and 2011. Highest emissions of both gases were observed during the growing season, often following a precipitation/irrigation event and shortly after N fertilization. Residue return increased cumulative CO_2 emissions each year, probably due to increased microbial activity. Nitrogen addition significantly increased cumulative emissions of N_2O both years, but only impacted cumulative CO_2 emissions in 2011. While crop rotation impacted biomass yield, it had no significant effect on cumulative CO_2 or N_2O emissions.

Introduction

The global push for biofuel production is transforming both agricultural and energy industries worldwide. Emphasis on producing biofuels is largely due to concerns about energy security and sustainability of the petroleum industry and reliance on foreign petroleum-based fuels. Recent attention on the impacts of GHGs, such as CO_2 and N_2O, on global climate change have further increased interest in biofuels as a means of reducing GHG emissions compared to petroleum-based fuels. The world population is expected to reach 9 billion in less than 40 years and with it, demand for food, water, energy and natural resources are expected to climb. Biofuel production, however, must be performed in a way that minimizes impacts to both food crop production and natural resources.

How bioenergy cropping systems will impact net GHG emissions is still not fully understood. Bioenergy sorghum (*Sorghum bicolor* L.) represents a crop which is not utilized as a food source, is an annual crop (single year land commitment for production), can produce large yields (minimizing land use requirements), has suitable biomass chemical compositional properties for fuel conversion, and requires relatively fewer inputs than many other bioenergy crops. The warm temperatures and moderate rainfall (>750 mm) associated with climates in the southeastern and south central U.S. may be particularly well-suited for bioenergy sorghum production. However, these climatic characteristics may also be particularly favorable for large GHG emissions from soil. The impact of bioenergy sorghum cropping systems on GHG emissions has not been thoroughly examined. Furthermore, little data exists on GHG emissions from cropping systems of any kind in central Texas.

Agronomic management practices, such as rotation and cropping intensity, can affect soil C and nutrient cycling, soil microbial activity, and GHG emissions. Emissions of N_2O from agricultural soils generally increase with increasing additions of N fertilizer. Nitrogen fertilization is frequently a large factor in GHG emissions, particularly of N_2O, but crop residue return can be a major contributor as well. Soil respiration can be impacted by a number of agronomic management practices including cropping rotation, N fertilization, and addition of organic C substrates, such as crop residues. Each agronomic management practice may have a primary effect, but may often have more complex interactive effects since biogeochemical cycling is connected through dynamic soil and microbial processes. Furthermore, site-specific factors such as soil properties, vegetation type, and climate further confound the complex effects of various management practices. Thus, various agronomic management practices utilized to produce bioenergy crops can significantly impact GHG emissions from soil, but further information is necessary to determine the potential effects of bioenergy sorghum production on GHG losses.

If the production of bioenergy crops increases net GHG emissions, producers could be undermining one of the overall central goals for producing biofuel crops. Thus, the impact of various agronomic management practices on bioenergy crop production should be examined.

Objectives

The objective of this project was to determine the effect of N fertilization, residue management, cropping sequence, and their interactions on growing season fluxes and cumulative annual CO_2 and N_2O emissions from bioenergy sorghum production scenarios in central Texas.

Methods

The experimental site was located at the Texas A&M Agrilife Research Farm, approximately 8 km southwest of College Station, Texas, USA (30˚ 32’ 15”N, 96˚ 25’ 37”W), which is situated within the Brazos River floodplain in south-central Texas. The soil used was a calcareous (pH 8.2) Weswood silty clay loam (100 g sand kg^{-1}, 560 g silt kg^{-1}, 340 g clay kg^{-1} in top 15-cm) and is classified as a fine-silty, mixed, superactive, thermic Udifluventic Haplustept. This study was conducted within a larger study investigating the yield potential and agronomic responses of bioenergy sorghum (*Sorghum bicolor* L.) to various integrated management practices. The bioenergy sorghum cropping system study utilized four row (1.02-m row centers) plots measuring 9.14 m long and 4.08 m wide. The larger study used a randomized complete block design with crop rotation, N fertilization rate, and biomass return rate as the three major factors. The study reported herein was limited to two levels of each of the three major factors and was replicated 3 times. The eight experimental treatments included every combination of crop sequence [corn-sorghum (CS) or sorghum-sorghum (SS)], N fertilization [0 kg N ha^{-1} (- N) or non-limiting rate of 280/168 kg N ha^{-1} for sorghum/corn (+ N)], and biomass return [0% (0%R) or 50% biomass returned (50%R)]. The bioenergy sorghum variety used, “4Ever Green”, was a photoperiod-sensitive, high-yielding hybrid forage sorghum (Walter Moss Seed Co, Waco, TX, U.S.A.). When corn was rotated with sorghum, Dekalb DKC68-05 was the corn variety utilized in the CS cropping sequence. Corn was planted in the CS sequence in 2008 and all following even-numbered years, whereas sorghum was planted in all treatments in 2009 and all subsequent odd-numbered years.

Treatments receiving N fertilization were side-dressed with subsurface banded granular urea at approximately the 4-leaf stage for sorghum and 6-leaf stage for corn. Corn grain and aboveground biomass were hand-harvested for yield on 2 July 2010. While corn was harvested from a randomly-selected 3 m segment of the middle two rows from each plot, remaining non-sampled portions of the corn plots were not harvested/incorporated into soil until at the later sorghum harvesting date. All corn residue was ultimately returned to each plot, regardless of residue return treatment. Sorghum harvest was performed with a silage harvester with an attached weigh-bucket and scale. Biomass yield was estimated from the entire length of the middle two rows of each plot and a random grab sample of chopped residue was captured for determining moisture and nutrient composition. The harvester mulched sorghum biomass into pieces approximately 2 cm by 2 cm. After biomass yield weights were collected, the 50%R treatments received the biomass residue returned from the middle two rows evenly distributed across the area of the entire plot. After harvest each fall, residue was disked into the field and the plots were bedded. Furrow irrigation was used sparingly as needed. Eleven cm of irrigation water were applied on 31 May 2010 and 12 April, 14 July, and 4 August in 2011, while 9 cm were applied on 9 May 2011.

Soil CO_2, N_2O, and CH_4 fluxes were directly measured using a static, vented chamber and a field photoacoustic gas analyzer. Specifically, the measurements were made by integrating a Li-Cor 20-cm survey chamber (model 8100-103, Li-Cor Inc., Lincoln, NE) with an INNOVA 1412 photoacoustic gas analyzer (Innova AirTech Instruments A/S, Denmark). The flux chamber was selected for its portability, adaptability, and ability to minimize pressure disequilibrium between the chamber headspace and ambient atmosphere. A PVC flux chamber soil-collar was installed to a depth of approximately 12 cm near the middle of each of the 24 sampled plots. The collars were placed on top of beds on level surfaces which were equidistant from the crop and injected fertilizer. Height measurements from the soil surface to the top of each collar were measured at four quadrants inside the collar and averaged to estimate height for periodic headspace volume calculation. To minimize disturbance effects, soil-collars were installed no less than 24 hours prior to the initial gas sampling event and remained in place through the entire growing and fallow seasons. Collars were only removed briefly in spring and fall to allow for field operations. Soil gas measurements were initiated shortly after N fertilization each year and were performed approximately weekly through the growing season and at a reduced frequency during the fallow period. Precipitation events and technical difficulties inhibited the uniform sampling frequency initially planned for 2010 and 2011. Cumulative annual emissions of GHG fluxes were calculated from 18 sampling events in 2010 and 20 sampling events in 2011. Measurements for year 1 (2010) occurred from 27 May 2010 through 3 March 2011 and for year 2 (2011) from 6 May 2011 to 1 March 2012.

Several environmental variables were monitored throughout the study to supplement trace gas flux measurements. Air temperature, relative humidity, wind speed, solar radiation, and rainfall were measured every half hour with a weather station approximately 400 m from the field plots. Soil temperature was measured hourly by type T thermocouples at 10-cm depth approximately 50 cm from gas sampling collars within each plot. A time domain reflectometry (TDR) system was utilized to measure soil moisture near each collar every 6 hours. The TDR array consisted of sensors installed approximately 50 cm from gas flux collars, using a TDR100, and five SDMX50 multiplexers (Campbell Scientific, Inc., Logan, UT). The volumetric water content was determined to a depth of 15 cm by analyzing TDR wave forms with PC-TDR (Campbell Scientific, Inc., Logan, UT) and Topp's equation (Topp et al., 1980). Soil temperature and moisture data from monitoring systems were collected within the field with a CR1000 data logger (Campbell Scientific, Inc., Logan, UT).

Three soil cores (4-cm diam.) were composited for analysis from each plot in the spring on 5 April 2010 and 14 March 2011, prior to each growing season. Cores were separated into depths of 0 - 5, 5 - 15, 15 - 30, 30 - 60, and 60 - 90 cm. Plant samples collected annually during harvest were oven-dried and ground to pass a 1-mm sieve. A subset of plant tissue samples were finely ground for elemental analysis of C and N by combustion methods.

Cumulative annual CO_2 and N_2O emissions were estimated by linearly interpolating between sampling events and integrating the underlying area. Annual N emission factors (NEF) were calculated. The mass of N_2O-N lost from [- N] treatments was subtracted from the mass of N_2O-N lost by [+ N] treatments, divided by the mass of fertilizer N applied, and multiplied by 100.

The effects of crop sequence, N fertilization, biomass return and their interactions on biomass yield, trace gas fluxes, and cumulative CO_2 and N_2O emissions were tested using a mixed ANOVA in SAS 9.2 using PROC mixed procedures. Crop sequence, N fertilization, and biomass return were considered fixed effects, while block was considered a random effect. The Shapiro-Wilk approach was used to test normality. A logarithmic transformation was applied to data with non-normal distributions prior to analysis. Statistical significance was determined at $\alpha = 0.05$ probability unless otherwise stated, and Fischer's LSD was used for means separation where significance was observed. Year was determined to be a significant ($P = 0.003$) effect, therefore all cumulative analyses were performed separately between 2010 and 2011.

Results and Outcomes

Aboveground Carbon

Crop biomass-C applied to the soil from the previous year's harvest may impact trace gas emissions and crop production in following years. Crop rotation increased biomass-C yield of sorghum in CS in 2009, resulting in greater biomass C being returned prior to the 2010 season in those treatments relative to SS. When corn was grown in CS in 2010, all corn residues were returned to the soil. However, the much smaller total mass of corn relative to sorghum caused residue C applied in CS treatments following the 2010 harvest to be less than 50% of sorghum biomass C returned from SS, 50%R treatments. Despite these differences, the rotation effect on sorghum in 2009 essentially negated the small mass of corn residue returned in 2010 because the cumulative mass of C applied to SS, 50%R and CS, 50%R was relatively similar across the two years.

Differences in aboveground biomass-C yield were observed in both 2010 and 2011, with cropping sequence having the largest impact on biomass-C yield. In 2010, CS produced less than one-third the biomass-C of SS, largely because it was in corn. However, in 2011 when sorghum was grown in all treatments, CS treatments produced 45% more biomass-C than SS. Addition of N fertilizer in 2010 and 2011 increased biomass-C yield by 50% and 26%, respectively. Biomass return did not have a significant impact on aboveground biomass-C yield in either 2010 or 2011.

Growing Season Gas Fluxes

Fluxes of CO_2 were usually higher when soil moisture was also relatively high, with peak fluxes generally observed after irrigation/precipitation events. As soil dried, fluxes decreased until a precipitation or irrigation event again increased soil moisture (Fig. 1). Early in the 2010 growing season, each of the treatments significantly affected CO_2 fluxes. Nitrogen fertilization significantly increased CO_2 flux during the first two sampling events, but had no effect on later CO_2 loss (Fig. 1a). On the third sampling event, both 50%R and SS significantly increased CO_2 flux. Treatments receiving biomass return also exhibited increased CO_2 losses on several other occasions during the middle of the growing season, but had no effect later in the season (Fig. 1b). Continuous sorghum exhibited greater CO_2 fluxes than CS early and in the middle of the growing season, but then showed little additional effect until the second to last sampling event

(Fig. 1c). All treatments exhibited increased CO_2 loss on the last two sampling dates, likely due to increased soil moisture associated with recent rainfall.

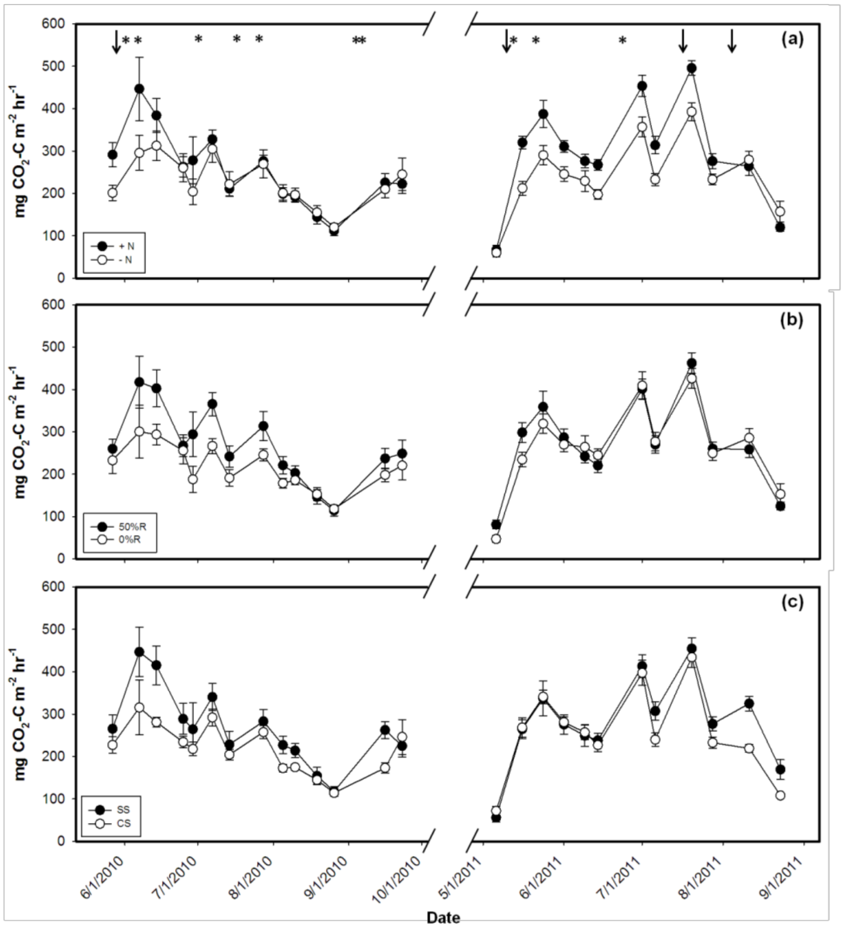

Figure 1. Mean CO2 flux rates from 2010 and 2011 growing seasons as affected by N fertilization rate (a), residue return rate (b), and cropping sequence (c) are reported with one standard error (n = 12). Arrow denotes irrigation event and asterisk denotes precipitation event >10 mm. SS, CS, +N, -N, and %R represent continuous biomass sorghum, corn/sorghum rotation, with and without N fertilization, and percent of previous crop biomass returned to soil, respectively (from Storlien et al., 2014).

The 2011 growing season included the period of most extreme drought on record. Carbon dioxide fluxes again followed soil moisture trends where highest flux rates were observed after rainfall or irrigation, but relatively low fluxes were observed across dry periods (Fig. 1). Because the fallow season from October 2010 through May 2011 experienced much lower than normal precipitation, gas sampling for the 2011 growing season was initiated under very dry soil conditions after fertilization. This likely explains why CO_2 fluxes were very low (< 100 mg CO_2-C m^{-2} hr^{-1}) across all treatments (Fig. 1). Following irrigation and rainfall in early May, CO_2 flux rates climbed dramatically and were more similar to early growing season fluxes observed in 2010. Nitrogen fertilization had the largest impact on CO_2 flux rates throughout the 2011 growing season, causing increased emissions on three-fourths of all sampling events (Fig. 1a). Biomass return (50%R) had relatively limited impact on CO_2 flux throughout the 2011 growing season, significantly increasing flux rate only on the second sampling event (Fig. 1b). Despite the entire study area being in sorghum in 2011, SS showed significantly higher CO_2 fluxes than CS treatments in the latter portion of the growing season (Fig. 1c). Nitrous oxide fluxes were somewhat sporadic in both 2010 and 2011, but also tended to increase following rainfall or irrigation (Fig. 2). As expected, N_2O fluxes were much higher following N addition, and fertilization was implicated as a significant driver of N_2O loss early in the 2010 growing season (Fig. 2a). By late June, soil moisture was lower and associated N_2O fluxes were also low. Shortly following precipitation events throughout the season, flux rates again increased, but never to the highest rates initially observed after fertilization. Similarly, after the initial peak early in the season, N fertilization no longer was a significant factor driving N_2O loss. Residue return had no observable impact on N_2O flux throughout the 2010 growing season (Fig. 2b). Beginning in mid-July, crop rotation became significantly associated with N_2O flux, but neither treatment consistently impacted flux rates.

Fewer detectable fluxes of N_2O were observed during the 2011 compared to the 2010 growing season. On the first gas sampling event following fertilization, 6 May 2011, no measurable N_2O flux was detected (Fig. 2). However, following irrigation and rainfall, subsequent sampling events yielded measurable fluxes. The highest measured individual flux rate of the study (~2,000 μg N_2O-N m^{-2} hr^{-1}) and highest mean flux rate (~1,000 μg N_2O-N m^{-2} hr^{-1}, in +N) were measured in late May. Nitrogen fertilization significantly increased N_2O flux early in the growing 2011 season, but the effect did not persist beyond mid-June (Fig. 3a). Biomass return appeared to have minimal effect on N_2O loss for most of the season, but 0%R exhibited greater fluxes on the last two sampling dates compared to 50%R (Fig. 2b). Cropping sequence significantly influenced N_2O flux at several points throughout the growing season, but impacts were not consistent (Fig. 2c). Continuous sorghum showed significantly higher N_2O loss near the beginning and at the end of the growing season, while rotated sorghum had higher flux near the middle of the growing season (Fig. 2c). This trend was also observed in 2010 despite CS being in corn in 2010 and sorghum in 2011.

While the most active period of trace gas emissions is typically during the growing season, the fallow period in central Texas may also contribute an appreciable portion of annual emissions because of relatively mild temperatures (commonly above freezing) and increased precipitation relative to the growing season. Although fallow season GHG fluxes are likely lower than those from the growing season (lower temperatures and lack of crop growth), these milder, wetter conditions may also be conducive to sustaining significant GHG fluxes. Thus, a complete analysis of GHG emissions from cropping systems in central Texas may necessitate accounting for both growing season and fallow season emissions.

Cumulative Annual Emissions

Analysis of cumulative annual CO_2 emissions revealed little difference among the treatments. The moderate drought during the summer of 2010 and sustained lack of moisture in the fall of 2010 through the winter of 2011 may have contributed to the low GHG fluxes through the 2010 fallow period. Visual field observations made in the spring of 2011 revealed little residue decomposition had occurred, likely due to the lack of soil moisture. Biomass return, the lone significant treatment effect in 2010, had 13% greater annual CO_2 loss with 50%R compared to treatments receiving no biomass return (Fig. 3b). The effect of biomass return on CO_2 emissions was anticipated, since the greater amount of organic C applied to the soil in these treatments led to greater soil microbial biomass (data not shown) and potentially greater CO_2

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flux via microbial decomposition. Continuous sorghum lost more cumulative annual CO_2 during 2010 compared to the rotated treatment, but the effect was not significant. Similarly, on average, the fertilized treatment had greater cumulative annual CO_2 loss, but again was not significant. Regardless of N fertilization, SS treatments with 50%R had 22% higher losses of CO_2 than CS, 50%R. Sorghum was photosynthetically active for a longer period of time (~3 mos.) relative to corn in 2010 (potentially greater cumulative CO_2 from root respiration and increased rhizodeposition) which may have combined with residue decomposition from the previous year for greater annual cumulative emissions.

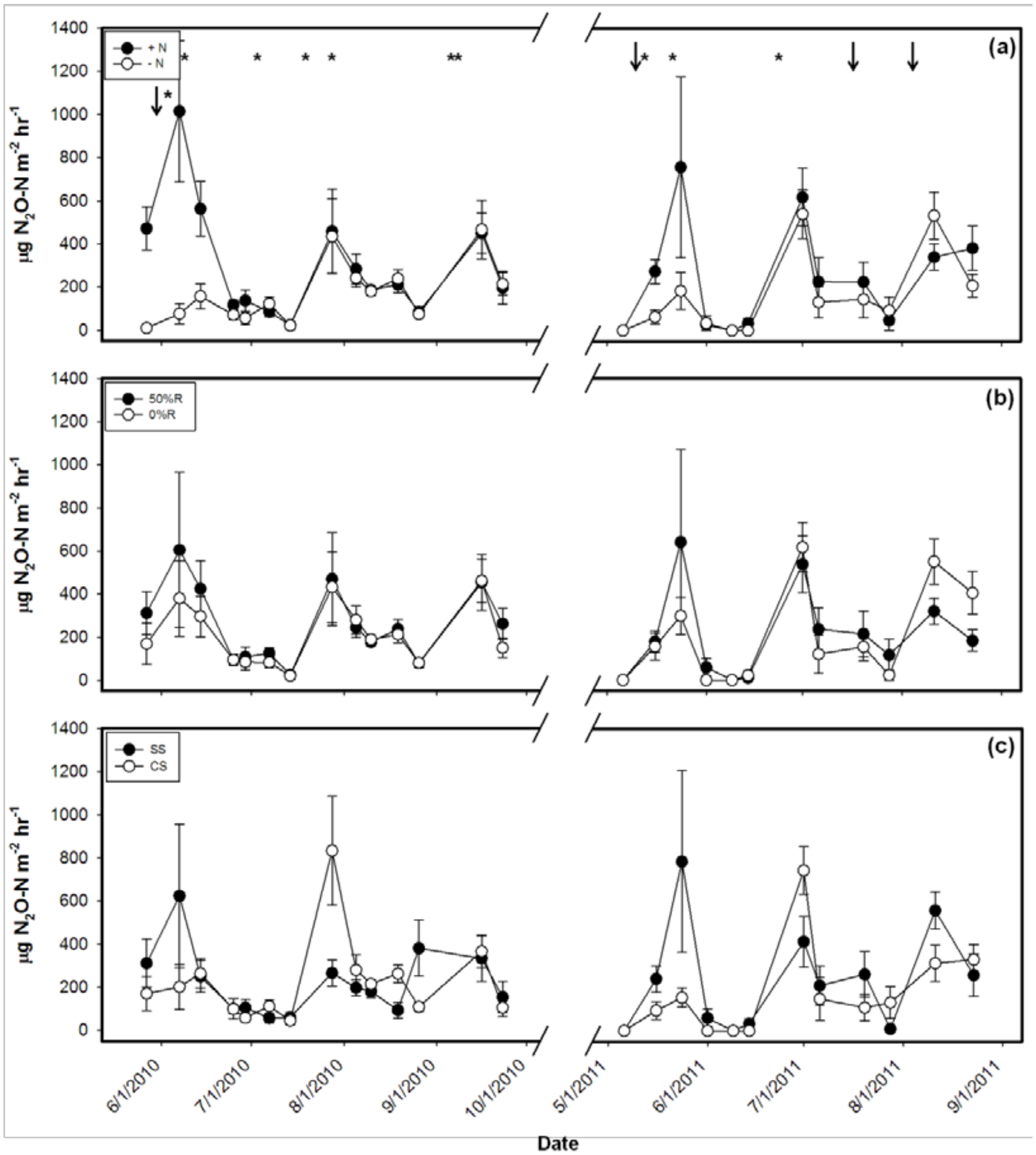

Fig. 2. Mean N_2O flux rates from 2010 and 2011 growing seasons for N fertilization rate (a), residue return rate (b), and cropping sequence (c) are reported with one standard error (n = 12). Arrow denotes irrigation event and asterisk denotes precipitation event >10 mm. SS, CS, +N, -N, and %R represent continuous biomass sorghum, corn/sorghum rotation, with and without N fertilization, and percent of previous crop biomass returned to soil, respectively (from Storlien et al., 2014).

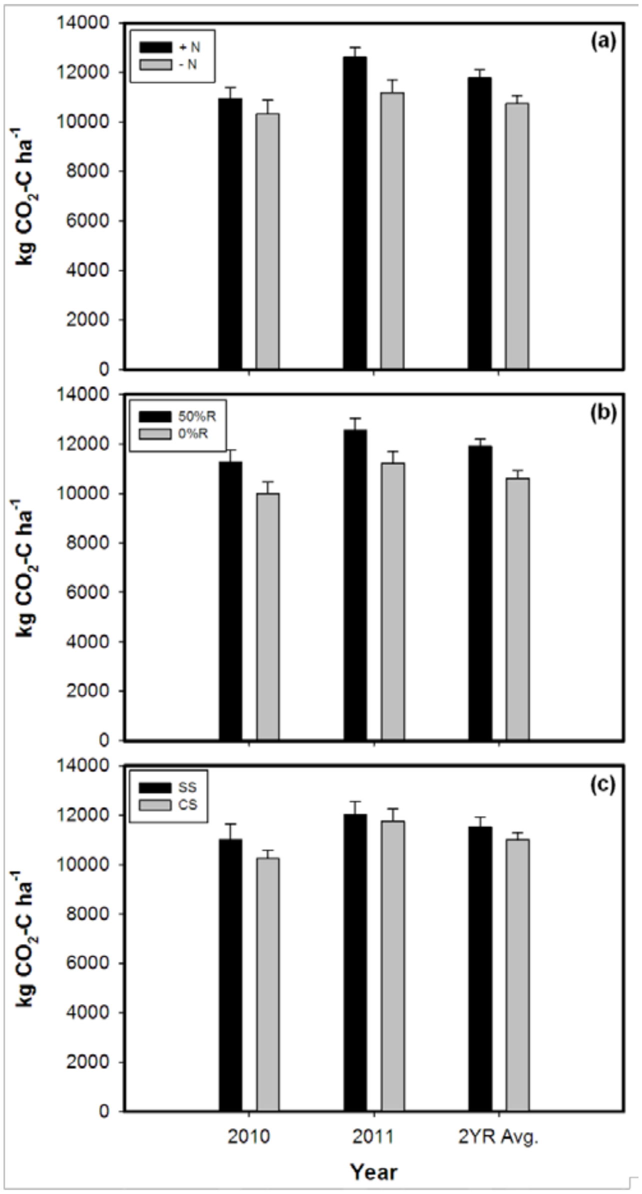

Fig. 3. Mean cumulative CO_2 emissions from the 2010 and 2011 sampling years and two-year average as affected by N fertilization rate (a), residue return rate (b), and cropping sequence (c) are reported with one standard error (n = 12). SS, CS, +N, -N, and %R represent continuous biomass sorghum, corn/sorghum rotation, with and without N fertilization, and percent of previous crop biomass returned to soil, respectively (from Storlien et al., 2014).

The entire study was planted to sorghum in 2011 and it was anticipated that less difference in annual CO_2 loss between treatments would be observed compared to 2010. However, both N fertilization and biomass return affected cumulative CO_2 loss in 2011. Nitrogen fertilization significantly increased annual CO_2 emission in 2011 by approximately 13% compared with unfertilized treatments (Fig. 3a). Biomass return also significantly increased

cumulative annual CO_2 emissions in 2011, with 50%R having 12% greater loss than 0%R (Fig. 3b). Both continuous and rotated sorghum lost approximately the same amount of cumulative CO_2 in 2011, nearly 12,000 kg CO_2-C ha^{-1}. A three-way rotation by N fertilization by residue return effect (P = 0.005) was also observed for cumulative CO_2 emissions in 2011.
On average each year, approximately 11,260 kg CO_2-C ha^{-1} was lost as soil respiration across all management practices. Residue return had the strongest impact on cumulative soil respiration across years, with 50%R having a 12% increase in respiration compared to 0%R (Fig. 3b). Cumulative respiration was also significantly (P = 0.015) impacted by N fertilization, with fertilized treatments having a 10% greater loss than those without (Fig. 3a). Cropping sequence did not have a significant (P = 0.197) effect on cumulative soil respiration across 2010 and 2011 (Fig. 3c).

Cumulative annual N_2O emissions were relatively high compared to those reported elsewhere. Nitrogen fertilization was the most significant (P = 0.018) treatment factor affecting cumulative N_2O emissions in 2010. Fertilized treatments had 53% higher annual N_2O emission than unfertilized (Fig. 4). In 2010, SS and 50%R treatments tended to have higher annual N_2O emission, yet neither effect was significant by itself (Fig. 4). However, a rotation by residue return interaction (p = 0.014) was observed in 2010, where, regardless of N fertilization, the SS, 50%R treatments had 66% higher cumulative N_2O emission than the CS, 50% treatments.
The greater variability in the 2011 N_2O data may have limited detection of significant differences between treatments, as subsequent measures of error were larger in 2011 than in 2010. The only treatment factor which significantly (P = 0.027) affected cumulative annual N_2O emission in 2011 was N fertilization. Fertilization results from 2011 were consistent with 2010 findings, where fertilized treatments again had 53% higher annual N_2O loss than unfertilized (Fig. 4). The rotation trend for annual N_2O loss remained consistent with 2010 findings, with mean emissions from SS being greater than CS, yet neither was significantly different. However, analysis of residue return found the trends differed between 2010 and 2011 (Fig. 4). In 2011, 50%R treatments had relatively lower annual N_2O losses than 0%R, but the effect was not significant.
The average cumulative loss of N_2O across 2010 and 2011 was 8.35 kg N_2O-N ha^{-1} yr^{-1}, across all treatments. Nitrogen fertilization was the only treatment to have a significant (P = 0.002) impact on N_2O emissions across years, where treatments with N addition exhibited a 53% increase in N_2O loss compared to those without (Fig. 4a). Neither cropping sequence nor residue return significantly influenced cumulative N_2O emissions across 2010 and 2011 (Fig. 4).

The nitrogen emission factor (NEF) was calculated to evaluate the fraction of applied N fertilizer lost as N_2O each year. The NEF varied between cropping sequence and year, but was generally higher in 2010 than in 2011, regardless of cropping sequence. In 2010, SS treatments exhibited an average NEF of 1.7%, while CS was higher at almost 2.5%. The NEFs associated with SS and CS in 2011 were 1.6% and -0.3%, respectively. Review of the data revealed that the highly sporadic N_2O fluxes measured during the 2011 drought made small fluxes of N_2O from unfertilized plots play a larger role in NEF when fertilized plots had no detectable fluxes during the same sampling event. High variability and relatively low N_2O flux associated with extreme drought conditions may explain the negative NEF observed in CS in 2011.

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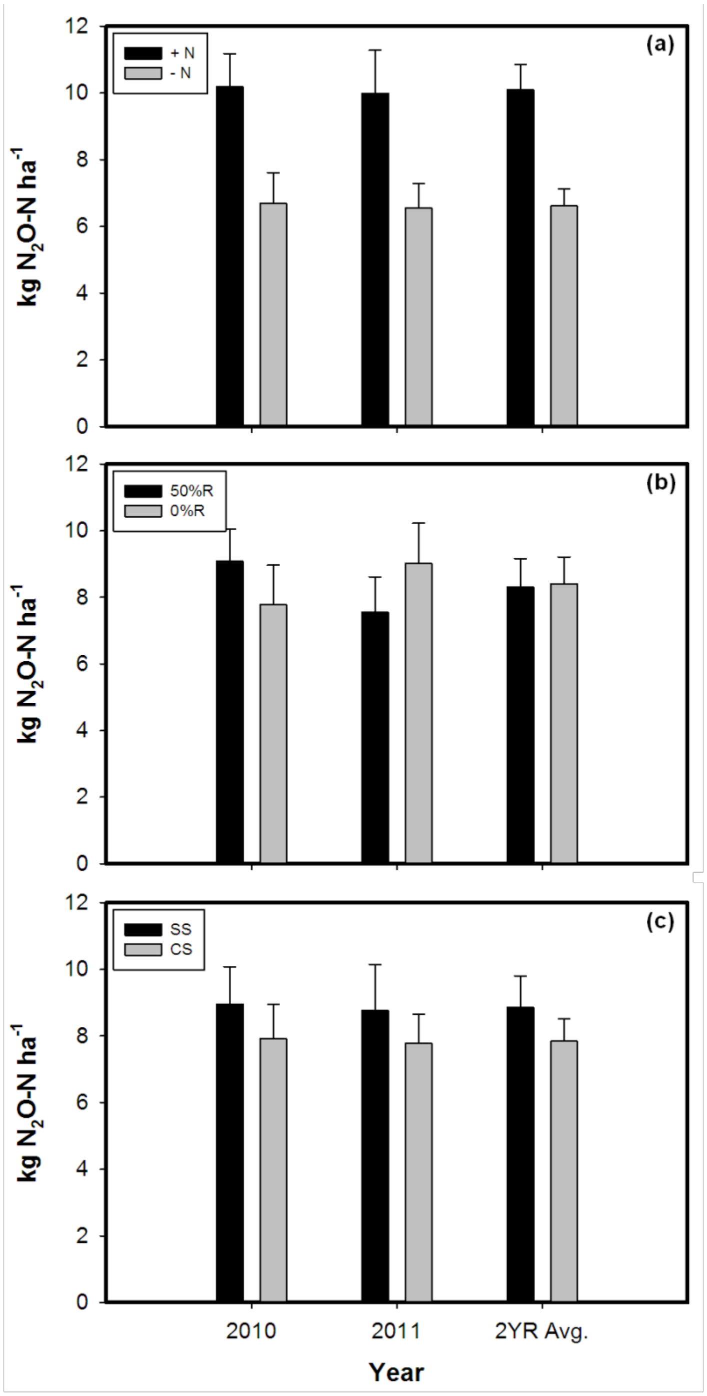

Fig. 4. Mean cumulative N_2O emissions from the 2010 and 2011 sampling years and two-year average as affected by N fertilization rate (a), residue return rate (b), and cropping sequence (c) are reported with one standard error (n = 12). SS, CS, +N, -N, and %R represent continuous biomass sorghum, corn/sorghum rotation, with and without N fertilization, and percent of previous crop biomass returned to soil, respectively (from Storlien et al., 2014).

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Key Outputs

Publications

Storlien, J. O., F. M. Hons, J. P. Wight and J. L. Heilman. 2014. Carbon dioxide and nitrous oxide emissions impacted by bioenergy sorghum management. Soil Sci. Soc. Am. J. doi:10.2136/sssaj2014.04.0176

Presentations

Storlien, J. O., F. M. Hons, J. P. Wight, F. Dou, T. J. Gentry and J. L. Heilman. 2012. Assessment of life cycle greenhouse gas emissions from bioenergy sorghum production in Central Texas. Annual Meeting of American Society of Agronomy.

Storlien, J., F. Hons, J. Wight, J. Heilman, T. Gentry and F. Dou. 2011. Impacts of biomass sorghum feedstock production on carbon sequestration and greenhouse gas emissions in the south central region. Annual Meeting of American Society of Agronomy.

Storlien, J., F. Hons, J. Heilman, J. Wight and I. Tokumoto. 2011. Emissions of carbon dioxide and nitrous oxide from bioenergy sorghum production under integrated agronomic management practices. Annual Meeting of American Society of Agronomy

Storlien, J., F. Hons, J. Wight, J. Heilman and T. Gentry. 2011. Carbon pool dynamics with bioenergy sorghum production systems in Central Texas. Annual Meeting of American Society of Agronomy.

Storlien, J., F. Hons, J. Heilman, J. Wright and I. Tokumoto. 2010. Greenhouse gas emissions from high biomass sorghum production systems in Central Texas. Annual meeting of American Society of Agronomy.

Conservation Reserve Program (CRP) Mixed Perennials

CRP Team: DoKyoung (DK) Lee, University of Illinois; Paul Adler, John Williams, USDA ARS; Keith Harmoney, Kansas State University; Chengci Chen, Montana State University; Dennis Hancock, University of Georgia; Robert Kallenbach, University of Missouri; Vijaya Gopal Kakani, Oklahoma State University; Ezra Aberle, North Dakota State University.

Summary

This farm scale field experiment demonstrated the potential of sustainable biomass feedstock production on Conservation Reserve Program (CRP) lands estimated in the 2005 Billion Ton Study. However, growing season precipitation was a critical factor for annual biomass feedstock production across all regions, and annual feedstock production was severely reduced when growing precipitation was below 50% of average. Nitrogen (N) fertilization significantly increased biomass feedstock production and adequate N application is crucial to obtaining the yields outlined by the Billion-Ton study, especially in systems where non-leguminous species are prevalent. However, economic analysis of N fertilization on biomass yield should be conducted to evaluate the potential of CRP lands for sustainable biomass production. Harvest management, or timing of biomass harvest, did not have much impact on stand health or long-term biomass production. Biomass yield was consistent under different harvest regimes. However, early season harvest for warm-season grass mixtures had adverse impacts on species composition and delayed harvest until after a killing frost or at the end of growing season is recommended for stand longevity and maintenance of desired species. By far, the greatest impacts on seasonal biomass production and changes in vegetation composition were due to location specific precipitation. Our six-year field research demonstrated the importance of long-term farm-scale research for accurate estimation of biomass feedstock production potential of CRP grassland.

Introduction

The Sun Grant Regional Feedstock Partnership and the United States Department of Energy (USDOE) identified grass mixtures planted in the Conservation Reserve Program (CRP) land as one of five herbaceous sources with potential for expansion over time as a sustainable bioenergy source. The CRP is a land retirement program established by the Food Security Act of 1985 (Food Security Act, 1985; Glaser, 1986), and administered by the U.S. Department of Agriculture's Farm Services Agency. The overall goals of this program are to protect environmentally sensitive land and, to a lesser extent, to reduce production of cash crops in order to stabilize commodity prices. These lands are potentially a major resource for cellulosic biofuel feedstock production. According to the 2005 Billion Ton Study, up to 10 Mha (25.4 million acres) of CRP grassland could be dedicated to bioenergy feedstock production from which total biomass production of approximately 50 million dry tons could be expected annually and 50% of the total biomass was assumed to be available for bioenergy production (Perlack et al. 2005). The Farm Security and Rural Investment Act of 2002 (a.k.a., 2002 Farm Bill) permitted managed haying, grazing, and biomass harvesting of CRP grassland in accordance with a conservation plan (Farm Security and Rural Investment Act, 2002; Mapemba et al, 2007). These harvests, however, were subject to limitations in frequency and timing during the year. The Food, Conservation, and Energy Act of 2008 (a.k.a., 2008 Farm Bill), Title II, Subtitle B allowed harvests for forage or biomass after the primary nesting season for grass-nesting birds (USDA, 2008). A recent announcement by the USDA stated that $328 million is being invested through the Agricultural Conservation Easement Program (ACEP), established in the Agricultural Act of 2014 (a.k.a., 2014 Farm Bill), Title II, Subtitle D (Agricultural Act, 2014), to “help landowners protect and restore key farmlands, grasslands and wetlands” (USDA, 2014).

Total land enrolled in the CRP has decreased by nearly 4.5 Mha (11.1 million acres) since 2007 (Fig. 1). In the Western Corn Belt alone (ND, SD, NE, MN, and IA), total conversion of grassland to conventional cropping systems from 2006 to 2011 was estimated to be 530,000 ha (1.3 million acres) (Wright & Wimberly 2013). Recent annual wetland loss rate in the Dakota Prairie Pothole Region, an area of particular concern for wildlife preservation, was estimated to be between 0.28 and 0.35% (5,203 to 6,223 ha yr^{-1}) due to row crop expansion (Johnston, 2013). One reason for these declines is the recent rise in corn and soybean prices which have lured growers away from CRP with the prospects of greater revenues. As corn prices increase, an incremental increase in CRP land leaving the program is predicted which could bring more environmentally fragile land into production with the likely outcome of reduced environmental quality (Secchi et al. 2009). Increased prices for corn may result in the decline in lands under CRP contracts in the Northern Great Plains (Fargione et al. 2009), although grassland conversion to corn and soybean cropping has exceeded the amount of land area lost from the CRP in the eastern portions of the Dakotas and Nebraska (Wright & Wimberly 2013). Total CRP enrollment as of July 2014 was 10.3 Mha (25.4 million acres), and contracts incorporating 8.8 Mha (21.7 million acres) of CRP land will expire between 2014 and 2022 (USDA-FSA 2014).

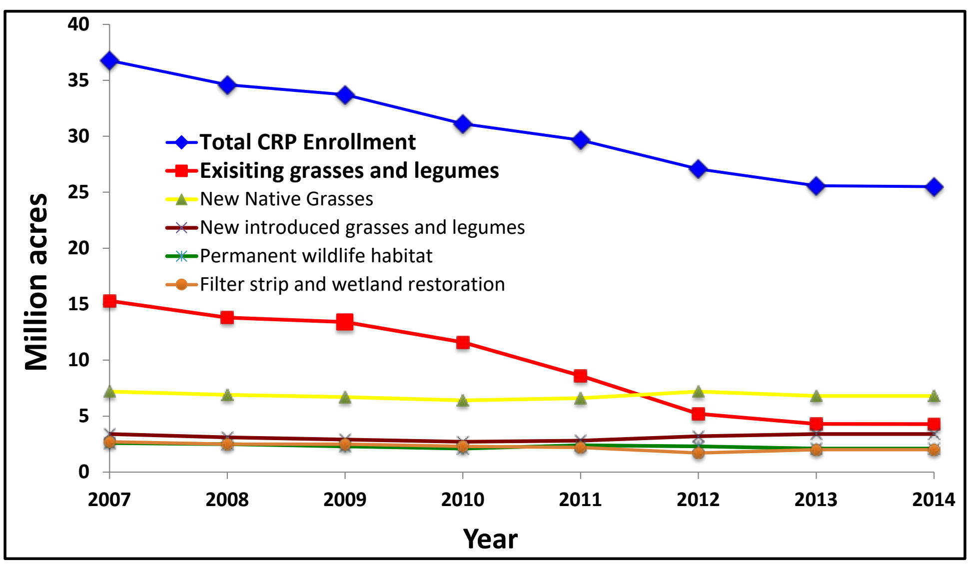

Figure 1. Change in enrolled Conservation Reserve Program (CRP) land and major resource land area from 2007 to 2013 in the U.S. Data were obtained from the Conservation Programs Statistics website (USDA-Farm Service Agency 2014).

There are numerous reported benefits of perennial bioenergy feedstock production over conventional row crop production systems. Impacts of conversion of CRP grasslands to conventional cropping systems on soil and water quality are more certain and assumed to be more negative than those from conversion to managed second-generation (i.e., cellulosic) bioenergy feedstocks based on existing assumptions in the literature to date (Clark et al. 2013). Periodic harvesting of biomass in CRP grasslands may be a beneficial method of removing litter buildup that can reduce the benefits to certain species, particularly if burning is not a viable option (Venuto & Daniel 2010). Other benefits include increasing soil organic matter (Burke et al. 1995; Lee et al. 2007a), increasing biodiversity and wildlife conservation (Fargione et al. 2009; Meehan et al. 2010; Wright & Wimberly 2013), and positive energy and greenhouse gas (GHG) balances (Lee et al. 2007b; Tilman et al. 2006; Schmer et al. 2008; Gelfand et al. 2011; Georgescu et al. 2011; Gelfand et al. 2013). Harvesting biomass on a successional old-field system with N fertilization achieved energy production rates comparable to those with no-till continuous corn cropping (62 GJ $ha^{-1}yr^{-1}$) with much better net GHG balances (-932 and -344 $g_{CO2e}m^{-2}yr^{-1}$, respectively) (Gelfand et al. 2013). Ruan and Robertson (2013) found that N_2O and CO_2 emissions in the initial period following conversion of CRP fields in Michigan to soybean were much higher using conventional tillage compared with no-till practices, and both systems resulted in substantially greater emissions than in the undisturbed CRP field. A carbon debt is expected to be incurred when converting CRP grassland to managed perennial grasses during the transition period only while the use of unconverted CRP grasslands for cellulosic biomass production would avoid any C debt associated with the change in land use (Gelfand et al. 2011).

A perennial biomass feedstock system consisting of a grass monoculture may produce higher yields, although a mixture of various native species may prove to be more resilient and provide greater biodiversity and benefits for wildlife (Venuto & Daniel 2010; Zilverberg et al. 2014). However, most CRP lands have been planted with indigenous species that are often not high yielding (Mapemba et al. 2007). Especially degraded and sensitive lands, particularly those in drier regions, are unlikely to produce appreciable biomass yields given long harvest intervals allowed under CRP contracts. Juneja et al. (2011) estimated that less than 183 l ethanol could be expected per hectare of CRP land in eastern Oregon and Washington per year if harvested every ten years, the allowed harvesting frequency on CRP land in this area due to the especially dry climate. Easing the restrictions on harvest frequency and widening the harvest window were modeled to greatly reduce feedstock production costs (Mapemba et al. 2007). Venuto and Daniel (2010) observed a linear decline in biomass production across a three-year study on CRP land in northwestern Oklahoma which was hypothesized to be due in part to repeated annual harvesting without the addition of fertilizer. Mapemba et al. (2007) assumed no fertilization was necessary to maintain biomass productivity on CRP lands when harvested every second or fourth year.

Production of perennial cellulosic bioenergy feedstocks on CRP land may provide a means to meet the goals of the US Biomass Program while providing landowners additional revenue. Economic viability of producing biomass on idle or marginal land would be linked to the expected price received for the biomass (Khanna et al. 2011), and decisions to convert CRP land would be based on expected biomass revenue minus income from program payments. Since CRP lands would require minimal changes in land use for biofuel production, a life cycle analysis of using these lands for biofuel production shows a substantial reduction in greenhouse gas production compared to first-generation biofuels. Therefore, these lands provide an excellent source of cellulosic feedstock without significant land-use changes while maintaining many of the original environmental benefits of the CRP (Lee et al. 2007a; Clark et al. 2013; Chamberlain et al. 2011).

Objectives

The overall goal of this study is to perform long-term, replicated field trials on CRP land to assess the yield potential and suitability of CRP grassland as a bioenergy feedstock source across logical regions of adaptation. The specific objectives of this project were to establish yield potential over multiple years for CRP grassland grown in different environments using farm-scale agricultural practices that are standard for each region in which there was a test and to determine species compositional changes under agronomic management practices including N fertilization and harvest timing. To implement farm-scale management practices, the experiment was designed with the minimum of 0.5 hectares for an individual plot.

Methods

Six field research locations were identified based on CRP grassland distribution in the United States in 2008 (Figure 2). Each site consisted of predominantly cool-season grasses (those species that grow best during cool, moist periods of the year) or warm-season grasses (those species that grow best during warm periods of the year) (Barnes et al., 2003). The established CRP stands were located at the following sites: Foster County, North Dakota (ND, 47.5° N 99.2°W); Ellis County, Kansas (KS, 38.8°N 99.4°W); Jackson County, Oklahoma, (OK, 34.7°N 99.3°W); Judith Basin County, Montana (MT, 47.6°N 110°W); Boone County, Missouri (MO, 39°N 92.2°W); and Oconee County, Georgia (GA, 33.8°N 83.4°W). The map shown in Figure 2 shows the six sites.

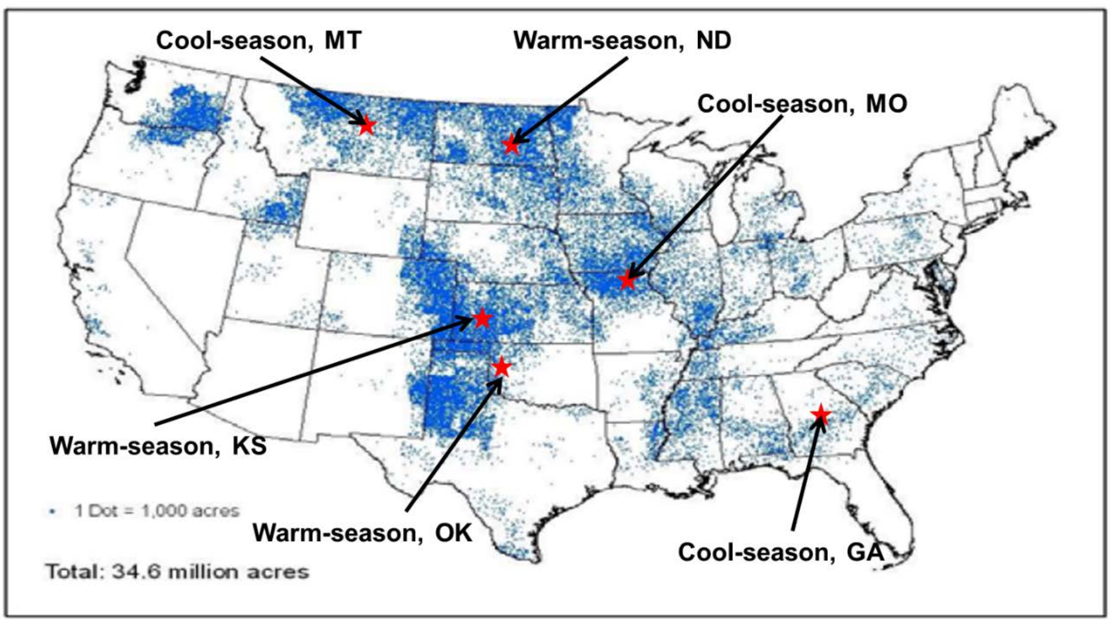

Figure 2. The U.S. Map of Conservation Reserve Program (CRP) enrollment in 2008 and field research locations. One dot equals 405 ha (1000 ac). (CRP enrollment map source, USDA Farm Service Agency).

The selected soil chemical properties in the top 15 cm for each location are shown in Table 1. The predominant plant species varied among the six locations; C_4 grasses at the ND, KS, and OK sites, C_3 grasses at the MT, MO and GA sites (Table 1). In addition to the C3 grasses, alfalfa was also a predominant species at the MT site. All locations had been managed in accordance with CRP regulations, including no N fertilization and/or aboveground biomass harvest since the start of the contract until fall 2007. All field sites were selected in spring 2008 and mowed at a 10- to 15-cm height in the spring before imposing fertilization treatments.

The experimental design was a factorial arrangement of three N rates and two harvest dates within a randomized complete block with three replicates at each location. The plot size for treatments was approximately 0.5 ha. Urea nitrogen (N) fertilizer was annually broadcasted with the rates of 0, 56, and 112 kg N ha^{-1} onto each plot using a farm-scale fertilizer spreader on the dates shown in Table 2. No other fertilizer was applied as a treatment, however phosphorus was added to some of the sites early in the experiment to increase deficient levels. The fertilizer spreader was calibrated for the rate of 56 kg ha^{-1} and applied to that respective treatment, while for the 112 kg N ha^{-1} treatment plots, the spreader went over the plots twice, each at 56 kg ha^{-1}.

To monitor the effect of agronomic practices on changes in species composition, species composition at each site was estimated annually, during June and July according to the dry-weight–rank procedures described by Gillen and Smith (1986).

Table 1. Selected soil chemical properties in the top 15 cm for each location in the Regional Feedstock Partnership (from Lee et al., 2013).

State	CRP Established	Soil pH	SOC§	T-N†	Soil Classification	Predominant Species*
			-----g kg^{-1}------			
ND	2001	7.8	20.8	2.90	Haploborolls	BB, SW
KS	1988	7.6	24.3	1.90	Argiustolls	SO, SW, LB, YS
OK	1998	6.6	5.9	0.64	Haplustalfs	SW, LB
MT	2008	7.4	22.0	3.22	Calciboroll	A, PW
MO	2004	5.0	19.0	2.12	Epiaqualfs	RC, TF
GA	1986	6.2	10.4	0.81	Kanhapludul	TF, OR

§ SOC: soil organic carbon; † T-N: soil total nitrogen; * BB: big bluestem, SW: switchgrass, SB: smooth bromegrass, SO: sideoats grama, LB: little bluestem, YS: yellow sweetclover, A: alfalfa, PW: pubescent wheatgrass , RC: red clover, TF: tall fescue, OR: orchardgrass

Biomass yield was determined from a whole plot harvest with a farm-scale harvester at a cutting height of 10- to 15-cm on the dates listed in Table 2. For warm-season CRP sites, biomass was annually harvested either at the anthesis (peak standing crop, PSC) or after a killing frost (AKF). For cool-season CRP sites, biomass was annually harvested either at the anthesis (peak standing crop, PSC) and/or at the end of growing season (EGS) depending on location. PSC harvest timing was determined at each location by the predominant species (as listed in Table 1) reaching anthesis. The MT site had a single cut system at either PSC or EGS. For the GA site, biomass at PSC treatment was harvested only in the spring; however, EGS treatment had two cuts and was actually a combined mass of both spring (PSC) and fall (EGS) harvests.

For the MO site, biomass in both treatments was harvested in early and late in spring, respectively and both treatments were harvested again at the end of year, and early harvest was considered as PSC and late harvest was considered as EGS for data presentation. The detailed harvest timing and frequency information is described in Table 2. Above ground biomass for each plot was baled with a large round baler, weighed, and then sub-sampled. Subsamples were collected from the bales using a core sampler (5-cm diameter and 50-cm long) attached to an electric drill and were dried at 60^{o}C for 48 h in a forced-air oven in order to determine dry matter concentration and feedstock chemical composition. No harvest was conducted in OK in 2011 due to insufficient biomass production caused by drought. Final harvests were made in 2012 in GA and MT and in 2013 at the remaining sites.

Table 2. Harvest and fertilizer application dates for each CRP research site. PSC-peak standing crop: date at which the predominant species reached anthesis; KF/EGS- date which killing frost (KF) occurred at the sites with warm-season mixtures or the end of the growing season (EGS) at the cool-season sites (from Lee et al., 2014).

Year	State					
	ND	KS	OK	MT	MO*	GA**
	PSC					
2008	7-Sep	29-Aug	n/a	8-Jul	16-May/15-Jul	8-Jun
2009	3-Sep	13-Aug	9-Sep	26-Jun	18-May/21-Jun	27-Apr
2010	24-Aug	22-Jul	7-Sep	29-Jun	27-May/23-Jun	24-May
2011	24-Aug	30-Jul	n/a	5-Jul	2-Jun/27-Jun	24-May
2012	9-Aug	25-Jul	25-Sep	5-Jul	14-May/22-Jun	8-May
2013	9-Aug	23-Jul	12-Oct	n/a	4-June/27-Jun	n/a
	AKF-EGS					
2008	31-Oct	31-Oct	27-Oct	2-Oct	9-Oct	3-Oct
2009	23-Oct	21-Oct	4-Dec	23-Oct	18-Oct	20-Oct
2010	23-Oct	5-Nov	11-Nov	28-Oct	1-Nov	4-Oct
2011	1-Nov	29-Oct	n/a	27-Oct	17-Oct	26-Oct
2012	1-Oct	26-Oct	22-Dec	18-Oct	29-Oct	22-Oct
2013	9-Oct	12-Nov	14-Jan	n/a	8-Nov	n/a
	N-Application					
2008	2-Jun	1-Jul	15-Jul	20-Apr	13-Mar	n/a
2009	15-Jun	24-Mar	24-Jun	13-May	16-Mar	16-Apr
2010	21-May	21-Apr	28-Jun	24-Apr	19-Apr	6-Apr
2011	11-Jun	22-Mar	5-Jun	13-May	18-Mar	8-Apr
2012	22-May	28-Mar	18-Jun	20-Apr	15-Mar	22-Mar
2013	12-Jun	19-Mar	18-Jun	n/a	18-Mar	n/a

n/a- not applicable
*MO: both early and late harvest treatments at PSC were harvested again at EGS. Late harvest treatment in spring was indicated as EGS.

** GA: EGS treatment had two cuts at both PSC and EGS

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Normality of the residuals was evaluated using box plots in the UNIVARIATE procedure and equality of the variances was evaluated using plots of the observed versus predicted residuals with SAS software (SAS Institute, 2012. The SAS System for Windows, Version 9.4. SAS Institute, Inc., Cary, NC). Statistical analyses were performed using the PROC MIXED procedure in SAS at α=0.05. Year, nitrogen rate, harvest timing and the N rate x harvest timing interaction term were considered fixed variables while block was considered a random variable. Locations were analyzed separately due to the differences in predominant grass species and harvest timing protocols. Single degree-of-freedom contrast statements were used to determine significant differences among treatments. Pearson product–moment correlation was used to identify significant correlations between biomass yield and precipitation levels using the CORR procedure in SAS.

Results and Outcomes

Conservation Reserve Program land is a potentially important land resource for sustainable biomass feedstock production. Based on the 2005 Billion Ton study (USDOE, 2005), 10.3 million ha of CRP land could annually provide about 50 million tons of dry biomass with an annual yield of 4.3 Mg ha^{-1} (1.9 T ac^{-1}). Our study indicated that the maximum biomass yields occurred when 112 kg N ha^{-1} was applied annually and biomass was harvested after a killing frost. Yields using these practices ranged from 3.4-6.0 Mg ha^{-1} (156-2.7 T ac^{-1}) for the three cool-season CRP sites and from 4.0-7.2 Mg ha^{-1} (1.8-3.2 T ac^{-1}) for the three warm-season CRP sites (Lee et al., 2013).

N fertility and harvest timing

The data in Figure 3 show that nitrogen fertility had a significant effect on biomass yield at all locatio Maximum Yield Mean ss all years of the study. The N rate of 112 kg N ha^{-1} produced the highest biomass yield at each location (Fig. 3a). Biomass yields with 112 kg N ha^{-1} were significantly higher than those with 56 kg N ha^{-1} at all locations except KS and MT. Yields increased from 0 to 56 kg N ha^{-1} at all sites although the difference was not significant at GA (Fig. 4a). Yield response to N fertility rate was generally low at CRP locations where legumes comprised a relatively high percentage of the mixture (i.e., KS, MT and MO according to Table 1). Optimal harvest timing was site-specific and was a function of predominant grass type, seasonal precipitation regime, and number of harvests taken per year (Fig. 3b). Interaction between Year and N rate was significant in KS, ND and OK because the yield response at both 56 and 112 kg N ha^{-1} was lower from 2011-2013(data not shown). This is likely due to dry conditions during this period. The AKF/EGS harvest timing produced the highest yields at GA and MO among cool-season grass sites and at ND and OK among warm-season sites. Yields at MT were significantly higher with the PSC harvest timing (Fig. 4b). Although no obvious pattern was observed over time between yields harvested at PSC and AKF/EGS for all locations, delayed harvesting until after a killing frost or at the end of growing season (AKF/EGS) secured maximum biomass and persistence of desirable species (Figs. 5 and 6).

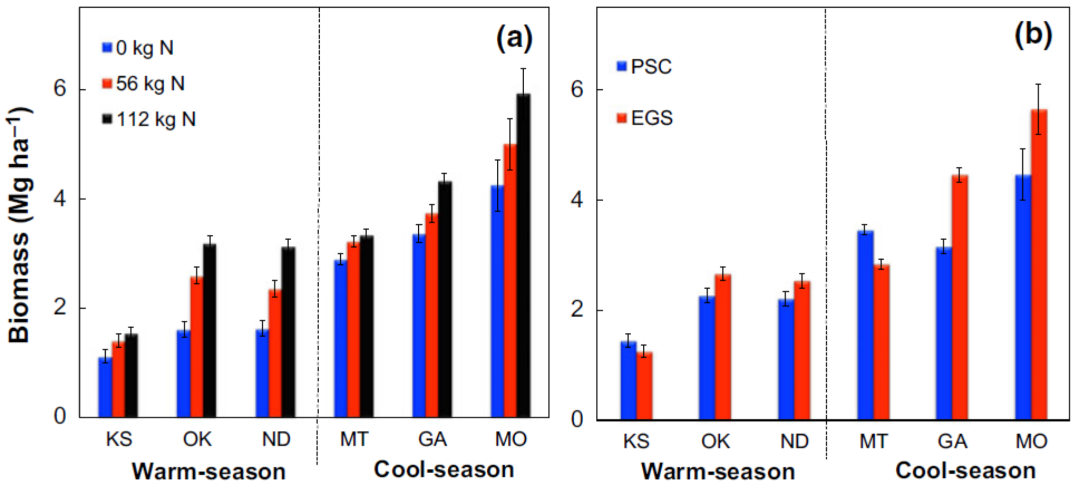

Figure 3. Biomass yield as affected by (a) N rate and (b) harvest timing for CRP sites with predominantly warm-season or cool-season grass mixtures. Yields were averaged across all harvest years. PSC: peak standing crop, at anthesis; EGS: end of the growing season. Harvests designated as EGS include those conducted at warm-season grass locations after a killing frost (AKF) and cool-season grass sites with one- and two-cut systems where the last harvest was conducted at the end of the growing season. Bars represent standard errors of the differences of means when analyzed by location (α=0.05) (From Lee et al., 2018).

Mean biomass yields under the N rate of 112 kg N ha^{-1} and delayed harvest (AKF/EGS) which were considered to be best management practices (BMP), and were significantly greater (range of 25% -114% higher) than yields under control (0 kg N ha^{-1} and AKF/EGS harvest) at each location (Fig. 2). Warm-season mixture CRP lands produced a biomass yield of 2.1 Mg ha^{-1}, and cool-season mixture CRP lands produced a biomass yield of 3.8 Mg ha^{-1} when averaged over time and across treatments. Under BMP, biomass yields of 2.9 and 5.1 Mg ha^{-1} were recorded for warm- and cool-season mixture CRP land, respectively, when averaged over time. As we observed, N fertilization was a key management factor for biomass production on CRP land. However, higher N fertilization for maximum biomass production may not be best economically or environmentally (Anderson et al., 2016).

Long-term yield trends

Biomass yields tended to increase with increasing precipitation at most sites (Figs. 4), although the critical period for precipitation differed among sites. Precipitation timing and amount at each site were primary factors in annual biomass yields. At MO, ND and GA, the highest correlation between yield and precipitation was observed for the April through June period. At KS, MT and OK, highest correlation was observed for the April through August, September and October period, respectively.

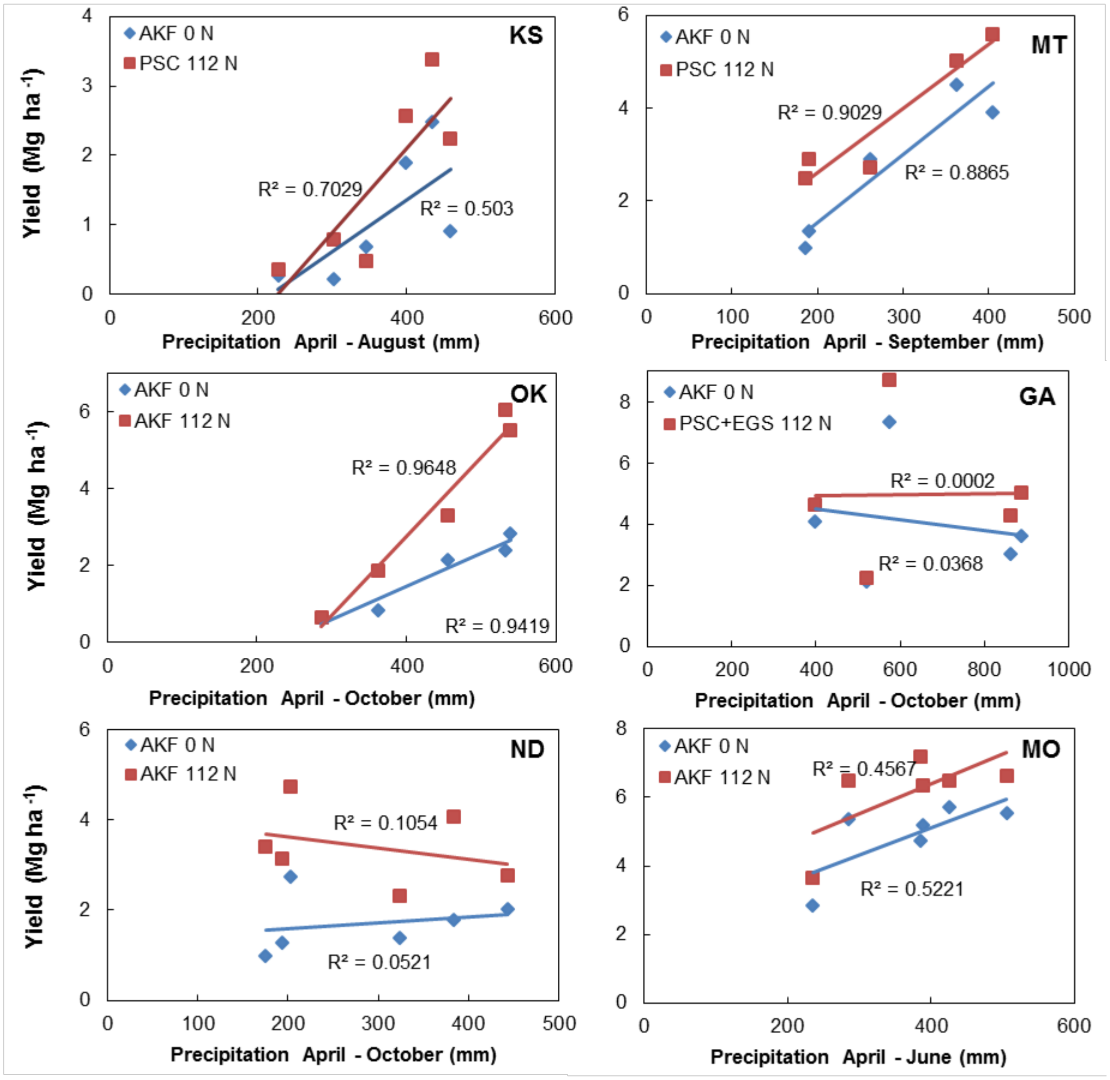

Figure 4. Yield response with control (♦, 0 kg N ha^{-1}) and management regimes for maximum biomass (■, 112 kg N ha^{-1}) at each CRP site with predominantly warm-season grass mixtures (KS, OK, ND) and cool-season grass mixtures (MT, GA, MO) and the correlation with precipitation during the most critical part of the growing season. Each data point represents a mean for an individual year. AKF: after a killing frost; PSC: peak standing crop, at anthesis; PSC+EGS: two harvests conducted annually, the latter occurring at the end of the growing season. (Figure adapted from Anderson et al., 2016).

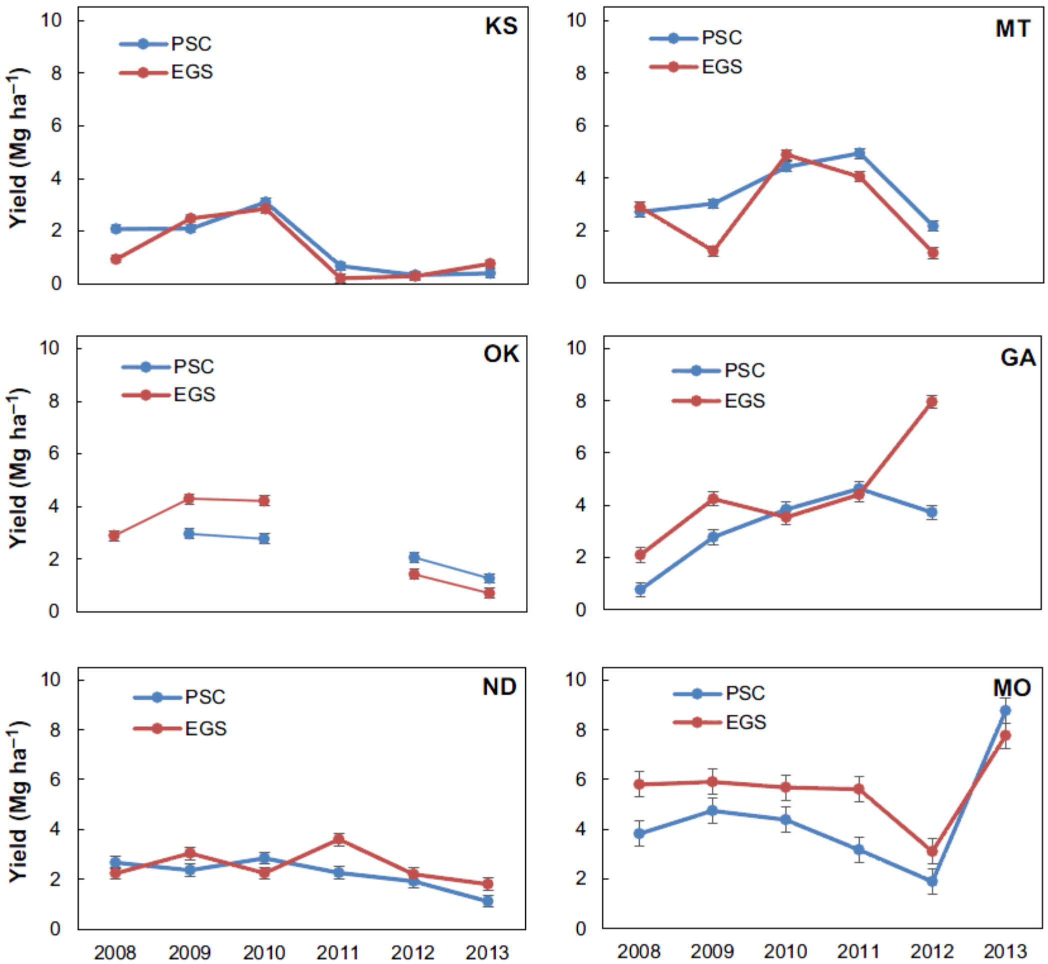

Figure 5. Biomass yields from 2008 to 2013 averaged across N rates at warm-season (KS, OK, ND) and cool-season (MT, GA, MO) grass locations. Bars represent standard errors of the differences of means when analyzed by location (α=0.05). AKF: after a killing frost; PSC: peak standing crop, at anthesis; PSC+EGS: two harvests conducted annually, the latter occurring at the end of the growing season (from Anderson et al., 2016).

Aside from the decline in yields during drought years, no trend was observed with respect to yields over time (Fig. 5). Biomass yields during the first three years (2008-2010) were much higher than those during the last three years (2011-2013) for all locations exception with GA. The main reason of low biomass yield during the period was lack of precipitation during the growing season (April through October) (Fig. 4). In particular, limited biomass yields in KS and OK during 2011 through 2013 were caused by severe drought in the Great Plains region.

The PRISM-ELM map of feedstock production potential of the CRP grassland was created based on data generated from the Feedstock Partnership field trials (Figure 6). The PRISM-ELM model well represented the biomass yield potential of the CRP grassland estimated from the Feedstock Partnership field trials. As the CRP grasslands were not established for biomass production, data from both the field trials and the PRISM-ELM model indicated the feedstock production potential of the CRP grassland is less than 4 Mg ha^{-1}.

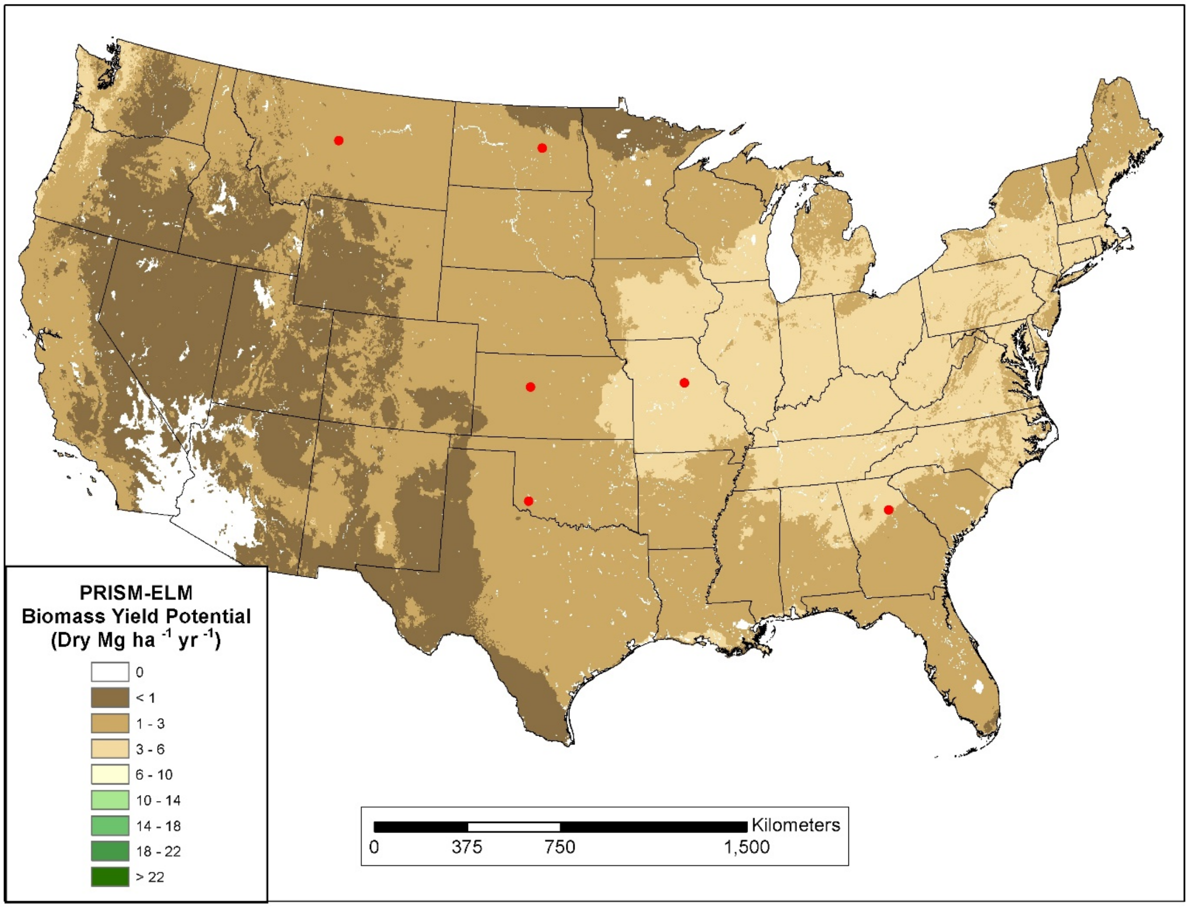

Figure 6. Biomass yield potential of mixed grasses in the Conservation Reserve Program (CRP) lands for the U.S. generated using the PRISM-ELM model and based in part on Regional Feedstock Partnership Field Trials (red dots) (from Lee et al., 2018).

Species composition

Three of the sites representing variable climatic parameters were analyzed for vegetation composition trends over the course of six growing seasons (2008-2013). For the other three locations, neither changes in species composition was observed nor was data collection completed due to drought and early termination. The two locations with legume components, Missouri (Table 3) and Kansas (Table 4), responded similarly to fertilization treatments, with greater legume composition at the low fertilization levels. Sweetclover in Kansas and red clover in Missouri were greatest in plots without added nitrogen. Legume composition was lower at the Missouri site after only one season of high 112 kg N ha^{-1} fertilization. Average legume composition and the linear trend, or slope of the lines for legume composition across years, were different for the low and high fertilization treatments in Missouri and in Kansas. The two locations with warm-season grasses, Kansas (Table 4) and North Dakota (Table 5), showed that individual species responded to harvest date to a greater extent than fertilizer treatment. Harvests at PSC were detrimental to individual grass species, especially switchgrass at both locations. Even big bluestem composition declined with PSC harvests during the growing season in ND,

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while harvests at AKF had little change in individual species composition. Average composition of most warm-season grasses were different between harvests, and so were the linear trends over time for each harvest in ND. When harvested at PSC, as in this study, most warm-season grasses are fully active and in reproductive stages of development, during which most non-maintenance photosynthate is allocated to the developing seed. A decrease in tallgrass species and an increase in annual cool-season grass weed species composition occurred over time regardless of fertilization at the Kansas site. Different linear trends for switchgrass, sideoats grama, and little bluestem were detected between harvest timing in Kansas. A large part of the weed species increase in Kansas in the last 3 years could be attributed to late winter precipitation utilized by Japanese brome, and a lack of May through June precipitation for warm-season grass growth.

Table 3. Species composition responses to nitrogen fertilization (kg N ha^{-1}) in cool-season mixture Conservation Reserve Program (CRP) land at Missouri.

Species	Treatments	Avg.†	Year						P<0.05 linear trend
			2008	2009	2010	2011	2012	2013	
	N Rate								
Legumes	0	34a	38	47	34	31	27	26	a
	50	26b	26	35	28	28	24	13	a
	100	19c	16	25	22	19	19	12	b
Cool-season grasses	0	56c	56	45	58	60	57	60	ab
	50	67b	69	58	66	66	68	77	a
	100	76a	82	71	73	75	73	83	b

*Within a vegetation class, different letters indicate statistical difference at P<0.05 between fertilization rates.

Table 4. Species composition responses to nitrogen fertilization (kg N ha^{-1}) and harvest timing (AKF: after a killing frost; PSC: peak standing crop at anthesis) in warm-season mixture Conservation Reserve Program (CRP) land at Kansas.

Species	Treatments	Avg.†	Year 2008	2009	2010	2011	2012	2013	P<0.05 linear trend
	N Rate								
Legumes	0	17a	23	17	59	1	1	1	a
	50	11b	24	8	32	0	0	0	ab
	100	8b	23	5	17	0	0	0	b
	Harvest								
Switchgrass	PSC	10b	15	19	9	5	4	6	a
	AKF	15a	15	17	13	14	12	17	b
Sideoats Grama	PSC	25a	20	24	15	41	24	24	a
	AKF	22a	22	25	15	31	22	18	b
Little Bluestem	PSC	8b	19	12	8	6	3	2	a
	AKF	15a	20	19	12	20	10	9	b
Indiangrass	PSC	9a	10	16	9	17	2	2	a
	AKF	9a	14	13	9	14	3	3	a
Japanese brome*	PSC	20a	0	6	12	8	54	39	a
	AKF	14b	0	1	9	3	41	31	a

†Within a species, different letters indicate statistical difference at P<0.05 between harvest timing or fertilization rate, respectively.
*Average Japanese brome composition for PSC and KF were statistically different at P<0.09.

The results presented here demonstrate, using field scale agricultural practices, that CRP land will shift vegetative composition over time based on harvest and fertilization management of CRP land for biomass feedstocks. Any shift by mismanagement over time to less desirable or less productive species will hinder the ability of CRP land to adequately provide a sustainable or reliable resource for bioenergy feedstock production. Cool-season species dominant mixtures did not have much impact of harvest and N fertility management on changes in species composition other than declining of legume species under N fertilization. However, warm-season species dominant mixtures had significant impacts of harvest timing management with over time declining of desirable species by early harvesting.

All field data and management practice information during 2008-2013 are available for use by others researching this area (Lee et al., 2018). Also a series of outputs including peer reviewed manuscripts, outreach publications, proceedings and presentations are available for scientific, ag-professional, and farming communities.

Key outputs

Peer Reviewed Publications

Anderson, E. K., E. Aberle, C. Chen, J. Egnolf, K. Harmoney, G. Kakani, R. L. Kallenbach, M. Khanna, W. Wang, and D. K. Lee. 2016. “Impacts of Management Practices on Bioenergy Feedstock Yield and Economic Feasibility on Conservation Reserve Program Grasslands.” *GCB Bioenergy* 8 (6): 1178–90. doi:10.1111/gcbb.12328.

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Chen, C., J. Heser, T. Porter, and D. K. Lee. 2012. “Nitrogen Application and Harvest Timing Affect Biomass Yield and Composition on CRP Grassland.” In *Proceedings of the 2012 Sun*

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Lee, D. K., E. Aberle, C. Chen, J. Egnolf, K. Harmoney, V. G. Kakani, and R. L. Kallenbach. 2009. "The Regional Biomass Feedstock Partnership: Herbaceous Energy Crops and CRP Land for Biomass Production across Environmental Gradients, CRP Land for Biomass Energy Production in the U.S." Presented at the Sixth Annual World Congress on Industrial Biotechnology and Bioprocessing, Montreal, Canada, July 19–22.

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Where do we go from here?

The Conservation Reserve Program was originally established for soil and water conservation, not biomass production. However, CRP land is a potentially important land resource for sustainable biomass feedstock production. Accordingly, in order for CRP to be a reliable source of sustainable biofuel feedstock, management considerations must be taken into account that can produce sustainable stands of desirable species and provide ongoing conservation services.

This study evaluated grasslands planted under the CRP retirement program as a herbaceous biomass source with potential use as a dedicated bioenergy feedstock and well covered the range of CRP land distribution and long-term trend. The results presented here demonstrate, using farm scale agricultural practices, that CRP land is a potential resource for bioenergy feedstock production if the appropriate management practices are followed under normal precipitation during the growing season. However, CRP lands could increase biomass production through renovating CRP grassland to high yielding species and/or cultivars recently developed for biomass feedstock production, since current species and cultivars are not necessary for high biomass yield.

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Energycane

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Summary

Energycane germplasm tested as part of the Sun Grant/DOE Regional Feedstock Partnership has shown that energycane can produce 22 – 25 Mg ha^{-1} yr^{-1} dry matter (DM) yields (or 9.8 – 11.1 tons ac^{-1} yr^{-1}) at the most northern locations (33°N latitude) and in excess of 45 DM Mg ha^{-1} yr^{-1} (i.e., 20.1 tons ac^{-1} yr^{-1}) at the southern locations. Maximum yields were generally achieved in the fourth year (third ratoon) at all locations, except St. Gabriel, LA and College Station, TX. At the latter two locations, maximum yields were observed during the second ratoon crop. At a majority of the locations, the genotypes Ho06-9001 and Ho06-9002 had the greatest mean dry matter yield over the seven years included in this study (2009-2015).

Energycane has significant sugar in the sap of the stem. °Brix (a measure of soluble carbohydrates in the sap) is highly affected by weather, especially rainfall. When measured after a rainfall event it would be lower, and after a significant period of drought, higher. Based on mean °Brix, there was an estimated 8 to 12% of dry weight of the stalk in sugar in the extracted sap. Genotypes had different levels of extractable sap, which relates directly to the moisture content of harvested stalks. The woody types (Ho06-9001 and Ho06-9002) and the intermediate type (Ho02-144) were all lower in extractable sap compared to the pithy types (Ho 72-114 and Ho72-147).

While it is possible at 33°N latitude to sustain growth of the energycane genotypes tested here, mean dry matter yields of other bioenergy crops (e.g., lowland switchgrass and giant miscanthus) are equal to or greater than these energycane genotypes for the same northern locations without the need for specialized infrastructure.

Introduction

Sugarcane is bred for large stalk diameter, low fiber content and high sugar content under Louisiana and Florida growing conditions. This limits the northern expansion of sugarcane varieties. During the 1960s mosaic virus threatened the sugarcane industry in Louisiana. USDA-ARS Sugarcane Research Unit at Houma (LA) had wild germplasm (*Saccharum spontaneum*) that would cross with sugarcane genotypes it had obtained that originated from the Himalayas. Testing indicated the Himalayan accession was resistant to mosaic virus (Hale, personal communication). In addition to mosaic virus resistance, this parent was cold tolerant. Crosses of this Himalayan parent with sugarcane caused the offspring to be more cold tolerant; however, the progeny are nearly always lower in sugar, making them useless for the sugar refining industry. During the “oil shocks” of 1973 and 1979 Louisiana State University (LSU) selected one of the hybrid progeny of a sugarcane by Himalayan parent cross. The reasoning was to capitalize on the high biomass and high fiber content of this progeny. LSU eventually released L79-1002, a cane specifically targeted to biomass/bioenergy uses (Bischoff et al. 2008), which is recognized as the first energycane variety made available for commercial uses. USDA-ARS Sugarcane Research Unit at Houma (LA) continued a small program on energycane development throughout the 1990s, but started to focus on cold hardiness as a desirable trait. In fall of 2007, Drs. Thomas Tew and Ed Richard sent billets of 11 genotypes north to Mississippi State University (Starkville, MS) for general assessment and winter hardiness screening. Five genotypes were deemed suitable for continued testing at latitudes north of 32°N latitude (roughly Interstate 20).

Energycane, like sugarcane, is a tropical perennial that is vegetatively propagated. A crop can be harvested and the subsequent year's crop grows back from the surviving crown (ratooning). Unlike most other summer crops, energycane is established in the fall from mature canes of existing plants. Because energycane is vegetatively propagated, vigor observed in F_1 hybrids of the sugarcane x *S. spontaneum* cross is maintained from field to field.

Establishment of a field follows the same process as commercial sugarcane. Mature canes (seedcane) of the desired genotype are harvested in August/September. The apical meristem is removed to stimulate shoot growth from lower nodes. New fields are plowed to create beds and furrows. Canes are placed horizontally in the furrow overlapping by one-third, and the soil from the bed is cast down over the canes to bury them. In roughly two to three weeks shoots emerge. These shoots are killed by the first fall frost, but the cane and crown of the new shoots remain protected underground. In spring new shoots emerge in the spring and will grow through the summer. Being tropical in origin, energycane doesn't undergo a natural senescence. Growth slows in the fall because of cooler temperatures, but a killing frost is required to stop growth. Failing natural senescence, a frost-kill traps nutrients in the above-ground plant parts. Removal of this material removes the minerals trapped in it. Immediately following the killing frost, moisture levels initially rise in the plant (root pressure keeps water flowing into the plant, but there isn't transpirational loss). Thus harvest is made on "wet" material (60-70% moisture) altering preservation/storage requirements. Unlike switchgrass and giant miscanthus, which can be baled, energycane must be consumed directly from the field, or ensiled anaerobically for storage.

Energycane was selected by the DoE to be investigated further because biomass yields of sugarcane, and energycane in the cane-growing regions of Louisiana can approach 67 Mg ha^{-1} yr^{-1} dry matter (stalks + sugar). This biomass production exceeds by nearly double, the other biomass crops currently being investigated (switchgrass, giant miscanthus and sorghum). At the onset of this project, there was limited energycane germplasm and no testing for yield outside of southern Louisiana. Hence, the objective below.

Objectives

The objective of this project was to evaluate energycane hybrids for biomass yield across fairly wide geographic range. Replicated field trials of five genotypes common to all eight locations across five states (Georgia, Hawai'i, Louisiana, Mississippi and Texas) to evaluate the potential production and sustainability of energycane as a bioenergy feedstock in diverse environments. Additionally, it is desirable to know the duration of productivity of a plant stand.

Methods

Five energycane lines that were tested at Starkville from 2006-2008 were selected for broader testing across the Southeast and on Hawai'i as part of the Regional Biomass Feedstock Partnership. Little was known concerning the area of adaptation and cold hardiness of this germplasm. Some may find it odd to be testing cold hardy energycane in Hawai'i, but as the population increases, the agricultural lands near the coast are being converted to housing, pushing cane production up the mountainsides into colder clines. The genotypes were: Ho 02-147; Ho 02-144, Ho 72-114. Ho 06-9001, and Ho 06-9002 (Table 1).

Table 1. Identity, pedigree and fiber percentage of five energycane genotypes originating from USDA-ARS Sugarcane Research Unit (Houma, LA); tested at all eight locations between 2009 and 2015.

Germplasm Line	Pedigree	Mean Fiber (%)
Ho 02-147	F_1 (*S. spontaneum* x domestic sugarcane)	12
Ho 02-144	F_1 (*S. spontaneum* x domestic sugarcane)	8
Ho 06-9001	F_1 backcrossed with *S. spontaneum*	11
Ho 06-9002	F_1 backcrossed with *S. spontaneum*	11
Ho 72-114	F_1 backcrossed with domestic sugarcane	8

In August and September of 2008, seed cane was distributed to seven test sites (Tifton, GA; Auburn, AL; Raymond and Starkville, MS; St. Gabriel, LA; Beaumont and College Station, TX) (Figure 1). Crop failure at the Auburn site caused an alternate site to be selected at Athens, GA. Waimānalo, HI was added in 2009. Athens, GA and Starkville, MS were the most northern locations (33°N latitude). Sites at each location were chosen as “prime farmland” indicating the ability to produce at least average yields of other area crops. Planting was accomplished at all locations within three days of seed cane delivery. Some sites included other sugar or energycane genotypes, but all locations had the same five genotypes in common. Because germplasm was new, plot size was limited. Individual genotypes were planted in plots 9 m long x 3 rows (16 m) wide. Two rows would be harvested for yield estimates, the third would be used for growing season data. Plots of all five genotypes occupied a space 9 m by 30 m; this was replicated four times within a field at each location. Field and plot size were limited by the amount of seedcane that could be obtained from existing fields in Starkville, MS and Houma, LA. During the following spring (2009) emergence data (shoots/plot), date of 50% emergence, and soil temperature at 15 cm was monitored. Over the course of the growing season, mean height and °Brix (a measure of soluble carbohydrates in the sap) were recorded. Site scientists recorded major factors potentially impacting yield (drought, hurricanes, and extremes of temperature, insects or disease). Harvest date varied by location, depending on frost date and weather conditions. At the end of each growing season, stalk count, final mean height, a general frost damage rating, final °Brix, fresh harvest weight were recorded. From the sacrifice row, sap yield, stalk moist weight (after pressing sap out) and dry weight were recorded. Dry pressed stalks were ground and analyzed for structural carbohydrates (cellulose, lignin, sugar). During summer 2015 the Continental sites were in their sixth ratoon crop (seven years of data). Hawai’i, which joined the program in 2009 and had a one year quarantine is reporting its fourth ratoon crop after the 2015 growing season. Because Waimanalo, HI, and St. Gabriel, LA, are located in sugarcane production areas, yields (taken on site as cane weight) at these locations were converted from fresh (wet) weight to a dry weight estimate by multiplying the cane weight by percentage fiber. Dry matter yields at other sites were calculated by determining dry matter content of fresh cane.

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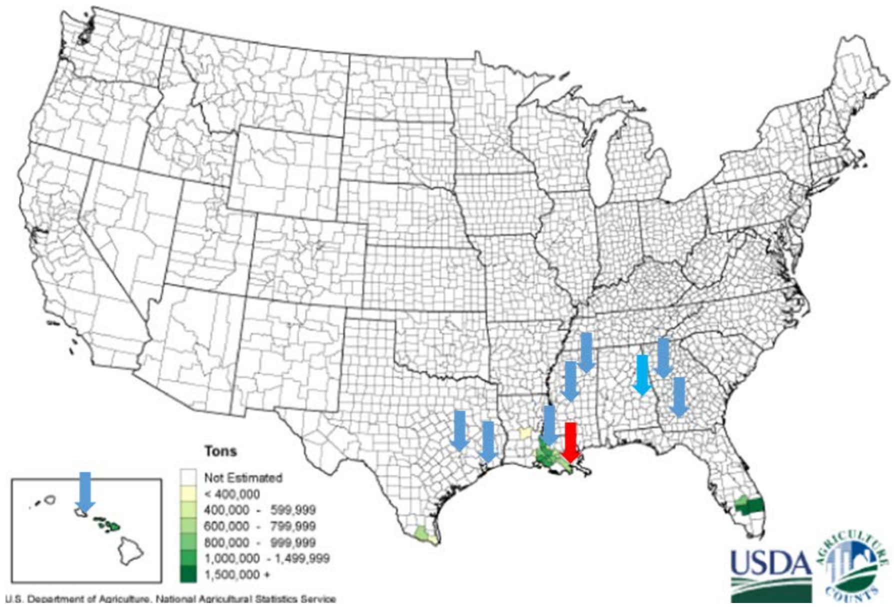

Figure 1. Location of test sites for energycane (*Saccharum* spp. (blue arrows) as part of the Regional Feedstocks Partnership (2009-2015). Green shaded areas indicate location of commercial sugarcane production in the U.S. Light blue = Auburn, AL; replaced starting in 2010 with Athens, GA. Red arrow = Houma, LA, source of all germplasm used in this testing. (Source of image: USDA-National Agricultural Statistics Service)

Results and Outcomes

As expected, most characteristics varied by variety and location. Height of all germplasm increased throughout the summer into the fall. Height measurements indicated onset of “grand growth” (the point at which growth accelerates rapidly) occurred in June or July. The date of onset differed for each of the sites. Regardless of location or year, the onset of grand growth corresponded to a mean ambient air temperature of 30°C. Grand growth ceased (heights stopped increasing) after mid-September, which corresponds to daytime temperatures cooling to below 30°C. There was a significant location by cultivar interaction, which means that the best performing cultivar may vary by location. Average yield for each genotype at each location across all years is presented in Table 2 while average yield at each location across years and genotypes is presented in Table 3.

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Table 2. Average annual dry biomass yield (Mg ha^{-1}) of five energycane genotypes tested at eight locations between 2009 and 2015 (from Lee et al., 2018).

Location	Variety				
	Ho 02-144	Ho 02-147	Ho 06-9001	Ho 06-9002	Ho 72-114
Athens, GA	19.7 a*	15.9 b	20.8 a	20.6 a	14.0 c
Tifton, GA	23.8 c	25.5 c	31.0 a	32.1 a	28.5 b
Waimānalo, HI	38.6 a	38.0 a	39.2 a	35.1 a	39.7 a
St. Gabriel, LA	20.1 b	22.2 ab	23.7 a	20.5 b	22.3 ab
Raymond, MS	12.1 c	15.5 ab	15.2 abc	17.8 a	13.3 bc
Starkville, MS	16.9 b	12.9 c	20.2 a	18.9 ab	12.6 c
Beaumont, TX	32.8 b	38.8 ab	42.1 a	39.5 ab	43.5 a
College Station, TX	22.4 ab	19.2 b	20.5 b	21.1 ab	24.4 a
Variety average	23.3 B†	23.5 B	26.6 A	25.8 AB	25.0 AB

*lowercase letters indicate significant differences among variety means within location at α=0.05.
†UPPERCASE letters indicate significant differences among variety means across all locations at α=0.05.
‡Hawaiian location mean is a five-year average.

Table 3. Average annual dry biomass yield (Mg ha^{-1}) of five energycane genotypes by year for eight locations between 2009 and 2015 (from Lee et al., 2018).

Location	Year							***Location average***
	2009	2010	2011	2012	2013	2014	2015	
Athens, GA[a]	8.3 e*	24.4 b	6.2 e	22.8 bc	30.5 a	13.9 d	21.3 c	18.2 CD†
Tifton, GA	29.2 c	29.5 c	34.0 b	39.8 a	22.1 d	14.7 e	27.9 c	28.1 AB
Waimānalo, HI	-‡	-	37.4 ab	45.2 a	37.2 bc	41.0 ab	29.7 c	38.1 A
St. Gabriel, LA	16.6 e	17.5 de	31.4 a	18.1 de	19.5 d	21.7c	27.5 b	21.8 BC
Raymond, MS	17.5 a	12.3 b	11.6 b	13.7 ab	14.1 ab	17.5 a	16.8 a	14.8 D
Starkville, MS	17.2 c	16.9 c	21.6 b	26.7 a	22.7 b	6.7 d	2.4 e	16.3 D
Beaumont, TX	-	46.3 a	30.7 b	30.0 b	28.5 b	50.3 a	50.2 a	39.3 A
College Station, TX	13.6 d	22.9 bc	16.1 d	26.8 b	22.3 c	17.6 d	31.3 a	21.5 BCD

[a] Athens was planted in 2009.
*lowercase letters indicate significant differences among year means within location at α=0.05.
†UPPERCASE letters indicate significant differences among location means across all years at α=0.05.
‡Hawaii entered the Feedstock Partnership late (2009) due to legislation and quarantine.

Athens, GA

Across all years and locations, biomass yield for the genotypes ranged from 13.96 to 20.55 DM Mg ha^{-1} yr^{-1} (6.23 to 9.16 tons ac^{-1} yr^{-1}) and was significantly different between the five cultivars (Figure 2). The pithy type, Ho 72-114 had the lowest average biomass yield across the seven years. Similarly, across all years, the woody types, Ho 06-9001 and Ho 06-9002, had the highest average biomass yield along with Ho 02-144 (Table 2). The intermediate true hybrid (Ho 02-147) had a lower average yield than the woody types, but was higher than the pithy type (Ho 72-114).

There was a very strong year by genotype interaction effect on biomass yield. Most of the interaction appears to be due to annual differences in biomass yield. Biomass yields were the lowest in 2011 due to an extremely cold event during the winter of 2010/2011. A cold front passed over the South, but stalled over northern Georgia (8-11 January, 2011). Low temperatures reached -7°C over the course of four days. The extreme winter cold coupled with a relatively dry spring and summer worked together to limit growth and thus yield. Biomass yields were highest in 2013, where spring and summer were unusually wet. The mean rainfall for the months of June and July at this location were more than twice the normal (87 vs 41.2 mm).

There was a slight change of biomass yield ranking among the five cultivars over the years, especially between the true intermediate hybrid, Ho 02-144, and the two woody hybrid backcrosses (Ho 06-9001 and Ho 06-9002; Table 3). The pithy genotype Ho 72-114 had the lowest yield every year, except for the establishment year 2009 when it exceeded all five cultivars in biomass yield (Figure 2).

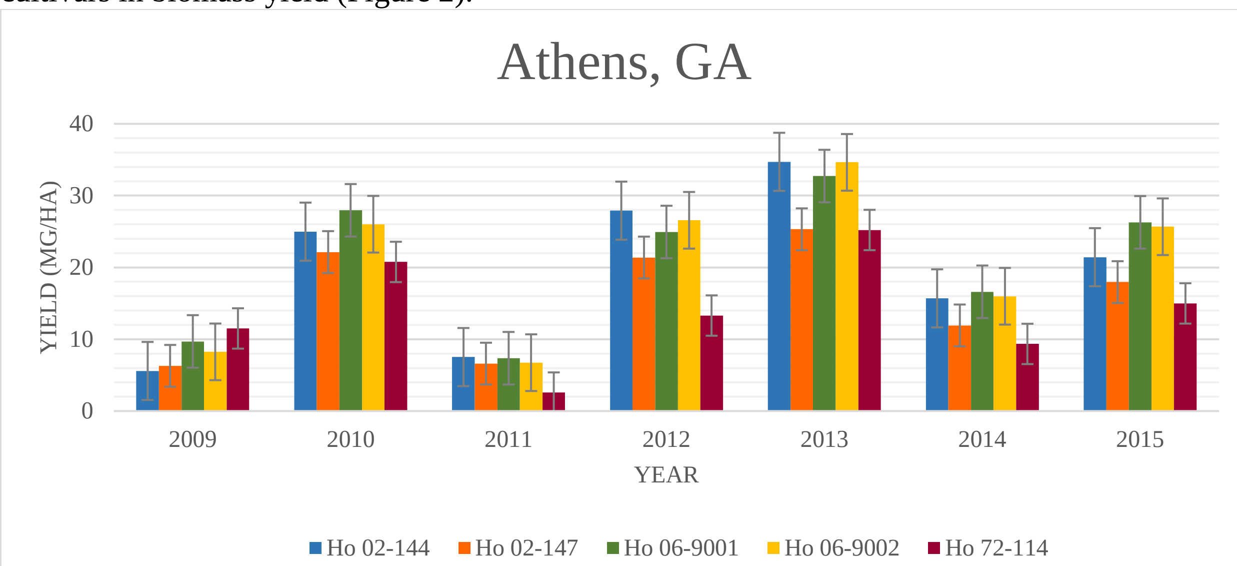

Figure 2. Dry weight yield of five energycane genotypes from 2009 – 2015 at Athens, GA.

Starkville, MS:

There was a significant effect of year on the five genotypes tested as well as an interaction, indicating that all genotypes did not respond the same in any given year. In the first year there were no significant differences among the genotypes in the test (Figure 3). After the first year of testing, the woody types (Ho 06-9001 and 9002) maintained the greatest DM yields for four of the seven test years. In 2010, dry matter yields of the two woody types (Ho06-9001 and Ho 02-9002) were greater than the other energycane genotypes (Figure 3). For the next two years (2011 & 2012) the woody types and Ho02-144 (an intermediate type) had the greatest yields (23 – 25 Mg ha^{-1} in 2011, and 31 to 31.75 Mg ha^{-1} in 2012). Averaged across all 5 genotypes, dry matter yields reached their peak in 2012 or 2013 and declined thereafter (Figure 3). Severe DM yield decline of all genotypes was observed in 2014, after a colder, wetter than average winter. The decline continued in 2015. Soil pathogens are believed to have taken hold in

2014 and continued to manifest in 2015. Sugarcane aphid (*Sipha flava*) was present at Starkville, but it preferentially infested sweet sorghum.

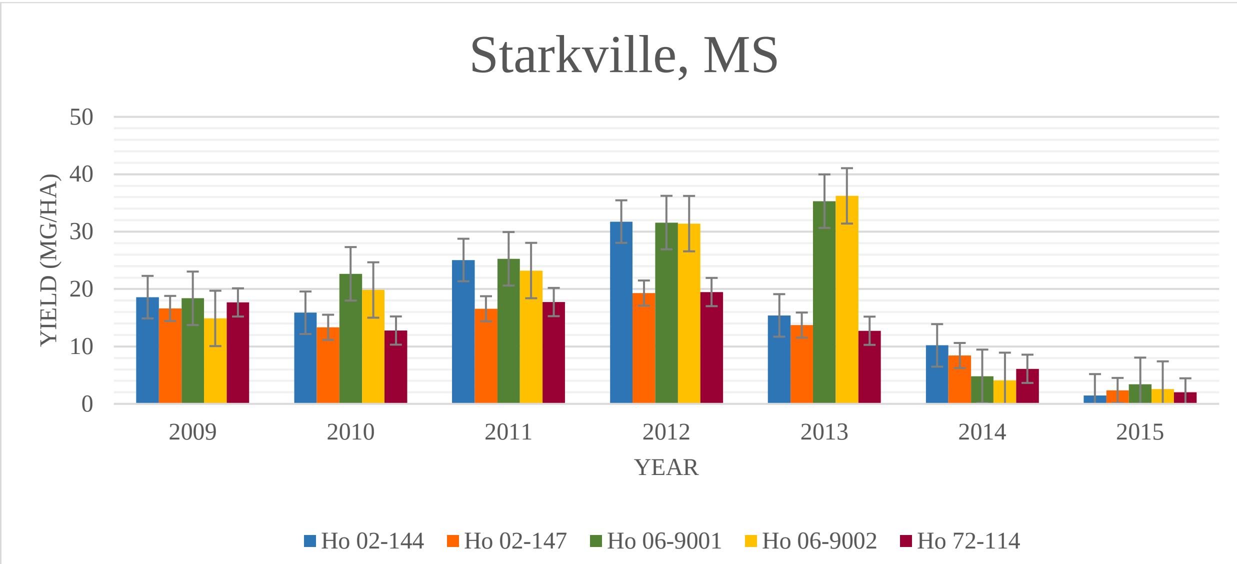

Figure 3. Dry weight yield of five energycane genotypes from 2009 – 2015 at Starkville, MS.

Raymond MS

As at the other locations, there was an effect of year on DM yield. The establishment year, 2014 and 2015 produced the greatest mean DM biomass yields (Figure 4; Table 3). Mean DM yields by genotype ranged from 12.1 (Ho 02-144) to 17.8 (Ho 06-9002) Mg ha^{-1} yr^{-1} (Table 2). The genotype Ho 06-9002 yielded significantly more biomass in 2014 and 2015 than other genotypes at this site (Figure 4).

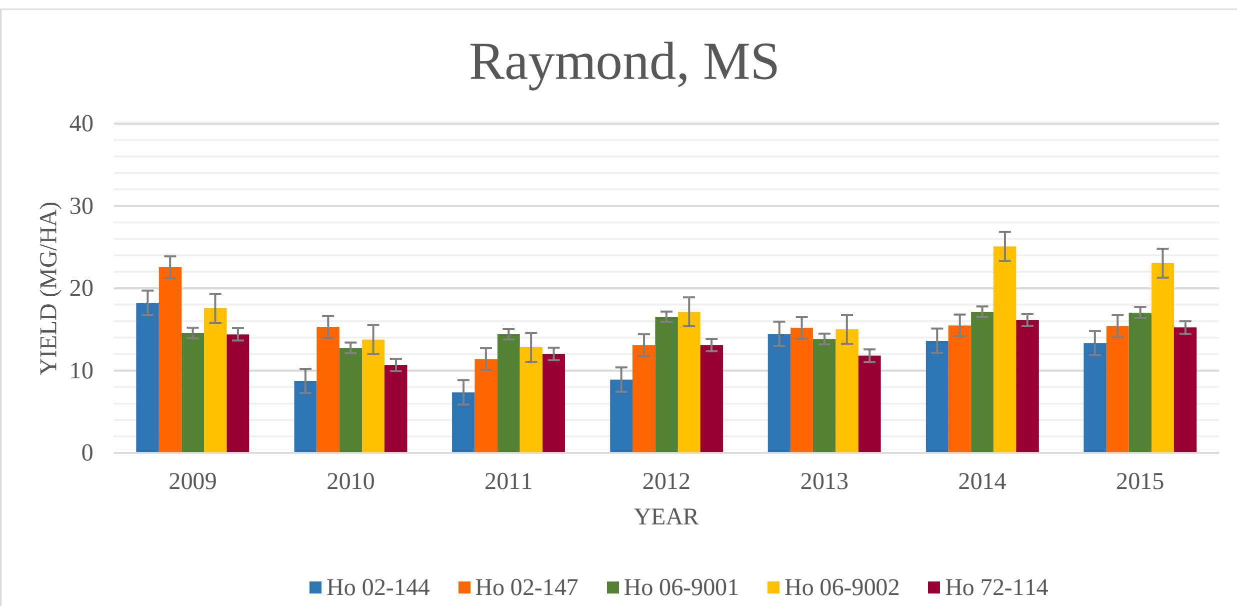

Figure 4. Dry weight yield of five energycane genotypes from 2009 – 2015 at Raymond, MS.

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College Station, TX

At College Station, establishment was hampered by drought. During the establishment year (2008), and the subsequent growing season (2009) a 7 cm irrigation was applied to the field to ensure establishment. There was a significant effect on biomass yield due to year, most likely attributable to rainfall amounts. Lowest yields were observed in 2009 and 2014 (Figure 5), which coincide with low rainfall years. During the March to September 2009 growing season College Station received 510 mm of rain, and none of it occurred in June nor July. Conversely, the greatest yield was observed in 2015 corresponding to record high rainfall for east Texas and Oklahoma. Cumulative March to September rainfall was 762 mm, effectively doubling mean DM yields compared to the yields during drought years. Averaged across all years at College Station, the genotype Ho 72-114 had the highest DM yields (Table 2), as it did at Beaumont, TX.

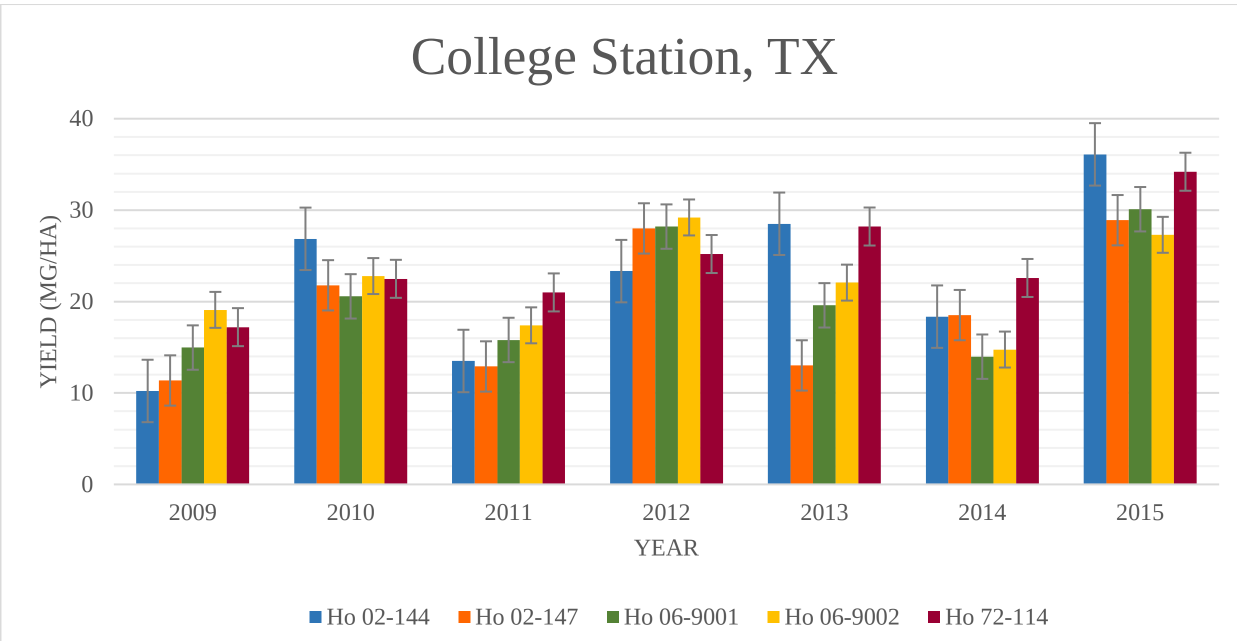

Figure 5. Dry weight yield of five energycane genotypes from 2009 – 2015 at College Station, TX.

Tifton, GA

There was a significant entry by year interaction for yield at Tifton. There were also significant year and genotype effects. The two entries that were hybrids back-crossed to wild *S. spontaneum* (Ho 06-9001 and Ho 06-9002) had the highest DM yields from 2010 to 2015 (Figure 6). Among all genotypes, yields decreased from 2012 through 2014 and then recovered in 2015. The reduction observed in 2013 may have been partially due to a much higher amount of rainfall and lower than normal temperatures. Heavy rains, cloudy weather and unseasonably cool temperatures in July and August 2013 may have retarded growth. The exceptionally cold winter of 2013-14 appeared to have delayed spring regrowth in 2014 and affected plant growth throughout the year which was reflected in the lower 2014 DM yields. This reduction in yield was also observed at Athens, GA and Starkville, MS. All genotypes recovered in 2015 after slow early growth. This site ranked third regarding total mean DM yield (Table 3).

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Energycane plots were monitored for pests. Tip smut (*Sporisorium scitaminea*) was observed only one year (2011), and only on a single variety of energycane (L79-1002), which was not a part of the testing reported here.

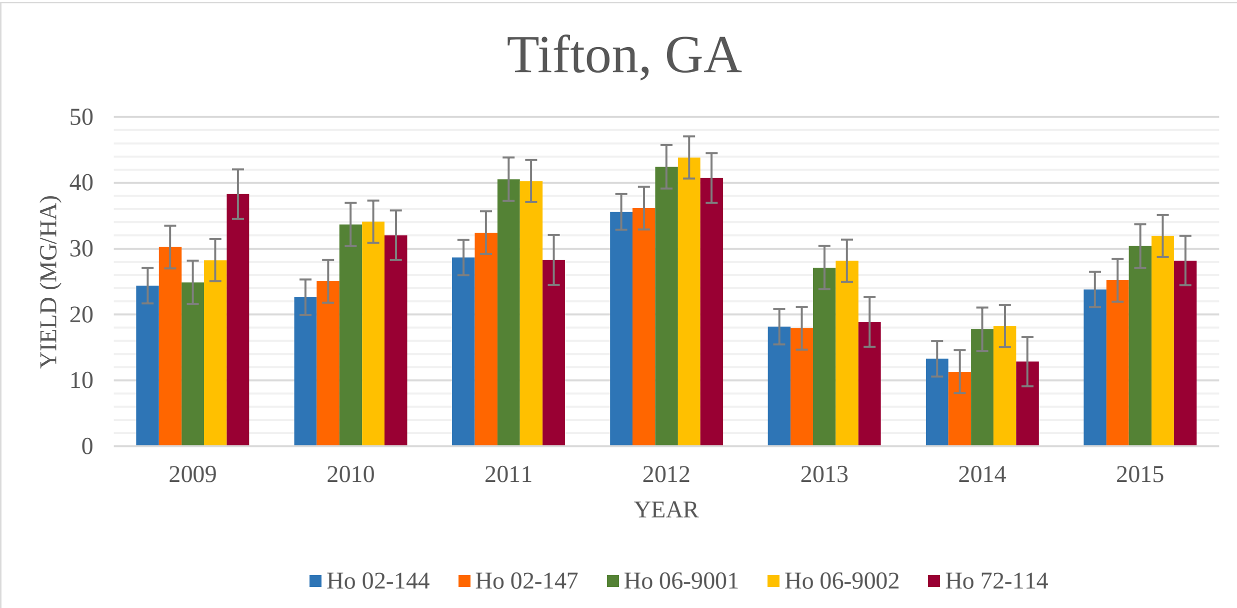

Figure 6. Dry weight yield of five energycane genotypes from 2009 – 2015 at Tifton, GA.

Beaumont, TX

Harvest reporting was delayed a year because tornadoes spawned by Hurricane Rita (22 September 2009) caused the entire crop to lodge. Data analysis indicated significant year and year by genotype interaction. Year had the greatest effect on energycane DM yield. The 2014 harvest produced the highest overall average yield observed at any site (50.3 Mg ha^{-1}) followed by 2015 (50.2 Mg ha^{-1}) and 2010 (34.4 Mg ha^{-1}) (Figure 7; Table 3). Increased yields in 2014 and 2015 are probably a result of an extra accidental fertilization (2.5 times normal application of N) in 2014. During 2009 through 2013 and 2015, 112 kg N/ha was applied in March and 225 kg N/ha was applied in April. In 2014, the crop received 112 kg N/ha and 225 kg N/ha both applied in March, with a third application of 225 kg N/ha also applied in April. In 2015 there was greater than average rainfall and carryover fertilizer likely contributed to greater than average yields that year. Beaumont was one of the three top yielding sites. It should be noted that even without the high yields of 2014 and 2015 induced by fertilizer, mean yields at Beaumont would still rank it in the top three.

The energycane research plots were not treated for insects; however, 10 to 20 percent of energycane internodes at this site typically had symptoms of stem borer damage by the end of the growing season. This exceeds current sugarcane treatment thresholds.

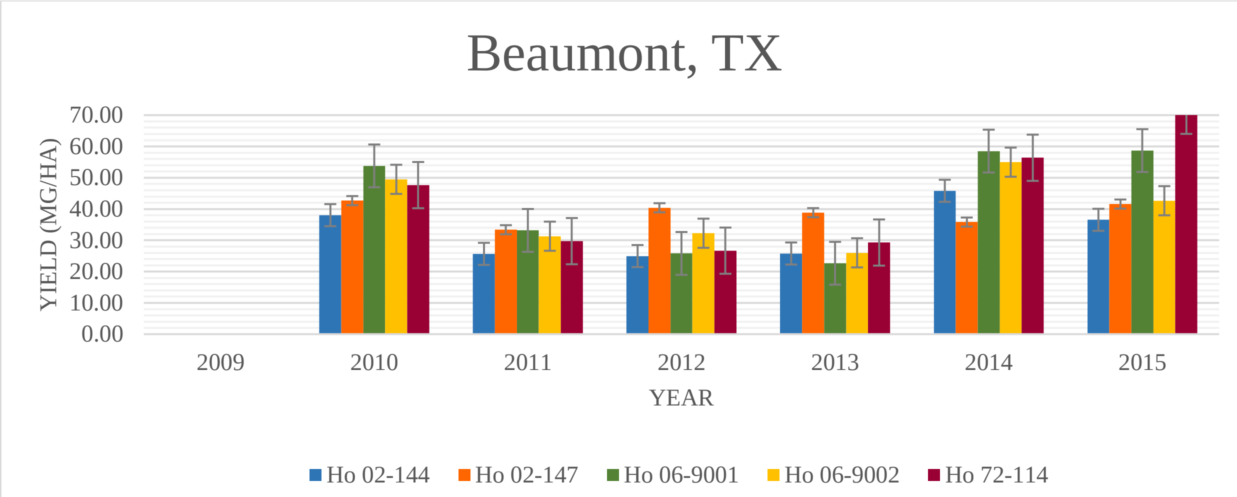

Figure 7. Dry weight yield of five energycane genotypes from 2009 – 2015 at Beaumont, TX.

St. Gabriel, LA

There was a significant entry by year interaction for yield at St. Gabriel. As expected, the weather conditions across the course of the study were variable. Because St. Gabriel is located in a sugarcane production area, yields (taken on site as cane weight) were converted from fresh (wet) weight to a dry weight estimate by multiplying the cane weight by percentage fiber. Greatest genotype yields were observed in 2011 while 2015 ranked second (Figure 8). The lowest average yields were observed during the establishment year. At St. Gabriel, when averaged across all years the genotype Ho 06-9001 out yielded Ho 06-9002 and Ho 02-144 (Table 2). While St. Gabriel is in the heart of Louisiana sugarcane production land, disease and insect pressure within the energycane yield trial was reported to be negligible.

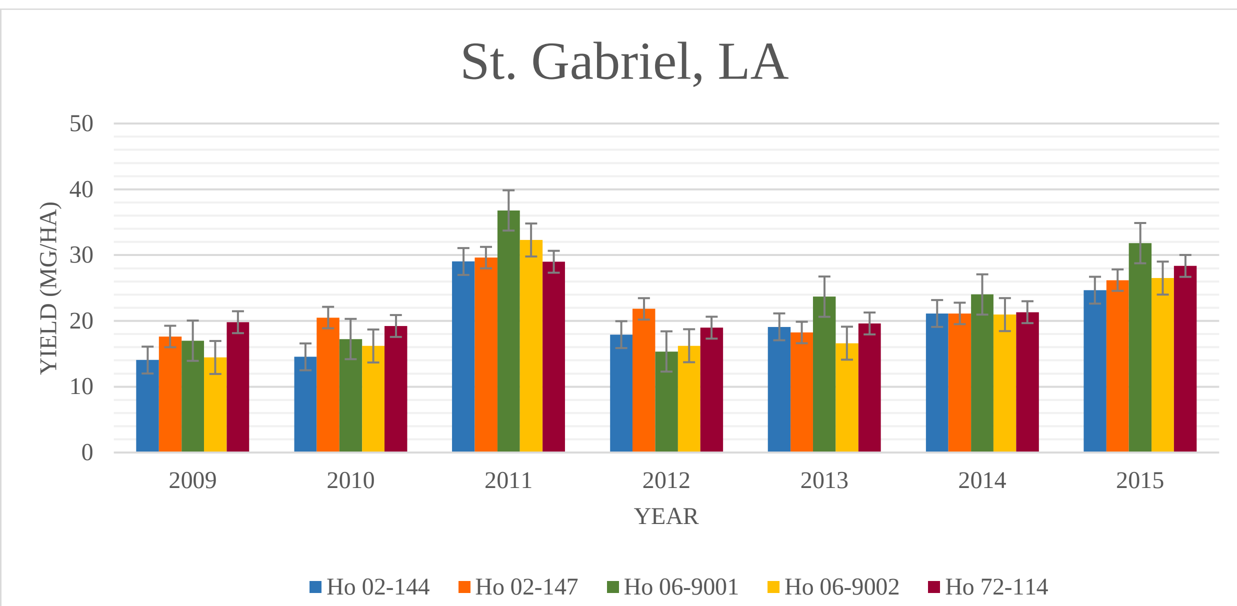

Figure 8. Dry weight yield of five energycane genotypes from 2009 – 2015 at St. Gabriel, LA. Dry weights were calculated by multiplying wet cane weight by fiber percentage at this location.

Waimānalo, HI:

This location was delayed entry to the national test because Hawai'ian law prevented the importation of sugarcane and its relatives to the islands. In 2009 that law was repealed and energycane billets were shipped directly from the USDA-ARS Sugarcane Unit (Houma, LA) in November. Mandated hot water sanitation treatment damaged the nodes on the billets and set back emergence and growth. During the first year (2009-2010) the plants we grown under quarantine to screen for sugarcane mosaic and ratoon stunting viruses, making the first harvest year 2011 There was a genotype effect as well as a genotype by year effect. Among genotypes, when averaged across all years there were no significant differences in DM yield (Table 2). Across years, average DM yields ranged from 29.7 to 45.2 Mg ha^{-1} (Table 3). Greatest average yields were recorded in 2011, 2012, and again in 2014; there was a steep decline in DM yield for all but one genotype in 2015 (Figure 9). As expected, Waimānalo was among the top three yielding energycane sites, second to Beaumont (Table 3).

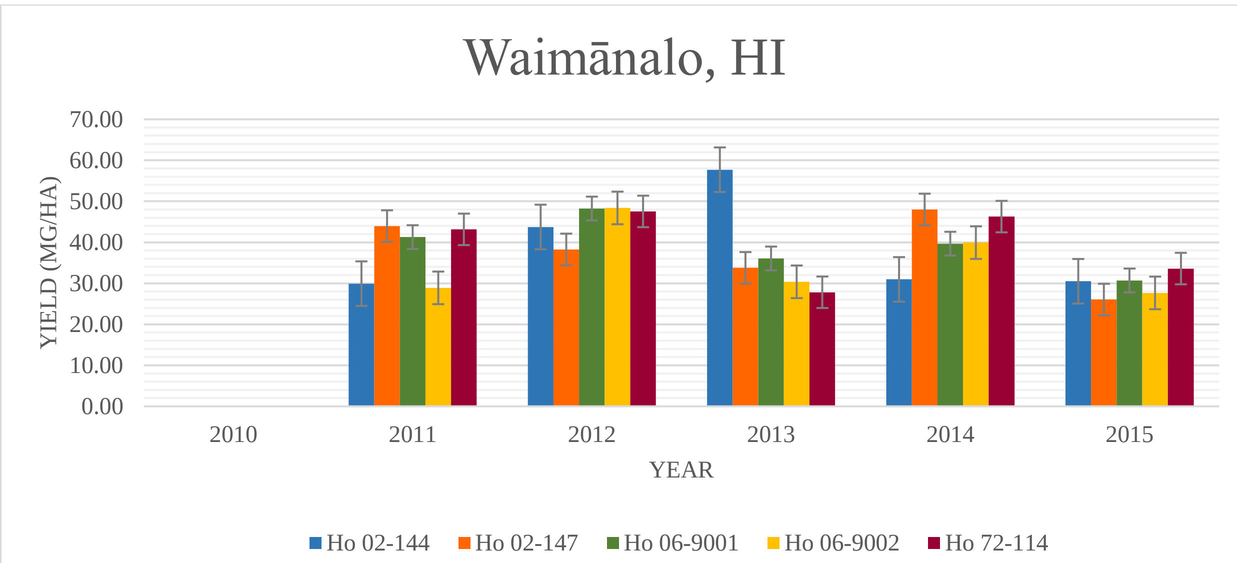

Figure 9. Dry weight yield of five energycane genotypes from 2011 – 2014 at Waimānalo, HI. Dry weights were calculated by multiplying wet cane weight by fiber percentage at this location.

°Brix varied by location, variety, weather and time of growing season. °Brix values at all locations dropped substantially two weeks after frost, but remained relatively stable until harvest, presumably due to cooler temperatures. Germplasm differences for °Brix were not uniform across locations. Some genotypes accumulated more sugar than other genotypes at a one location, and less sugar than other genotypes at other locations.

Like sugarcane, energycane stores a significant amount of carbohydrates in the stem. Efforts to make sugarcane cold hardy by crossing it to *S. spontaneum* increase cold hardiness at a cost; namely, a decrease in the ability to store simple carbohydrates/sugars. While sugarcane is generally reported to have a °Brix of 12 – 15, sap extracted from the five energycane genotypes tested ranged from 8.8 – 11.7 (data not shown). The volume of extractable sap also varied by genotype. Genotypes Ho 06-9001 and Ho 06-9002 had less extractable sap than Ho 02-147 and Ho 02-144, which had less than Ho 72-114. The differences noted are due to hybridization and backcrossing to the woody parent, *S. spontaneum*. Genotypes Ho 06-9001 and Ho 06-9002 are hybrids that were backcrossed to *S. spontaneum*, making them 75% *S. spontaneum* and 25%

sugarcane. While this increased cold tolerance, it strongly reduced stem sugar storage and extractability. The remaining genotype, Ho 72-114 is genetically 75% sugarcane: 25% *S. spontaneum*; the hybrid backcrossed to sugarcane in an effort to increase sugar storage potential in the stem. This sugary genotype performed poorly at Athens, GA; Starkville, MS and Raymond, MS, the northern-most test sites, likely due to the limited genetic contribution (25%) of cool hardiness of the *S. spontaneum* parent.

°Brix is highly variable, but fiber percentage is not. Mean fiber percentages are generally fixed (± 1-2%) for the five genotypes across locations. Mean fiber percentages were; 8% for Ho 02-144, Ho 72-114 and Ho 06-9001; 11% for Ho 06-9002 and 12% for Ho 02-147 (Table 1).

While soluble carbohydrates in the sap would be considered a beneficial attribute, dry matter yield is the most important attribute. Of the eight locations; highest yields were observed at Beaumont, TX; Waimānalo, HI and Tifton, GA. This was followed by St. Gabriel, LA; and College Station, TX. Lowest yields were observed at the three most northern sites; Athens, GA: Starkville, MS and Raymond, MS. Yields at Beaumont in 2014 and 2015 were artificially inflated due to an accidental duplication of nitrogen fertilizer applied in 2014.

In 2011, Hawai'i's yields for the five common energycane genotypes had a mean of 37.4 DM Mg ha^{-1}, similar to Beaumont, TX (30.7 Mg ha^{-1}) (Table 3). It is important to note, that this was Waimānalo's first year of growth, and that is being compared to sites on the mainland in their third year of growth.

After the second and third ratoon crop yields started to decline, with the exception of the Raymond site (which always had modest yields). This decline coincides with observations in sugarcane fields suggesting a maximum of three to four productive years before replanting is necessary. It should be noted, if the test had not continued into 2014 (fifth ratoon crop) yield declines would not have been noticed at Starkville, MS, nor Athens, GA.

Because of the longer growing season, plant height and generally plant yield were greater at southern locations compared to northern locations. Energycane is highly responsive to nitrogen fertilization and rainfall as evidenced by changes in DM yield at Beaumont when 2.5 times the nitrogen was applied accidentally, and at College Station in 2015 when rainfall exceed twice average amounts.

At the more northerly locations, extremely cold winters limited and caused production to decline, especially if the cold front persisted for more than two days. However, inclusion of these locations allow the breeders at USDA-ARS, Houma, LA to differentiate between lines that are more cold-hardy than others.

Energycane field scientists from all sites and modeling scientists from Oregon State University's PRISM Climate Group, as well as Oak Ridge National Laboratory assembled together to generate the PRISM-ELM yield potential map for energycane (Figure 10). Yield data from each location, as described above, was combined with climatic parameters to determine an assessment of yield at locations across the southern U.S.

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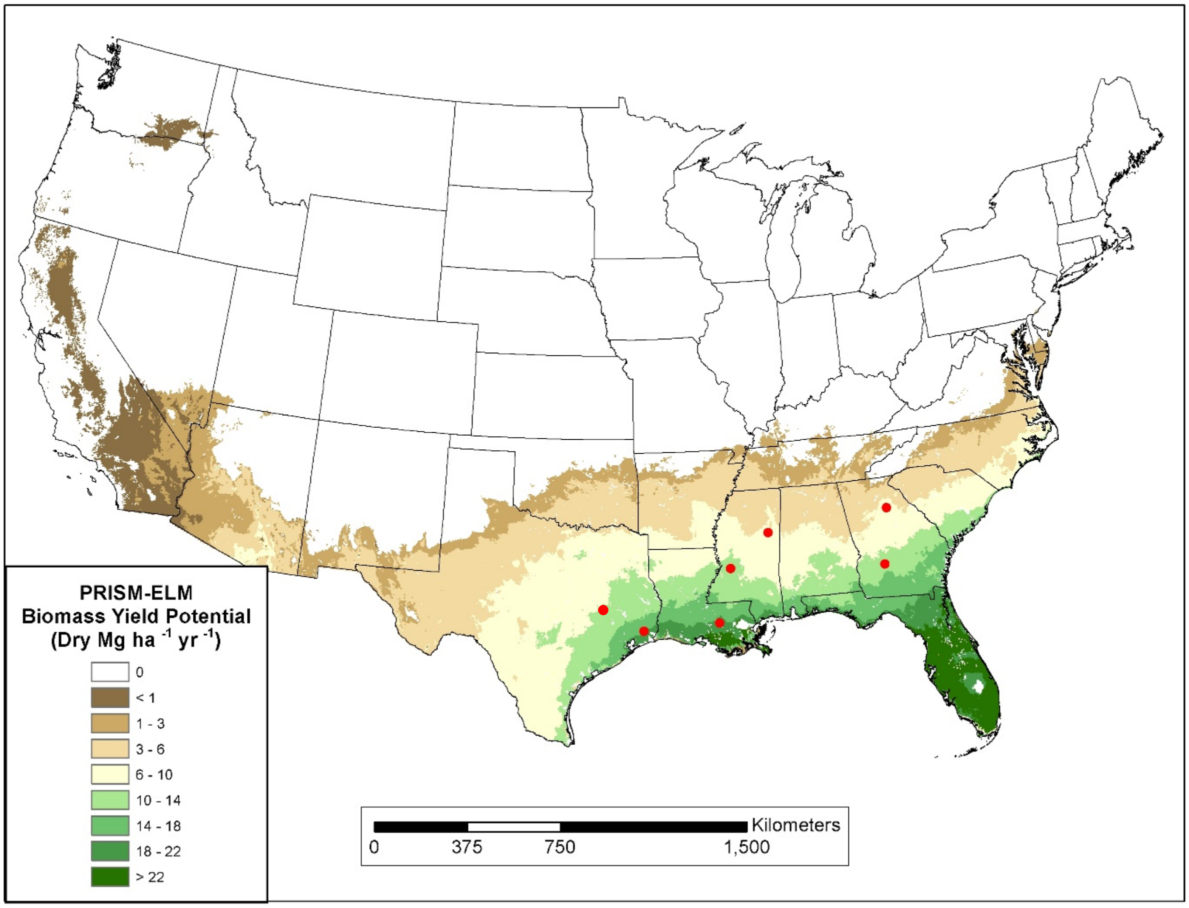

Figure 10. Biomass yield potential of energycane for the U.S. generated using the PRISM-ELM model and based on in part on Regional Feedstock Partnership Field Trials (red dots). An eighth site was located in Waimānalo, HI (from Lee et al., 2018).

The presence of potentially troublesome insects; sugarcane borer (*Diatraea saccharalis*), Mexican rice borer (*Eoreuma loftini*) were reported at Beaumont, TX and Raymond, MS. Sugarcane aphid (*Sipha flava*) was noted at several of the locations as far north as Starkville. While it was noted that these insect pests were above the economic thresh hold for sugarcane, they were considered negligible on energycane. Sugarcane smut (*Sporisorium scitaminea*) was reported at Tifton, GA in 2011, but only in a single variety (L79-1002) which was not part of the five genotypes in common to all locations. Discussion among scientists has raised the potential concern that growing energycane across the entirety of the South would provide pests from western cane production areas a migration route to Florida production areas. Given the frequency of examples of pest migration in the literature, this thesis should not be ignored.

In summary, the new germplasm/varieties tested in this work have shown that energycane can produce 13.3 – 18.8 DM Mg ha^{-1} yr^{-1} yields at the most northern locations (33°N latitude) and in excess of 30 DM Mg ha^{-1} yr^{-1} at the southern locations. However, at locations from Raymond, MS north, other biomass crops (lowland switchgrass and giant miscanthus) produce similar or greater yields. These two temperate grasses have an advantage in that they don’t require the specialized infrastructure for harvest and planting.

Key Outputs

Peer reviewed manuscripts

Lee D.K., E. Aberle, E.K. Anderson, W. Anderson, B.S. Baldwin, D. Baltensperger, M. Barrett, J. Blumenthal, S. Bonos, J. Bouton, D.I. Bransby, C. Brummer, P.S. Burks, C. Chen, C. Daly, J. Egenolf, R.L. Farris, J.H. Fike, R. Gaussoin, J.R. Gill, K. Gravois, M.D. Halbleib, A. Hale, W. Hanna, K. Harmoney, E.A. Heaton, R.W. Heiniger, L. Hoffman, C.O. Hong, G. Kakani, R. Kallenbach, B. Macoon, J.C. Medley, A. Missaoui, R. Mitchell, K.J. Moore, J.I. Morrison, G.N. Odvody, R. Ogoshi, J.R. Parrish, L. Quinn, E. Richard, W.L. Rooney, J.B. Rushing, R. Schnell, M. Sousek, S.A. Staggenborg, T. Tew, G. Uehara, D.R. Viands, T. Voigt, D. Williams, L. Williams, L.T. Wilson, A. Wycislo, Y. Yang, V. Owens*. 2018. Biomass Production of Herbaceous Energy Crops in the United States: Field Trial Results and Yield Potential Maps from the Multiyear Regional Feedstock Partnership. GCB Bioenergy, doi: 10.1111/gcbb.12493.

Knoll, Joseph E., William F. Anderson, Edward P. Richard Jr., Joy Doran-Peterson, Brian Baldwin, Anna L. Hale, and Ryan P. Viator. 2013. "Harvest Date Effects on Biomass Quality and Ethanol Yield of New Energycane (*Saccharum* hyb.) Genotypes in the Southeast USA." *Biomass and Bioenergy* 56: 147–56. doi:10.1016/j.biombioe.2013.04.018.

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Proceedings

Baldwin, B., W. Anderson, J. Blumenthal, E. C. Brummer, K. Gravois, A. Hale, J. R. Parish, and L. T. Wilson. 2012. "Regional Testing of Energy Cane (*Saccharum* spp.) Genotypes as a Potential Bioenergy Crop." In *Proceedings for the Sun Grant Initiative National Conference*, Vol. 1, Ch. 1.4. New Orleans, LA: Southeastern Sun Grant Center, October 2–5. http://sungrant.tennessee.edu/NR/rdonlyres/40B6A4BE-C9A0-4A32-BBD0-8D5A2CF0D436/3688/14Baldwin_Brian.pdf.

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Baldwin, B. S. 2010. "Biomass for Bioenergy: Where the Industry Appears To Be Going." Presented at Seventh Eastern Native Grass Symposium, Knoxville, TN, October 5–8.

Baldwin, B. S. 2012. "Factors Impacting Feedstock and Bio-Based Fiber Composition." Keynote Presentation. Presented at the 2012 BioEnvironmental Polymer Society Conference, Denton, TX, September 18–21.

Baldwin, B. S., W. Anderson, J. Blumenthal, E. C. Brummer, K. Gravois, A. Hale, and L. T. Wilson. 2013. "Energycane (*Saccharum* spp.) Sugarcane Goes North." Presented at 245th

National Meeting of American Chemical Society, Carbohydrates Division: Special Session on Biofuels, Bioproducts, and Biomass from Sugar Feedstocks, New Orleans, LA, April 8.

Baldwin, B. S., W. Anderson, C. Brummer, J. R. Parish, K. Gravois, L. T. Wilson, J. Blumenthal, and A. Hale. 2013. "Southeast Regional Evaluation on Energy Cane (*Saccharum* spp.) Genotypes as a Potential Bioenergy Crop." Presented at Southeastern Conference Symposium, Atlanta, GA, February 10–12. http://www.youtube.com/watch?v=nIkf8NZSzwc.

Baldwin, B. S., D. K. Lee, V. Owens, W. Rooney, and T. Voigt. 2009. "U.S. Dept. of Energy Regional Biomass Feedstocks Partnership." Presented at 21st Annual Meeting for the Association of Advancement of Industrial Crops, Termas de Chillán, Chillán, Chile, November 14–19.

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Knoll, J. E., W. F. Anderson, B. Baldwin, and E. Richard. 2010. "Harvest Date Effects on Biomass Yield and Quality of New Energycane (*Saccharum* hybrid) Genotypes in the Southeastern USA." Presented at American Society of Agronomy, Crop Science Society of America, Soil Science Society of America Annual Meeting, Long Beach, CA, October 31–November 4.

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Lee D.K., E. Aberle, E.K. Anderson, W. Anderson, B.S. Baldwin, D. Baltensperger, M. Barrett, J. Blumenthal, S. Bonos, J. Bouton, D.I. Bransby, C. Brummer, P.S. Burks, C. Chen, C. Daly, J. Egenolf, R.L. Farris, J.H. Fike, R. Gaussoin, J.R. Gill, K. Gravois, M.D. Halbleib, A. Hale, W. Hanna, K. Harmoney, E.A. Heaton, R.W. Heiniger, L. Hoffman, C.O. Hong, G. Kakani, R.

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Miscanthus x *giganteus*

Miscanthus Team: Thomas Voigt, University of Illinois; Adam Davis, USDA ARS; Stacy Bonos, Rutgers University; David Williams, University of Kentucky; John Fike, Virginia Tech; Roch Gaussoin, University of Nebraska.

Summary

From planting in 2008 through harvest following the 2015-growing season, *Miscanthus* x *giganteus* 'Illinois', a perennial C4 grass of Asian origins, was the focus of bioenergy-related studies in Illinois, Kentucky, Nebraska, and New Jersey, and in Virginia from 2010 through 2015 using replicated plots. Biomass productivity was measured following plateau yields starting in the third growing season following planting at three nitrogen fertility levels (0, 60, and 120 kg N ha^{-1}). Across years and N applications, biomass yields averaged 18.0, 16.1, 24.7, 17.4, and 17.3 dry Mg ha-1 in Illinois, Kentucky, Nebraska, New Jersey, and Virginia, respectively, over 28 harvests. While not universal, yields were variously affected by N fertility levels, winter weather, spring freezes, drought, disease, and soil conditions. This study also evaluated several environmental factors at the same study sites and found that the nitrogen applications, especially at the 120 kg N ha^{-1}, can result in nitrous oxide emissions and nitrate leaching that are greater than the 0 kg N ha^{-1} levels. Overall, yields were impressive when fertilized at 0 and 60 kg N ha^{-1}, and it can be concluded that this grass is an attractive bioenergy feedstock.

Introduction

Miscanthus x *giganteus* Greef & Deuter ex Hodkinson & Renvoize is a large-statured (up to 4 m in height), warm-season, perennial grass (family – Poaceae). This sterile triploid hybrid was originally discovered in Japan in 1935, following a spontaneous mating between fertile diploid *M. sinensis* and tetraploid *M. sacchariflorus* (Hodkinson and Renvoize 2001). The hybrid was later introduced and spread throughout Europe and the United States initially as an ornamental landscape plant and later as a bioenergy crop. Miscanthus 'Illinois', the clone chosen for this study, is referred to as 'Illinois' because UIUC has conducted significant research using this clone. It was originally obtained in 1988 from established plants at the Chicago Botanic Garden (Glencoe, IL), vegetatively propagated, and planted in demonstration plantings at UIUC (Maughan et al. 2012). In this study, annual average miscanthus yields beginning in the third growing season and later have ranged from less than 10 dry Mg ha^{-1} in VA in 2011 to more than 30 dry Mg ha^{-1} in NE, also in 2011.

Uncommon among warm-season species, miscanthus has the ability to photosynthesize at cool temperatures and typically emerges in April taking advantage of snowmelt and spring rainfall in Central Illinois (approximately 40° N latitude) (Pyter et al., 2009). Most years, it reaches two meters by early June, grows vegetatively through summer, and reaches peak biomass production in September. Miscanthus reaches 3-to-4 meters, usually flowers in late September or early October, and begins to senesce and drop foliage at the onset of freezing temperatures (Pyter et al., 2009). Harvest takes place from mid-December through late March when the stems are completely senesced and the ground is frozen and without snow cover (Pyter et al., 2009).

Environmentally, miscanthus requires ample precipitation and soil moisture to be productive; Richter et al. (2008) wrote that the most limiting factor for miscanthus growth may be water availability. It has been reported that, based on European field studies, between 80 and

300 liters of water was needed to produce one kg of dry miscanthus biomass (Beal et al., 1999; Dressler, 1993). Maughan et al. (2012) reported that below-average precipitation, sandy soils, and a shallow root zone resulted in less miscanthus biomass production in New Jersey in the third growing season than in the second growing season. Once established, drought can negatively impact biomass production, but the grass is normally able to survive dry periods and regrow acceptably the following season as occurred in Illinois in 2013 following the 2012 drought (author observations). It is recommended that without supplemental irrigation, ‘Illinois’ miscanthus be planted in areas that receive at least 75 cm of precipitation annually.

Miscanthus tolerates a wide range of soil types, organic to sandy, with pH levels that range from 5.5 – 7.5 (Caslin et al., 2010). Williams and Douglas (2011) wrote that for best production with the fewest inputs, plant miscanthus on USDA NRCS capability class I and II soils. Pyter et al. (2009) reported that it commonly takes at least three years to reach full establishment and plateau biomass production, but full establishment can be delayed when planted on infertile sites.

Established miscanthus is quite cold tolerant for a C4 species and have survived, temperatures lower than -20° C in Central Illinois (Pyter et al., 2009). First-year plants, however, were less tolerant of low winter temperatures in 2009 in Illinois (Maughan et al., 2012, Anderson et al., 2011), and it has been reported that soil temperatures of -3.4° C at the 5-cm level can kill 50% of newly planted rhizomes (Clifton-Brown and Lewandowski, 2000). Optimal miscanthus photosynthesis occurs at temperatures above 12° C (Long, 1983), and thus, spring growth commences earlier than many other warm-season grasses. In addition, miscanthus also continues growth longer than many other C4 grasses into the late summer and early autumn (Dohleman and Long, 2009). The early start and late finish contribute to its annual productivity.

Miscanthus has been highly productive in these trials, with mean plateau yields (third growing season and beyond; 2010 through 2015 for IL, KY, NE, and NJ; 2012 through 2015 for VA) ranging from 5.8 Mg ha^{-1} from the unfertilized IL plots in 2012, a growing season of extreme drought, to a high of 34.1 Mg ha^{-1} from the plots receiving 120 kg N ha^{-1} in NE in 2011. Over the entire study, the mean annual biomass production from plateau yield growing seasons was 18.9 Mg ha^{-1} with the unfertilized plots averaging 16.7 Mg ha^{-1}, plots receiving 60 kg N ha^{-1} averaging 19.4 Mg ha^{-1}, and the plots receiving 120 kg N ha^{-1} averaging 20.0 Mg ha^{-1}.

Objectives

The objective of this project was to establish and manage replicated field trials of miscanthus to gather biomass production, yield response to fertility treatments, and sustainability data across five locations in the Eastern and Midwestern U.S. to assess its potential as a bioenergy feedstock by collecting long-termed survivability and biomass productivity data and also measuring nitrate and ammonium leaching and nitrous oxide and carbon dioxide gas emissions from the IL plots and nitrate and ammonium leaching from the KY, NE, NJ, and VA sites.

Because available vegetative miscanthus propagation material was limited at the beginning of this study in 2008, the five field trials were limited to 12, 10 m x 10 m plots per location. Miscanthus plants were propagated at UIUC, and 25 cm potted plants were distributed to each site for planting on one meter spacing; thus each 10 m x 10 m plot was planted with 100 plants.

Methods

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To evaluate miscanthus, collaborator sites through the central portion of the eastern half of the U.S. were selected. The five locations spanned 22° of longitude and 4° of latitude, representing a wide variety of environmental conditions (Table 1 and Figure 1) with locations chosen based on assumed good growing conditions for this species. At the initiation of the project in 2008, the five participating collaborators and research sites were the University of Nebraska, Mead, Nebraska; the University of Illinois, Urbana, Illinois; Purdue University, West Lafayette, Indiana; the University of Kentucky, Lexington, Kentucky; and Rutgers University, Adelphi, New Jersey. However, due to great miscanthus mortality and collaborator turnover, Purdue University dropped out of the study in 2009 and was replaced in spring 2010 by a Virginia Tech University collaborator using a study site near Gretna, Virginia.

Figure 1. Map showing Sun Grant Regional Feedstock Partnership *Miscanthus* x *giganteus* evaluation sites.

The soil at the Illinois site is classified as a very deep, moderately well-drained Dana silt loam (fine-silty, mixed, superactive, mesic Oxyaquic Argiudolls) and a very deep, poorly drained Drummer silty clay loam (fine-silty, mixed, superactive, mesic Typic Endoaquolls); the upper 30 cm of soil is dominated by a sandy loam. At Kentucky, the soil is classified as very deep, well-drained Maury silt loam (fine, mixed, active, mesic Typic Paleudalfs) and a Bluegrass silt loam (fine-silty, mixed, active, mesic Typic Paleudalfs). At Nebraska, the soil is classified as a very deep well-drained Tomek silt loam (fine, smectitic, mesic Pachic Argiudolls); however, this specific site is dominated by a silty clay loam soil texture. At New Jersey, the soil is a Holmdel sandy loam (fine-loamy, mixed, active, mesic Aquic Hapludults) with a restrictive soil layer or a bedrock layer between 50 cm and 80 cm in depth, depending on the plot. At Virginia, the soil is a very deep, well-drained Cecil sandy loam (fine, kaolinitic, thermic Typic Kanhapludults). Being predominantly sandy, the Illinois and New Jersey soils were less fertile and held less water than the soils at the other sites.

Once participants were identified, miscanthus plants were propagated at UIUC for distribution and planting at each site following a coordinated protocol. *Miscanthus* x *giganteus* is a sterile triploid (3n) hybrid that resulted from a spontaneous mating between fertile diploid (2n) *M. sinensis* and tetraploid (4n) *M. sacchariflorus* (Hodkinson and Renvoize 2001). Because it is sterile, this study used miscanthus plants that were clonally propagated at UIUC using rhizome fragments. The resulting plants were shipped to collaborators for planting. Although several related *Miscanthus* varieties have now been developed and tested (e.g., ‘Amuri,’ ‘Freedom,’ and ‘Nagara’), the ‘Illinois’ clone was chosen because of direct experience with it (it has been grown at the UIUC for almost 30 years) and because it is the most commonly available type being grown across the country and world. This clone continues to show promise as a feedstock, but its cold tolerance and response to nitrogen fertilizer are variable (Maughan et al. 2012). In addition, it is difficult to imagine a successful bioenergy crop emerging from a single genotype. Collaborators were responsible for planting, managing, applying the research treatments, and collecting the data for the miscanthus field trial at their site. At each location, miscanthus was grown using a randomized, complete block design with three nitrogen fertility treatments (0, 60, and 120 kg ha^{-1}) replicated four times in 12, 10 × 10 m^2 test plots.

Site location and soils and annual precipitation and temperature information is presented in Table 1. Annual temperature and precipitation information for each location is in Tables 2 and 3 and Figures 2 and 3. All data were subjected to ANOVA and means separated using the Student’s t-test at p=0.05. Finally, the field trial data from each site were collected and assembled from all participants (Tables 7 – 11).

Table 1. Collaborator, location, date of trial initiation, and environmental conditions at the five research sites testing ‘Illinois’ *Miscanthus* x *giganteus* in the 2008-2015 North Central Sun Grant Regional Feedstock Partnership.

Collaborator/ Location/Date of Trial Initiation	Latitude (N)	Longitude (W)	Soil	Mean Annual Temp (°C)[§]	Mean Annual Precip. (cm)[§]
U. of NE/Mead, NE/2008	41.17	-96.46	Tomek silt loam	9.9	74.4
U. of IL/Urbana, IL/2008	40.06	-88.19	30-cm cap of sandy loam sediments over Dana silt loam and Drummer silty clay loam	10.8	100.9
U. of KY/Lexington, KY/2008	38.12	-84.50	Maury and Bluegrass silt loams	13.1	114.7
VA Tech/Gretna, VA/2010	36.93	-79.39	Cecil sandy loam	14.5	114.1
Rutgers U./Adelphia, NJ/2008	40.22	-74.24	Holmdel sandy loam	12	119

[§] 30-year (1981-2010) average weather data collected from National Oceanic and Atmospheric Administration sites closest to trials atUniversity of Illinois Willard Airport, Champaign, IL; Lexington Bluegrass Airport, Lexington, KY; Mead, NE.; Freehold Marlboro, NJ; and Danville Regional Airport, Danville, VA.

Table 2. 2010-2015 annual temperature (°C) and change from the 30-year (1981-2010) average at five Miscanthus field trials.						
	2010	2011	2012	2013	2014	2015
30-Year Average Temperature (°C)	Change From 30-Year Average Temperature (°C)					
Champaign, IL						
10.8	+0.4	+0.8	+2.2	-0.1	-1.1	+0.6
Lexington, KY						
13.1	0.0	+0.4	-1.1	+0.1	-0.5	+0.4
Mead, NE						
9.9	-0.3	-0.2	+2.0	-0.7	-0.7	+0.7
Freehold Marlboro, NJ						
12	+0.8	+0.9	-1.4	0.0	+0.5	+0.1
Danville, VA						
14.5			+1.0	0.0	-0.2	+1.1

Table 3. 2010-2015 annual precipitation (%) deviations from the 30-year (1981-2010) average at five Miscanthus field trials.						
	2010	2011	2012	2013	2014	2015
30-Year Average Precipitation (cm)	Percent of 30-Year Average Precipitation					
Champaign, IL						
100.9	86.1	86.6	73.7	83.7	100.2	110.3
Lexington, KY						
114.7	84.3	147.0	94.5	131.8	120.1	132.6
Mead, NE						
74.7	128.6	107.1	62.9	96.6	117.3	143.8
Freehold Marlboro, NJ						
119	93.6	129.4	80.5	88.0	112.2	89.2
Danville, VA						
114.1			64.8	115.2	96.1	115.9

Environmental sustainability

Two published sustainability trials were conducted from 2009 through 2013 in IL, and 2011 through 2013 at the other four sites. In 2008 and 2012, soils were sampled to measure changes in soil organic matter. Inorganic N leaching was measured using resin lysimeters buried at 50 cm soil depth. The lysimeters were replaced and analyzed annually each spring in IL from 2009-2013 and from 2011 through 2013 at the other four sites. Nitrous oxide (N_2O) gas samples in IL were collected year round from 2009-2013. Nitrous oxide sampling was conducted from 10:30 AM to 12:30 AM when soil temperatures are typically near average for the day and more frequently in spring and summer months when fluxes were greater with increased variation. Nitrous oxide gas samples were analyzed in a gas chromatograph, and carbon dioxide was measured using a LI-COR® LI-8100 Automated Soil CO_2 Flux System. Carbon dioxide measurements were taken during the same sampling period as N_2O. Significant difference was calculated for cumulated daily fluxes using pairwise t-tests through Fisher's least significant difference procedure. Significance between treatments was measured at $\alpha = 0.05$.

Results and Outcomes

Productivity

This grass has been studied for its bioenergy potential in European trials since 1983 (Lewandowski et al. 2000) and in U.S. trials since the early 2000s (Heaton et al. 2004). Early results promised high biomass yields of as much as 39.9 Mg ha^{-1} in some European locations (Miguez et al. 2008), with mean yields of 22 Mg ha^{-1} across European test sites (Heaton et al. 2004). Yields in the U.S. have ranged from 4.5 to 35 Mg ha^{-1} (Lee et al. 2014; Heaton et al. 2008), and have been extrapolated to as high as 63 Mg ha^{-1} from small-scale test plots (Smith et al. 2015). It is unknown whether field-scale plantings could reach these yields in the U.S., particularly across varied environmental conditions, but the potential for high yields exists, at least in some locations. Additional data are needed to compare years, regions/environments, and agronomic practices for miscanthus plantings in the U.S..

This research showed that miscanthus can achieve high productivity in five distinctly different environments. The overall average annual yield over 28 growing seasons - six growing seasons (2010-2015) in IL, KY, NE, and NJ, and four growing seasons in VA (2012-2015) - over the three nitrogen fertility regimes was 18.8 dry Mg ha^{-1}. Unfertilized plots across all locations and years produced an average of 16.9 dry Mg ha^{-1}, and the application of 60 kg N ha^{-1} increased mean yields across all locations and years to 19.4 Mg ha^{-1} (Table 4).

Table 4. 'Illinois' *Miscanthus* x *giganteus* productivity at three N fertility levels starting in the third growing season averaged across five eastern U.S. sites (2010-2015).*

Fertilizer Rate (kg N ha^{-1})	Average Yield (Mg ha^{-1})
0	16.9
60	19.4
120	20.1
$LSD_{(0.05)}$	NS

*Because of joining the study late, the third growing season for the Gretna, VA, site is 2012, not 2010.
NS = not significantly different at p=0.05.

Annually, across all sites and fertilizer levels, mean miscanthus biomass production in 2010, 2012, and 2014 was less than in 2011 and 2015 (Table 3). In 2010, the IL, KY, NE, and NJ sites were in the third year of production and had likely not reached full plateau productivity levels. In addition, Emerson et al. (2014) described the negative effects of the 2012 drought in much of the eastern U.S. on miscanthus productivity and quality during the 2012 growing season.

Table 5. Average annual 'Illinois' *Miscanthus* x *giganteus* productivity over three nitrogen fertility levels starting in the third growing season at five eastern U.S. sites (2010-2015).*	
Year	Average Yield (Mg ha^{-1})
2010	16.9 a
2011	22.4 b
2012	15.8 a
2013	18.9 ab
2014	17.1 a
2015	22.0 b
$LSD_{(0.05)}$	2.7

*Because of joining the study late, the third growing season for the Gretna, VA, site in 2012, not 2010. Different letters in the Average Yield column indicate statistical differences among the means at $P < 0.05$.

With data collection beginning in the third year after planting (when productivity usually reaches plateau productivity) and across all fertilizer levels, mean miscanthus biomass yields from 2010-2015 in NE were greater at 24.7 dry Mg ha^{-1} than in IL (18.0 dry Mg ha^{-1}), NJ (17.4 dry Mg ha^{-1}), VA (17.3 dry Mg ha^{-1}), and KY (16.1 dry Mg ha^{-1}) (Table 8).

There were no statistically significant biomass yield differences among the three fertilizer application levels across all sites and years of the study (Table 4). However, there were statistically significant miscanthus biomass yield differences among years and across fertilizer treatments in IL and VA (Tables 10 and 11). In both cases, the nitrogen fertilized plots were more productive than the unfertilized plots. In KY, NE, and NJ (Table 10), there were no statistically significant productivity differences among nitrogen application treatment over the years of the trial. It is not clear why nitrogen application increases productivity at some sites and not at others.

The effects of N fertilization were most notable in IL (Table 11), likely due to the sandy, infertile site and the removal of N in harvested biomass. Low yields in IL and KY in 2012 can be attributed to the effects of two events. An April freeze occurred when the miscanthus was approximately 50 cm tall, killing the aboveground shoots and forcing regrowth. In addition, a pronounced summer drought was experienced at all sites besides KY (Table 3), but yields were only significantly impacted at IL and NJ (Table 6). New Jersey's shallow, sandy soil may have contributed to its lower productivity under the 2012 drought conditions experienced there.

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Table 6. 'Illinois' Miscanthus x giganteus biomass yield (Mg ha^{-1}) starting in the third growing season at five eastern U.S. sites. Values are averages of four replicate plots for each treatment.

N Rate (kg ha^{-1})	2010	2011	2012	2013	2014	2015	Average
Champaign, IL							
0	14.9	17.3	5.8 a	15.3 a	8.5 a	16.6	13.1 a
60	16.1	23	12.2 b	27.1 b	25.9 b	19.4	20.6 b
120	16	21.5	11.9 b	28.3 b	25.2 b	20	20.5 b
$LSD_{(0.05)}$	NS	NS	2.6	5.2	5.0	NS	6.8
Lexington, KY							
0	12.1	19.3	12.7	19.2	13	18.3	15.8
60	11.9	20.8	11.9	20.3	13.5	19.2	16.2
120	15.3	17	14	20.3	12.7	18	16.2
$LSD_{(0.05)}$	NS	NS	NS	NS	NS	NS	NS
Mead, NE							
0	26.9	31.6	23.9	18.2	16.4	27.8 a	24.1
60	28.2	28	23.4	15.7	15.7	30.5 ab	23.5
120	28.1	34.1	23.8	18.6	19.7	34 b	26.4
$LSD_{(0.05)}$	NS	NS	NS	NS	NS	4.3	NS
Adelphia, NJ							
0	12.4	18.8	14	18.8	14.7 a	20	24.1
60	11.6	18.8	16.9	16	17.1 a	26.4	23.5
120	9.9	18.3	16.9	17.1	21.9 b	23.5	26.4
$LSD_{(0.05)}$	NS	NS	NS	NS	2.5	NS	NS
Gretna, VA*							
0			16.3	14.4	13.6 a	12.5 a	14.2 a
60			16.7	18.1	18.8 b	20.8 b	18.6 b
120			17.1	16.5	19.6 b	23.4 b	19.1 b
$LSD_{(0.05)}$			NS	NS	5.0	4.3	2.5

*Because of joining the study late, the third growing season for the Gretna, VA, site in 2012, not 2010.
NS = not significantly different at P = 0.05.
Different letters within a column indicate statistical differences among the means at P < 0.05.

The only pest found in these plantings was a leaf blight (*Pithomyces chartarum*) in KY (Ahonsi et al., 2010) that began soon after planting and reoccurred in additional years. This leaf blight may have limited productivity, but the KY plantings still averaged yields of more than 16 Mg ha^{-1}. In NE, the high biomass production throughout the study eventually led to a nitrogen yield response in 2015 (Table 11). This study has determined that miscanthus can be productive over a variety of much of the east-central U.S. locations and environments.

PRISM-ELM maps were created by using a four-year average yield for the years 2010-2013 and regressed against the actual yield values (Figure 2; Lee et al 2018). Although yield data were collected beyond 2012, meetings with the PRISM-ELM personnel and initial map development occurred before this time. Our field data is in good agreement with the model results, although we did measure higher yields than the model predicted in some years and locations (e.g., 2012 NE and 2014 NJ). However, it is important to note that the PRISM-ELM models are based on 30 years of climate data through 2010, and that any spikes in particular years during that time period will be smoothed out due to averaging, which would tend to moderate maximum and minimum values generated by the model.

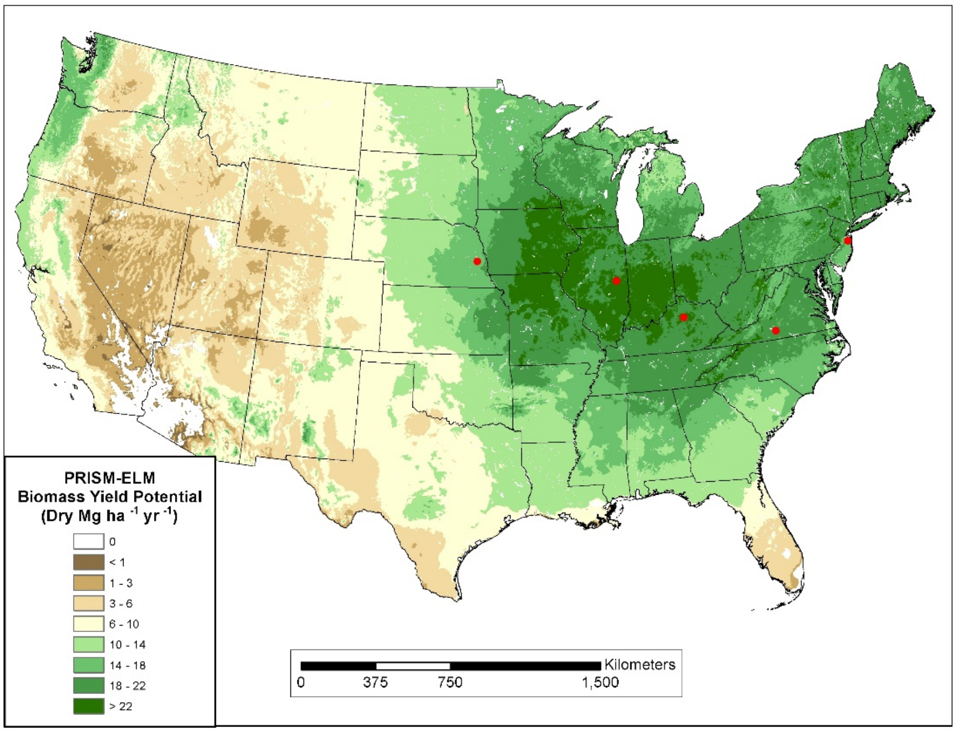

Figure 2. Biomass yield potential of Miscanthus for the U.S. generated using the PRISM-ELM model and based in part on Regional Feedstock Partnership Field Trials (red dots) (from Lee et al., 2018).

Sustainability

Two sustainability studies were conducted and published using Partnership sites. In the first, Behnke et al. (2012) evaluated the effects of the three urea nitrogen fertilizer rates (0, 60, and 120 kg N ha^{-1} $year^{-1}$) applied to 1- and 2-year old (2009 and 2010)miscanthus crops on nitrous oxide (N_20) and carbon dioxide (CO_2) emissions, nitrogen leaching, and biomass yields. We found no statistically significant yield response to nitrogen fertilizer at any site in either 2009 or 2010 (Table 7); however, the amount of nitrogen in the harvested biomass in 2010 was significantly greater for both fertilizer treatments than for the unfertilized treatment (Table 7). Behnke et al. (2012) found no carbon dioxide emission differences based on N fertilization levels in 2009 or 2010 (Table 8; however, Davis et al. (2014) did see differences among treatments in 2011 and 2013. Nitrous oxide emissions were affected by N-fertilization rate in all years, with N_2O emissions increasing as fertilizer N rates increased (Table 8). There were also no inorganic nitrogen (nitrate and ammonium) leaching differences among nitrogen fertilizer treatments at a

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depth of 50 cm in 2009, but in 2010, however, there was more nitrate (NO_3) leached from the 120 kg N ha^{-1} treatments than from the 0 N kg ha^{-1} treatments (Table 9).

Table 7. Miscanthus yield in 2009 and 2010 and biomass N in 2010 by N treatment in Illinois (with standard deviations) (from Behnke et al., 2012)

Year	N Rate (kg N ha^{-1})	Yield (Mg ha^{-1})	Biomass N (kg N ha^{-1})
2009	0	1.1 (0.7)	
	60	4.1 (3.7)	
	120	4.0 (2.2)	
2010	0	14.9 (2.9)	44.9 (9.0)
	60	15.8 (1.8)	53.5 (5.3)
	120	17.0 (1.4)	66.6 (1.3)

Table 8. Annual cumulative carbon dioxide (CO_2) and nitrous oxide (N_2O) fluxes from 2009 to 2013 in Illinois (adapted from Davis et al., 2015).

		Nitrogen Application Treatment (kg N ha^{-1})		
Greenhouse Gas	Year	0	60	120
CO_2 (Mg-C ha^{-1})†	2009	6.3	6.6	6.7
	2010	7.9	7.7	7.0
	2011	5.0 b	4.5 ab	4.1 a
	2012	5.7	5.0	5.2
	2013	7.2 b	6.1 a	5.8 a
N_2O (kg N ha^{-1})†	2009	0.54 a	0.87 ab	0.92 b
	2010	0.33 a	0.76 a	13.6 b
	2011	0.10 a	0.47 b	1.08 c
	2012	0.39 a	3.67 b	4.32 b
	2013	1.19 a	1.64 ab	2.39 b

† Cumulative fluxes were calculated from measured fluxes corrected for temperature variations using a Q_{10} = 2.
* Differences are only within the year and measurements values with same letter are not significantly different at the 0.05 level.

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Table 9. Trimmed mean annual leaching of nitrate, ammonium, and total inorganic nitrogen observed from the ion exchange resin lysimeters at 50 cm soil depth under the *Miscanthus* (Behnke et al., 2012).

Year	Treatment (kg N ha^{-1})	Leached NO_3^- (kg N ha^{-1} yr^{-1})	Leached NH_4^+ (kg N ha^{-1} yr^{-1})	Leached total Inorganic (kg N ha^{-1} yr^{-1})
2009	0	6.4 (3.2) a‡	6.8 (0.8)	13.3 (2.8) a
	60	7.1 (6.7) a	5.6 (0.5)	12.6 (7.1 a
	120	13.3 (7.0) a	7.1 (1.3)	20.5 (8.0) a
2010	0	8.9 (5.9) a	2.3 (2.1)	9.1 (7.3) a
	60	15.3 (7.1) ab	3.0 (1.9)	18.3 (7.9) ab
	120	28.9 (6.1) b	6.1 (4.3)	34.9 (8.5) b

‡ Values in parentheses are standard deviations.
***** values with same letter are not significantly different at the 0.05 level

In Davis et al., (2015), miscanthus sustainability trials were continued by measuring the N fertilization effects on biomass production, soil organic matter, and inorganic nitrogen leaching in Illinois, Kentucky, Nebraska, New Jersey, and Virginia. In addition, they continued to measure nitrous oxide and carbon dioxide emissions at the Illinois site. There were yield responses to N fertilization in Illinois (2012, 2013, and 2014), Nebraska (2015), New Jersey (2014) and Virginia (2014 and 2015) while miscanthus at the Kentucky trial had showed no response to nitrogen fertilizer in any year (Table 6). Potentially mineralizable nitrogen (POM) in the upper 10 cm of soil (the soil surface layer) increased across all fertilizer treatments and sites, indicating that the soil organic matter composition was altered after just four years of miscanthus production (Figure 3). On the other hand, this effect was not observed at any site at the 10-30 cm soil depth (Figure 3).

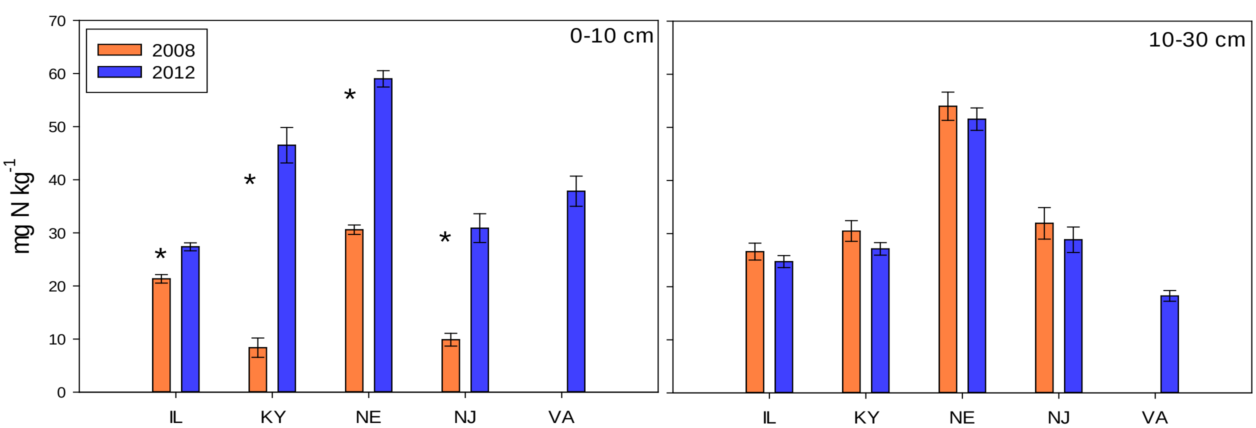

Figure 3. Potential mineralizable N at soil depths of 0-10 cm and 10-30 cm in 2008 and 2012 at five miscanthus locations. Data are averaged across N rate at each location. * indicates statistical differences within a location between 2008 and 2012 at P = 0.05. Bars represent standard errors (from Davis et al., 2015).

Even though biomass yields were not universally improved by fertilization, we found that applying N did increase both leaching and N_2O emissions at all sites and all years. Inorganic N leaching (at 50 cm soil depth) was measured at the Illinois site beginning in 2009, and at the other sites starting in 2012. The fertilized plots leached more nitrate compared to the unfertilized plots every year after 2009 in Illinois and across all sites in 2012 (Table 9, Figures 4 and 5). The 120 kg N ha^{-1} plots leached the most annual nitrate every year and across all sites during the study period (ranging from 10 to 39 kg N ha^{-1} yr^{-1}). Leaching from the 0 kg N ha^{-1} plots was less than 10 kg N ha^{-1} yr^{-1}, the primary source of which is likely to be the decomposition of soil organic matter. The unfertilized plots had decreases nitrate leaching as the miscanthus established at the Illinois site (Table 9, Figure 4).

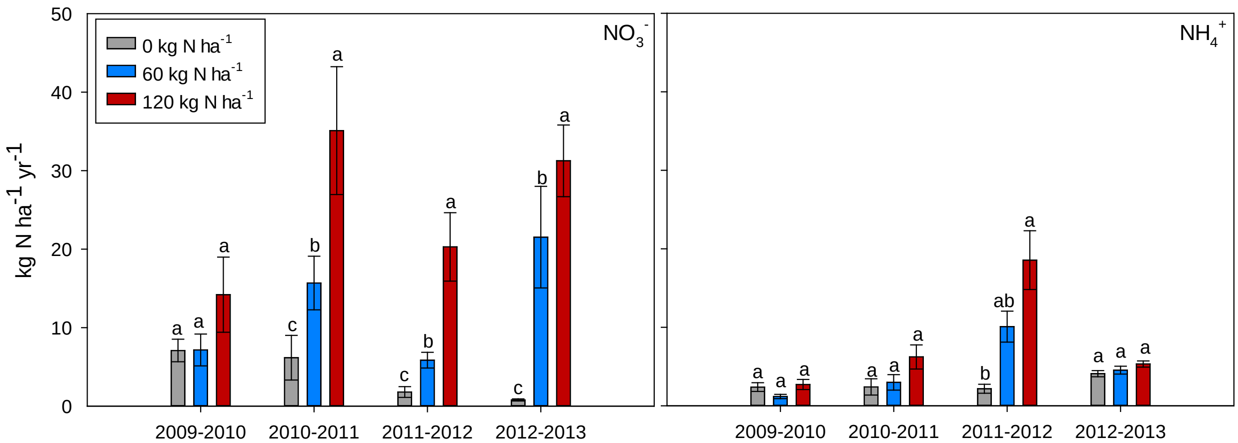

Figure 4. Inorganic N leached from the Illinois site from 2009-2013, as both nitrate and ammonium. Leachate was collected at a depth of 50 cm. (Davis et al., 2015). Different letters above the bars within a time period indicate statistical differences among N rates at P = 0.05. Bars represent standard errors (from Davis et al., 2015).

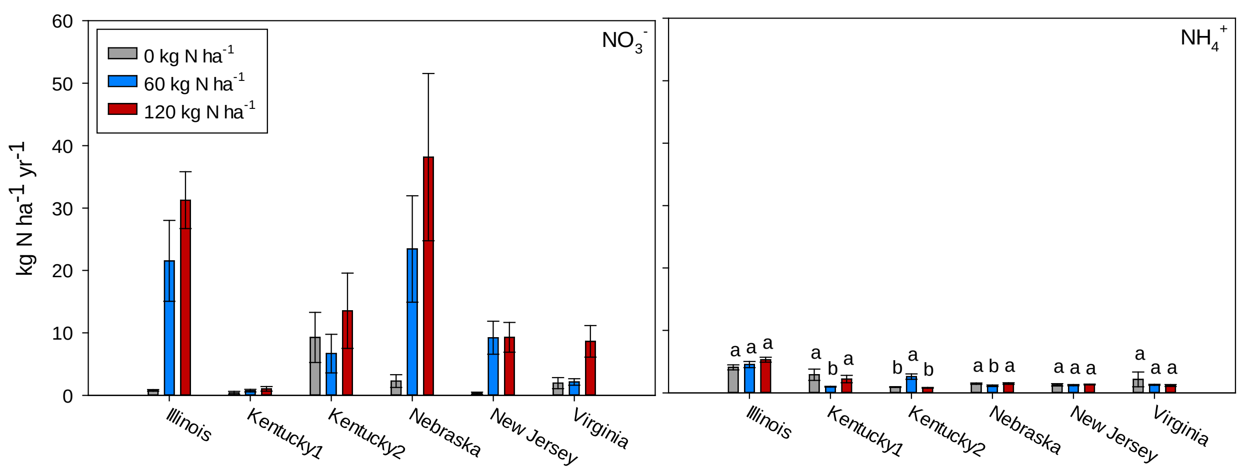

Figure 5. Nitrate and ammonium leaching from miscanthus produced in IL, KY, NE, NJ, and VA at 50 cm for three N application rates from April 2012–2013. Kentucky1 are the two southern plots, Kentucky2 the northern plots. Nitrate and ammonium bars designated with the same letter within a growing site are not significantly different (P = 0.05). Bars represent standard errors (from Davis et al., 2015).

Nitrous oxide emissions were measured from 2009-2013 at the Illinois site only and were found to be greater on fertilized plots. The 60 and 120 kg N ha^{-1} plots ranged in annual N_2O emissions from 0.5 to 4.3 kg N ha^{-1} yr^{-1}, and the 0 kg N ha^{-1} plots ranged from 0.1 to 0.5 kg N ha^{-1} yr^{-1} (Table 12).

Carbon dioxide emissions were not found to be affected by fertilizer treatments (Table 12). In summary, N fertilization of miscanthus led to N_2O releases, increased fluxes of inorganic N (primarily NO_3^-) through the soil profile, and increased harvested N in biomass without a significant increase in biomass production (Behnke et al., 2012; Davis et al., 2015). From these observations, we conclude that N fertilization is generally not necessary to achieve high yields of miscanthus during the first several years of production, which helps to make the crop more economically viable, while also avoiding increased GHG emissions (i.e., N_2O) and nitrate leaching into ground and surface waters. As with other feedstocks however, N application rate should be determined on a case-by-case basis.

There are several barriers to the successful adoption of miscanthus as a bioenergy crop. First, the lack of a market for miscanthus biomass limits its production. At present, the grass is best used for combustion to produce heat and electricity and as a source of cellulosic ethanol in small-scale applications. Because there are few commercial furnaces or boilers in the U.S. designed and available to successfully burn miscanthus for long time periods on a large scale, and because there are no commercial-scale cellulosic ethanol plants using miscanthus, there is no market for the crop. Producers will not invest in large-scale production without long-term markets for the biomass, and potential biomass users will not invest in facilities that use miscanthus until adequate biomass supplies are available. Additional markets and uses for miscanthus need to be developed to enable producers to have a suitable income stream while the bioenergy market develops. At present, miscanthus has been used on a limited basis for combustion, animal bedding, and landscape mulch.

Second, because ‘Illinois’ miscanthus is sterile, vegetative planting costs are high, and plateau production usually occurs in growing season three and beyond, farmers (and probably agricultural bankers) are reluctant to take on the high upfront costs of planting and establishment. These costs can be amortized over a productive lifespan that appears to be much longer than 10 years – a 1988 UIUC landscape demonstration planting is still very productive. Without favorable and sustained long-term markets, growers will not take on the up-front establishment costs. In addition, farmers are not able to easily switch from miscanthus to another crop to take advantage of shifts in the marketplace because of the time required to reach plateau production and recoup up-front planting and establishment costs. Identifying fast developing, seeded miscanthus varieties can potentially reduce up-front costs and make producing this crop more attractive. Third, the majority of miscanthus planted in the U.S. has been the sterile ‘Illinois’ and ‘Freedom’ clones. These two types provide limited genetic diversity. Additional germplasm with diverse genetics needs to be developed to expand the geographic range for successfully growing miscanthus and resist potential disease and insect pest invasion.

Finally, while the Feedstock Partnership provided a great opportunity to study miscanthus in five locations over eight growing seasons, additional long-term studies in additional geographic regions is necessary to hone in on the region of best miscanthus and *Miscanthus* spp. production. It is especially important to learn its tolerances to suboptimal or marginal soils, and how to successfully manage the crop in those locations.

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Where do we go from here?

There is additional research needed in order for miscanthus to become a commercial feedstock. First, based on biomass productivity, grasses in the genus *Miscanthus* are essentially unimproved; the most widely planted bioenergy type, *Miscanthus* x *giganteus* 'Illinois', resulted from a naturally occurring cross in Japan. Recognizing the production potential of sterile and fertile *Miscanthus* spp. germplasm, is necessary to develop new types that can be successfully produced over a broader geographic area and have identified pest resistance. A goal of *Miscanthus* spp. breeding programs should be to develop types with greater tolerance to cold, heat, and drought. This requires creation of additional crosses of *M. sinensis* and *M. sacchariflorus*, as well as biofuel-oriented *M. sinensis* and *M. sacchariflorus* types. Because invasiveness is an issue, new types of *Miscanthus* spp. germplasm should be sterile or late flowering for cold regions.

Second, few miscanthus studies have been conducted in marginal or sub-optimal soils. Thus, there is a need to determine if the grass will grow in infertile soils, as well as soils that are wet, dry, cold, hot, erodible, and saline, in order to avoid competition with food, feed and fiber crops in high-quality soils. Additionally, new varieties of *miscanthus* and agronomic practices will need to undergo long-term testing to optimize productivity in these settings.

Finally, as we have learned that 'Illinois' miscanthus is likely to require nitrogen application as stands age, so that productivity can be optimized. Long-term studies that fine-tune N applications based on location, as well as evaluate nitrogen replacement fertilization levels are needed. Additional fertility studies that identify the productivity effects of other nutrients are also needed.

Other *Miscanthus* spp. have been considered for bioenergy deployment. Along with *M.* x *giganteus* 'Illinois', 'Freedom'™ miscanthus is a sterile hybrid released by Dr. Brian Baldwin of Mississippi State University (Heaton et al. 2010). With *M. sinensis* and *M. sacchariflorus* as parents, 'Freedom'™ is very similar to *M.* x *g.* 'Illinois', but was selected for the Southeastern U.S. (Anonymous, 2012). New Energy Farms of Leamington Ontario, Canada markets 'Nagara' miscanthus, another sterile type with N with *M. sinensis* and *M. sacchariflorus* as parents. 'Nagara' has been advertised to be extremely cold tolerant (*http://www.newenergyfarms.com/crops/miscanthus/*). University of Illinois Urbana-Champaign perennial grass breeder, Dr. Erik Sacks, is working to advance the cold hardiness of individual species and crosses of *M. sinensis* and *M. sacchariflorus*; there are no reports, however, of commercially available bioenergy types at this writing (Author observation).

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Switchgrass

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Summary

Switchgrass has been the primary herbaceous species studied for bioenergy production in the USA. The plant has been a model crop because of its potential high productivity with limited inputs, adaptability to marginal sites, and potential conservation services provisioning. Despite numerous and widespread studies, switchgrass production has received little exploration in field settings using the complement of typical establishment and harvest systems. This study is unique in providing long-term production data (typically six or seven years' worth) at diverse sites around North America under field settings. The data suggest that when grown and harvested for biomass on marginal lands, switchgrass yields will be less than typically reported in small plot studies in the regions of interest. Response to N generally was limited under the end-of-season harvest management scenario, but in some cases, the benefits of N application began to be evident over time.

Ultimately, the value of a ton of switchgrass will remain the key driver for feasibility for marginal land use and fertilization inputs. Thus, while the general recommendation has been to fertilize the crop to meet replacement needs, this research suggests that generalized N fertilizer recommendations will not be sufficient to provide optimum fertility management across multiple agro-ecoregions. The Feedstock Partnership generated a number of important pieces of information, including the switchgrass database in the Knowledge Discovery Framework (KDF; website in preparation) and PRISM models of switchgrass production. A number of publications and presentations on, or related to, this work have also entered the public sphere as a result of partnership efforts.

Feedstock system development in the future will benefit from new management strategies that incorporate environmental conservation. Perhaps more important will be the direct comparison of the various energy crops suited to a region. For example, both miscanthus and switchgrass are suited to many of the sites used in the feedstock partnership, but there has been little effort directed at assessment of their relative productivity, profitability or conservation benefits in side-by-side trials. At present (and aside from stover and other crop residues), switchgrass remains the primary herbaceous biomass feedstock of interest across much of the United States and newer more productive varieties offer promise of greater productivity. Future work assessing switchgrass varieties and crop management practices will be important for optimizing opportunities for the developing bioenergy industry.

Introduction

Switchgrass (*Panicum virgatum* L.) has been the principal perennial herbaceous crop investigated for bioenergy production in North America (McLaughlin and Kszos, 2005; Parrish and Fike, 2005). The outpouring of interest and research effort on this North American native species arose from its high productivity, broad adaptability, and suitability to marginal sites. Low nutrient input requirements further add to the attractiveness of switchgrass for limited-input bioenergy systems (Wright and Turhollow, 2010). Potential to grow switchgrass and other "second generation" perennial bioenergy crops on marginal land has been a particular point in their favor, as many consider these crops a way to avoid competition for arable lands which could be used to grow food, feed and fiber crops (Gopalakrishnan et al., 2011; Hill et al., 2006).

Switchgrass is a perennial native of North America with an expansive range (Hitchcock, 1971). The species is generally divided into two ecotypes (lowland and upland), typically of southern and northern adaptation, respectively. Lowland ecotypes are larger, more robust plants with bunchgrass characteristics and which often reach heights > 3m. These ecotypes generally are more productive on deeper soils, in wetter conditions, and at lower latitudes (Brunken and Estes, 1974; Casler et al., 2004; Porter, 1966; Sanderson et al., 1996). Upland ecotypes generally are finer-stemmed and shorter, with thicker roots and longer root internodes. These root morphological traits leave upland ecotypes appearing more as a sod-forming grass. The upland ecotypes generally are better suited to higher elevation, drier land forms, and higher latitudes. Because of their greater productive potential, lowland ecotypes are of interest where they are adapted for bioenergy production. However, upland ecotypes may be better suited for much of the available production area in North America, which is typified by cooler temperatures and drier conditions.

Among potential cellulosic energy crops, switchgrass may be one of the easiest species to utilize for biomass-to-bioenergy systems given its high production potential with modest agricultural inputs. Economic assessments at large scale have been limited, however. Results from one such study in the Great Plains indicated that $66/Mg would be a reasonable breakeven price for the farm side of a switchgrass-based energy production system (Perrin et al., 2008). This assessment was based on an annualized average production of 5 Mg/ha, and higher production levels have been anticipated in the Southeast where more productive lowland switchgrass would be the ecotype of choice. External factors such as fossil energy prices and the competitive value of other crops will determine whether there are sufficient available acres and profit potential to propel a switchgrass-based biomass industry.

Objectives

The primary objectives of this study were to determine switchgrass response to N and corresponding estimates of production costs in field-scale studies located on marginal sites with diverse soil and climatic conditions. Environmental impacts (e.g., management effects on soil carbon and N and on feedstock quality) were also measured (see Hong et al., 2014; Owens et al., 2013). Rather, we report findings on establishment, crop yield, and switchgrass production costs over 2009-2015 on six selected sites in Alabama, Iowa, New York, Oklahoma, South Dakota, and Virginia, USA.

Methods

Biomass Production

This synopsis describes findings on establishment and crop yield over 2009-2015 crop years on selected marginal production sites in Elmore County, AL; Story County, IA; Tompkins County, NY; Muskogee County, OK; Day County, SD; and Pittsylvania County, VA (Figure 1). Unless irrigation is applied, switchgrass is best adapted east of the 100th meridian where precipitation is generally greater; hence, our choice of locations east of this line. These sites are all within zones of high productivity for switchgrass and they represent a broad range of diversity in terms of climatic and edaphic conditions. Switchgrass stands were established at five of the sites in 2008 (2009 at Iowa). Land management practices and cultivar selections were based on regionally-appropriate guidelines for switchgrass production, including use of the best available adapted cultivars. Upland cultivars ‘Cave-In-Rock’ (used at sites in Iowa and New York) and ‘Sunburst’ (used in South Dakota) were planted at more northern latitudes. The lowland cultivar ‘Alamo’ was planted in Alabama and Virginia. ‘Blackwell’, a locally-adapted upland cultivar was grown in Oklahoma because Alamo seed could not be procured for that site. Soil series at sites in Alabama, Iowa, South Dakota and Virginia ranged from moderately well drained to well drained, while production fields in Oklahoma and New York generally had poorly drained soils. A detailed description of these soils can be found in Fike et al. (2017). Prior to establishment, most sites used in these studies had been in perennial sod cover or a typical crop rotation. At establishment, the previous crop typically was killed with herbicide and following seeding the fields were treated with herbicides to control annual (usually grass) weeds. Additional herbicides for vegetation control were applied as needed by site in subsequent seasons.

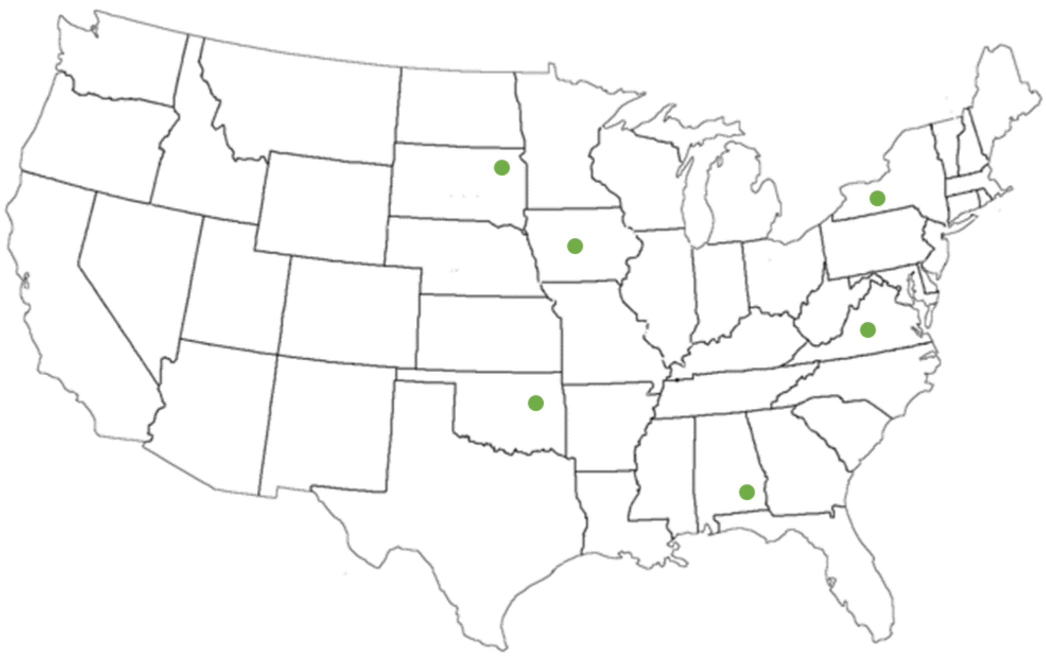


Figure 1. Map showing approximate locations of six Sun Grant Regional Feedstock Partnership switchgrass evaluation sites.

Details regarding planting time, seeding rate, land preparation, cultivar, previous crop, N application date, and herbicide rate, timing, and date can be found in Fike et al. (2017). Fields were not fertilized or harvested in the establishment year. Beginning the year after planting (i.e., 2009 at all sites except Alabama and Iowa; 2010 at Iowa and 2012 in Alabama), switchgrass plots (four replicates per site; minimum plot size = 0.39 ha) were fertilized using local farm or commercial application equipment and locally available inorganic N sources. (Production in Alabama did not begin until 2012 due to stand establishment failures in 2009 and 2010.) Nitrogen was applied as ammonium sulfate in New York and Virginia and as urea in Alabama, Iowa, Oklahoma and South Dakota at rates of 0, 56, or 112 kg N ha^{-1}. At all sites, plots received additional herbicide treatment during the first crop year (2009 at all sites except Iowa; 2010 at

Iowa and 2012 at Alabama). Herbicides also were applied in 2010 and 2011 in Oklahoma and South Dakota. A broadleaf herbicide was applied at Alabama in 2012. No site received herbicides during the 2013-2014 cropping seasons. A broadleaf herbicide was applied in 2015 at the Iowa site (Fike et al., 2017).

Plot harvest dates also varied by site. Harvests began as early as October, following the first killing frost (New York). Harvests were planned for January in Virginia but occurred as late as March in order to have sufficiently firm (i.e., dry or frozen) ground. Entire plots at all sites were harvested with conventional hay making equipment, but harvest equipment and practices varied by state (Fike et al., 2017).

Establishment-year stand percentages were determined by collecting four random measures per plot with a 0.75-m × 0.75-m metal grid following the procedure of Vogel and Masters (2001). Stand percentages for each plot were measured at the start of the 2009 growing season (Table 5). Yield data for this study cover the 2009 to 2015 crop years (Table 1). At most sites, all bales from each experimental field were weighed and the biomass yields were calculated as total bale weight per plot × percent dry matter of the plot subsample. In South Dakota, yields were determined by mowing and baling a strip through the middle of each plot (approximately 5.5 m wide and 300 m long) with standard agricultural equipment available on the farm. Bales were weighed at the field edge for yield measurements. At all sites, switchgrass subsamples (approximately 2 kg) from each plot were collected from within the row (prior to baling), or from the bale (South Dakota). Subsamples were weighed, dried at 55 °C for a minimum of 48 hr, then re-weighed for moisture determination. Based on common experience, a correction factor of 0.92 was applied to all dry matter values to adjust yields to an estimated bone dry level.

The MachData (Lazarus, 2013) model was used in order to have consistent, engineering-based estimates of per-hour and per-acre costs of labor, tractor and equipment use. This machinery cost estimator is used widely by farm management advisors and farm managers (Maung and Gustafson, 2013; Myhre, 2010; Venuto and Daniel, 2010). The results indicate a representative farm cost per activity rather than that specifically incurred in the field – or in this case, on the research plot. With this approach, machinery and equipment used in the research could be matched closely with MachData to provide an estimate of field activity costs at a commercial farm scale, emphasizing the relative agronomic impacts of N fertilization and economic contrasts between states (Fike et al., 2017).

Other costs of selected activities were estimated as the price of custom contracted activities. Nitrogen fertilizer applications were charged at custom rates, and baling was charged at a per-bale rate, both of which were set equal to the midpoint of custom rates reported in Edwards and Johanns (2012). Additional costs that must be considered are farmland cash rent, operating loan expenses, and labor cost. Farmland opportunity cost was estimated by annual own-county cash rent survey value (USDA-NASS 2013b). To reflect the marginal nature of these sites, the rental rate was estimated at the midpoint between reported county cropland and pastureland rates. Operating loan interest expense was estimated for all fertilizer and chemical purchases for a term of six months at the assumed interest rate of 6% per annum. Finally, skilled labor for machinery operation was priced at $15/hour (Fike et al., 2017).

As described in Fike et al. (2017), the economic assessment provided in this report is designed to estimate switchgrass per-ha and per-Mg production. The format of the enterprise budgets follows Mooney et al. (2009) and Miranowski et al. (2010). The principal factors affecting production cost, land cost, the cost of operating inputs, the cost of power equipment and implements, and other costs reflect management and owner investments, risk, and opportunity costs. Machinery and equipment costs reflect both operating and overhead charges. Establishment-year costs are prorated over 11 years (estimated establishment and production period until reseeding). The costs of harvest staging, storage, and transport are not considered, as the focus of the current research is only on production costs.

Sustainability

Three key parameters (nitrate [NO_3^-] leaching and soil surface CO_2 and N_2O fluxes) were selected for evaluating switchgrass sustainability at the South Dakota location only. Water samples (leachate) from the unsaturated soil were collected using porous stainless steel suction lysimeters installed at 100 cm depth at 36 positions across this study site. To collect leachate, vacuum was applied to the lysimeter through a sealed tubing system from the lysimeter to the soil surface using a hand pump. Leachate collected from the lysimeters was transferred to collection bottles following which the lysimeters were completely emptied using a pump in preparation for next sampling time. Water samples were tested for NO_3^- using a Dionex DX500 analyzer following the EPA 300.1 method.

Soil CO_2 and N_2O emissions were monitored using the vented PVC static flux chambers (25 cm diameter × 15 cm height) (Hutchinson and Mosier, 1981), which were installed and fixed in the field from 2010 to 2015 according to the guidance of Parkin and Venterea (2010). Before taking gas samples, a PVC cap with a vent tube and sampling port were placed on anchors. Gas samples inside the chambers were collected with a 20 mL syringe at 0, 20, and 40 minutes. Using the syringe, the gases were transferred into 12 mL evacuated glass vials sealed with butyl rubber septa. Concentrations of CO_2 and N_2O were measured using a Gas Chromatograph [(Shimadzu 14B with a CombiPal AOC-5000 autosampler, 2-ml injection loop, Porapack Q precolumns, a 1/8" stainless steel Porapack Q (80/100 mesh) column, a Haysep-D column (columns operated at 60 °C), and a flame ionization detector (FID), and an electron capture detector (ECD) both at 260 °C)]. Soil CO_2 and N_2O fluxes were calculated as the change in headspace gas concentration over time within the enclosed chamber volume and the average of two chambers was used to represent each plot for further analysis (Mbonimpa et al., 2015).

Results and Outcomes

Biomass Production

Description of yield and production costs largely are taken from Fike et al. (2017). Stand percentages (Table 1) were determined before the initiation of fertility treatments and ranged from 76 % (Iowa) to 28% (Virginia). Stand percentages in South Dakota and Virginia were low (<30%) compared to recommendations for successful biofuel crop establishment (such as ≥40% in Schmer et al., 2006). This likely was a factor in the relatively low yields produced during the first harvests in 2009 at all sites except New York. However, there may be some question about the effect of initial stand frequency on total productivity over time, given the limited effects of wide row spacing reported on biomass yield (Foster et al., 2012; Ma et al., 2001).

Production responses were affected by significant interactions among variables and thus were analyzed and presented by year within sites. Yield response to increasing N applications was not observed at any site during the first production season. Over all growing seasons, yield responses in Alabama, Iowa, South Dakota and Virginia were linear or quadratic or both. At most sites the quadratic response indicated more limited response to N at higher application rates. In contrast, biomass yields were largely unresponsive to N in Oklahoma and negative in New York.

Alabama: Crop yields were highest in Alabama and not responsive to N. However, there was less opportunity to gauge the value of N applications over time because of stand failures in the first two years. It is suspected that residual herbicide may have played a role in those failures. The study site was moved in 2010 and successful establishment was achieved, although the N protocol was not introduced until 2012. As a result, the effect of N application was only observed for four growing seasons (2012-2015) at this site.

Iowa: Our approach was to use regionally specific best management practices as guidelines for establishment. In Iowa, seeding switchgrass with maize both allowed the use of atrazine, an herbicide labeled for maize (as per Hintz et al., 1998), and provided for some productivity from the site during the period of establishment. Responses to N were significant in crop years 2011 through 2015, with stronger response to N the last two years. , Similar yield responses to N inputs were observed by Lemus et al. (2008a) in a field-scale study in southern Iowa. Data from small plot studies in the region have indicated yields could be much higher (12.5 Mg ha^{-1}) than these results (Heggenstaller et al., 2009), and yields likely would have been greater with use of an adapted lowland switchgrass variety (Lemus et al., 2002).

New York: Yields in New York generally were uniform across treatments, although there was about a 7% yield reduction with N application. Over all years and treatments, mean switchgrass yields were about 6.9 Mg ha^{-1}, which is similar to other data from studies in the region.

Oklahoma: Yields largely were insensitive to N treatment. After the first crop year, yield increases averaged about 1.3 Mg ha^{-1} relative to average yields from the previous cropping season. Yields in 2013, 8.74 Mg ha^{-1}, were 5.29Mg ha^{-1} (153%) greater than yields of biomass produced in 2009 (3.45 Mg ha^{-1}). Yields in this study likely would have been higher had a lowland cultivar been available based on data from other studies evaluating end-of-season harvest systems within the region.

South Dakota: Yields from South Dakota averaged 3.94 Mg ha^{-1} across years and treatments and were comparable to yields from other regional studies. Yields nearly doubled from year one to year two, and aside from the first cropping season, yields were consistently positive in response to N treatments. For most years, however, there were no differences in yield between the 56 and 112 kg ha^{-1} application rates.

Virginia: As with South Dakota, yield gains were almost 100% from 2009 to 2010. The Virginia site also was the most responsive to added N fertility. Averaged over crop years, yield increases in response to N fertilizer application rates of 56 and 112 kg ha^{-1} were 41% and 77% above the control. This response to fertility likely reflects the fact that the Virginia site had more marginal soil with lowest soil N to depth (Owens et al., 2013).

Table 1. Establishment year stand estimates and crop year yields in response to nitrogen (N) application (adapted from Fike et al., 2017).

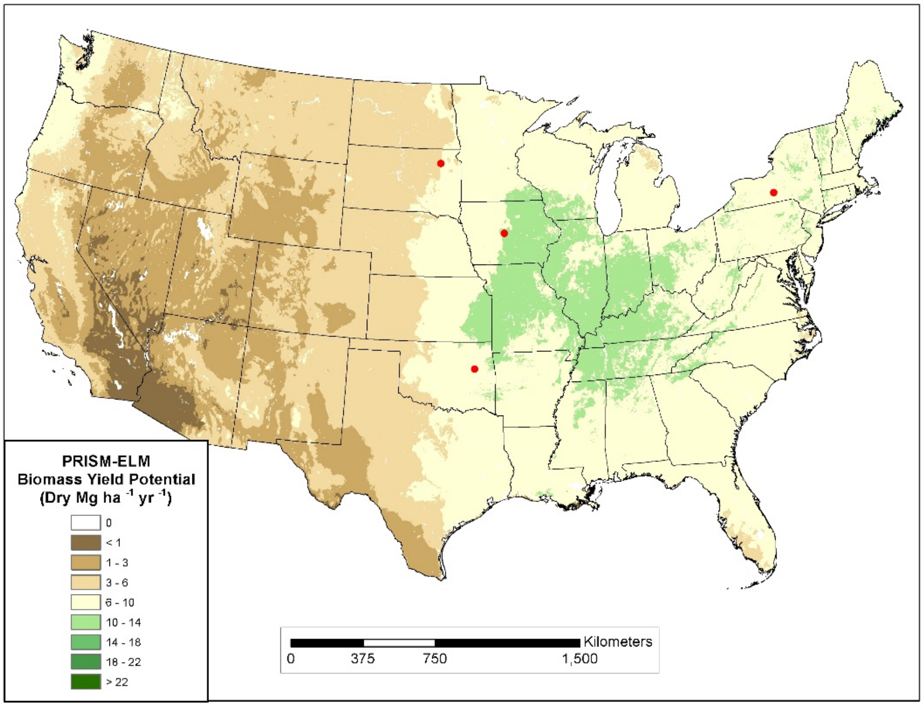
State	Stand	N	2009	2010	2011	2012	2013	2014	2015	Mean
	%	kg ha^{-1}				Mg ha^{-1}*				
Alabama	--	0	--	--	--	7.22	8.95	12.27	8.27	9.18
		56	--	--	--	7.40	9.06	11.58	8.50	9.14
		112	--	--	--	8.19	9.78	13.90	9.78	10.41
		S.E.	--	--	--	0.65	1.32	1.42	1.01	0.89
Iowa	75.9	0	--	6.96	6.41b	6.15b	6.72b	3.82c	5.95c	6.00b
		56	--	6.73	7.38a	7.99a	9.14a	5.52b	8.26b	7.50a
		112	--	7.35	7.05ab	8.25a	10.22a	7.16a	9.64a	8.28a
		S.E.	--	0.38	0.28	0.40	0.64	0.36	0.397	0.28
New York	60.1	0	6.19	6.72a	7.81a	6.66	9.42	7.04	6.93a	7.25a
		56	6.11	6.14b	6.10b	6.96	8.84	6.28	6.40b	6.69b
		112	6.4	5.71c	6.79ab	6.74	8.6	6.01	6.67ab	6.70b
		S.E.	0.35	0.13	0.46	0.13	0.68	0.411	0.141	0.24
Oklahoma	47.3	0	3.13	4.62	7.34ab	7.82	8.81	7.66a	5.86	6.46
		56	3.29	4.97	6.11b	8.71	9.11	7.36ab	7.16	6.67
		112	3.92	4.84	7.36a	7.93	8.31	6.93b	6.09	6.48
		S.E.	0.39	0.18	0.399	0.29	0.39	0.244	0.404	0.36
South Dakota	29.0	0	1.82	3.59b	3.29b	3.48b	3.04c	2.82b	2.54c	2.94b
		56	2.48	4.98a	4.42a	5.02a	4.57b	4.66a	3.71b	4.26a
		112	2.87	4.82a	4.59a	5.41a	5.40a	4.94a	4.30a	4.62a
		S.E.	0.35	0.46	0.214	0.32	0.17	0.266	0.145	0.19
Virginia	27.8	0	2.73	4.82b	4.71b	5.63b	4.23b	6.40b	5.42b	4.85c
		56	3.82	6.67ab	6.39b	8.37a	6.78ab	8.29ab	7.73a	6.86b
		112	4.14	9.05a	8.48a	10.53a	8.38a	10.23a	9.07a	8.56a
		S.E.	0.53	1.06	0.70	1.08	1.01	1.10	0.76	0.66

*Means within columns for each location with different letter designations are significantly different (*P*<0.05)

Although climate, soil drainage class, switchgrass ecotype, initial stand establishment and N source, all impacted switchgrass yields at the five sites, a couple of across-site observations can be noted (Table 1). South Dakota and Virginia, sites with good soil drainage, had the lowest initial plant stands, but yields increased over three years (a typical length of time for establishment of mature stands) and had significant yield increases with N application. The combination of good soil drainage and late planting date in Virginia likely limited seedling establishment in the planting year. Sites with poor soil drainage (Iowa, New York, and Oklahoma), had good initial plant stands, with little or no yield increase from the first to the second crop year (yields were not measured in the establishment year). This observation points to the importance of the establishment year and having many seeds germinate and many seedlings survive. Fields that were well drained may have been more susceptible to seedlings dying from moisture stress, which is an important factor affecting seedling survival (Hsu and Nelson, 1986). Therefore, well drained sites are likely more sensitive to planting prior to extended dry periods, so planting dates should be selected that provide the greatest probability for regular precipitation to promote rapid establishment. These sites also displayed no positive yield response to N application in that time period. It appears that if the initial stand establishment is good (and thus plant and tiller density are high), then adding N does not increase yields in the short term. On fields where plant and tiller density are low, added N may improve yields. At one location in Texas, Muir et al. (2001) reported tiller mass of the lowland switchgrass Alamo increased with increasing N fertility. The limited response to N inputs generally observed here is characteristic of switchgrass, particularly under single, end-of-season harvest management. Indeed, this has been an important criterion for choosing switchgrass as a potential energy crop. Several factors may contribute to this apparent lack of response, including an ability to mobilize large quantities of N from belowground storage (Dohleman et al., 2012; Lemus et al., 2008b; Wayman et al., 2014) and capacity to attain large amounts of N from soil pools (Stout et al., 1991). In addition, N from atmospheric deposition (Coulston et al., 2004) and contributions of N from fungal and bacterial symbionts also may affect shoot N uptake and increase biomass production (Ghimire and Craven, 2011; Ker et al., 2012; Schroeder-Moreno et al., 2012).

Switchgrass yields on these marginal sites generally were well below those reported elsewhere. Khanna et al. (2011) predicted peak yields in the Midwest ranging from 9.9 Mg ha^{-1} (Minnesota) to 15.5 Mg ha^{-1}. In contrast, mean yields obtained in these studies at 112 kg N ha^{-1} range from 4.6 Mg ha^{-1} (South Dakota) to 10.4 Mg ha^{-1} (Alabama). Quite apart from N response, switchgrass yield of currently available cultivars on such marginal sites may not be sufficient to warrant establishment for purposes of supplying a biofuel or bioenergy facility, given the increased per-unit logistics costs associated with low yields or limited land base available (Fike et al., 2007).

In addition to field studies, switchgrass field researchers and scientists from Oak Ridge National Laboratory met with the Oregon State University PRISM-ELM Climate group to develop maps of switchgrass yield potential across the U.S. based on data gathered from these field trials and from previous work (Figure 2). Average relative maximum yield for lowland ecotypes was 22 Mg ha^{-1} and 13 Mg ha^{-1} for upland ecotypes. Modeled yields confirm the yield advantage of lowland ecotypes, specifically in the southeastern U.S. They also demonstrate the wide adaptability of upland ecotypes east of the 100^{th} meridian.



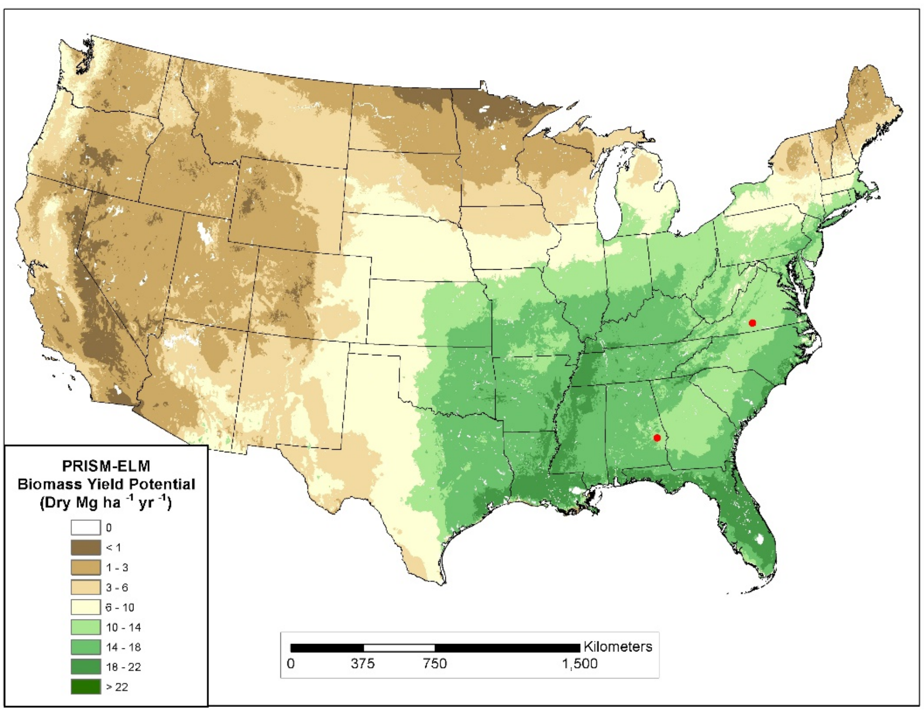


Figure 2. Biomass yield potential of upland (top) and lowland (bottom) switchgrass for the U.S. generated using the PRISM-ELM model and based in part on Regional Feedstock Partnership Field Trials (red dots) (from Lee et al., 2018).

Using the ethanol efficiency reported by Schmer et al. (2008) of 0.38 L kg^{-1}, a relatively small 100 M L yr^{-1} ethanol refining facility would require from 26000 ha (Virginia) to 57000 ha (South Dakota) of similar farmland to supply it with sufficient switchgrass. The cultivars used in the current study were all released between 1944 (Blackwell) and 1998 (Sunburst) and do not represent yield gains made in cultivars such as 'Liberty' (Vogel et al., 2014) released specifically for bioenergy. Gains in switchgrass biomass yield of up to 4% per year have been achieved through intrapopulation improvement methods (Casler and Vogel, 2014). Thus, more genetically improved cultivars are needed for other ecoregions to significantly reduce the land base needed for a bioenergy facility.

As described in Fike et al. (2017), economic results include production cost ha^{-1} (Figure 3) and production cost Mg^{-1} (Figure 4). Data for Alabama were incomplete and could not be included here. Mean total production cost ha^{-1} in 2015 dollars averaged \$452 ha^{-1} and ranged from \$394 (South Dakota) to \$536 ha^{-1} (New York), which had the lowest and highest harvest costs, respectively, primarily because of higher harvests costs associated with greater yields in New York compared to South Dakota. Production costs are determined not only by production activities, but also by establishment costs, land rent and yields. Per hectare, the highest cost in New York was 36% greater than in South Dakota. New York had the highest pro-rated establishment costs (\$64 ha^{-1}), largely due to high seed costs, and the highest harvesting costs (\$267 ha^{-1}) among sites. Both land charges and pre-harvest operating expenses were greatest in South Dakota, but these were more than offset by the very low harvest charges for that site. Although the South Dakota location was on marginally productive cropland, land rent for the area was higher than the other locations, with the exception of Iowa, due to competition for annual crops. On the other hand, labor is typically lower than in other areas which helped to reduce overall harvest charges. For comparative purposes, the mean weighted average annualized cost of production reported in Perrin et al. (2008) was \$453 ha^{-1} (\$2015), almost identical to the mean production cost reported here. However, the per Mg production cost of biomass in this study is higher than that of Perrin et al. (2008) because their estimates included staging and storing costs, which were not estimated in this study.

Mean production costs per bone dry Mg dry matter varied widely: from \$65 Mg^{-1} in Oklahoma to \$99 Mg^{-1} in South Dakota. Costs per Mg in Oklahoma, New York and Virginia were intermediate (\$65 to \$73 Mg^{-1}). Although the New York site had the highest production costs per hectare, these were offset by relatively high yields, resulting in a per Mg cost of (\$73). South Dakota county land rental rates were much higher than in other states, likely reflecting competition for land from corn production, and switchgrass yields were relatively low (3-4.6 Mg ha^{-1}). Thus, the South Dakota unit cost of production was 66% greater than that of Oklahoma, which benefited from greater average yields (6.5 Mg ha^{-1}).

The key questions to be explored in the data from these sites is whether there is economic justification for application of N fertilizer, and if so, how much? As noted in the discussion of yields, observed evidence of yield response to N fertilization was relatively weak and sporadic, and at one site (New York) the yield response to N was sporadically negative.

The economically efficient management rule is to increase input use until the value of production from the marginal input equals the price of that input (including application cost), or in other words, until marginal revenue equals marginal cost. The results for New York and

Oklahoma (poorly drained sites), are clear – there is little or no apparent economic justification for any N application on these sites at any currently expected switchgrass price. In South Dakota, there was statistical evidence of increased yields from application of 56 (or more) kg N ha^{-1} compared to the control treatment. However, the breakeven switchgrass price at the farmgate to justify such an application would have to be over \$70 Mg^{-1}. Data from Virginia indicate significant yield increases when responses to N applications of 112 kg ha^{-1} are compared to the control treatment. While there is some economic evidence to warrant such N application rates at switchgrass prices above \$63 Mg^{-1}, it remains unknown whether a bioenergy industry can support such a price.

The production costs and associated switchgrass yields reported here indicate the need for further production economic research on N response. Based on these results for marginal sites, it could be suggested that typically-promoted agronomic recommendations include costly and economically unjustified N application rates. Typical recommendations for N fertilization in published switchgrass budgets often range from 56 kg ha^{-1} to 112 kg ha^{-1}. At N prices used here, such applications add \$37-\$74 ha^{-1} to production costs, with sparse evidence of an economically profitable response with the currently available cultivars. Mitchell et al. (2013) suggested applying 5.4 to 6.3 kg of N ha^{-1} for each Mg ha^{-1} of expected DM yield when harvesting after frost. At these rates, using the mean yield for the 0 N treatment as the expected yield for the Iowa, New York, and Oklahoma sites, suggested N fertilizer rates would range from 32 to 45 kg N ha^{-1}, significantly reducing N application costs.

Data from these studies provide greater understanding of the year-to-year and site-to-site variability in switchgrass production than is available with other research. The multiple years encompassed by this work also show the changes in production and nitrogen utilization that would not have been observable with shorter-term research. Data in the literature (largely from small plot studies) regarding switchgrass response to nitrogen are highly variable, and our data indicate that response to nitrogen occurs primarily on soils that are nitrogen limited (Owens et al. 2013). In addition, our data indicate that with soils of even moderate fertility, it may take several years of harvesting to reach a point at which response to nitrogen applications becomes economical.

The Partnership has had a history of good leadership, and communication and coordination with the research team worked well. In the field, the research was faced with some initial crop failures, but all sites achieved adequate stands of switchgrass for the desired research. In general, the outputs of these trials are indicative of a strong and cohesive research effort and provide better insight into realistic production estimates from field-scale operations. These trials have also developed a large database available through the KDF that will be mineable for researchers and modelers in the future and were informative for the PRISM modeling efforts.

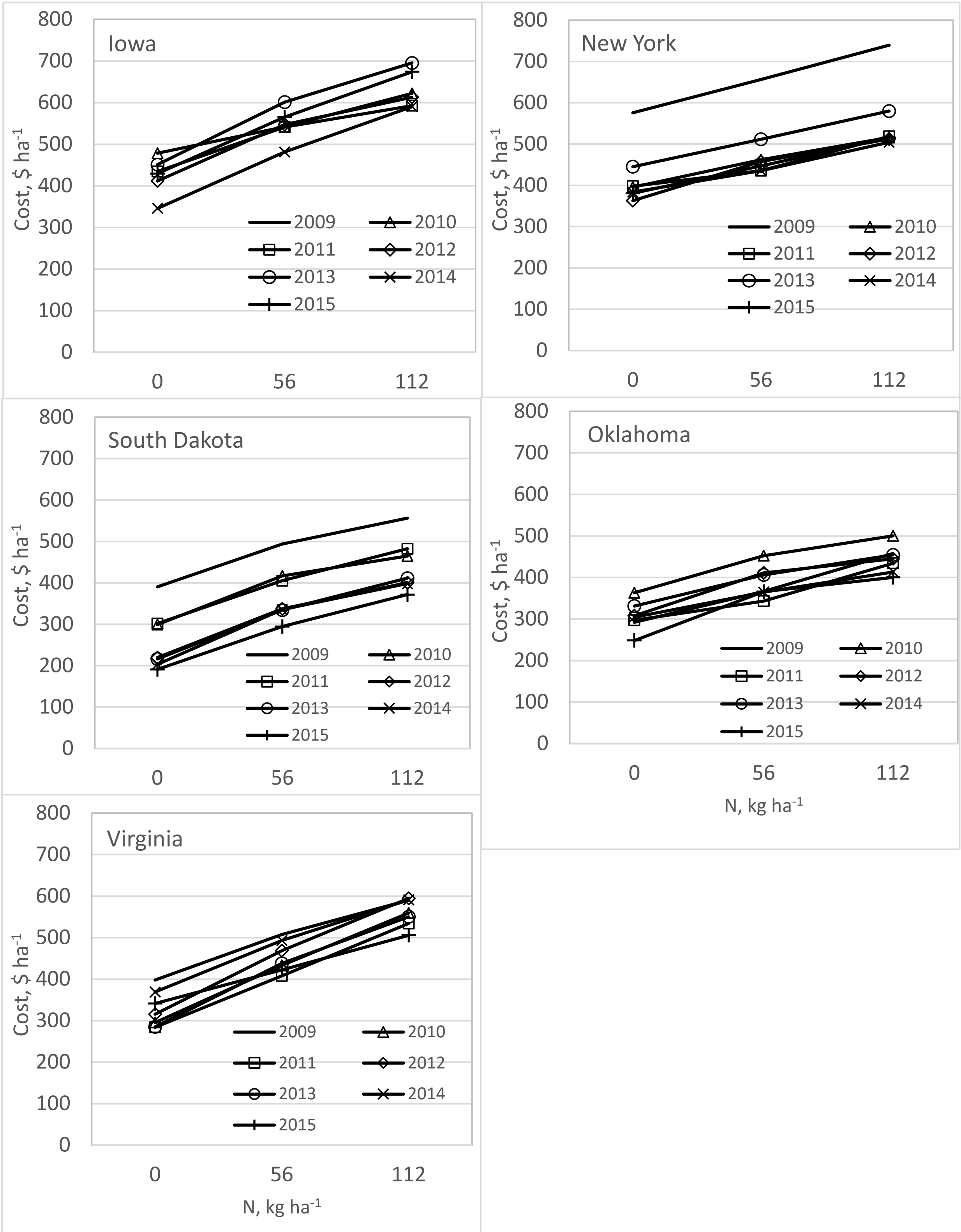


Figure 3. Annual switchgrass production costs per hectare in 2015 dollars at five diverse sites in the USA. Establishment costs are prorated over 11 years (adapted from Fike et al., 2017).

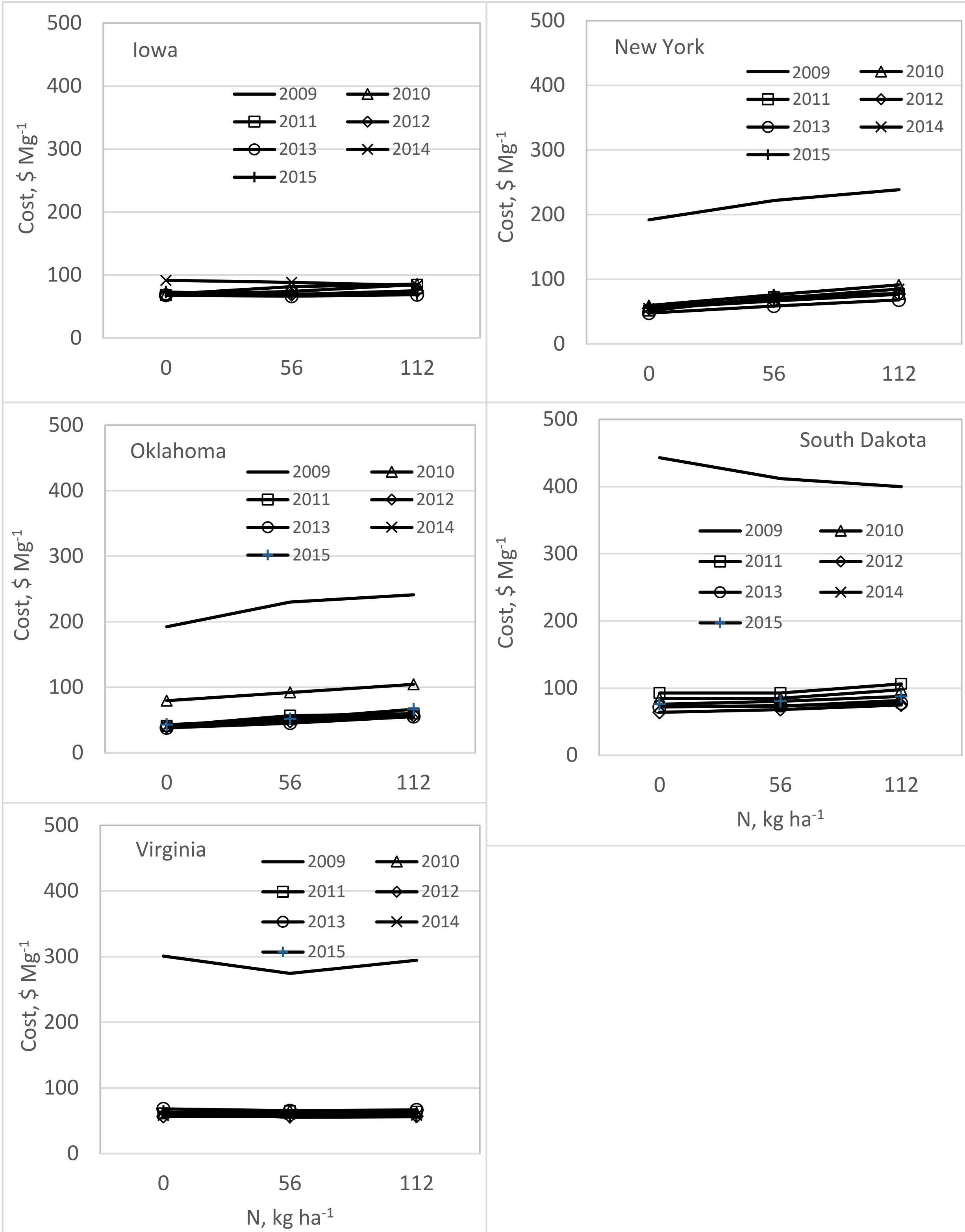


Figure 4. Annual switchgrass production costs per bone dry megagram in 2015 dollars at five diverse sites in the USA. Establishment costs are prorated over 11 years (adapted from Fike et al., 2017).

Sustainability

The trend for annual average soil surface N_2O fluxes (g ha^{-1} d^{-1}), averaged across the three N rates (0, 56, and 112 kg N ha^{-1}) from 2010 to 2015 at the South Dakota location are presented in Figure 5 (Lai et al., 2018). There was a significant downward trend in soil surface N_2O fluxes over time. This could mainly be attributed to the fact that switchgrass has a deep root system which adds organic matter to the soil, decreases soil bulk density, and increases soil porosity; subsequently decreasing denitrification and hence the N_2O emissions. In addition, soils at this site were not disturbed (not cultivated) since the switchgrass was established in 2008, thus further lowering soil bulk density (Clark et al., 1998) over years. Soils with lower bulk density generally have less water filled pore space (WFPS), which can decrease denitrification rates (Parton et al., 2001). Denitrification is the principal pathway that produces N_2O emissions to the atmosphere (Bouwman, 1990; Weier et al., 1993). Therefore, we suspect that the decreasing N_2O fluxes over the course of this study were primarily a result of reductions in the rate of denitrification resulting from the decreased soil bulk density. Furthermore, deep switchgrass roots can promote increased water infiltration rate over time (Katsvairo et al., 2007), resulting in more leaching of NO_3^- in top soil over time. This reduces the source of N for denitrifying bacteria over time (Hofstra and Bouwman, 2005), resulting in a downtrend of the soil N_2O fluxes over the years. Although deep rooting systems are inherently important, they may not be able to reduce N_2O emissions indefinitely, as noted by the leveling off of N_2O fluxes in later years of this research (Figure 5).

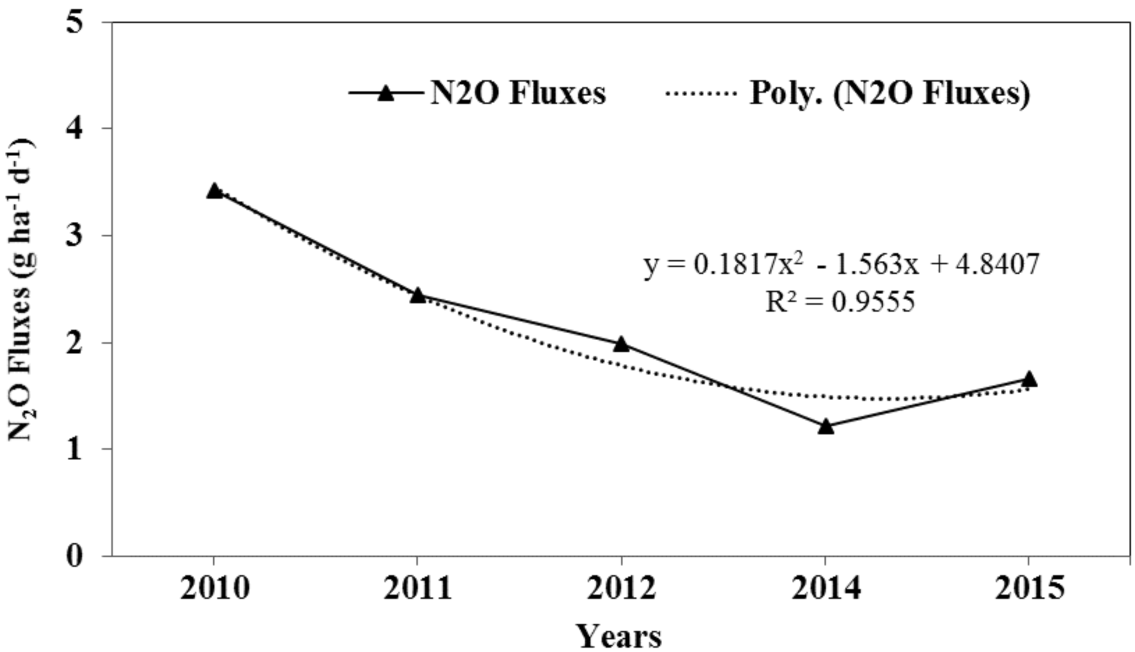


Figure 5. Average annual soil N_2O fluxes (g ha^{-1} d^{-1}) averaged across three N rates (0, 56, and 112 kg N ha^{-1}) from 2010 to 2015 in a switchgrass field in South Dakota. Switchgrass was planted at this location in 2008 (adapted from Lai et al., 2018).

Similar to N_2O emissions, average annual NO_3^-, averaged across N rates, leaching decreased from 2009 to 2015 (Figure 6) (Lai et al., 2018). This was primarily attributed to changes in annual precipitation over time and the potential for an abundance of arbuscular mycorrhizal (AM) fungi. First, annual precipitation followed a slight downtrend from 2009 to 2015. Lower precipitation could result in less soil moisture, which can decrease denitrification rate, reducing the nitrate available for leaching (McIsaac et al., 2010). Secondly, the abundance of AM fungi in switchgrass would enlarge the nutrient interception zone, prevent nutrient loss after rain-induced leaching events, reduce the volume of soil leachate (Asghari et al., 2005; van der Heijden, 2010), and enhance rates of N immobilization thus reducing the risk of N loss via leaching (Cavagnaro et al., 2015).

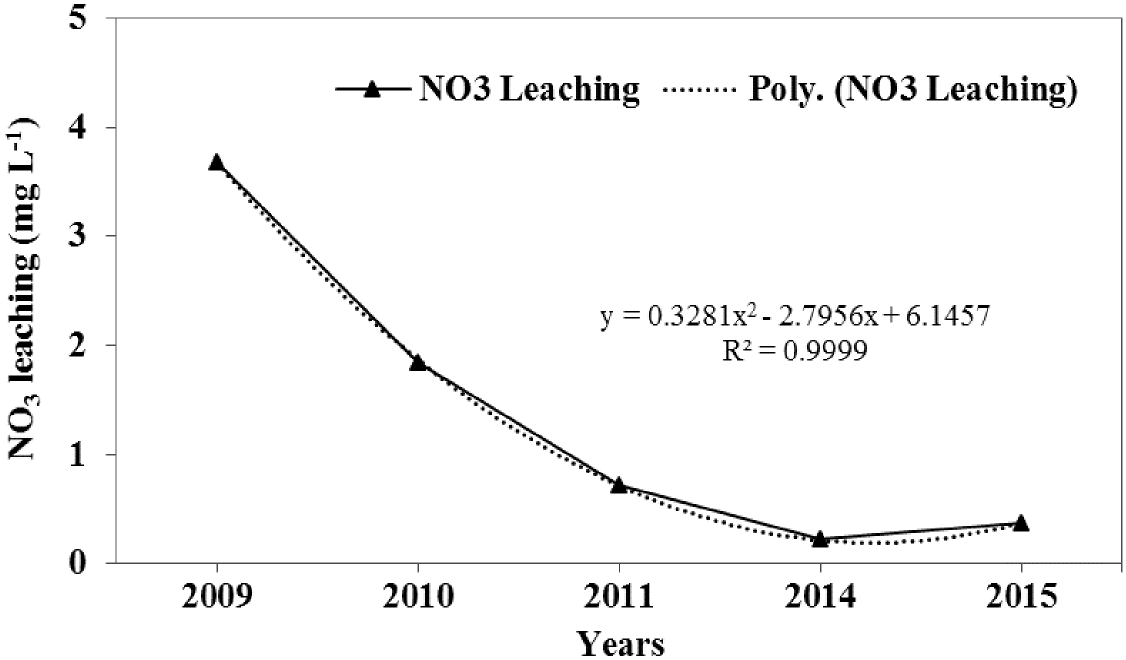


Figure 6. Trends of annual mean soil NO_3^- leaching contents (mg L^{-1}) averaged across the three N rates (0, 56, and 112 kg N ha^{-1})from 2009 to 2015 at Bristol site in South Dakota (adapted from Lai et al., 2018).

Average annual soil surface CO_2 fluxes (kg ha^{-1} d^{-1}) from 2010 to 2015 are presented in Figure 7 (earlier parts of this work were published in Mbonimpa et al. (2015). There was an upward trend in soil surface CO_2 fluxes over this time period. Switchgrass has an extensive fibrous root system that contributed to increases in soil organic matter (SOM). This was primarily due to increased soil organic matter (SOM) decomposition made possible by the addition of SOM from the extensive fibrous root system of switchgrass (Brown et al., 2000; Frank et al., 2004). These findings indicate that the growing switchgrass can benefit soil fertility but increase soil surface CO_2 emissions over the observed years. However, soil CO_2 emissions appear to level off after several years of switchgrass growth and development (Figure 7).

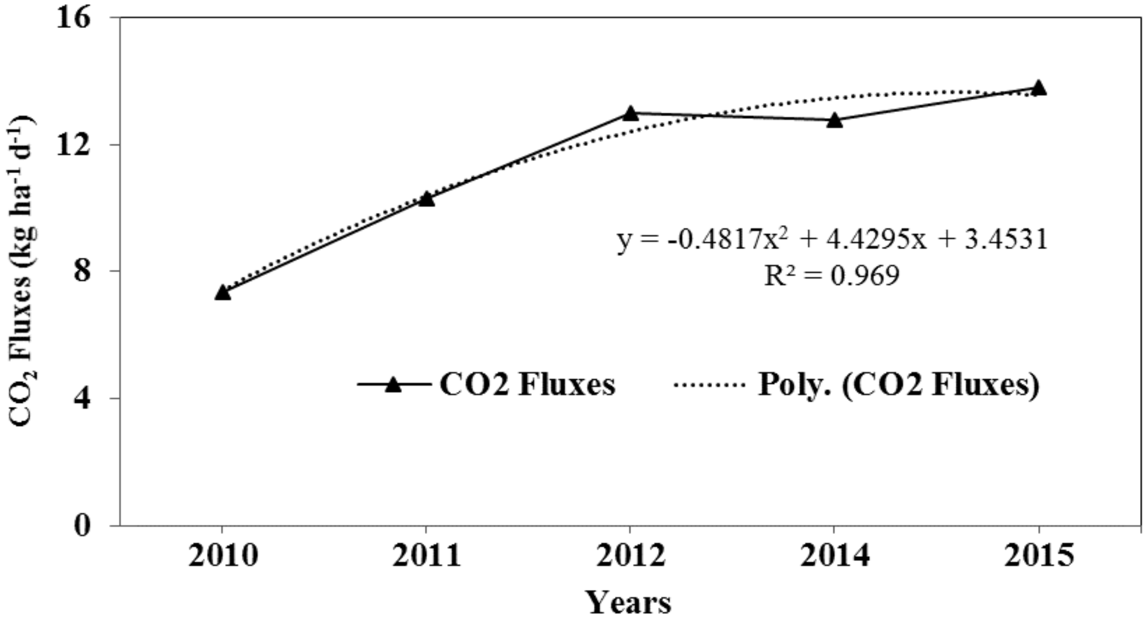


Figure 7. Trends of annual average soil CO_2 fluxes (g ha^{-1} d^{-1}) averaged across N rate (0, 56, 112 kg N ha^{-1}) from 2010 to 2015 at Bristol site in South Dakota (adapted in part from Mbonimpa et al., 2015).

Key outputs

Peer-Reviewed Publications

Lee D.K., E. Aberle, E.K. Anderson, W. Anderson, B.S. Baldwin, D. Baltensperger, M. Barrett, J. Blumenthal, S. Bonos, J. Bouton, D.I. Bransby, C. Brummer, P.S. Burks, C. Chen, C. Daly, J. Egenolf, R.L. Farris, J.H. Fike, R. Gaussoin, J.R. Gill, K. Gravois, M.D. Halbleib, A. Hale, W. Hanna, K. Harmoney, E.A. Heaton, R.W. Heiniger, L. Hoffman, C.O. Hong, G. Kakani, R. Kallenbach, B. Macoon, J.C. Medley, A. Missaoui, R. Mitchell, K.J. Moore, J.I. Morrison, G.N. Odvody, R. Ogoshi, J.R. Parrish, L. Quinn, E. Richard, W.L. Rooney, J.B. Rushing, R. Schnell, M. Sousek, S.A. Staggenborg, T. Tew, G. Uehara, D.R. Viands, T. Voigt, D. Williams, L. Williams, L.T. Wilson, A. Wycislo, Y. Yang, V. Owens*. 2018. Biomass Production of Herbaceous Energy Crops in the United States: Field Trial Results and Yield Potential Maps from the Multiyear Regional Feedstock Partnership. GCB Bioenergy, doi: 10.1111/gcbb.12493.

Fike, J.H., J.A. Pease, V.N. Owens, R.L. Farris, J.L. Hansen, E.A. Heaton, H.S. Mayton, R.B. Mitchell, D.R. Viands. 2017. Switchgrass Nitrogen Response and Cost of Production on Diverse Sites. GCB Bioenergy. 9: 1526–1542. doi:10.1111/gcbb.12444.

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Mitchell, R., V. N. Owens, N. Gutterson, E. Richard, and J. Barney. 2010. Herbaceous Perennials: Placement, Benefits and Incorporation Challenges in Diversified Landscapes. In *Sustainable Alternative Fuel Feedstock Opportunities, Challenges, and Roadmaps for Six U.S. Regions*, edited by R. Braun, D. Karlen, and D. Johnson. Ankeny, IA: Soil and Water Conservation Society. http://www.swcs.org/roadmap.

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Owens V. N., C. O. Hong, D. Bransby, R. Farris, J. Fike, E. Heaton, H. Mayton, R. Mitchell, and D. Viands. 2012a. Switchgrass Response to N Fertilizer across Diverse Environments in the USA: A Regional Feedstock Partnership Report. In *Proceedings of the 2012 Sun Grant National Conference: Science for Biomass Feedstock Production and Utilization*, Volume 1. New Orleans, LA, October 2–5. https://ag.tennessee.edu/sungrant/Documents/2012%20National%20Conference/ConferenceProceedings/Volume%201/Vol1final.pdf.

Presentations

Burke R.H., K.J. Moore, M.J. Shipitalo, F.E. Miguez and E.A. Heaton. 2016. Simulated Rainfall Events Induce Real, but Minimal, Foliar Nutrient Leaching in Senescing Switchgrass Biomass in Iowa. Presented at the American Society of Agronomy, Crop Science Society of America, and Soil Science Society of America International Annual Meeting, Phoenix AZ, Nov. 6-9.

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Kumar, S., C. O. Hong, E. Mbonimpa, V. N. Owens, R. M. Lehman, S. Osborne, T. Schumacher, and D. Clay. 2014. Soil Carbon Dioxide and Methane Fluxes Influenced by Nitrogen Rates and Landscape Positions from Switchgrass Land of South Dakota, USA. Poster presented at the 20th World Congress of Soil Science, Jeju, South Korea, June 8–13.

Kumar, S., C. O. Hong, V. N. Owens, D. E. Clay, M. Lehman, S. L. Osborne, T. E. Schumacher, and E. G. Mbonimpa. 2013. Soil Carbon Dioxide Fluxes from Switchgrass Land under Nitrogen Fertility Management in South Dakota. Presented at the American Society of Agronomy, Crop Science Society of America, and Soil Science Society of America International Annual Meeting, Tampa, FL, November 3–6. https://www.researchgate.net/publication/268116297_Soil_Carbon_Dioxide_Fluxes_From_Switchgrass_Land_Under_Nitrogen_Fertility_Management_In_South_Dakota.

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Where do we go from here?

Several questions affecting economic outcomes could not be addressed by this research. This includes factors such as harvest method, storage, and supply logistics. For example, the large square and large round bales used for these systems could require very different field-to-factory storage and handling systems. These issues were beyond the scope of this research but have further economic implications for switchgrass to bioenergy/bioproduct systems. From an industry perspective, the economics behind these production systems will present the greatest barriers to development and deployment. Developing plant materials that are more productive on marginal sites may be an important next step for moving forward if marginal land is to be used for these systems.

Initially, data collection was challenged in part by the lack of stand establishment in Alabama, hence, we were only able to collect yield data over the last few years (2012–2015) of the trial. In addition, greater yield potential would be expected from the Oklahoma site if a southern lowland cultivar had been available for planting at the start of the study. To address the issues of upland versus lowland switchgrass production in Oklahoma, we began a cooperation with a second site to gather some of the lowland production data. These data have been made available for the modeling efforts that are part of the Partnership. One of the challenges facing the modeling team is the limited data from sites on the periphery of the switchgrass production range. Therefore, initiating trials in these areas would provide critical data for the next iteration of yield potential maps.

Whether switchgrass response to nitrogen should have been the preferred treatment has been a question for discussion at times. Other treatments that warrant exploration include use of species mixtures vs. monocultures, and how best to extend harvest windows for these systems in order to reduce logistics costs; and these systems will likely be different depending on location. For example, harvest timing may vary from summer to the following spring to explore variation in moisture concentration of just-in-time harvested biomass as well as to evaluate harvest timing impacts on other ecosystem goods and services (e.g., wildlife habitat, soil health, etc.).

Although such questions are important, perhaps the most productive line of inquiry for the future would involve the direct comparison of the various energy crops suited to a region. For example, both miscanthus and switchgrass are suited to many of the sites used in the feedstock partnership, but there is little assessment of their productivity in side-by-side trials, something which should be included in future work.

Aside from stover and other crop residues, switchgrass remains the primary biomass feedstock of interest across much of the United States. This is due to its high productivity, broad adaptability, perenniality, and the biofuel community's high level of familiarity with the crop. Although not part of this work, new switchgrass varieties have been developed or are in the development pipeline, and they promise to increase yields by several percentage units. Future work assessing these materials will be important for optimizing opportunities for the nascent bioenergy industry.

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Poplar

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Summary

Poplar is one of the energy crops selected for further research under the Sun Grant Regional Feedstock Partnership. Poplar is known to be one of the most productive tree species in the world adapted to a range of climatic conditions globally. Commercial production of poplar in plantations has taken place in many regions of the world where the combination of selected superior genotypes and economic conditions facilitate production of competitively-priced wood for a variety of uses, including structural lumber, pulp and papermaking, fuel for heat and power generation, and biofuel production. While there are a wide variety of species classified in the *Populus* genus including aspens and cottonwoods, the species selected for research include *Populus deltoides*, *P. trichocarpa*, *P. nigra* and *P. maximowiczii* with hybrids among these species generally referred to as “hybrid poplar”. These species were selected due to a number of attributes that lend themselves to commercial culture and yield improvement. First, unlike most trees, these species can be propagated through dormant hardwood cuttings. This enables cost-effective commercial deployment of clonal populations with attributes specifically selected for a variety of industrial uses. Due to the fact that poplar can be clonally deployed, variation in feedstock chemical and physical properties can be reduced, thus enhancing the efficiency of industrial conversion to fuels and chemicals. Second, these species can be readily hybridized to combine complementary attributes useful for commercial culture and industrial processes. For example, combining traits of *P. deltoides* with those of *P. nigra* makes it possible to increase disease resistance and rooting of hardwood cuttings in commercial plantations over what might be possible with pure-species genotypes. Also, the demonstrated high natural variation in growth rate and disease resistance within parental populations provides significant opportunity to increase biomass yield through multiple cycles of breeding, field testing and selection.

The Sun Grant Regional Feedstock Partnership provided the opportunity to consolidate a wide array of field trials and genetic improvement research within one national framework. The members of the Poplar Team consist of academic institutions and industry partners, including the University of Minnesota, Michigan State University, Mississippi State University, the University of Tennessee, ArborGen (Southeast U.S.) and GreenWood Resources (Pacific Northwest). Research was oriented toward commercial production in close cooperation with industrial partners. The combined legacy of field sites and breeding activity pre-existing the Partnership, as well as new field tests and breeding activity done by the Poplar Team enabled us to consolidate, leverage and expand the ongoing research and development activity. Without the funding from the Partnership, many of the longer-term legacy sites would likely have been abandoned with no measurements taken. As a result, prior investment by universities and industry across the country combined with the DOE/Sun Grant funds made possible an unprecedented program with federal funds adding needed research infrastructure to a foundation of existing sites. The resulting program has produced significant progress in nationally-coordinated poplar research related to advanced breeding, field testing, yield analysis and evaluation of wood characteristics of poplar.

Estimates of biomass yield of poplar were developed using the nationwide network of yield tests. Field sites used in this work included a combination of clone tests, large yield-block experiments and commercial plantations. Due to the lack of yield data from large-scale

plantations on uplands in the Southeast U.S., work was done to develop reference curves based on height growth to estimate yields from clone tests in the Southeast region. Yield estimates in the Midwest, Pacific Northwest and Mid-South regions were done using data collected from large-block field experiments or sampling in commercial plantations. Biomass yield ranged from 7.8 to 13.5 Mg per hectare per year of total oven dry biomass, with higher yields in the Pacific Northwest, Mid-South and Southeast due to a longer growing season.

Economic analysis of poplar production was done using cash flow analyses with cost input and yield information derived from commercial programs in Minnesota and the Pacific Northwest. Based on our estimates of production costs, including stand production and harvest and transport economics, the DOE's delivered price target range of $92 per dry Mg appears to be achievable on many sites in the Midwest. In the Pacific Northwest, the estimated breakeven production cost using prevailing land values ranged from $77.00 to $110.00 assuming unirrigated land and current yields. Spreadsheets of our cash flow analyses were provided to staff working on the recently completed Billion Ton Update (U.S. DOE, 2016).

Introduction

Poplar is one of the energy crops selected for further research under the Sun Grant Regional Feedstock Partnership. Poplar is known to be one of the most productive tree species in the world adapted to a range of climatic conditions globally (Zamora et.al.2015). Commercial production of poplar in plantations has taken place in many regions of the world where the combination of selected superior genotypes and economic conditions facilitate production of competitively-priced feedstock (Stanton et.al. 2014, Berguson et.al. 2010, Lazarus et.al. 2015). While there are a wide variety of species classified in the *Populus* genus including aspens and cottonwoods, the species selected for research include *Populus deltoides*, *P. trichocarpa*, *P. nigra* and *P. maximowiczii*, with hybrids among these species generally referred to as "hybrid poplar". These species were selected due to a number of attributes that lend themselves to commercial culture and yield improvement strategies. First, unlike most trees, these species can be propagated through dormant hardwood cuttings. This enables cost-effective commercial deployment of clonal populations with attributes specifically selected for a variety of industrial uses. Due to the fact that poplar can be clonally deployed, variation in feedstock chemical and physical properties can be reduced, thus enhancing the efficiency of industrial conversion to fuels and chemicals. Second, these species can be readily hybridized to combine complementary attributes useful for commercial culture and industrial processes. For example, combining traits of *P. deltoides* with those of *P. nigra* makes it possible to increase disease resistance and rooting of hardwood cuttings in commercial plantations over what might be possible with pure-species genotypes. Also, the demonstrated high natural variation in growth rate and disease resistance within these populations provides significant opportunity to increase biomass yield through multiple cycles of breeding, field testing and selection.

A significant benefit to the Regional Feedstock Partnership program is the large infrastructure of trials and effort that has been expended that predates the Partnership, which began in 2008. All of the cooperators in the Regional Feedstock Partnership poplar team have been conducting research and commercial application over the previous two decades to enhance poplar production. Even in the case of programs managed by academic institutions, such as the University of Minnesota, Michigan State and Mississippi State University, the majority of the effort was oriented toward commercial production with research often being done in close cooperation with industrial partners. In addition to the three academic institutions involved in the Sun Grant Poplar Team, two industrial research programs run by ArborGen in the Southeast and

GreenWood Resources in the Pacific Northwest were members of the Poplar Team. As a result, there existed a large network of field tests and genetic collections that enabled us to consolidate and expand the ongoing research and development activity under the Partnership. Without the Sun Grant Partnership, many of these legacy sites would likely have been abandoned with no measurements taken. As a result, prior investment by universities and industry across the country combined with the DOE/Sun Grant funds made possible an unprecedented program with federal funds adding needed research infrastructure to a foundation of existing sites. The resulting program has produced significant progress in nationally-coordinated poplar research related to advanced breeding, field testing, yield analysis and evaluation of wood characteristics of poplar.

The goal of the research program is to improve growth and reduce production risk and costs of biomass produced in dedicated feedstock production systems. In the case of poplar, the need exists in all regions of the United States to develop a suite of genetically diverse clones with demonstrated high yield, disease resistance and growth stability. Due to the wide array of genotypes available in each region generated through ongoing selection and breeding programs, a major emphasis of research is the development of new clones through breeding and testing of these new materials leading to increased biomass production and reduced cost.

The extreme genetic diversity within and among *Populus deltoides*, *P. nigra*, *P. trichocarpa*, and *P. maximowiczii* presents great opportunity to capitalize on this variation to improve yield and disease resistance of poplar as an energy crop. However, no method currently exists to estimate *a priori* performance of clones in a given region and circumvent the process of planting regional field trials to observe growth rate and disease resistance under field conditions over time. While alternate methods are being explored, disease resistance of poplars can change through time as pathogen abundance and virulence changes (Dunnell 2016). As a result, clone tests are a necessary part of research to identify the subset of clones from a larger collection that could be considered for commercial release in operational biomass production as well as the next generation of parents to be used in further breeding efforts. Also, identification of the best genotypes suited to a region is critical to deciding the subset of clones to be used in more intensive research (such as enhanced yield analysis under various management scenarios using different stand spacing) or in coppice management (Miller and Bender, 2012). An additional consideration is that the phenotype of growth rate and disease resistance is not immediately evident, and growth ranking among clones can change significantly over time. In light of this reality, clone trials must be done in the target regions and be maintained over a sufficient time period to identify those clones that are most promising for commercial production. Further, clone performance at one site within a region may or may not be stable across other sites within that region. This significant “genotype-by-environment interaction” and changes in clone ranking over time necessitate intensive testing across multiple sites within a region once a subset of superior material has been identified.

It should be noted that a large pool of clones suited to a region can only be derived through a breeding program. Initially, clonal material can be selected from wild populations, but further progress can only be made through breeding. Collection of populations from the wild is a first step in the process but cannot be the final step. Breeding both within and among candidate species must be done to improve yield, disease resistance, and other characteristics such as rooting ability from hardwood cutting, wood characteristics, and tree form. The phases of genetic improvement and field testing will be discussed in greater detail in their respective sections.

Field Trials in the Sun Grant Poplar Program

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The field testing program contains a range of yield studies, clone tests, and larger scale family field trials underway at a variety of locations ranging from the Pacific Northwest, upper Midwest, alluvial Mid-South, and Southeastern regions. Figure 1 shows the location of 45 poplar study sites across the United States that were included in the Poplar Team's field testing network.

Figure 1. Location of 45 poplar field tests in Sun Grant Regional Feedstock Partnership trial network.

For clarification, the Family Field Test (FFT) sites are located in Minnesota and are part of the breeding and field testing program with a specific purpose to estimate variance components and additive and non-additive genetic effects, as well as the selection of new clones for more intensive testing. To our knowledge, no trials of this type and scale have been done on poplar prior to the Sun Grant program. While family field tests contain many clones-within-families (i.e., a family includes the progeny resulting from a sexual cross between the same two parents and typically includes 30 full-sib hybrid families each represented by 20-30 individual progeny), clone tests typically include fewer clones that have been selected from a larger population with these tests containing more replicates of each clone (four to six single-tree replicates). Clone tests are planted at multiple sites within a region with the primary purpose to evaluate stability in growth across sites. Once a reduced set of superior clones has been selected for growth rate and disease from clone tests, yield tests are planted to evaluate growth of single clones under conditions similar to commercial-scale planting. Clone trials (consisting of 50-60 clones)are planted in three replicated blocks of sufficient size to eliminate edge-effects and estimate biomass yield in absolute terms (e.g., Mg ha^{-1} yr^{-1}) as opposed to relative terms, as is the case in family field tests and clone tests. Figure 2 details the typical poplar breeding and selection process. Data from a combination of yield block tests and commercial plantations provided the foundation for the national maps of expected biomass yield developed by the Oregon State University Parameter-Elevation Relationships on Independent Slopes Model (PRISM) group. Tables 1 and 2 show the current Sun Grant field tests underway by state and trial type.

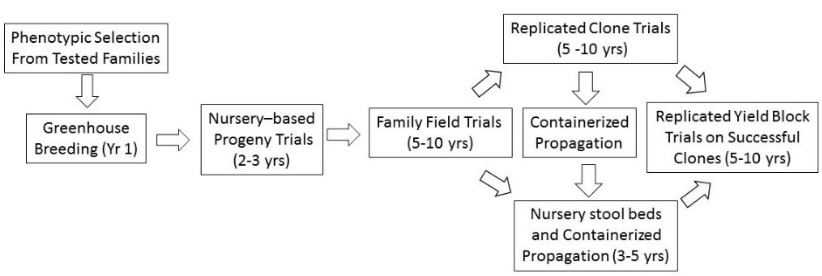

Figure 2. Typical selection and breeding process for poplar for biomass.

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Table 1. Sun Grant Populus Field Trials by Establishment Year, State, Study Design with Family and Clone Composition, Number of Sites, and Study Size. All field sites were maintained at least through 2013. In Minnesota, only the 2008 and 2009 trials continue to present.

Year	State	Study	Source	Families	Clones	Sites	Hectares
1999	MN	Family Field Trial	1996 CP[a]	21	563	1	0.65
1999	MN	Family Field Trial	1996 OP[b] *P. deltoides*	78	1170	1	1.09
2000	MN	Family Field Trial	1996 CP & 1997 CP	38	684	1	0.81
2000	MN	Family Field Trial	1997 OP *P. deltoides*	50	750	1	0.69
2001	MN	Family Field Trial	1998 CP	69	1725	1	5.30
2002	MN	Family Field Trial	1999 CP	33	899	1	2.99
2003	MN	Family Field Trial	1999 CP	27	907	1	2.99
2004	MN	Family Field Trial	2002 CP	35	785	2	4.37
2005	MN	Family Field Trial	2003 CP	33	511	2	6.64
2006	MN	Clone Trial			70	2	0.81
2006	MN	Yield Blocks			22	2	6.60
2007	MN	Family Trial	2003 CP & 2004 CP	40	672	2	4.13
2007	MN	Clone Trial			70	2	0.81
2007	MN	Yield Blocks			12	2	3.28
2008	MN	Family Field Trial	2005 CP & 2006 CP	45	400	1	1.86
2008	MN	Clone Trial			70	6	2.43
2009	MN	Clone Trial			70	3	1.21
2009	MN	Clone Trial	2005 OP *P. nigra*	10	46	3	0.85
2009	MN	Yield Blocks			10	3	3.04
2010	MN	Family Field Trial	2007 CP	30	400	1	1.86
2010	MN	Clone Trial			70	2	0.81
2010	MN	Yield Blocks			10	3	2.06
2011	MN	Clone Trial			98	2	1.05
2011	MN	Yield Blocks			12	2	1.62
2008	MI	Yield Trial			7	1	
2010–2012	MI	Yield Test			16	5	
2010	GA	Yield Block			7	1	0.81
2003	GA	Clone Trial			120	2	1.05
2010	GA	Yield Block			2	1	0.24
2008	SC	Clone Trial			243	1	0.62
2011	SC	Clone Trial			84	1	0.40
2009	SC	Clone Trial			162	1	0.85
2013	SC	Clone Trial	*P. nigra*		690	1	1.13
2009	AL	Clone Trial			162	1	0.85
2009	AL	Clone Trial			124	1	0.36
2010	NC	Clone Trial			87	1	0.20
2010	NC	Yield Block			9	1	0.85
2010	NC	Yield Block			10	1	0.09
2013	TN	Clone Trial	*P. nigra*		670	1	0.61
2013	VA	Clone Trial	*P. nigra*		690	1	0.57
Totals					13,109	68	66.60

[a] CP designates a controlled cross or controlled pollination.
[b] OP designates an open pollination or plant collected from the wild.

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Table 2 shows sites included in Sun Grant tests under management by GreenWood Resources. These trials include a range of studies, including Bioenergy Trials, which are single-tree plots replicated three or four times containing 80 to 89 clones; Consolidated Clone Trials, which contain a complement of clones from the Sun Grant cooperators with 20 clones each of four sources in a single tree design with six replicates; Stage I Trials, which are tests of multiple seedlings with no replication (initial observation of adaptability); Stage II trials, which are clone trials containing eight, single-tree replicate plots of many clones; and Stage III tests, which are block plantings embedded in operational acreage typically having four replications of 8 to 20 clones depending on year planted. Also, nursery tests and orchards are comprised of collections of a range of genotypes for purposes of propagation (nurseries) or further breeding (orchards).

Table 2. GreenWood Resources—Sun Grant Populus field trials by establishment year, study name, and location.

Year	Name	Location	Families	Clones
2009	SG Bioenergy Trial	Boardman, Oregon	NA	89
2010	Consolidated Clone Trial	Boardman, Oregon	NA	80
2011	Consolidated Clone Trial	Boardman, Oregon	NA	80
2011	Stage I Trial	Boardman, Oregon	46	2,275
2012	Stage I Trial	Boardman, Oregon	46	3,045
2012	Stage II Trial	Boardman, Oregon		
2013	Orchard	Boardman, Oregon	NA	568
2013	Stage I (*P. deltoides*) Trial	Boardman, Oregon	37	786
2013	Stage I (*P. nigra*) Trial	Boardman, Oregon	12	778
2014	Stage I (*P. tricho., P. nigra*)	Boardman, Oregon	28	558
2014	Stage II Trial	Boardman, Oregon	37	458
2015	Stage I (*P. deltoides*) Trial	Boardman, Oregon	35	753
2011	Stage III Trial	Boardman, Oregon	NA	7
2012	Stage III Trial	Boardman, Oregon	NA	16
2013	Stage III Trial	Boardman, Oregon	NA	11
2014	Stage III Trial	Boardman, Oregon	35	41
2015	Stage III Trial	Boardman, Oregon	26	54
2009	Sun Grant Bioenergy Trial	Westport, Oregon	NA	168
2010	Stage II Trial (*P. maximowiczii*)	Westport, Oregon	30	183
2010	Consolidated Clone Trial	Westport, Oregon	NA	38
2011	Consolidated Clone Trial	Westport, Oregon	NA	82
2007	LCTF Stage III Trial	Clatskanie, Oregon	5	9
2008	LCTF Stage III Trial	Clatskanie, Oregon	6	6
2009	LCTF Stage III Trial	Clatskanie, Oregon	8	9
2011	LCTF Stage III Trial	Clatskanie, Oregon	7	7
2012	LCTF Stage III Trial	Clatskanie, Oregon	7	7
2013	LCTF Stage III Trial	Clatskanie, Oregon	8	6
2014	LCTF Stage III Trial	Clatskanie, Oregon	17	19
2015	LCTF Stage III Trial	Clatskanie, Oregon	15	38
2014	2014 Clonal Screening Trial	Fitler, Mississippi	184	1,223

Objectives

The overall goal of the research is to improve growth and reduce production risk and costs of biomass produced in dedicated feedstock production systems. In the case of poplar, the need exists in all regions of the United States to develop a suite of genetically diverse clones with demonstrated high yield, disease resistance and growth stability on many sites within a region. Due to the wide array of genotypes available in each region through ongoing selection and breeding programs, a major emphasis of the research is oriented toward development of new clones through breeding and field testing of these new materials.

Methods

The research program concentrated on three main avenues of research including, 1) Field Clone Testing and Breeding, 2) Genetic Improvement and, 3) Yield Analysis. The methods used in conducting research in these areas differ significantly and will be discussed separately.

Field Clone Testing

Family Field Tests (FFTs)

Due to the fact that we have a continuing breeding program in Minnesota with all plant material under our control, we were presented with a unique opportunity to investigate the sources of genetic variance within populations of poplar using locally-derived plant material in a series of test referred to as Family Field Tests (FFTs). The purpose of the family-field-trial network is twofold; 1) to identify clones that should be propagated for more intensive yield testing on the path to commercial deployment and, 2) to understand the underlying genetic effects at the family- and clone-level. FFTs can be viewed as a very large clone test with a unique family and clone-within-family structure.

A debate has existed within the poplar community regarding the most efficient method that can be used to test clonal material as well as the underlying genetic mechanisms affecting growth rate and yield of an individual clone. The practical issue as it relates to the breeding program is that if the genetic system is dominated by non-additive effects with very little additive effects, then little justification exists for testing parental performance prior to using selected parents in breeding. In other words, if ultimate field performance depends entirely on the specific genetic combination residing in a specific clone, then parental makeup has little influence and all clones resulting from the breeding program must be maintained in order to evaluate the population to identify potential new commercial clones – all clones have an equal chance of being the next commercial clone. On the other hand, if additive effects are known to be in operation and of practical significance, then the contribution of the parents does indeed “carry over” to the next generation. Families and all full-sib members of that family share a commonality of a trait to some degree. In our case, the primary trait of interest is growth rate or yield. Answers to these questions are important to us due to the fact that a better understanding of the underlying genetic effects has direct bearing on the design of the breeding strategy and the expected rate of genetic improvement in yield that can be expected with each generation. These questions have practical importance to those directly involved in genetic improvement of poplars, those funding the research, and those potential users who are looking for a reliably suitable and affordable feedstock material. The implications of understanding genetic effects can help answer the questions: what is the magnitude of improvement in yield that can be expected with a sustained level of funding? In addition, what methods are to be employed to improve poplar yield and disease resistance in the most efficient manner possible?

The typical planting design for a FFT involves thirty controlled-pollinated families (i.e., known male and female parents) represented by up to thirty individual clones per family, for a total of 900 new clones. Replication of each individual clone ranged from three to five at each site. As mentioned, this experimental design facilitates analysis of sources of genetic variation at the following levels: among families; among full-sib individuals (within family); and within clone (cloned ramets). The FFTs were established on operational fiber farms (formerly Verso Paper Company) in central Minnesota and were measured annually through age five. Many trials remain in place and served as a source of breeding materials for second-generation crosses.

We established four field tests in 2001, 2002, 2007 and 2008, which were measured during the period of the Sun Grant program. The test design was randomized complete blocks with a single ramet of each genotype per replication with three to five replications at each site. Plant spacing was 3.0 x 3.0 meters. Each family was represented in at least two field tests. We tested 41 full-sib (full sibling, i.e., same parents) families of *P. deltoides x P. nigra*; 28 full-sib families of *P. deltoides x P. maximowiczii*, 40 full-sib families of various F_2 and first-generation backcross advanced-generation pedigrees, and 40 full-sib families of *P. deltoides x P. deltoides.*

In 2005, a large population of *Populus nigra* L. was obtained for evaluation in Minnesota prior to the initiation of the Partnership. Open-pollinated seed collections (i.e., seed collected from the same mother plant, each of which may have originated from a different, and unknown, pollen donor individual) were obtained from cooperators throughout Europe ranging from Italy in the south to Belgium in the north with an east to west range extending from France to Turkey. A progeny trial was established in 2006 and maintained for 3 years (2006-2008). We reared 38 families, 2,712 progeny, which were clonally replicated and established in field tests as described above for the controlled-pollinated families. All trees were measured for diameter at breast height (dbh) at age 3 (two years of initial seedling growth, 1 year of rising coppice) to the nearest 0.1 cm and converted to basal area (nearest 0.1 cm^2) (West, 2015). Overall, we have planted, maintained, and annually measured nearly 10,000 *P. nigra* trees at four test locations through age three years.

Using these data we evaluated statistical variance components attributable to among-families, among clones-within-families, within-clone and experimental error. We then used the appropriate published formulas (Berguson et al., 2017) of genetic expectation to estimate lower and upper bounds of the additive effect. Due to the unknown effect of higher-order interactions (epistatic effects), the estimate of the additive effect can only be bounded, not precisely calculated. To our knowledge, the manipulation of these formulas in this context is unique to the field of applied genetics and the availability of estimates of variance components in clonal populations of poplar allowed us to gain knowledge heretofore unavailable to the research community. Results of the FFT tests on multiple sites after five growing seasons are discussed in the Results and Outcomes section of this report.

Clone Tests

Clone tests are done for the purpose of identifying those genotypes having desirable attributes in the field over a multi-year period. Prior to the breeding effort supported by the Sun Grant program, the collection of clones available for field testing was limited. Clones used in early field tests were usually obtained from older collections from programs, many of which are now discontinued (e.g., USFS Stoneville, MS; Oxford Paper). However, in the case of the Sun Grant program, we were able to use a combination of pre-existing clones from other programs and those obtained through the current breeding program. Clone trials typically consist of a collection of 80 to 100 clones planted in single-tree replicates with three to six replications per site. Plant spacing varied from region to region but generally conformed to a spacing representative of a commercial plantation, usually between 1235 and 2224 trees per hectare. Data are collected on growth rate annually through measurement of tree height and diameter. Also, disease susceptibility is monitored throughout the five-year period.

GreenWood Resources - Quantification of clonal variation in plot biomass yield

In the Pacific Northwest, two clone trials were established in 2009 at GreenWood's Westport and Boardman research facilities. The purpose of the trials was to identify vigorous bio-energy selections under coppice management for coastal and continental sites. This trial is unique in that stands were coppiced from established stools and measured over a three-year period similar to what is commonly practiced in willow biomass production. One hundred and sixty-eight (168) clones were included in the coastal trial on the lower Columbia River floodplain (Westport, Oregon.) Eighty-nine (89) clones were tested at the continental trial in the mid-Columbia River basin (Boardman, Oregon.) Both trials were planted at an equivalent planting density of 3,605 trees-per-hectare, and established with hardwood cuttings. The experimental design utilized four randomized complete blocks within which the clones were set out in single-tree plots. The genetic material originated in GreenWood's proprietary breeding program. The trials were harvested following their first growing season (2009) and allowed to coppice for the next two seasons (2010 and 2011), before they were harvested the second time in the winter of 2011. They were then coppiced for three growing seasons (2012, 2013 and 2014) after which all sprouts 10 mm and larger in breast-height diameter were counted for each stool and individually measured for breast-height diameter. Stool basal area was then calculated. A subset of sprouts were weighed and stem dimensions measured from which yield equations were developed to estimate the yield of each clonal variety. Data were also taken on moisture content, leaf-to-stem weight ratio, and stem specific gravity. One of the clones identified in the 2009 Westport bioenergy trial, clone 6320, was also deployed to GreenWood's 2012 AFRI AHB coastal biomass yield trial at Jefferson, Oregon and Pilchuck Washington. At this location, it was established in 2.43 hectare monoclonal plots at a stocking of 3,605 stems-per-hectare. The plots were harvested following a two-year establishment cycle and regenerated by coppicing. The coppice stands were carried for a three-year cycle at which time they were inventoried and harvested. Clone 6320 grew at the rate of 15.8 and 13.8 DMT/ha/yr respectively during the coppice cycle at Jefferson and Pilchuck respectively.

Genetic Improvement - Breeding

It should be emphasized that without a breeding program using locally adapted parental material, there is no opportunity to continually increase yield or reduce production risk associated with growing poplar commercially once readily available and adapted genotypes have been screened for a region. When considering a national crop development program, variation in climatic adaptability dictates that genetic development programs consider a range of parent material originating from the region targeted for commercial production. Adaptation and change of disease organisms is fluid and constant and, as a result, it is imperative that genetic diversity be maintained. As will be discussed in the Results and Outcomes section, the considerable variation in growth and demonstrated potential to increase gains in growth and disease resistance argues for a continued breeding program with a national scope.

Breeding of poplar involves a multi-step process including planting of parental archives, maintenance of these archives to flowering age (typically year 7), collection of flowers, forcing of male flowers to produce pollen and preparation of female branches for breeding. Pollination is done in a greenhouse equipped with enclosures to isolate each cross to ensure the genetic purity of the cross. While successful production of viable seed occurs at a rate of roughly 40 to 50 percent, seed is collected from viable crosses and sown in containers for outplanting in nurseries. These seedlings are then grown for a period of three years with stoolbeds cut to produce coppice growth leading to the production of sufficient quantities of hardwood plant material for deployment into replicated field tests. The entire process from planting of archives to the production of cuttings for field testing can take 10 to 12 years. For this reason, genetic improvement is viewed as a long-term process which requires planning and an infrastructure of parental archives, greenhouse facilities and nursery acreage.

During the period of the Sun Grant Regional Feedstock Partnership program, we have conducted repeated breeding efforts capitalizing on the existing infrastructure and expertise in Minnesota and Oregon. Breeding in any given year typically involved 80 to 160 cross combinations in mating or cross breeding matrices. The female half of the cross is dominated by *P. deltoides* selected from local field tests while the male pollen sources are obtained from cooperators or local sources. The number of crosses done in any year is dictated by state of maturation and availability of flowers in parental archives. While our resources are not unlimited, the scope of the breeding program is very large compared to other programs worldwide and typically results in 5,000 to 10,000 new seedlings being produced in each breeding cycle.

Breeding in Minnesota relied on a combination of local sources of *P. nigra,* as well as pollen from collections maintained by GreenWood Resources in Oregon. In the case of *P. deltoides*, female branches were obtained from local sources in Minnesota for the Minnesota breeding effort, while archives in the southeastern U.S supplied flowers for breeding done by GreenWood Resources. As will be discussed further in the report, one of the goals of research supported by the Feedstock Partnership is to distribute unique collections of *P. nigra* maintained through the Sun Grant program for field testing in various environments across the United States. Based on a trial established in 2009 in Minnesota using a large collection from Europe, it appears that *P. nigra* has the potential to have a greater range of cross-regional adaptability than *P. deltoides* due to the fact that a high percentage of this collection have survived for several years under the relatively severe conditions of mid-continental winters of Minnesota. In light of this, we viewed the distribution of this *P. nigra* collection as an important component of a long-term national poplar improvement program. As noted in the Results and Outcomes section on clone screening in the Mid-South, development of a set of *P. nigra* parental stock adapted to that region is viewed as a critical part of future breeding for that region. Collections of *P. deltoides* are available from institutions such as ArborGen LLC (Dr. Mike Cunningham) and Mississippi State University (Dr. Randy Rousseau) but flowering collections of *P. nigra* with proven regional performance have been lacking and therefore effort was put into distribution of this species across the United States.

Once seed from successful capsules are sown, seedlings are maintained and overwintered for establishment of progeny trials in the nursery the following year. All seed lots are carefully tracked to ensure that all progeny can be accurately traced back to the specific cross. Individual seedlings are grown in nurseries to provide a sufficient supply of plant material for propagation and eventual planting in replicated field trails.

Yield Analysis

Previous work done to map poplar yield nationally such as that published recently by Wang, LeBauer and Dietze (2013) used a physiologically-based approach with the database of poplar yield constructed from studies published approximately a decade ago. The wide geographic range of test sites established by Sun Grant cooperators and new genetic composition of these tests provides the opportunity to revisit the topic of polar yield. The Sun Grant Poplar Team has undertaken the task of assembling a database of poplar yield from this network of research sites to facilitate the national mapping project. This database serves as the foundation for development of model estimates of poplar biomass yield nationally using indices of site productivity generated by the PRISM Climate Group at Oregon State University.

For purposes of this study, stand biomass is the total aboveground leafless biomass of all tree components including bole wood, bole bark, branches and top expressed on an oven-dry basis. The ultimate metric used in this study is the mean annual biomass increment expressed as the total stand biomass divided by stand age. In the case of poplar, the stand age includes all years since stand establishment with no age deduction for stand establishment. This distinction is important when comparing poplar yields to that of other perennial energy crops in which annual yields are typically reported during the period after the full-production phase has been reached. As a result, the early years of low production are not included in calculations of mean annual increment. This is particularly applicable to annually-harvested crops such as switchgrass in which yield during the period of steady-state production is usually the value of interest. For sake of clarity, we used all years, including the establishment period, in our calculations of mean annual increment in this report and in data provided to the PRISM yield mapping project.

Poplar-related activity funded by the Sun Grant Regional Feedstock Partnership began in 2009 with most of the new research sites being established in 2010 and 2011. Data from recently planted field tests are generally too young to be directly used in yield analyses.

A number of factors affect poplar yield and interpretation of data. A major factor influencing yield is plantation age and the interaction of plantation spacing and yield through time. Plantation spacing has a direct effect on early-rotation yields as canopy closure and maximum light interception, a dominant driver of productivity, is delayed with wider plant spacing. Once full canopy closure has been achieved, differences in periodic annual increment among stands of various spacing are minimal. In other words, maximum leaf area has been attained and no additional increase in canopy density is possible, all other factors being equal. However, the longer time period required for a widely-spaced plantation to attain full canopy development delays the point when the plantation is at full production. As a result, curves of plantation yield with time are dependent on stand spacing and time-of-measurement is a critical factor in measurement of poplar yield.

Yield Dataset Development Field Sites

The characteristics of the datasets used in the development of yield estimates varied by region. In those regions with a pre-existing research program in place, it was possible to continue measurement of established yield studies under the Sun Grant program and generate yield data from plantations either at maturity or nearing maturity. In the case of nearly-mature plantations with a history of several years of full-canopy growth, steady-state production, projections to maturity were done using annual incremental growth in the most recent years to estimate the stand conditions at maturity. In this instance, full-rotation yields are expected to be reasonably accurate as the projection period is relatively short and annual growth of biomass or total volume has been demonstrated to remain relatively constant in the years prior to maturity. This assumes the stand is harvested prior to incidence of disease or mortality due to extreme overcrowding. In other situations, early stand growth rate and incremental height growth was used to estimate final-rotation yield using a reference model developed for that purpose.

Yield data used in this project ranged from measurements in fully mature plantations with large, replicated, single-clone blocks to clone trials where data of height growth in single-tree plots is measured. In those cases where full-rotation yield data are available, yields were simply calculated at maturity with no need for additional extrapolation or adjustment. Obviously, this is the ideal case. In the case of clone trial data, height growth of top-performing clones was used to estimate mature plantation yield.

We did not have a comprehensive set of stands in all regions that were at full rotation using clones that could be considered commercially relevant. Many of the sites in Minnesota were at the point of 2/3 rotation age. As a result, final rotation yield was estimated assuming steady-state annual production from the point of current measurement to final harvest. In other cases, pre-existing stands at maturity had not been measured due to discontinuation of measurements. This was the case with the Alluvial Mid-South data where commercial plantations existed that were established under the Mead Westvaco program but were not continued to be measured. In that case, we made an effort to remeasure these sites to develop yield estimates for that region. The methodology used is described by region below.

Regional Poplar Yield Datasets

Sites in the poplar database are located in four major regions, the Southeastern United States (ArborGen), the alluvial South/Mid-South (USDA Forest Service, Mississippi State University), the Midwest (University of MN, Michigan State University) and the Pacific Northwest (GreenWood Resources). Sites included in the yield database for the Midwest are comprised of Sun Grant research sites measured by the University of Minnesota, Duluth and Michigan State University . The datasets used to develop yield estimates are discussed by region below.

Alluvial Southeast

When considering yield estimates for the Southern United States, we made the distinction between two physiographic zones; South/Mid-South alluvial sites and Southeast uplands. Alluvial sites are very different in their characteristics from upland sites in that they are concentrated along the Mississippi River and, as such, are not as geographically dispersed as uplands across the South and Southeast. Also, water demand by crops on alluvial sites is supplied in part by virtue of a high water table. This is not the case with upland sites where soil texture and inherent water holding capacity likely plays a greater role. The alluvial sites in the poplar yield database are located primarily along the lower Mississippi River Valley and include sites planted in commercial programs operated by MeadWestVaco in Kentucky and Illinois as well as a site in Mississippi. The Poplar Team reviewed available information on yields in all areas of the South and Mid-South. Apart from work done by the US Forest Service at Stoneville, Mississippi, very little long-term data had been published on poplar biomass productivity in the region. At the same time, commercial poplar plantations established through Mead Westvaco's program in Kentucky and Illinois presented an opportunity to measure existing stands that were at or near maturity.

Through the assistance of the current landowners, we located stands and established multiple measurement plots in a range of clone/site combinations. A total of 302 plots were established on 30 clone/site combinations. Of these, we selected 25 clone/site combinations to be included in the yield dataset with the final dataset being collapsed to eight sites. The average spacing in these stands is 667 trees per hectare or 14.96 square meters per tree with the average age of measurement being 8 years ranging from 5 to 11 years of age. In those cases where stands were less than age 9, growth was estimated to age 9.

Southeast Region Uplands - ArborGen Field Tests

The case of developing poplar yield data for upland sites in the Southeast presented a particular challenge. While other areas of the country have a history of commercial production, upland sites in the Southeast are dominated by loblolly pine production in the region due to production economics driven by attractive markets for sawlogs in the region and proven genetics and high yield. While there is a history of cottonwood selection and clone testing in the region, very little information is available on large-block yield tests using selected clones. We were not able to locate published studies that used clones known to be high-yielding in the region that had been planted on representative sites in sufficiently large yields blocks. Research done at the USFS Savanna River site was designed to evaluate the effect of irrigation and fertilization on a particularly drought-prone site (Kaczmarek et.al. 2013). Also, in communication with the authors, the effect of irrigation was not thought to be optimal in all years of the study. As a result, height growth of the best clones in this test was relatively low at 10 meters for the best clones after ten years. The lack of poplar production data in the region remains an issue, particularly data derived from proven high-yielding clones in more densely spaced plantings.

Due to the limited number of such stands in the Southeast and the importance of estimating yields on upland sites in the region, work was done to develop a reference yield model to estimate final-rotation yield from a set of geographically-dispersed clone trials using height measurements. The challenge was to use clone height data from the multi-site network of ArborGen clone trials and estimate yields expected at harvest. While mean tree diameter is known to be affected significantly by stand spacing, tree height is generally understood to be unaffected. This assumption forms the basis for the use of site index for estimating stand productivity. The intention was to use observed tree height at the current age from a subset of clones in these tests as an indicator of stand productivity and compare this measurement to the expected value generated from a reference model. The ratio of observed/expected tree height was then used to decrease or increase the estimated final-rotation mean annual biomass increment.

In order to build the most comprehensive and geographically-dispersed dataset possible for yield estimation, we sought to develop a reference model using data from published literature and from sites measured by the Sun Grant poplar team that included a wide range of stand densities measured over a sufficiently long time period to allow development of growth curves to final rotation age. Towards this end, we used datasets from the published literature and those supplied by GreenWood Resources that would allow us to put early rotation performance of clones in younger clone tests into a context of these long-term studies. Considering the fact that the available field study dataset consisted primarily of clone trial data at various tree spacings, an attempt was made to maximize the use of tree height data and estimate whole-stand parameters such as stand basal area and biomass from these data.

We were able to locate three datasets that had multiple observations of tree height, stand basal area and mean tree diameter across multiple ages on which to establish a reference poplar production model. The first dataset was derived from a paper published by Roger Krinard in 1985 describing results of a cottonwood growth study employing a Nelder-spoke design measured over a nineteen year period. This study was done near Stoneville, Mississippi using a single cottonwood clone (genetic makeup not described) on an alluvial site (Commerce silt loam). We used data from measurement years four, five, eight and nine with multiple spacings. This study included spacings of 3.53, 4.83, 11.33, 20.07, 35.77, 64.19 square meters per tree or 2805, 2224, 880, 497, 279, 156 trees per hectare, respectively. In total, twenty-four age-spacing combinations were included in this dataset. Multiple linear regression analysis was done to estimate total tree height, diameter and stand basal area at any age-spacing combination. Total stand basal area was then used to estimate stand volume and biomass. Our analysis included a model using stand age and spacing and the interaction of these two variables assuming that the effects of spacing may be curvilinear through time. In this way, the reduced effect of inter-tree competition at earlier ages is accounted for.

The second dataset used in the development of the reference model was provided by Rich Shuren and Brian Stanton at GreenWood Resources. This dataset was developed from an eight year study of multiple spacings of a single clone (Hybrid 11) on a site near Westport, Oregon in the lower Columbia River drainage in an area of high rainfall and was not irrigated. This study featured a wide array of spacing treatments measured over an eight year period with tree spacing ranging from 0.84 to 13.38 square meter per tree in a 5x5 factorial design of 0.91, 1.2, 1.8, 2.4, 3.7 within-row spacing times the same spacing arrangement between-row for a total of 25 treatments. Treatments were replicated three times with measurement periods from age two to age eight for a total number of observations of 575. The size, range of treatments and length of time over which this experiment was measured is unprecedented (Ritters et al., 1989).

Estimation of Production of Southeast Sites

The ArborGen dataset consists of clone trials on a variety of sites with ages ranging from three to nine with most sites falling near the younger end of the age range. In the exceptional case of the nine-year-old Floyd Tract site, we were able to use the mean height and diameter of all clones in the trial as a conservative estimate of closed-canopy, long-term yield.

Having constructed a set of reference production curves using the method described above, we estimated the expected mean annual increment of younger sites. We assumed age 7 as our rotation age for faster growing plantations and age 8 for slower growing stands for biomass-oriented plantations in the Southeast. The method is as follows; using the age and stand spacing of the site in question, we estimate the expected mean tree height at the current study age based on the average of the two methods described above. Results are then compared to the measured height at the current stand age based on the models and a ratio of the actual to expected height is calculated. Using this ratio, we adjust the estimated mean annual biomass increment at age 7 accordingly to arrive at an estimated expected annual increment for the site. For example, if the age-four height is 70% of the model, the estimated mean annual increment of the site is 70% of 13.0 Mg hectare^{-1} year^{-1} or 9.0 Mg hectare^{-1}. In simple terms, we are using the early-rotation height growth of the best clones in the clone trials as an indicator of site potential.

There were a total of seven sites managed and measured by ArborGen in the Southeastern upland region that are included in the yield database. Most of these sites are clone tests with the oldest trial, the Floyd Tract, being nine years of age. In this case, the actual height and total stand basal area of the entire study was used to estimate biomass production using the formula described above with a factor of 1.2 times the average accounting for selection of the best clones and elimination of holes due to poor survival of a portion of clones from planting. The Moultry, Eastover, Randolph and Wooten Farm sites are clone trials from which the mean of the top ten clones in height was used for comparison to expected height based on the mean of the GreenWood-1984 and Krinard-Nelder models.

A legitimate criticism of this methodology is that future growth patterns are unknown. Future growth may or may not continue at the current rate and growth curves may be affected by site. However, based on height growth patterns of the oldest site at Floyd, Georgia, height growth did continue and accelerated later in the rotation. The average height of the ten tallest clones at age four in the Floyd study was 6.71 meters compared to an expected height of 12.19 meters, 56% of expected height based on model estimates. However, the average height of the ten tallest clones was 17.7 and 21.6 meters at ages eight and nine, respectively, approximately 90% of expected height. The question of future growth patterns can only be answered by continued measurement of the existing Sun Grant network of trials and establishment of yield studies across the region that would accommodate measurements over a rotation. Continued measurement of these tests will improve estimates and reduce uncertainty associated with yield estimates.

Midwest - University of Minnesota and Michigan State University

The history of poplar development in the Midwest includes work done since the mid-1980s by the US Forest Service at Rhinelander, Wisconsin, the University of Minnesota and Michigan State University in plantation management, clone testing and yield evaluation. The Midwest poplar yield database is comprised of Sun Grant research sites measured by the University of Minnesota-Duluth and Michigan State University. A limited number of clones have the complete set of important commercial traits such as high growth rate, disease resistance and a high rooting rate from hardwood cuttings. Those clones exhibiting these traits were selected from the trial network for longer-term observation in closed-canopy, replicated, pure-clone blocks to evaluate biomass yield under conditions resembling commercial production acreage. Data collected from this network of longer-term yield tests was included the yield database.

The University of Minnesota dataset includes a number of clones planted in replicated yield blocks, typically in a 7-tree by 7-tree configuration, with the outermost two rows serving as buffer rows to eliminate edge effect in yield measurements. In most cases, data have been collected annually on these sites and growth curves over the complete rotation are described. In those cases where the stand has not reached full rotation age of age ten, we estimated the final yield based on the annual steady-state growth rate after full canopy closure has occurred and extended that growth rate to full rotation. The median period of extrapolation of growth to full rotation is three years with a total of 17 clone/site combinations. In many cases there are multiple clonal yield blocks at each site and data were ultimately consolidated to produce a single yield value for each site for use in the OSU-PRISM yield mapping effort by calculating the mean yield value of the most productive clones at each site.

Hybrid poplar trials have been underway at Michigan State University for decades. Until recently, these trials have been conducted using older clones developed by US Forest Service, and regional university breeding programs. Trials of newer clones from the University of Minnesota breeding program began in 2008 in Michigan with yield trials of selected clones established between 2010 and 2012. However, these trials are too young to produce reliable yield data for these models. Yield data from Michigan is limited to five older trials comprised of clones that pre-dated the regional breeding program.

All Michigan yield trials are composed of replicated 64-tree plots. Measurements were taken from the inner 16 trees, leaving a 2-tree border around the measurement plot. The first of these was a replicated trial comparing five poplar varieties harvested in 2008 after 11 growing seasons. Biomass growth was estimated in years 4 – 10 and actually measured in year 11 at final harvest. A second poplar trial reported here is a spacing/clonal yield trial containing 7 clones planted at 3 spacings (1920, 2241, and 2691 trees per hectare). Biomass yields in this trial were estimated after 5 growing seasons. Annual biomass increment estimates are also provided from three additional poplar yield trials containing between 9 and 14 clones. These trials are located at three locations in Michigan's Upper Peninsula. Biomass estimation of standing trees in Michigan trials was done using an allometric equation developed by Wang and MacFarlane (2012).

Pacific Northwest

GreenWood's commercial hybrid poplar plantations are situated in two distinct physiographic regions of Oregon and Washington, the lower Columbia River floodplain and the mid-Columbia River Basin. Drainage is the prominent plantation management feature. F_1 varieties of the *P. ×generosa* (*P. deltoides* × *P. trichocarpa* and reciprocal) and *P. deltoides* × *P. maximowiczii* taxa are used in lower-Columbia operations. In contrast, the annual rainfall on the eastside of the Cascades is 200 millimeters with these plantations being irrigated. Following preliminary clone screening trials, GreenWood Resources conducts yield testing of selected varieties within each deployment region. There are 60 varieties currently in yield trials established in operations along the lower Columbia floodplain over the period 2007-2012, and 44 varieties being followed in yield trials established in the mid-Columbia River basin 2010 through 2013. Sixty-seven (67) varieties were used in planting the mild/mesic trials at Westport, Oregon, Puyallup, Washington, and Newberg, Oregon in 2005 and at Acme, Washington in 2006. After reviewing of the potential dataset for the Pacific Northwest, the decision was made to eliminate those sites that were under irrigation and restrict the estimation of poplar yield exclusively to unirrigated sites receiving sufficient natural rainfall to support poplar production. As a result, estimates of poplar yield in the Pacific Northwest are only done in those areas that could support unirrigated production.

Biomass Estimation

Due to the rarity and value of existing yield studies, it is impossible to destructively sample all clone/site combinations and develop clone-specific equations. In light of this issue, we decided to use an equation based on the average taper and density of poplar to estimate the mean annual incremental biomass production over time. The formula used is:

[Eqn. 1] Total Stand Basal Area * Tree Height * 0.4085 * 337 * 1.2 = total dry weight per hectare in kilograms and stand basal area in ^{m2} per hectare and height in meters

The above equation uses the standard formula of the product of tree basal area and height (a cylinder) reduced by a taper factor (0.4085) to estimate total stand volume outside bark. The total outside-bark tree volume is then multiplied times the assumed average poplar specific gravity of 0.337 (337 kg per cubic meter). This sum is then multiplied times 1.2 to add 20% additional top- and limb-biomass, a generally agreed value found in studies of biomass components within the Sun Grant poplar team (B. Berguson, unpublished, personal communication).

We tested our method of volume estimation against an equation developed from cottonwood trees published by Krinard (1988). Estimating tree volume in trees between 8.9 and 25.4 centimeters DBH, the mean difference between Equation 1 and that predicted from Krinard is less the one percent. We chose to use equation 6 to estimate the total tree volume with the tree volume then multiplied by the average combined wood and bark density plus a 20% factor to account for additional top and limb biomass.

Results and Outcomes

Clone Testing

<u>*Cooperative Clone Tests*</u>

Cooperative Clone Tests were one avenue of clone testing pursued by the Poplar Team. These tests were planted at various locations in 2009 and 2010. Because research partners had access to or owned unique collections of poplar that warranted further testing, we were in a unique position to begin the process of interregional exchanges of clones with selections from four distinct collections. Clones native to the South and Mid-South were supplied by ArborGen and Mississippi State, those of more westerly origin by GreenWood Resources and northern sources were supplied by the University of Minnesota. Twenty clones from each of four collections for a total of 80 clones were planted at four locations to evaluate clone growth rate and adaptability across a wide geographic range.

Analyses of the four Cooperative Clone Tests show that species composition and source of material are significant factors influencing clone growth and disease susceptibility in all regions. As expected, clones of northern origin planted at southerly locations, while surviving, showed reduced growth compared to those clones derived from collections native to the respective region. Figure 3 shows the distribution of volume growth, which is linearly related to tree biomass, at the New Madrid, Missouri Cooperative Clone Test after three years. As shown in this graph, clones of more southern origin are clearly more productive while those derived from northern locations (Minnesota) are significantly less productive. The average ratio of tree volume index to the test mean among the four sources were 1.5, 1.39, 0.66 and 0.44 for clones derived from ArborGen, Mississippi State, GreenWood Resources and the University of Minnesota, respectively. Obviously, clones contributed by ArborGen and Mississippi State that are native to the region and have undergone some degree of prior selection are far superior to non-native sources. This test demonstrates the effect of photoperiod response inherent to northerly-derived material whereby trees from this region cease active growth too early in southern environments. Conversely, clones of southern origin did not survive the cold winters of Minnesota. However, statistically significant correlations between clone ranks in the Consolidated Clones Tests in Minnesota and Oregon indicate that clone exchanges and testing of material between these regions may have merit. Overall, performance among clones in this test is quite variable with volume and biomass of the ten fastest-growing clones ranging from 1.3 to 1.5 times the test mean.

As suspected prior to planting these tests, our results demonstrate the need for region-specific breeding using locally-tested sources as well as a network of clone tests done in the region of commercial interest. Importation of clones from vastly differing environments is not recommended and likely an unproductive route to produce fast-growing, robust plant material for a given region.

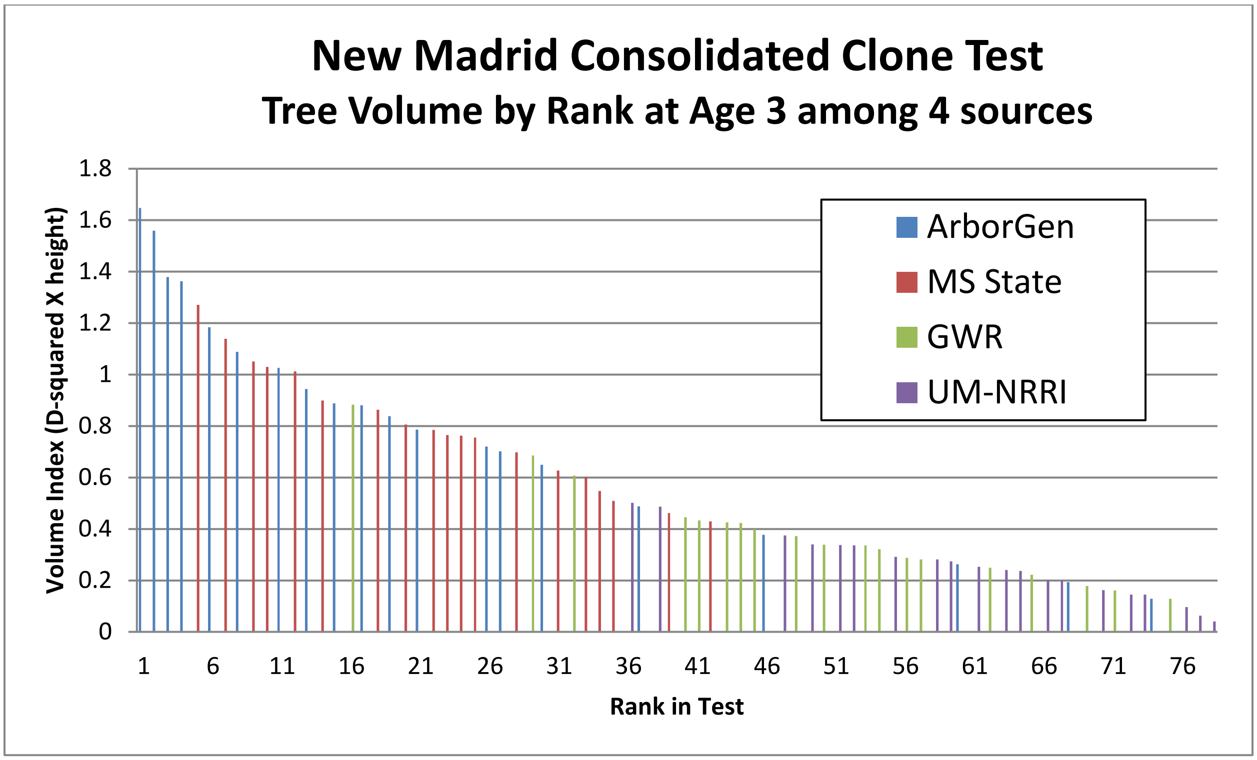

Figure 3. Distribution of volume index by clone origin at the New Madrid test site after three years.

Disease resistance in poplars is extremely important and clone selection must be done carefully to ensure proper selection. The incidence of stem canker was measured in all of the tests but the direct comparison is best made with the 2010 test sites. Analysis of branch and stem canker prevalence shows that, when planted in humid southerly locations, some hybrid clones containing *P. trichocarpa* may exhibit increased susceptibility to *Septoria* canker.

The age-four incidence of stem canker by taxa on the 2010 alluvial site in Missouri showed that the eastern cottonwood (DxD) exhibited no stem canker, while all of the hybrid taxa exhibited various level of stem canker. This ranged from a low of 21.4% for the DT (deltoides x trichocarpa) taxon to a high of 100% for the DM (deltoides x maximowiczii) taxon. Although, all of the hybrid taxa exhibited incidence of stem canker, there was some variability among clones within taxa. At age-four the presence of stem canker did not result in mortality at that time. Ten of the 24 DN (deltoides x nigra) hybrid poplar clones (41.7%) exhibited no stem canker. Only one other hybrid clone, 4491, a DT taxon was also free of stem canker. In most cases the majority of the hybrid clones demonstrated poor growth as compared to pure eastern cottonwood at the alluvial site. Field observations suggest that trees start developing cankers as early as the second growing season. While there are exceptions, as a general rule it appears that the native *P. deltoides,* and possibly *P. nigra,* may be the species of primary interest in the southern regions of the United States. When examining the 2010 upland site, the incidence of stem infection of the hybrid poplars is lower than seen on the alluvial site ranging from a low of 31.2% for the DN taxon to a high of 91.2% for the DM taxon. Surprisingly, the DT and DN taxa showed lower percentage of disease detected at the alluvial site rather than the upland site. The top performing clone (8019) on the upland site was one of the DM taxon clones. The results showed survival at age-one in eastern cottonwood are related to reduced rooting while survival of the hybrids through age-four is related to lack of adaptability or susceptibility native diseases. These results point to the potential of DN hybrids in the region if selections of European black poplars show suitability to southern US sites.

This information has helped shape the field testing program and provided the impetus to accelerate testing of pure-species *P. nigra* in the alluvial South and Southeast. Results of these tests have helped identify those clones to be included in further yield tests in the respective regions, which aim to answer questions related to yield potential in each region using superior genetic material. Table 3 below shows the taxa, number of clones per taxa, percent stem infection and clones that showed resistance at age four of the 2010 Consolidated Populus Trial, with test sites located on a Mississippi River alluvial soil and an upland Gulf Coastal soil.

Table 3. Percent stem infection and number of canker-resistant clones at two sites in the Mid-South.

		Percent Stem Infection		No. of Resistant Clones	
Taxa[1]	Number of Clones	Alluvial (%)	Upland (%)	Alluvial	Upland
DD	36	0	0	36	36
TD	8	75 - 100	17 - 100	0	0
DT	3	0 - 50	0 - 100	1	1
DM	6	100	67 - 100	0	0
DN	25	0 - 100	0 - 83	10	5

Regional Clone Tests

In addition to the Cooperative Clone Tests, a number of tests were planted that contained only clones that were derived from local sources. These tests typically included a greater number of regionally-derived clones in a replicated design. These tests were planted at sites in the Southeast, Midwest, and the Pacific Northwest. These tests typically included clones that had not undergone extensive testing in the region previously. Pooled results of tests in the Southeast showed a gain of up to 35% in tree volume and biomass relative to the standard clone S7C8. In Minnesota and Michigan, clone tests typically showed that mean biomass growth of the ten best clones in an eighty-clone test exceeded that of the regional commercial standard, NM6, by an average of 1.5 times (i.e. 50%). Across all regions, clone tests demonstrated that testing of new genotypes has significant potential to increase growth rate and genetic diversity of poplar for commercial planting. Results of these trials have identified the subset of clones suitable for more extensive clone and yield testing in the respective regions.

Clone Testing in Minnesota – University of Minnesota

A particularly vexing problem identified in our work in Minnesota and in the Midwest generally is the lack of site-to-site stability in clone performance. Stability in growth across environments is often referred to as *plasticity*. Stable performance across sites is a critical attribute of new clonal material as a commercial program will ultimately include a large amount of acreage with a wide variety of site types included in the land base. As noted in previous clone tests done across the region, the lack of stability in growth performance is an unavoidable fact in conducting this research and has a direct effect on clone recommendations. A very fast-growing clone at one site exhibiting high site-to-site variability would not be as desirable as a stable clone with a slightly reduced growth rate due to increased risk of poor growth on some sites associated with planting an unstable clone. In statistical parlance, the presence or absence of stable performance across sites is evaluated by the inclusion of a clone-by-site interaction term in a statistical model. This clone-by-site interaction is identical to the more generalized genotype-by-environment interaction often cited in similar studies. These analyses will be discussed further in this section.

In light of the observed potential for significant genotype-by-environment interaction and the orientation to develop new commercially useful clones, a focus of our program has been establishment of clone trials containing an identical set of clones at multiple sites. Beginning in 2006, we began the process to propagate material to allow establishment of clone tests at multiple sites across Minnesota (Figure 4). This effort involved propagation of sufficient material to allow six clone tests to be planted at separate sites with six replications of each clone contained in each test. In 2008, multiple tests of identical material were planted with the total number of trees in these tests being 2,160 (60 clones 6 replications/site X 6 sites). From previous stages of testing at the FFT level (under NRRI's testing program), Clone Trials are made up of the next round of "promising clones" for future testing – thus, based on Age 5 data and on a clone-mean basis, candidate clones are identified and moved to the Clone Trial phase of testing. This study provides data to evaluate correlations among and within sites through time, clone growth rates and stability of growth of individual clones across sites and years.

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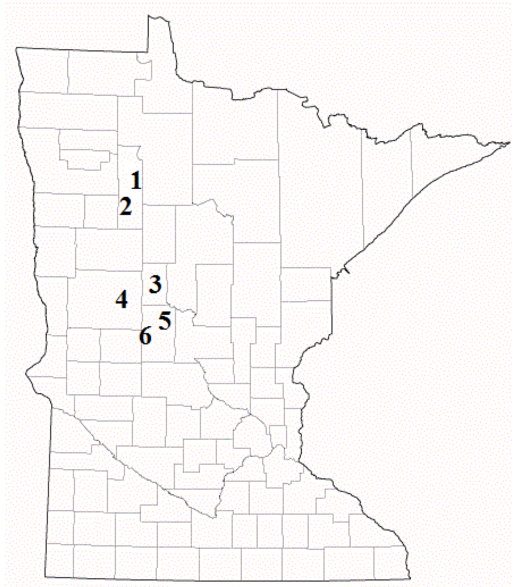

Figure 4. Location of clone tests in Riewer (1), Crabtree (2), Wheeler (3), Koljonen (4), Olander (5), and Wandershceid (6), Minnesota.

Figure 5 shows the distribution of tree growth within these tests after five growing seasons in the field sorted from largest mean tree size to smallest. In this case, individual tree basal area, or cross-sectional area, is the metric of interest as tree basal area is linearly related to total tree volume and weight. As shown, average tree growth differs among sites with the Wheeler site located in central Minnesota significantly higher in overall growth than the other sites. Average growth across all sites is 69.7 square centimeters per tree with the four sites, Crabtree, Kiljonen, Olander and Wanderscheid within ten percent of the grand mean across all sites. However, tree growth at the Wheeler site was found to be significantly higher than the grand mean of the other sites with the average tree basal area being 1.35 greater than the grand mean including all clones at all sites. Conversely, the Riewer site, located in northern Minnesota near Bagley, Minnesota was 0.74 of the grand mean. As can be seen in Figure 3, the variation in growth of clones within sites is significant. The mean of the top 10 clones compared to the average for each site is 1.35, 1.38, 1.48, 1.5, 1.51 and 1.45 at the Crabtree, Koljonen, Olander, Riewer, Wanderscheid and Wheeler sites, respectively. The mean of the bottom 10 clones compared to the average for each site is 0.41, 0.33, 0.37, 0.36, 0.31 and 0.22 at the Crabtree, Koljonen, Olander, Riewer, Wanderscheid and Wheeler sites, respectively.

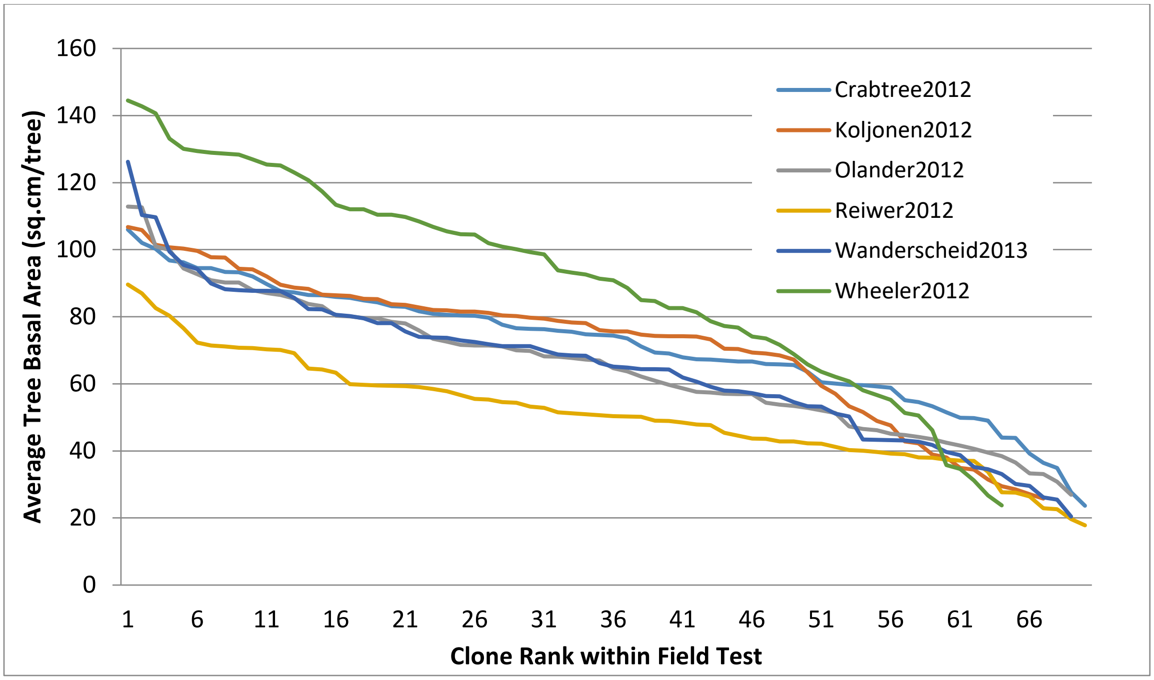

Figure 5. Distribution of individual tree basal area at age five in six clone tests established in 2008 at sites in Minnesota.

Statistical analyses were done using a subset of clones in this study. In order to generate values to estimate site-by-clone interactions, a complete set of all clones at all sites is required. After reviewing the data, some clones did not survive at all sites which preclude analysis of site-by-clone interaction effects using all clones. After eliminating those clones that did not appear at all sites, 55 of the original 70 clones remained in the dataset. Analysis of variance components showed that all sources of variation, site, clone and site-by-clone interaction are statistically significant (p-value<0.05) with variance components being 21, 22, 16 and 41 percent for site, clone, site-by-clone interaction and error, respectively. A review of the data shows that few clones exhibit a high degree of site-to-site stability. The greatest site-to-site stability occurs at the bottom of the distribution where maladapted clones perform consistently poorly at all sites. This underscores the need for a concerted breeding effort to generate clones that have both traits of high growth rate and site-to-site stability and further highlights the rarity of those clones that possess both traits. In light of the need for stability and high growth rate, a suggested direction of future research is to test parental sources (pure *P. deltoides* and *P. nigra* collections) for stability by establishing similar multi-site trials of this material prior to their use in a breeding program. While unproven at this time, it may be possible to specifically develop progeny that have these traits occurring simultaneously through the selection of parental stock exhibiting these traits. Due to past investment in development of pure-species collections, we have the necessary plant material that would allow us to establish field tests to further explore this issue.

Correlation analysis of the clone ranks at each site was done to gain insight into the need to conduct multiple-site clone tests and the transferability of data from one site to another. Table 4 shows the correlation coefficients for clone ranks in 2012 (five growing seasons) among all six sites in the 2008 trial network. Although not shown, the p-values of all correlation coefficients are less than 0.01 in all cases. It should be noted that the correlation coefficients are simple correlation coefficients and the square of these values is the percent of variation explained using ranks at one site to predict performance at another site. Overall, rank-order correlations are relatively low and do not provide a great deal of insight regarding the number of clone tests that should be planted in a given region. It appears that two or three sites could be planted to identify poorly adapted material with those clones selected from the upper part of the growth distribution marked for expanded testing on a greater number of sites.

Using the clone rank data across all sites, we calculated the coefficient of variation among clone ranks across all sites by clone. In this case, the coefficient of variation is the standard deviation of clone rank as a percentage of the mean clone rank including all six sites. This analysis shows that the average coefficient of variation among the top 50th percentile is 0.65 versus that of the bottom 50th percentile at 0.30. In other words, those clones occupying the bottom of the distribution tend to be near the bottom at all sites (consistently poor) while the variation in rank at the top of the distribution is greater. The practical implication of this result is that we can feel confident that eliminating poorly-performing material based on a limited number of sites can be done with little expectation of losing a potentially high-performing clone. However, in the case of clones in the upper half of the distribution with respect to growth, all clones should be retained for further multi-site clone testing due to the potential for significant changes in rank across sites. Based on our results, we recommend that a clone testing program be done by conducting clone trials on a limited number of sites and the bottom of the distribution be culled with only the top 50th percentile retained for further testing; however logistics (e.g., land availability) may limit selection to an even greater extent.

Table 4. Correlation coefficients of clone ranks among pairs of sites in 2008 trials after 5 years. See Figure 3 for a map of these locations.

	Crabtree	Koljonen	Olander	Riewer	Wanderscheid	Wheeler
Crabtree						
Koljonen	0.408					
Olander	0.392	0.565				
Riewer	0.678	0.305	0.454			
Wanderscheid	0.563	0.606	0.616	0.357		
Wheeler	0.537	0.561	0.619	0.439	0.588	

A useful extension of the above discussion is the issue of age-age correlations and the amount of time that is necessary before clone selection can be done. The practical implication of this from a program management and financial standpoint is that the older that woody crop trials get, the more difficult and costly they are to remove and restore the land for a future use. If the incremental value of additional annual growth data is shown to decrease significantly over time, it makes sense to run these tests only to the earliest possible point when a decision can be made regarding selection of a subset that warrants more intensive testing on multiple sites. We evaluated correlations among ages three, four and five years on the six-site, 60-clone test network. R-squared values are 0.486 and 0.85 for relationships between the clone growth ranks between ages 3-to-5 and ages 4-to-5, respectively, with the p-values for these correlations highly significant at less than 0.01. Based on this analysis, there is enough rank change between ages three and five that warrant continuation of clone trials to at least age four in our region.

Taking this information together, the recommended strategy is to conduct field trials of the same set of clones at a minimum of three sites in any given year and continue these tests to age four. We have learned that one site is not enough, two sites may be worse for interpretation if one has high yields and the other has low yields; therefore, to do the stability analyses, a minimum of three sites improves evaluation of regression slopes of the set of clones over three sites. At this point, the bottom half of the distribution could be eliminated and additional clone tests could be planted at a greater number of sites with a reduced number of clones. This procedure appears to be the most efficient process to retain potentially useful genotypes while still maintaining a sufficient number of clones to ensure that the majority of elite genotypes are retained. In the case of elite clones that appear to exhibit consistently high rank in the first set of tests, nursery propagation could begin to prepare for testing in yield blocks.

Clone Testing – Michigan

Disease evaluation in clone trials in Michigan was done using two poplar hybrid clone trials at the Escanaba, Michigan test location. Measurements of DBH, height, and score of *Septoria musiva* infection (causes leaf spot and stem canker in poplars) were made in the winter of 2014-15. Analysis shows a high proportion of the variation in DBH, height, and disease susceptibility is attributable to the hybrid cross and therefore heritable. *Septoria* incidence is highly heritable in the 2008 test but not in the 2009 test. This may be due to real differences between the set of clones in each test, as the 2009 test has more infection, which may mask differences among hybrids. It is possible that the general prevalence of disease in the 2009 tests affects moderately-resistant clones due to higher overall levels of ambient inoculum in the study.

Table 5. Analysis of variation among clones and replications in two hybrid poplar trials in Escnaba, MI. Broad sense heritability estimates are unitless, thus these data represent the percentage of total observed variation attributed to replication, clone, and error. DBH=diameter at breast height.

2008, 56–hybrid Clonal Trial			
	Broad Sense Heritability (H^2)*		
	DBH	Height	Cankering**
Replication	0.03	0.1	0.01
Clone	0.48	0.32	0.68
Unexplained	0.49	0.57	0.32
Plantation Mean	2.9"	30.1'	1.9*
2009, 70-hybrid Clonal Trial			
	DBH	Height	Cankering
Replication	NS	NS	NS
Clone	0.3	0.29	0.04
Unexplained	0.7	0.71	0.96
Plantation Mean	2.7"	26.5'	2.4*
* DBH: the degree to which a trait is genetically determined, expressed as the ratio of the total genetic variance to the phenotypic variance. ** Note: *Septoria musiva* cankering score from 1 = no infection to 5 = dead from cankering. NS = not significant			

Pacific-Northwest - GreenWood Resources - Quantification of clonal variation in biomass yield

The average cumulative clone dry weights for the entire trials at Westport, OR (not irrigated) and Boardman, OR (irrigated) following three years of coppice production are equivalent to area yields of 43.9 and 50.4 dry tonnes per hectare. These convert to mean annual increments of 14.6 and 16.8 dry tonnes per-hectare per-year and are representative of the growth of a polyclonal stand. Thus, even a polyclonal mixed stand of poplar in this region could be expected to yield in the range of 13-16 DMT ha^{-1} yr^{-1}. Monoculture plots of irrigated switchgrass yielded 15 DMT ha^{-1} yr^{-1} over a three-year period at Boardman (Kimua et al. 2018).

The monoclonal block of clone 6320 was established at GreenWood's 2012 AFRI AHB, funded by USDA NIFA, coastal biomass yield trial at Jefferson, Oregon (not irrigated). Clone 6320 was the topmost clone of 11 in the Jefferson trial, producing 47 DMT ha^{-1} after three years of coppice growth. In terms of tree basal area, at the 2009 Westport bioenergy trial, clone 6320 produced 95 cm^2/stool of basal area. It is noteworthy that the top 10 clones in the Westport trial exceeded the basal area of clone 6320 basal area by an average of 36% (i.e. 129 cm^2 versus 95 cm^2). Similar to clone trials in other regions, our results indicate the significant potential to select highly productive clones to increase the mean annual increment in biomass production in monoclonal stands under operational conditions.

Wood Characteristics

In addition to evaluating growth rate, variation in wood characteristics such as specific gravity and chemical composition were also studied. Work done by ArborGen on a collection of 26 clones showed little variation between pure *P. deltoides* and hybrids with a mean specific gravity of 0.355 (355 kg ha^{-1}). Also, research showed significant variation in the average wood moisture content at harvest among a selection of clones (data not shown). University of Tennessee research on hybrids grown in Minnesota clone tests helped to quantify chemical constituents of hybrid poplar (data not shown). This information is valuable in yield analyses as well as to inform potential conversion technologies regarding issues related to process suitability and ultimate fuel product yield.

Genetic Improvement Research

Genetic improvement research involves several phases of research. These include: (1) clone testing of potential pure-species parental stock typically from wild populations, (2) inter- and intra-specific breeding of selected parents to produce the next generation of improved genetic material, and (3) field testing of progeny resulting from the breeding program to understand fundamental genetic mechanisms and identify the next set of promising clones for inclusion in a new round of genetic crosses and field clone tests. We make the distinction between (a) clone tests of pure-species collections with the primary aim of identifying new parents for breeding and (b) clone tests of a subset of hybrids and pure-species clones with the near-term goal of identifying new material for commercial development.

Breeding

Breeding has been ongoing throughout the duration of the Sun Grant program at locations in Oregon (GWR) and Minnesota (U MN). The legacy of refined parental populations and expertise available at the start of this project allowed us to begin breeding under the Sun Grant program. Together, the two programs have produced a large collection of clones that will serve as the source of new genetic material for future testing in clone trials and yield blocks. The

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history of poplar breeding in Minnesota by taxon is shown in Table 6 below. These materials are planted in nurseries in Minnesota, Mississippi, and Oregon and are ultimately propagated for field tests. Also, in Minnesota, populations resulting from the breeding program are planted in Family Field Trials containing a large population of genotypes with a threefold aim: (1) to increase biomass growth and disease resistance in the next generation, (2) to enhance genetic diversity and reduce commercial risk, and (3) to provide insight into the underlying genetic mechanisms operating within these populations, to allow for optimal design of the breeding program so that we can accelerate future progress in genetic improvement.

Table 6. NRRI breeding activity (i.e., breeding attempts by taxon group or species type over time) during the period 1996 through 2016 by cross type.

CP Cross	YEAR					
Taxon type[1]	1996 - 2000	2001 - 2005	2006 - 2011	2012	2016	Totals
D x D	90	41	478	77	121	807
D x M	69	19	4			92
D x N	62	72	628	236	40	1,038
D x T	9	64	37			110
N x T		10				10
N x N		4			120	124
N x D		5			20	25
N x M		6	22			28
N x M	*GreenWood Resources Breeding*		*20*			*20*
DM x (D)[2]	0	1				1
DM x (DM)	8					8
DM x (M)	2					2
DM x (N)	1					1
DN x (D)	0	0				0
DN x (DM)	3					3
DN x (DN)	2					2
DN x (M)	3					3
DN x (N)	5					5
DN x (T)	1					1
D x (DM)	47	13				60
D x (DN)	5	10				15
D x (TD)		43				43
TD x (D)		53	1			54
NM x (D)	1					1
NM x (DN)	2					2
NM x (N)	2		5			7
NM x (T)	1					1
Totals	313	341	1,175	313	301	2,443

[1] D: *Populus deltoides;* M: *Populus maximowiczii*; N: *Populus nigra*; T: *Populus trichocarpa*; DM: *P. deltoides x P. maximowiczii* hybrid; DN: *P. deltoides x P. nigra* hybrid; NM: *P. nigra x P. maximowiczii* hybrid; TD: *P. trichocarpa x P. deltoides* hybrid

[2] Parentheses indicate male parent.

GreenWood Resources – Hybridization and varietal selection

GreenWood Resources conducted an intensive poplar hybridization and varietal selection project over the period 2008-2016 that was partially supported by the Sun Grant Partnership program. The intent was to develop energy varieties for the renewable biomass market focusing mainly on the first-generation of the *P.×generosa* (aka *P. deltoides × P. trichocarpa*) and the *P. deltoides × P. maximowiczii* taxa for coastal regions and the *P. ×canadensis* inter-specific taxon for continental regions. Improvement of the hybridizing value of the key parental species ‒*P. deltoides, P. trichocarpa, P. nigra, P. maximowiczii, P. fremontii, P. simonii* ‒was also pursued. Finally, a program in the southeast focused principally on intra-specific *P. deltoides* breeding. GreenWood Resources is now moving the top tier germplasm from the three project regions into conservation gardens at its Westport Tree Improvement Center in view of diminished prospects for near-term company investment opportunities into commercial biofuel assets, a reflection of low of crude oil prices. The conservation gardens will be maintained in a state of readiness for hybridization and deployment to commercial biomass plantations.

A total of 663 genotypes will be conserved from the coastal program in breeding orchards and clone banks, the latter managed by annual coppice as detailed in Table 7. Also, 1,385 genotypes from GreenWood's continental hybridization program will be relocated from its Boardman test site to Westport (Table 8). Finally, a total of 1,232 genotypes from the southeastern *P. deltoides* breeding program at Fitler, Mississippi will be repatriated to Westport (Table 9.) When completed, the Westport Tree Improvement Center will host the following conservation gardens.

- Breeding Orchards:
    - *P. maximowiczii*
    - *P. trichocarpa*
- Clone Bank I: coastal material
    - *P. maximowiczii*
    - *P. trichocarpa*
    - *P. ×generosa, P. deltoides × P. maximowiczii* and *P. trichocarpa × P. maximowiczii*
- Clone Bank II: continental material
    - *P. ×canadensis, P. deltoides, P. fremontii, P. nigra, P. simonii.*
- Clone Bank III: southeastern material
    - *P. deltoides* 1^{st}, 2^{nd}, 3^{rd} generation selections from southern provenances.

Site preparation for the new Westport gardens and orchards was completed in September 2016. Data collection at Westport, Boardman and Fitler was finished in December 2016. Data analyses and identification of conserved material were finalized in January 2017. Propagation material was collected during February–March 2017, along with tissue collections for DNA identification for archival purposes. All clone bank plantings and orchard conversions were completed by mid-April 2017.

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Table 7. Inventory of *Populus* genotype selections being conserved from coastal testing program.

Class	Current Inventory of Genotypes	Genotypes to be Conserved	Selection Intensity
P. trichocarpa orchard	317	150	0.47[2]
P. maximowiczii orchards	417	300	0.72[3]
Hybrid clone bank[1]	108	108	1.00
P. trichocarpa clone bank	317	317	1.00
P. maximowiczii clone bank	417	417	1.00
P. trichocarpa × *P. maximowiczii* clone bank	97	97	1.00
Commercial poplar propagation beds	8	8	1.00

1/ Predominantly *P.* ×*generosa* and *P. deltoides* × *P. maximowiczii*

2/ Selection intensity lower as this is the last culling in a multiple stage evaluation where genomic markers are used in selection (Brian Stanton, personal communication).

3/ The *maximowiczii* collection is quite expansive (China, Korea, Japan) and the desire is to conserve as much diversity as possible to enable breeding for multiple regions (Brian Stanton, personal communication.

Table 8. Inventory of *Populus* genotype selections being conserved from continental testing program.

Class	Number of Trials or Plantings	Current Inventory of Genotypes	Genotypes to be Conserved	Selection Intensity[1]
Orchard Genotypes[2]	1	568	535	0.94
Selected Hybrids[3]	2	102	102	1.00
Hybrid Test Populations	3	2,070	525	0.24
Parental Species Test Populations	8	4,797	223	0.07

1/ Selection intensity based on calculation of the top clones that were determined to move to next stage of testing (Brian Stanton, personal communication).

2/ *P. deltoides*, *P. nigra*, *P. deltoides var. wislizeni*, *P. fremontii*

3/ Predominately *P.* ×*canadensis*

Table 9. Inventory of *P. deltoides* selections being conserved from southeastern testing program.

Class	Current Inventory of Genotypes	Genotypes to be Conserved	Selection Intensity
2014 $2^{nd}/3^{rd}$ gen clones	1,248	125	0.10
2016 $1^{st}/2^{nd}/3^{rd}$ gen clones	890	301	0.34
Culled/untested 2^{nd} gen genotypes	348	0	0
Southwest *P. deltoides* Provenance collection	1,600	800	0.50

Family Field Tests

Berguson et al. (2017) published a paper describing the details of analysis and implications of the Family Field Tests. The following is a brief summary of the salient results. Variance components including the genetic components of among-family variance and clones-within-family variance among the FFT experiments were estimated using a random-effects statistical model by taxon group. We found that both genetic variance components were statistically significant and greater than zero. As shown in Figure 6, while absolute values vary depending on tree size and site, the proportion of family variance versus clone-within-family variance is remarkably consistent among the three full-sib families comprised of *P. deltoides* x *P. deltoides*, *P. deltoides* x *P. maximowiczii* and *P. deltoides* x *P. nigra.* The average percent of family variance of the total genetic variance is 36.2 with a minimum of 34.5 and a maximum of 36.8. Obviously, this value is remarkably consistent across taxa. As expected, the family variance within the *P. nigra* population was found to be a lower proportion of the total genetic variance due to the fact that this population is comprised of half-sibs and not whole-sibs as is the case with the other three taxon groups.

Using the values obtained above and formulas of genetic expectation for among-family variance and clone-within-family variance, we estimated the proportion of additive variance in the system. This value is then used to estimate the gain in yield expected at a chosen level of selection intensity. In our case we used a selection intensity of 1 in 10 in each family and clone-within-family (1 of 100 clones overall). Lower limit and upper limit additive response to among- and within-family selection is estimated to be between 17.5% and 27.2%, respectively in the case of full-sib populations across all taxa. In the case of the half-sib *P. nigra* population, the similar point-estimate value was 47.4%. Based on our analyses and the relatively early stage of poplar breeding overall, we estimate that gains in biomass growth of roughly 20 to 30 percent can be expected through each breeding cycle using selected parents. On an annualized basis, this value is roughly 2% per year which is similar to annual gains through breeding of many agronomic crops. Our results provide the case for a “ladder-and-rung” approach to breeding whereby the parents are continually improved through intraspecific breeding (ladder) and populations of potential new commercial hybrids are produced through interspecific breeding (rungs) using those improved parents. If funds are available for future poplar breeding work, our results argue for a specific structured program testing parental stock of all potential parental species in clone tests in each region, with interspecific breeding being done to capture yield gains and desirable commercial characteristics (e.g., rooting ability, tree form). To our knowledge, information of this type is a unique output of the Sun Grant program and is critically important in designing an effective future poplar breeding program and maximizing use of limited research funds.

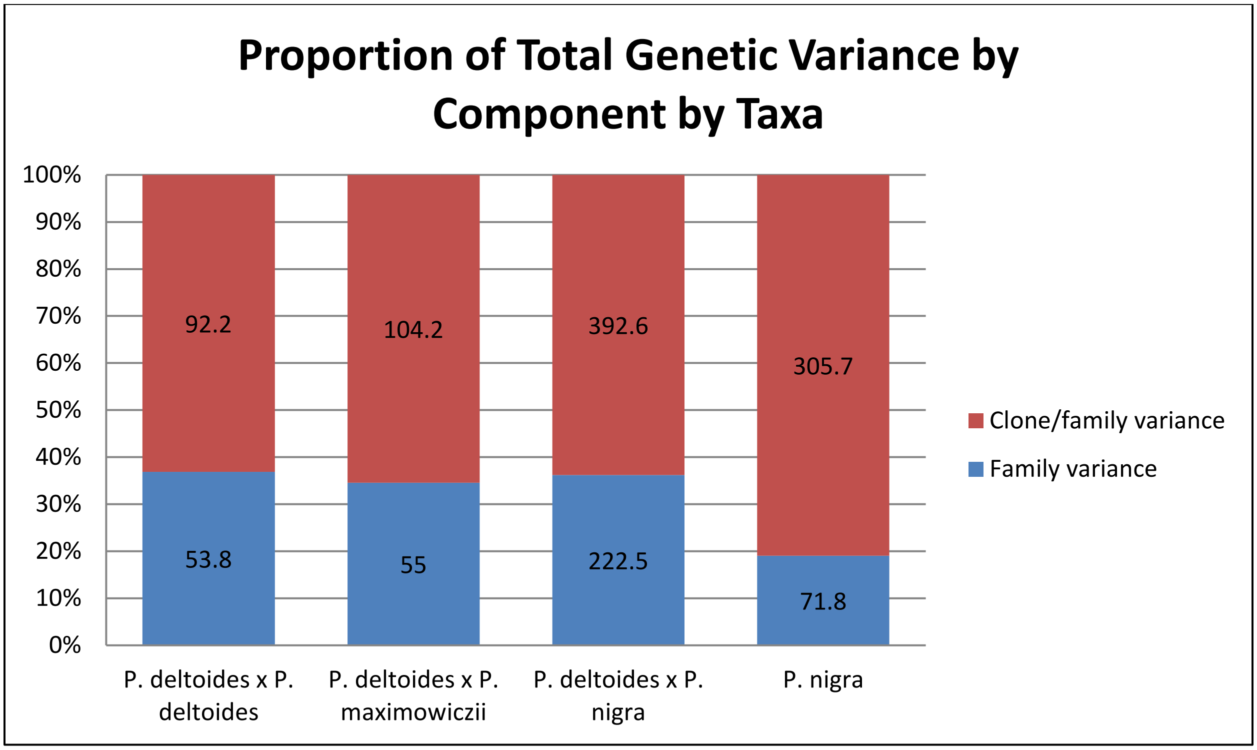

Figure 62. Proportion of total genetic variance by component based on tree basal area among four taxa after five years in Family Field Tests in Minnesota.

Parental Populations

As mentioned in the section on clone testing, disease incidence of *P. deltoides* and *P. nigra* was found to be significantly lower than those clones containing *P. trichocarpa* and survival was much higher in hybrids containing *P. nigra* and *P. trichocarpa* in crosses with *P. deltoides* as opposed to pure-species *P. deltoides.* This was found to be the case in all regions except the western sources where P. trichocarpa is native. Also, as mentioned previously, pure-species *P. deltoides* clones have shown poor survival from hardwood cuttings. This result argues strongly for development of new *P. nigra* that have been tested in the target environments to be crossed with s *P. deltoides* to increase in-field rooting and maintain disease resistance in the South, Mid-South and Northern environments.

Due to the interest in hybridization overall and specifically hybridization including *P. nigra* in crosses, we sought collections of *P. nigra* from native regions in Europe. Through the efforts of the programs at GreenWood Resources and University of Minnesota–Duluth, a large collection of *P. nigra* was obtained. Thousands of clones were procured and propagated for distribution to the Poplar Team members. The breadth and magnitude of this collection is unprecedented in North America. Distribution of *P. nigra* collections has continued, with new plantings of this species being maintained at a site in central Minnesota as well as sites in Washington, Mississippi, South Carolina, Tennessee, and Virginia.

Figure 7 shows results of a five-year clone tests in central Minnesota using the metric of diameter-squared, an index of total tree biomass and volume. As shown, considerable variation exists among this collection and opportunities exist to improve yield based on selection and further breeding within the selected clones. As a means of absolute rather than relative comparison, the top 30 clones at this site were found to be nearly identical in tree size to the same grouping in a clone trial in the area using pre-selected clones. This result indicates that *P. nigra* has the potential to be as productive as current hybrids in northern climates and the recommendation to employ the "ladder-and-rung" breeding approach is likely to result in significant improvements in the genetic diversity and growth of *P. nigra.*

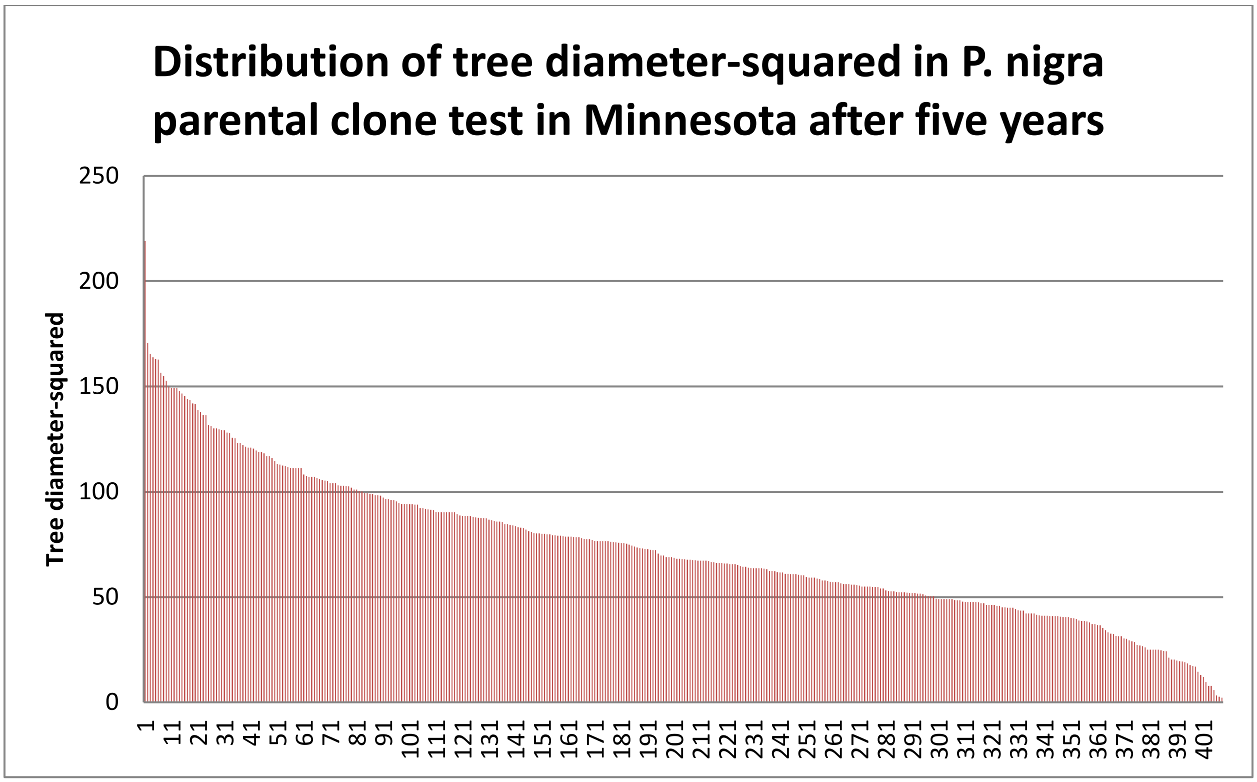

Figure 7. Tree size distribution among an open-pollinated collection of *P. nigra* at a site in central Minnesota.

It should be emphasized that the Sun Grant program has not only produced new knowledge, but has contributed significantly to the physical infrastructure of genetic resources—notably, parental populations that have not existed in North America prior to the program. The significance of these resources cannot be overemphasized. The current network of sites of unique parental populations puts the Sun Grant program in a position to conduct structured breeding in a manner that has never been done before. While funding restrictions are a constant reality, these resources are not static in time and may be lost if funding is not maintained. This could represent a setback of 15 years if allowed to lapse, not to mention the lost progress that could be made if the program were to continue.

Yield Analysis

Southeast Yield Dataset and Reference Model Development

As mentioned in the Methods section, due to the lack of large monoclonal blocks of clones proven to be high-yielding, limitations of younger ages and variable spacing in some of the available field measurements on uplands in the Southeastern U.S., work was done to develop a reference yield model to estimate final-rotation mean annual biomass increment (MABI) yield from mid-rotation measurements of clone tests over a range of plantation spacings in this region.

The first dataset used is that derived from Krinard (1988). A model estimating tree height from age, spacing and the interaction between these variables is as follows with height in meters and tree spacing in square meters per tree:

[Eqn. 1] Tree Height (m) = 5.61 + 1.68 StandAge - 0.0309 m^2-Tree (R-squared = 87.8%)

The model estimating total stand basal area (square meters per hectare) through time using a combination of stand age and spacing (m^2/tree) and their interaction was found to be:

[Eqn. 2] StandBA(m2/ha) = 2.95 + 1.15 StandAge - 0.118 m^2-Tree (R-squared = 79.7%)

P-values for all parameters in these equations are less than 0.05 in all cases. While statistically significant, Equation 1 showed a limited effect of stand age and spacing on tree height. The interaction of age and spacing affects tree height less than one foot in height at age 7 assuming a stand spacing ranging from 3.7 to 7.4 square meters per tree. While much of the literature of thinning studies of other species indicates little effect of stand density on tree height, our analysis shows a slightly negative response to higher density. This is thought to be due to the very wide range of spacings not typically found in other studies, particularly, at the very high density of some spacing treatments in the Nelder design used in this study.

Similar analyses as described above were done on the dataset supplied by GreenWood Resources with the following equations for stand height and stand basal area derived:

[Eqn. 3] AvgHt(meters) = 5.21 + 1.36 StandAge - 0.768 m^2perTree + 0.0632 AgeXSqm-tree (R-squared = 85.1%)

[Eqn. 4] StandBA = 0.14 +6.3 StandAge - 1.35 m^2perTree - 0.103 AgeXSqm-tree (R-squared = 79.3%)

The third dataset supplied by the staff at GreenWood Resources did not include measurement of height but was restricted to stand diameter measurements from which the total stand basal area was derived. The following equation describes the relationship between age and stand basal area:

[Eqn. 5] Stand BA (m^2/ha) = -3.45 + 2.75 Age - 0.085 Age-Squared (R-squared = 92.7%)

As shown, estimation of important stand parameters necessary to estimate yield over a range of age and spacing are generally useful with the R^2 of most of the models ranging from roughly 79 to 93 percent. As a result, we have a tool to reasonably estimate final-rotation stand

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parameters in those cases in which a complete growth history is lacking by comparing the observed height of the clone in question to that expected at the same age and spacing.

The following table shows predicted stand basal area and height through time from age four to age eight assuming two different spacing, 3.71 and 7.43 square meters per tree using the two equations for height and diameter and the GreenWood-1990 equation for stand basal area. Predicted values for the GreenWood-1990 dataset were done using only the 80 square foot treatment as the 40 square foot treatment was beyond the range of the treatments in this study. As can be seen in the following table, agreement is high among the various methods.

This method was applied to the stands shown in Table 11 to estimate the expected yield of a monoclonal block planting at each location using the observed stand height.

Table 10. Estimated height and stand basal area using equations developed from three long-term poplar growth datasets.

		GreenWood 1984		**Krinard Nelder**		**GreenWood 1990**
Stand Age	m^2/Tree	Height meters	Basal Area m^2/ha	Height meters	Basal Area m^2/ha	Basal Area m^2/ha
4	3.7	10.9	17.7	11.8	8.3	
4	7.4	11.1	12.1	12.8	6.5	15.2
5	3.7	13.0	23.4	13.2	9.8	
5	7.4	14.0	17.1	14.2	7.9	20.1
6	3.7	15.2	29.0	14.7	11.3	
6	7.4	16.9	22.2	15.7	9.4	24.6
7	3.7	17.3	34.7	16.1	12.7	
7	7.4	19.8	27.2	17.0	10.9	28.6
8	3.7	19.4	40.4	17.5	14.2	
8	7.4	22.8	32.2	18.5	12.4	32.3

Table 11. Location, plot size, planting year and estimated mean annual increment of the seven upland poplar sites in the southeast U.S. used in the yield database.

State	**Site**	**Plot Size (hectare)**	**Planting Year**	**Harvest Year**	**MAI (Mg/ha)**	**Comments**
SC	Moultry	0.045	2009	2016	6.7	Top 10 clones height, estimated MAI
GA	Floyd	0.036	2003	2011	12.8	MAI estimated from measured top 10 clone ht
SC	Eastover	0.022	2008	2014	12.6	Top 10 clone height, estimated MAI
AL	Randolph	0.045	2009	2016	4.3	Top 10 clone height, estimated MAI
AL	Randolph	0.027	2009	2016	4.3	Top 10 clone height, estimated MAI
NC	Wooten Farm	0.022	2010	2016	10.5	Top 10 clone height, estimated MAI
GA	Bellville	0.022	2010	2016	13.5	Hybrid 11 and 24-128, MAI estimated

Mid-South Alluvial Sites

Data used to produce yield estimates for the Mid-South region were derived from plantations that had been established by the now-discontinued Mead Westvaco Corporation. As mentioned, these plantations are comprised of *P. deltoides* planted at spacings with the average spacing being 667 trees per hectare or 3.96 X 3.96 meter spacing. Using plot-level summaries of tree measurements, linear regression analysis was done to evaluate the relationship of stand basal on stand age and average tree height on tree diameter and age. In the case of the tree height model, only tree diameter was found to be statistically significant (p-value < 0.05). Regression fits (R-squared) ranged from 60% for stand basal area on age to 72% for tree height on tree diameter. Ultimately, these data were converted to total dry biomass and the following equation used to standardize the yield data:

Dry Weight (Mg/ha) = - 32.6 + 15.67 StandAge (R-squared = 63%).

Similar to analyses described for the Southeast Uplands dataset, the ratio of regression-estimated values to actual measured values were used as an index to standardize the yield data to a common age. This was done by using the regression equation of total stand biomass on age and estimating the age-9 stand biomass of stands by developing a ratio of the estimated stand biomass to the measured biomass at the age of measurement.

We suspect that the wide spacing in these plantations is contributing to a reduction in our estimates of yield. This is particularly the case in younger stands. These plantations were originally intended for pulpwood production and not strictly energy and, as a result, the full expression of the productivity potential of poplar for energy is likely underestimated on these sites. After review of these data with the PRISM group, the data were ultimately collapsed to eight sites with the average of the clones occurring at those sites. The decision to eliminate two sites was based on a review of production as affected by stand management history; primarily a question of weed competition in the early years of stand establishment.

Minnesota Yield Data – University of Minnesota

As mentioned in the Methods Section of this report, the majority of yield blocks in Minnesota that were used to build the national yield dataset consisted of replicated, large-block plantings of selected clones that have grown over a time period that are at, or near, final rotation age. Trees included in measurements were interior trees with a minimum of a two-row buffer to eliminate edge-effect and the potential for upwardly biased yield estimates. Also, because we have measured these studies annually, we were able to track yields during the steady-state, full-canopy growth period in all stands. Using the measurements of annual yield during this phase, we calculated the periodic annual increment and, in those cases where the stands have not reached full-rotation age, estimated the final-rotation yield by assuming that annual observed yields would continue to rotation. The average projection period was three years (age seven to ten) and, as a result, we expect that most of the yield estimates reasonably reflect final-rotation yield.

As can be seen in Appendix A, there were 17 clone/site combinations measured on nine sites. The sites were all situated on previously-cropped land and, as such, were of moderate to high productivity for agricultural crops. The sites encompass a wide geographic range from northwestern to south central Minnesota. The average mean annual biomass increment observed is 9.0 Mg ha^{-1} yr^{-1} with a minimum of 7.6 and a maximum of 11.4 ovendry Mg ha^{-1} yr^{-1}. The

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coefficient of variation (standard deviation/mean) within these MABI values is relatively low at 15%. Yields observed in this study are slightly higher and more consistent than those reported previously in the region but generally agree with previous studies. We expect that the primary difference between our observations and prior studies is the greater degree of clone selection and the fact that we used yields from the best-performing clones at each site as an indicator of potential production, not the average of all clones regardless of suitability. Due to the history of breeding and clone testing that has been done in the region prior to the beginning of the Sun Grant Regional Partnership, we were able to use a new set of clones that were developed by our breeding program. The clones planted in yield blocks in our studies had undergone some degree of selection in clone trials in the region and are not the same as those used in previous studies and, as such, an improvement in yield and greater consistency in yield is to be expected.

Michigan State University Yield Tests

In addition to monoclonal yield blocks, a variety of studies of the effect of stand spacing and clone were conducted by Ray Miller and colleagues at the Michigan State University. These studies provide useful information on the effects of genetic composition, stand spacing and site quality on yield and are briefly summarized below.

Escanaba, MI Yield and Spacing Trial

A yield and spacing trial established in 2008 was harvested in the fall of 2014, after 7 growing seasons. Yield varied significantly by variety but was not found to be affected by either spacing or replication. The most productive hybrid was NM6, yielding approximately 67.3 dry tonnes/hectare of biomass or 9.5 dry Mg ha^{-1} yr^{-1} expressed as mean annual increment. The best three hybrids (NM6, NM2, & DN5) together averaged about 53.8 dry Mg ha^{-1} or a MABI of 7.6 Mg ha^{-1} yr^{-1}. From this test, biomass yield of hybrid poplar plantations in the Upper Peninsula of Michigan is expected to range between 6.7 and 9 Mg ha^{-1} yr^{-1} on a 7-year rotation when established at modest densities (i.e. 1,927 trees/hectare). We did not find any advantage to planting more trees at this rotation length. Yields found in this study are consistent with those shown in similar studies in Minnesota and further underpins our estimates of roughly 6.7 to 9 Mg ha^{-1} yr^{-1} on moderate sites in the northern region.

Michigan Biomass Equation Analysis

Yield determinations, like those made in the study above, require destructive sampling which is not always feasible. Standing tree parameters are often measured as surrogates or predictors of biomass. In the spacing trial described above, tree heights, diameters, and plot basal area were measured or calculated annually from the third year onward. The parameter most closely correlated with final biomass yield was “total plot basal area.” Tree heights are exceptionally difficult to accurately measure as plantations age and although correlated with final biomass yield do not add precision to biomass predictions based on basal area alone. Our results show that stool basal area is the best standing tree parameter for monitoring stand development and ranking hybrid biomass production. Measuring tree heights in hybrid poplar test plantations is not a productive use of time.

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Table 12. Correlations between tree parameters and final biomass yield of poplar hybrids after 7 years in a plantation in Escanaba, MI.

Correlations between tree parameters and final biomass yield of poplar hybrids after 7 years in a plantation in Escanaba, MI					
Measured or Calculated Parameter	Correlations with Actual Biomass Yield in Year 7 (Pearson Correlation α=0.01)				
	Growing season when parameter was obtained				
	3rd	4th	5th	6th	7th
Plot Average Height	0.892	0.902	0.892	0.866	0.743
Plot Maximum Height	0.862	0.888	0.872	0.814	0.742
Plot Average DBH	0.872	0.868	0.870	0.796	0.709
Total Plot BA	0.872	0.909	0.932	0.936	0.938

<u>Biomass Estimation Equation Testing</u>

Allometric equations are routinely developed to predict biomass using tree parameters. This is done by destructively sampling trees over a range of diameters and using regression procedures to construct predictor equations. We obtained data sets from the spacing trial described above as well as from a previous study in Escanaba and published studies in Minnesota. We developed biomass predictor equations from these three data sets. While the R^2 statistics for each equation were high, the less-reported RMSE statistics indicate that predicted tree weights are incorrect by an average of 15% to 20%. Further, when we compared the calculated plot weights against the actual measured plot weights in our spacing trial, we found these equations consistently underestimated plot weights by about 20%. The error was not random but systematic.

Our data indicate that predicting end-of-rotation biomass using stem height or diameter measurements or stool basal area is highly problematic and errors of 20% may occur. Care must be taken to ensure that equations used are calibrated using trees of the same clone and tree size being evaluated in a yield study. Variation caused by difficult-to-measure parameters like crown architecture and wood quality are probably causing this error.

Table 13. Biomass prediction equations developed from three datasets.

Biomass predictor equation developed from 3 separate datasets.				
Dataset	# trees	Best Fitting Equation Biomass in oven-dry pounds/tree, BA in square feet/stool	R^2	Root Mean Square Error over the Grand Mean
Netzer, 2002 (published)	152	Biomass = -0.255+477.907xBA+561.742xBA^2	0.983	15%
Fiber Farm, 2008 (MSU)	159	Biomass = 699.206x$BA^{1.143}$	0.915	19%
Spacing Trial, 2014 (MSU)	72	Biomass = 578.509x$BA^{1.08}$	0.978	20%

Yield Analysis on Multiple Sites – Michigan

Basal area growth was measured at the end of the 2014 growing season in each of five poplar hybrid yield trials across Michigan. Absolute growth varied considerably from site to site due to atmospheric, soil, cultural, and age factors, so relative performance (expressed as hybrid average BA divided by the site grand mean) was calculated for each hybrid at each site. Several hybrids appear to be good general performers – doing well everywhere they are planted. Other hybrids appear to be specialists – exhibiting strong genotype by environment interaction. Although absolute growth varied by as much as seven times, some poplar hybrids appear to be good general performers across sites while others exhibit strong genotype-by-environment interaction.

Table 14. Growth of poplar hybrids at 5 sites in Michigan (basal area increment in m^2 ha^{-1} yr^{-1})

Hybrid	Albion	Brimley	Escanaba	Lake City	Onaway	Hybrid Average
Dn170	2.0					2.0
Nm2	2.4		2.4		0.15	1.7
Nm5		0.82	2.3	1.1	0.37	1.7
Dm114	1.6		2.1	0.7	0.24	1.5
Nm6	2.1	0.66	1.8	1.1	0.06	1.5
I4551	2.4		0.9		0.02	1.3
Dn154	1.6		1.6	0.4	0.05	1.1
Dn2	1.8	0.14	1.7		0.11	1.1
Dn70	1.7	0.11	1.4	0.5	0.06	1.1
Dn177	2.1			0.1	0.01	1.0
Dn34	1.4	0.03	1.9	0.3	0.06	1.0
Dn5	1.4	0.08	1.6	0.5	0.12	1.0
Dn182	1.8	0.34	1.3		0.02	0.9
Dn164	1.8		1.3	0.1	0.03	0.9
NE222	1.1	0.22	1.4	0.3	0.05	0.7
Dn17	1.5	0.08	0.7		0.05	0.6
Site Average	1.8	.31	1.6	0.5	0.10	1.1

The variation in hybrid performance across sites contributes to production risk for growers. Genetic, cultural, climatic, and site factors all contribute to phenotypic variability. Unusual events like extreme drought or killing temperatures also cause variation. This was entirely expected. But, we have now had the opportunity to repeatedly examine selected hybrids and surprisingly find as much or even more variation within a site as we find among the sites. This leads us to conclude that within-site factors are causing as much, or more, uncertainty in biomass growth as general site conditions. For example, on three sites where NM6 grows particularly well, basal area growth after 4 growing seasons varied by 22% from site to site. However, basal area growth varied by as much as 35% among plots-within-sites. This variable

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performance remains unexplained and contributes substantially to the risk faced by poplar biomass growers. Our results show that both within-site variation and among-site variation is significant. This unexplained variation and significant GXE interaction represents a barrier to commercialization.

Table 15. Standardized growth of poplar hybrids at 5 locations in Michigan - basal area growth was standardized by dividing each plot's basal area by its site average plot basal area.

Standardized growth of poplar hybrids at 5 locations in Michigan Basal area growth was standardized by dividing each plot's basal area by its site average plot basal area.												
Hybrid	Test-Wide		Albion (year 4)		Brimley (year 6)		Escanaba (year 6)		Lake City (year 5)		Onaway (year 5)	
	# of plots	**Standardize d Growth**	# of plots	Standardized Growth	# of plots	Standardized Growth	# of plots	Standardized Growth	# of plots	Standardized Growth	# of plots	Standardized Growth
Nm5	19	2.44			4	2.67	5	1.41	5	2.08	5	3.65
Dm114	20	1.52	5	0.94			5	1.32	5	1.43	5	2.41
Nm2	15	1.47	5	1.43			5	1.47			5	1.52
Nm6	21	1.41	5	1.24	4	2.16	5	1.14	5	2.08	5	0.59
Dn170	5	1.16	5	1.16								
Dn2	18	0.97	5	1.07	3	0.44	5	1.05			5	1.10
Dn5	24	0.87	5	0.80	4	0.27	5	1.02	5	0.96	5	1.16
Dn70	21	0.82	5	0.98	2	0.37	5	0.90	5	0.93	4	0.59
Dn154	20	0.81	5	0.95			5	1.03	5	0.73	5	0.53
I4551	13	0.80	5	1.41			5	0.55			3	0.22
Dn182	19	0.78	5	1.08	4	1.11	5	0.79			5	0.20
Dn34	22	0.75	5	0.82	2	0.11	5	1.17	5	0.64	5	0.62
NE222	24	0.67	5	0.63	4	0.72	5	0.89	5	0.60	5	0.54
Dn177	12	0.65	5	1.22					5	0.29	2	0.12
Dn164	20	0.62	5	1.04			5	0.83	5	0.26	5	0.35
Dn17	19	0.54	5	0.90	4	0.26	5	0.45			5	0.48

Pacific Northwest Yield Dataset

Varietal site trials used in the development of the yield dataset were established at eight locations in Oregon, Washington, Idaho, and New Mexico in 2005. Sites were: Puyallup, WA; Pullman, WA; Westport, OR; Caldwell, ID; Boardman, OR; Klamath Falls, OR; Newberg, OR; and Farmington, NM. Each location was classified as either a mild/mesic or a cold/xeric habitat so as to better match the suitability of an experimental set of poplar clonal varieties that were tested at each location. Sixty-nine varieties were used in planting four of the trials classified as mesic sites, while a group of 65 varieties was used in planting trials at the eight trial locations considered xeric in nature. Thirty-six varieties were common to all mesic and xeric trials; thirty-two varieties were unique to the mesic trials while 28 varieties were unique to the xeric trials. Each clone was replicated four times per site and with each replication represented by a randomly located four-tree row plot.

Yields ranged widely depending on site. As expected, those sites having relatively lower natural rainfall were lower in yield. The decision was made to leave these sites in the analysis for the national mapping to capture the effect of drought on poplar production.

Final Dataset used for National Mapping

As shown in Table 16 below, MABI yields ranged from less than 2.2 Mg ha^{-1} yr^{-1} to a high of 15.0 Mg ha^{-1} yr^{-1}. The lowest yielding sites in the Pacific Northwest were generally on sites that were very low rainfall and, as such, drought stress accounted for low yields. Conversely, on those alluvial sites in the Mid-South with presumed optimal water availability, yields ranged from 10.3 to 15.0 Mg ha^{-1} yr^{-1}. Yield estimates for upland sites in the Southeast region show the potential for relatively high yields assuming that selection of further research is done to select appropriate clones and sites. As mentioned, yields in the Midwest range from 7.8 to nearly 11.2 Mg ha^{-1} yr^{-1}. In all regions, the need for further refinement of clone selection for the region is was noted as a factor limiting yield and commercial adoption of poplar culture.

After assembling the final dataset of poplar yield using data and the methodology described, a meeting was held with Oregon State University PRISM staff to evaluate the utility of data contained in the larger dataset (see Appendix A), explore issues related their suitability or limitations of use in the national yield mapping effort, and to develop the yield potential map shown in Figure 6. After reviewing model fits and outliers, the obvious choice was to eliminate those sites in the Pacific Northwest that were under irrigation (i.e., Boardman, OR; others?) and, as such, did not lend themselves to the development of climate and soil suitability indices being calculated by the PRISM group. Also, possible reasons for deviations from estimated yield were discussed but all sites remained in the dataset that didn't have an obvious reason for elimination. The decision was made to leave all sites in the dataset that were known to have a history of adequate management. After review of the data and discussion of the effect of genetic composition on yield, the decision was made to average all of the clones on those sites having multiple clones on a given site. As a result, the dataset was consolidated to those sites shown in the table below.

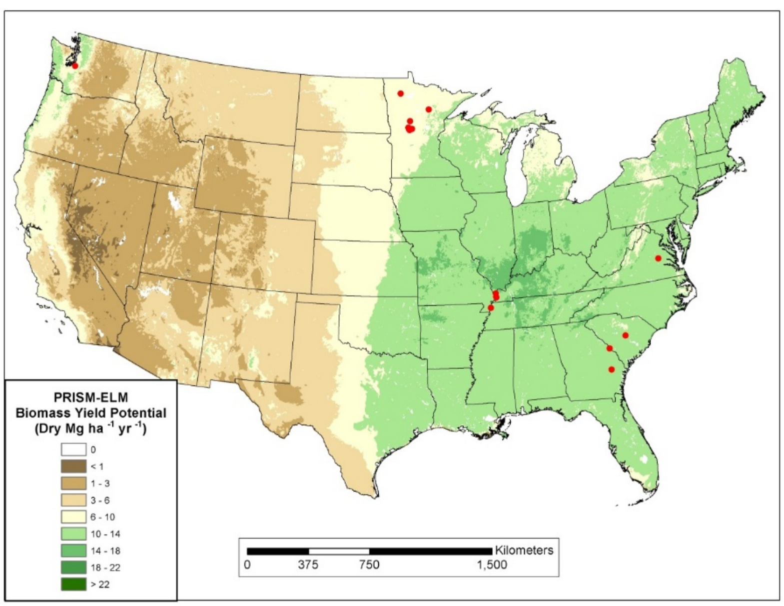

Figure 8. Potential poplar biomass yields for the US generated from the PRISM-ELM model and data from a network of 17 trials. Location of the poplar trials used for yield predictions are indicated with red dots (from Volk et al., 2017).

Table 16. Final poplar yield dataset used in the PRISM national yield mapping project.

NAME	Region	Yield ($Mg\ ha^{-1}\ yr^{-1}$)	comments
Angelo 1999	Mid-South	15.0	
Bellville 2010	Southeast	13.5	
DOE 2005 Caldwell	Pacific NW	1.2	
DOE 2005 Klamath Falls	Pacific NW	.04	
DOE 2005 Puyallup	Pacific NW	9.6	
DOE 2005 Westport	Pacific NW	13.9	eliminated - no soils grid data
Eastover 2008	Southeast	12.6	
Floyd 2003	Southeast	12.8	
Hansen 2006	Midwest	8.3	
Hemming 2005	Midwest	10.5	
Island 3 2005	Mid-South	10.4	
Joppru 1996	Midwest	10.1	
Kniesel 1999	Midwest	7.5	
Lansing Hi-Den 2002	Midwest	20.7	Eliminated – very hi-density
Grand Rapids 2007	Midwest	11.1	
Moultry 2009	Southeast	6.7	
Peck 2005	Mid-South	12.4	
Schultz 2007	Midwest	8.0	
Sebeka 1996	Midwest	7.6	
Wise 2003	Mid-South	14.0	
Woelfel 2001	Midwest	7.8	
Wolf Island 2005	Mid-South	10.5	
Wooten Farm 2010	Southeast	10.5	

Yield Conclusions/Comments

Obviously the ideal situation to develop comprehensive yield estimates for all regions of the U.S. is an extensive, long-term dataset of trials specifically designed to assess short-rotation biomass yield using a set of selected clones based on a network of clone tests in each region. Such a dataset does not currently exist. In the case of yield data for the Southeast region, we used the best available data to estimate future yield based on performance of a subset of selected clones at younger ages. In the case of alluvial sites in the Mid-South, the very wide plantation spacing undoubtedly contributes to an underestimate in yield. Also, the northeast United States is under-represented in the yield dataset even though it is known that poplar will survive in this region. To date, *Septoria* susceptibility and the need for comprehensive clone screening of regionally-adapted genetic material limits the availability of data from this region. While the Midwest has a longer and more extensive program including breeding and yield studies, the need continues for breeding and field testing to increase clonal stability and biomass across a wider area within this region. A similar situation exists in the Pacific Northwest where a commercial program has supported a long-term genetic improvement and yield testing program but opportunity exists for continued yield improvement.

The Sun Grant Biomass Feedstock Partnership has allowed establishment of a network of clone tests across the country, a critical first step. However, there is a clear need for continued work to evaluate the yield potential of poplar on a variety of sites in large-block trials using clones selected from local tests. Also, research in yield of poplar under higher-density, biomass-oriented spacings as well as coppice production under these conditions is needed in all regions of the U.S. Additional research is needed on cost-effective stand establishment and weed control on sites across the United States, particularly in the Southeast. Based on results in selected regions, continued breeding holds great potential to increase rooting potential, growth rate and site-to-site stability. This work is critical to development of poplar as a commercially viable biomass crop in the region. If biomass conversion technology continues to show progress and economics of fuel production are favorable, the regions holds promise for enhanced production of biomass in poplar plantations.

Production Economics

Over the past five years of activity, cash flow models were developed using input costs from the former Verso Paper commercial operation in Minnesota as well as the GreenWood Resources program. This information was put into a cash flow analysis where management inputs are identified and cost of those practices delineated on an annual basis through ultimate harvest. Breakeven costs are then calculated using a selected discount rate (in our case, a 3% real rate) and the sum of input costs throughout the life of the plantation. The reader is referred to a paper published by members of the Poplar Team (Berguson et.al. 2010) for specific analyses and methodology.

In addition to the fundamental cash flow analysis, work was done in Minnesota to estimate the opportunity costs associated with displacing an agricultural crop. While we don't necessarily advocate direct replacement of agricultural crops energy crops with energy crops, it is nevertheless instructive to consider the reality of displacing energy crops on land currently producing agricultural commodities and quantifying the delivered price that would likely need to be received in order to pay the farmer or landowner an amount that is cost-competitive with that associated with growing an agricultural crop. Based on our estimates of production costs, stand production and harvest and transport economics, the DOE's delivered price target range of $77 to $88 per dry Mg appears to be achievable on many sites in the Midwest. In the Pacific Northwest, the estimated breakeven production cost using prevailing land values ranged from $77 to $110 per hectare assuming unirrigated land and current yields. Spreadsheets of our cash flow analyses were provided to staff working on the recently completed Billion Ton Update (2016).

Key Outputs

- Clones performed and selected superior genotypes for each region
- Identified canker-susceptible clones in the Southeast
- Produced large quantities of next-generation materials for testing and yield improvement through breeding
- Produced unprecedented infrastructure of parent collections to support further breeding through genetic improvement research

- Enhanced understanding of genetic effects and "bang for the buck" in expected yield gain per breeding cycle (20% gain expected per cycle)
- Developed cash flow models and gained better understanding of production economics
- Developed a much more extensive dataset of yield estimates for all regions, with benefits to the national mapping effort
- Supported cooperative research in sustainability and carbon sequestration
- Facilitate international cooperation resulting in joint field tests in Europe.

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Where do we go from here?

Clone Testing

A particularly frustrating and puzzling aspect of poplar clone testing is the lack of site-to-site stability in growth rate within a region. The high degree of "genotype-by-environment interaction" associated with this work requires that a field testing program include many tests replicated within site and across sites within a region in order to have a level of confidence that a particular clone will perform consistently and reduce the risk of plantation failure or underperformance. While there are notable exceptions to this phenomenon, they are a very small subset of clones. Field testing of new clones at multiple sites within a region is necessary to identify those clones capable of adapting to a range of field conditions prevalent throughout the region. It may be possible to approach this problem through testing of parental stock in replicated field tests prior to breeding to determine if it is possible to "breed in" plasticity.

Breeding

Building on the results of the analysis of family and clone-within-family variance, coupled with the array of flowering collections of superior parents, we are in a unique position to conduct second-generation breeding and secure yield gains available to the program. Research done over the past 5 years in the development of parental collections, understanding of genetic effects, and the demonstrated success of the breeding programs suggests that additional funding could contribute to significant yield improvement and diversification. In order to continue to refine genetics and improve yield, further breeding and field testing of progeny is recommended.

Also, as highlighted in the discussion of disease by taxa in field experiments in the Mid-South region, new field trials of *P. nigra* have begun and should be continued. The low inherent rooting ability of *P. deltoides* could potentially be overcome assuming that proven adapted parents of *P. nigra* could be used to create a new generation of *P. deltoides x P. nigra* hybrids.

Fertility, Nutrition, and Nutrient Cycling

One aspect that the Sun Grant Poplar Team commonly discusses is the lack of information on responses (or lack thereof) of poplar plantations to fertilizer additions, particularly nitrogen fertilization. While it is axiomatic that high growth rates cannot continue without nutrient additions, the lack of response to nitrogen in many environments is puzzling. Our experience in research into nitrogen response has been mixed, with some sites exhibiting statistically significant response to nitrogen and others showing no response. In those cases where fertilization response was noted in Minnesota, the asymptote of the response curve occurred at relatively low rates (90 kilograms hectare^{-1} of elemental nitrogen) with no additional benefit of annual fertilization over biennial fertilization. There is a need to link site type, site management history, and nitrogen status (possible using chlorophyll meters calibrated for poplars) to identify those conditions where fertilization may prove to be cost effective. To date, that understanding is unclear and is a subject for more research. Related to this, life cycle analyses are heavily influenced by energy inputs, and nitrogen fertilization represents a potentially high energy input into these analyses. Thus, nutrient response and the need for fertilization have an effect on commercial performance and sustainability and energy efficiency.

Regional Yield Analysis and Verification on Multiple Sites

The effort to construct estimates of expected poplar yield for the regions as part of the national mapping effort highlighted the continued need to first identify promising high-yielding, disease-resistant clones, but then to plant and measure these trials on a wide array of potential site types within each region. This work is viewed as a logical continuation of genetic improvement research to verify yield performance of selected clones in a region. These data are an important part of analyses to estimate production costs and the optimal siting of plantations in a given region.

Coppice Management and Spacing Effects

Questions remain regarding the effect of repeated coppices on long-term production and the variation in suitability of clones in regrowth and maintenance of long-term productivity under a coppice system. While this system has been in place in Europe, and research into coppice systems is underway on both relatively large-scale (GreenWood Resources in the Pacific Northwest) and smaller research plots (University of Minnesota–Duluth Natural Resources Research Institute and Michigan State University), more intensive research on this topic over a longer time period is required before this system can be relied on to produce feedstock on a commercial scale.

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Poplar Appendix A. Original plot-level database of poplar yield developed for the national yield mapping project.

LAT	LON	County	State	Plot size (hectares)	Planting Year Coppice Year	Harvest Measured Year	Dry Weight tonnes per hectare	MAI dry tonnes per hectare per year
48.15106	-95.7606	Pennington	MN	.178	1996	2008	131.4	10.1
46.61604	-95.0081	Wadena	MN	.178	1996	2005	76.2	7.6
46.27225	-95.1459	Ottertail	MN	.096	1999	2008	75.1	7.6
46.26469	-95.1452	Todd	MN	.096	2001	2010	77.8	7.8
46.18593	-95.119	Todd	MN	.096	2005	2014	90.3	9.0
46.18593	-95.119	Todd	MN	.096	2005	2014	114.6	11.4
46.18593	-95.119	Todd	MN	.096	2005	2014	109.4	11.0
46.12928	-95.0578	Todd	MN	.137	2006	2015	81.6	8.1
46.12928	-95.0578	Todd	MN	.137	2006	2015	81.1	8.1
46.12928	-95.0578	Todd	MN	.137	2006	2015	84.7	8.5
46.12928	-95.0578	Todd	MN	.137	2006	2015	80.5	8.1
47.24922	-93.4902	Itasca	MN	.087	2007	2016	113.7	11.4
47.24922	-93.4902	Itasca	MN	.087	2007	2016	107.6	10.8
46.19521	-94.8618	Todd	MN	.137	2007	2016	82.5	8.3
46.19521	-94.8618	Todd	MN	.137	2007	2016	77.8	7.8
44.06203	-93.5437	Waseca	MN	.034	2007	2016	83.2	8.3
44.06203	-93.5437	Waseca	MN	.034	2007	2016	84.5	8.5
33.00189	-81.3828	Allendale	SC	.047	2009	2016	53.8	6.7
33.29955	-81.9026	Floyd	GA	.036	2003	2011	115.0	12.8
33.86487	-80.7207	Richland	SC	.022	2008	2014	87.4	12.6
32.92397	-86.8982	Bibb	AL	.047	2009	2016	31.4	4.3
32.92319	-86.8986	Bibb	AL	.027	2009	2016	34.1	4.3
37.70209	-77.5764	Pitt	NC	.022	2010	2016	73.8	10.5
32.13843	-81.9666	Evans	GA	.022	2010	2016	94.2	13.5
36.79672	-89.1629	Carlisle	KY	.040	2005	2010	61.9	12.3
36.21208	-89.5855	Lake	TN	.040	2005	2010	62.1	12.3
36.74464	-89.1709	Hickman	KY	.040	2005	2010	50.4	10.1
36.84222	-89.1292	Carlisle	KY	.040	2004	2010	72.0	12.1

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LAT	LON	County	State	Plot size (hectares)	Planting Year Coppice Year	Harvest Measured Year	Dry Weight tonnes per hectare	MAI dry tonnes per hectare per year
36.84222	-89.1292	Carlisle	KY	.040	2004	2010	71.1	11.9
36.68094	-89.1612	Hickman	KY	.040	2004	2010	49.1	8.1
36.68094	-89.1612	Hickman	KY	.040	2004	2010	63.2	10.5
36.68094	-89.1612	Hickman	KY	.040	2004	2010	92.6	15.5
37.03361	-89.1542	Ballard	KY	.040	2003	2010	52.9	7.6
37.03361	-89.1542	Ballard	KY	.040	2003	2010	54.2	7.8
36.75736	-89.1547	Mississippi	MS	.040	2003	2010	97.7	13.9
36.70922	-89.1629	Hickman	KY	.040	2003	2010	89.9	12.8
36.70922	-89.1629	Hickman	KY	.040	2003	2010	78.9	11.2
36.83356	-89.1608	Carlisle	KY	.040	2002	2010	74.2	9.2
36.83356	-89.1608	Carlisle	KY	.040	2002	2010	63.0	7.8
36.83356	-89.1608	Carlisle	KY	.040	2002	2010	80.7	10.1
36.83356	-89.1608	Carlisle	KY	.040	2002	2010	60.5	7.6
36.71406	-89.1842	Hickman	KY	.040	2002	2010	67.1	8.5
36.80458	-89.1604	Carlisle	KY	.040	2001	2010	108.0	12.1
36.21508	-89.587	Carlisle	KY	.040	2000	2010	82.9	8.3
36.97675	-89.1639	Alexander	IL	.040	1999	2010	183.1	16.6
36.97675	-89.1639	Alexander	IL	.040	1999	2010	153.3	13.9
36.97675	-89.1639	Alexander	IL	.040	1999	2010	193.9	17.7
36.97675	-89.1639	Alexander	IL	.040	1999	2010	138.1	12.6
36.80369	-89.1599	Carlisle	KY	.040	1999	2010	117.7	10.8
45.76678	-87.1901	Delta	MI	.040	1998	2008	88.8	8.1
45.7706	-87.1996	Delta	MI	.040	2008	2012	57.4	7.2
45.7706	-86.1996	Delta	MI	.040	2008	2012	48.4	6.1
45.76809	-87.1987	Delta	MI	.003	2002	2004	25.6	8.5
45.76809	-87.1987	Delta	MI	.003	2002	2004	24.2	8.1
45.76809	-87.1987	Delta	MI	.003	2005	2007	22.9	7.6
45.76809	-87.1987	Delta	MI	.003	2005	2007	23.5	7.8
45.76809	-87.1987	Delta	MI	.003	2008	2010	25.6	8.5
45.76809	-87.1987	Delta	MI	.407	2008	2010	25.6	8.5

LAT	LON	County	State	Plot size (hectares)	Planting Year Coppice Year	Harvest Measured Year	Dry Weight tonnes per hectare	MAI dry tonnes per hectare per year
42.82359	-84.385	Clinton	MI	.003	2002	2006	91.5	22.9
42.82359	-84.385	Clinton	MI	.003	2002	2006	74.4	18.6
45.76913	-87.2008	Delta	MI	.032	2009	2012	35.9	4.5
45.76913	-87.2008	Delta	MI	.032	2009	2012	34.1	4.3
46.36182	-87.2449	Marquette	MI	.032	2009	2012	7.2	0.9
46.36182	-87.2449	Marquette	MI	.032	2009	2012	5.4	0.7
46.40054	-84.4668	Chippewa	MI	.032	2009	2012	3.6	0.4
46.40054	-84.4668	Chippewa	MI	.032	2009	2012	3.6	0.4
42.82359	-84.385	Clinton	MI	.003	2002	2006	60.1	15.0
42.82359	-84.385	Clinton	MI	.003	2002	2006	29.6	7.4
42.82359	-84.385	Clinton	MI	.003	2002	2006	28.7	7.2
42.82359	-84.385	Clinton	MI	.003	2002	2006	26.0	6.5
42.82359	-84.385	Clinton	MI	.003	2002	2006	21.5	5.4
46.13129	-123.371	Clatsop	OR	.0006	2011	2012	1.5	0.8
46.13129	-123.371	Clatsop	OR	.0006	2011	2012	2.3	1.1
46.13129	-123.371	Clatsop	OR	.0006	2011	2012	7.3	3.6
46.13237	-123.367	Clatsop	OR	.0003	2009/2010	2011	32.8	16.4
46.13237	-123.367	Clatsop	OR	.0003	2009/2010	2011	40.8	20.4
46.13174	-123.367	Clatsop	OR	.001	2006	2011	80.3	13.4
46.13174	-123.367	Clatsop	OR	.001	2006	2011	114.8	19.1
46.13	-123.37	Clatsop	OR	.001	2005	2009	35.8	7.2
46.13	-123.37	Clatsop	OR	.001	2005	2009	68.9	13.8
45.85	-119.62	Morrow	OR	.001	2005	2009	22.0	4.4
45.85	-119.62	Morrow	OR	.001	2005	2009	46.0	9.2
36.73	-108.17	San Juan	NM	.001	2005	2009	18.5	3.7
36.73	-108.17	San Juan	NM	.001	2005	2009	39.7	7.9
42.20	-121.82	Klamath	OR	.001	2005	2009	0.4	0.1
42.20	-121.82	Klamath	OR	.001	2005	2009	1.9	0.4
47.20	-122.30	Pierce	WA	.001	2005	2009	32.2	6.5
47.20	-122.30	Pierce	WA	.001	2005	2009	47.7	9.5
46.73	-117.00	Whitman	WA	.001	2005	2009	32.9	6.6
46.73	-117.00	Whitman	WA	.001	2005	2009	41.9	8.4
43.66	-116.23	Canyon	ID	.001	2005	2009	2.0	0.4

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LAT	LON	County	State	Plot size (hectares)	Planting Year Coppice Year	Harvest Measured Year	Dry Weight tonnes per hectare	MAI dry tonnes per hectare per year
43.66	-116.23	Canyon	ID	.001	2005	2009	6.1	1.2
45.21	-122.987	Yamhill	OR	.001	2005	2009	2.6	0.5
45.21	-122.987	Yamhill	OR	.001	2005	2009	4.9	1.0
45.83217	-119.567	Morrow	OR	.0006	2011	2012	3.1	1.6
45.83217	-119.567	Morrow	OR	.0006	2011	2012	4.1	2.1
45.83217	-119.567	Morrow	OR	.0006	2011	2012	6.2	3.1
45.83349	-119.566	Morrow	OR	.001	2003	2007	54.8	11.0
45.83349	-119.566	Morrow	OR	.001	2003	2007	79.8	16.2
45.83232	-119.565	Morrow	OR	.0003	2009/2011	2011	28.2	14.1
45.83232	-119.565	Morrow	OR	.0003	2009/2011	2011	30.7	15.4

Willow

Willow Team: Timothy Volk, State University of New York; Lawrence Smart, Cornell University; Raymond Miller, Michigan State University; Julia Kuzovkina, University of Connecticut

Summary

Interest in shrub willows (*Salix* spp.) as a perennial energy crop for the production of biomass has developed in Europe and North America over the past few decades because of the multiple environmental and rural development benefits associated with their production and use. Initial trials with shrub willows as a biomass crop were conducted in the mid-1970s in Sweden, with the first trials in the United States starting in 1986. Since the initial trials in upstate New York, yield trials have been conducted in a number of locations in the northeastern and midwestern United States, as well as in several provinces in Canada. Willow shrubs have several characteristics that make them an ideal feedstock for biofuels, bioproducts, and bioenergy: high yields that can be sustained for over 25 years in 3- to 4-year rotations, ease of propagation from dormant hardwood cuttings, a broad underutilized genetic base, ease of breeding for several characteristics, ability to resprout after multiple harvests, and chemical composition and energy content similar to other northern hardwood species.

The objectives of the willow feedstock network were to (1) assess the current and future production potential of willow biomass crops across a wide range of sites in the Northeast and Midwest and (2) use the data from these trials to develop models to estimate yield potential of willow biomass crops across multiple regions. First-rotation yields were generated for a wide variety of cultivars across sites with a range of conditions across a broad geographical range. The current recommendation for large-scale plantings of willow biomass crops is that multiple cultivars should be planted at each site to minimize risk. Therefore, reporting yields of the top three or top five cultivars at each site is more representative. The yield of the top three cultivars across the sites ranged from 2.9 (Potsdam, New York) to 14.2 (at Middlebury, Vermont, and Storrs, Connecticut) dry Mg ha^{-1} yr^{-1}. The mean across the 19 sites was 9.7 $\pm$ 0.9 dry Mg ha^{-1} yr^{-1}. The yield of the top five cultivars across the 19 sites ranged from 2.7 (Potsdam, New York) to 13.9 (Middlebury, Vermont) dry Mg ha^{-1} yr^{-1} with a mean across all the sites of 9.2 $\pm$ 0.9 dry Mg ha^{-1} yr^{-1}.

Willow biomass crops are cultivated in a perennial system that is typically harvested multiple times on 3- or 4-year rotation cycles, and they have projected lifespans of over 25 years. However, data on the long-term production potential of willow biomass crops are very limited. The trials in this project have provided valuable results on the production of willow over multiple rotations. These long-term data begin to provide verification that willow can be productive over multiple rotations and provide a basis for modeling these systems over 25 or more years. This network of field trials with the large number of cultivars has provided essential information on potential yield increases associated with breeding and selection efforts. Yield increases associated with new cultivars have typically ranged from 15–25%, with some variation across sites. Since breeding and selection work is still at an early stage for willow, these results suggest that significant gains can still be realized by developing improved cultivars.

For perennial crops like willow, projection of yields over two or more decades is an important factor that influences key attributes, including the economic viability of these systems.

The yield data collected across a range of sites and over multiple rotations as part of this project have provided a solid foundation for improving economic models of this system. In addition, this yield data, and seven other additional trials outside of the network, were used to develop yield models using PRISM-EM across multiple regions of the United States.

Introduction

Interest in shrub willows (*Salix* spp.) as a perennial energy crop for the production of biomass has developed in Europe and North America over the past few decades because of the multiple environmental and rural development benefits associated with their production and use (Börjesson 1999; Rowe et al. 2008; Volk et al. 2014). Initial trials with shrub willows as a biomass crop were conducted in the mid-1970s in Sweden, with the first trials in the United States starting in 1986 (Volk et al. 2006). Since the initial trials in upstate New York, yield trials have been conducted in a number of locations in the northeastern and midwestern United States, as well as in several provinces in Canada.

Willow shrubs have several characteristics that make them an ideal feedstock for biofuels, bioproducts, and bioenergy: high yields that can be sustained for over 25 years in 3- to 4-year rotations, ease of propagation from dormant hardwood cuttings, a broad underutilized genetic base, ease of breeding for several characteristics, ability to resprout after multiple harvests, and chemical composition and energy content similar to other northern hardwood species (Stoof et al. 2015).

The shrub willow cropping system consists of planting genetically improved cultivars in prepared open land where weeds have been controlled. Willow can be grown successfully on marginal agricultural land across the Northeast, Midwest, and parts of the Southeast. Weed control usually involves a combination of chemical and mechanical techniques and should begin in the fall before planting if the field contains perennial weeds, which is often the case with marginal land. Willows are planted as unrooted, dormant hardwood cuttings in the spring as early as the site is accessible at about 15,000 plants ha^{-1} using mechanized planters that are attached to farm tractors and operate at about 0.8 ha $hour^{-1}$. Following the first year of growth, the willows are typically cut back (coppiced) close to the ground level during the dormant season to force coppice regrowth, which increases the number of stems per stool from 1–2 to 8–13, depending on the genotype (Tharakan et al. 2005). After an additional 3 to 4 years of growth, the stems are mechanically harvested during the dormant season after the willows have dropped their leaves. The coppiced plants sprout again the following spring when they are typically fertilized with about 100 kg nitrogen ha^{-1} of commercial fertilizer or organic sources like manure or biosolids. Further research is underway to refine these recommendations for new willow cultivars across a range of sites. The willows are allowed to grow for another 3- to 4-year rotation before they are harvested again. Projections indicate that the crop can be maintained for seven 3-year rotations before the rows of willow stools begin to expand to the point that they restrict access to harvesting equipment and thus need to be trimmed back with a heavy disk or mower. After 22 years in cultivation, some cultivars will need to be replaced by improved cultivars developed through breeding. This is easily accomplished in one season by killing the existing stools with herbicides after harvesting and then chopping the killed stools with a heavy disk and/or grinding machine, followed by planting the same year or the following year.

The large genetic diversity across the genus *Salix* and the limited domestication efforts to date provide tremendous potential to improve yield and other characteristics, such as insect and disease resistance, and growth form of willow biomass crops. Worldwide there are over 350 species of willow (Kuzovkina et al. 2008; Smart and Cameron 2008), with growth forms ranging from prostrate, dwarf species to trees with heights of greater than 40 m. The species used in woody crop systems are primarily from the subgenus *Vetrix*, which has over 125 species worldwide (Kuzovkina et al. 2008). While these species have many characteristics in common, their growth habits, life history, and resistance to pests and diseases vary, which are important considerations in the successful development of woody crops. The ability for vegetative propagation of most willow genotypes means that once superior individuals are identified, they can be maintained and rapidly multiplied for deployment.

As willow breeding programs in North America and Europe have advanced in the last decade, interspecific hybridization has proven to be a very effective strategy for capturing heterosis for yield in combination with pest and disease resistance, yet we know little about the genomic basis for heterosis in interspecific hybrids. More specifically, a trend that has emerged that is predominant in *Salix* is the consistent success of crosses between diploid species and tetraploid species in generating triploid progeny that outperform their parents (Serapiglia et al. 2014, 2015). This phenomenon is not a major component of breeding in poplar, but it is critical to future cultivar breeding in willow. These triploid genotypes also have reduced reproductive fertility, helping to allay concerns about potential invasiveness. Since USDA's Animal and Plant Health Inspection Service has recently banned the import of *Salix* cuttings into the United States, it is imperative to maintain a strong willow breeding and conservation program in North America and to expand existing *Salix* germplasm collections through seed import, if possible.

Objectives

The objectives of the willow feedstock network were to (1) assess the current and future production potential of willow biomass crops across a wide range of sites in the Northeast and Midwest and (2) use the data from these trials to develop models to estimate yield potential of willow biomass crops across multiple regions.

Methods

This project included 19 trials across six states (Connecticut, Illinois, Michigan, Minnesota, New York, and Vermont) planted between 1993 and 2010 (Figure 1 and Table 1). Two trials with older cultivars were included to provide data on the long-term productivity of shrub willow systems over multiple rotations. An additional eight trials with new cultivars bred in New York, which were established before the start of this project, were included. Finally, eight additional trials were established during this project and included some of the most recently developed cultivars. In addition to the data from these trials, results from seven other trials that were not formally part of this project were included in the data set used to develop regional yield estimates in conjunction with Oregon State University using their PRISM Environmental Limitations Model (PRISM-ELM) (Daly et al., 2018). The trials were monitored and measured for most of this project, resulting in data from harvests of one 6th rotation, two 5th rotations, one 4th rotation, two 3rd rotations, eight 2nd rotations, and fifteen 1st rotations. This network of trials is providing essential data on long-term production of willow biomass crops as well as yield information for new cultivars across a range of sites.

All yield trials in this project were relatively small in scale, consisting of between 4 and 30 genotypes at each site. Individual field plots typically contained three double rows of willow with 10 to 18 plants in each row. Plots were typically 6.9 m in width and 6.0 to 7.9 m long. Most of the trials included four replications of each genotype, but in a few of the older trials, only three replications were available. In the vast majority of cases, the trials were coppiced after the establishment year and then were harvested on 3-year rotations. Site characteristics for the trials varied widely from some better site conditions, particularly at university research stations, to truly marginal conditions at other sites. USDA National Resources Conservation Service (NRCS) land capability class varied from 1 to 4 and the National Commodity Crop Production Index ranged from 0.17 for a site in the Upper Peninsula of Michigan (Skandia) to 0.98 at the site in Illinois (Savoy). Based on NRCS soils data, drainage conditions at the sites varied from moderately well drained to poorly drained.

Overall, 94 different willow genotypes were included in these trials, representing more than 10 different diversity groups (a diversity group represents a willow species or particular hybrid). Trials planted before 2005 included older genotypes that were either acquired from the University of Toronto or were collected from the wild in the northeastern United States. Following 2005, the majority of genotypes in trials were based on breeding work that had been done at the State University of New York College of Environmental Science and Forestry. Only one cultivar is present in all 18 trials: *Salix* × *dasyclados* 'SV1'. Three cultivars are present in 17 of the 18 trials (*S. eriocephala* 'S25,' *S. miyabeana* 'SX61,' and 'SX64') and eight cultivars are present in 14–16 trials in the network.

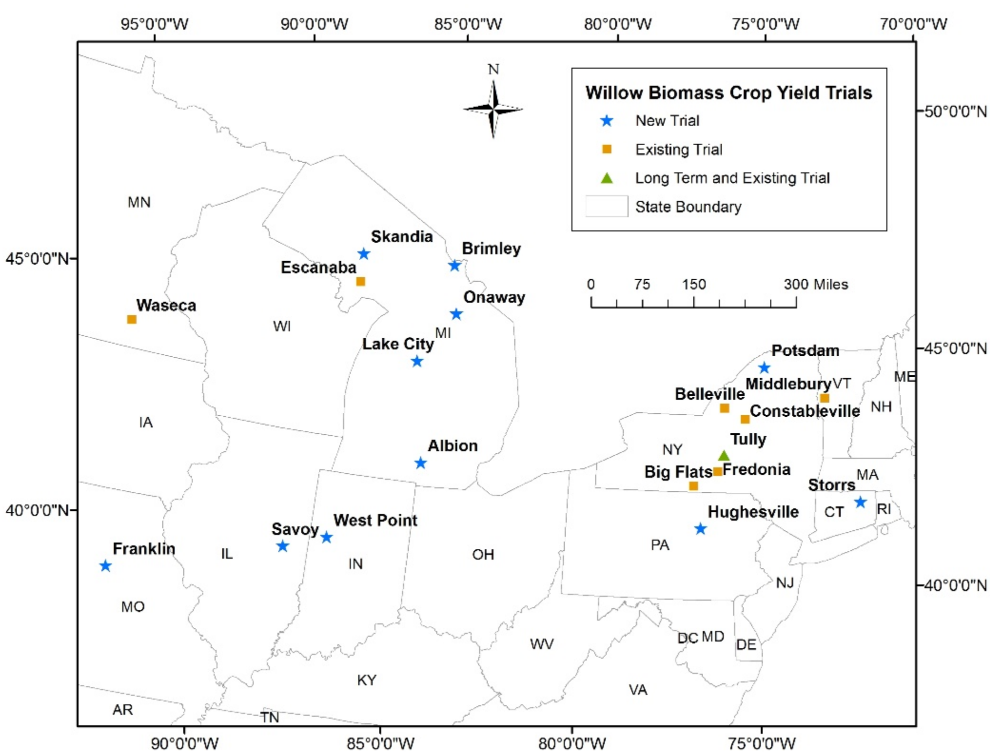

Figure 1. Map showing Sun Grant Regional Feedstock Partnership willow evaluation sites.

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Table 1. Existing and New Willow Biomass Crop Yield Trials Included in the Willow Biomass Crop Feedstock Project under the Sun Grant Feedstock Development Program

Trial name	Number of cultivars	Rotations harvested	Soil type[a]	Drainage[b]	Land capacity class[c]	NCCPI[d]
Existing willow biomass trials with older cultivars						
1993 Tully, New York	19	5, 6	Palmyra gravelly loam, 0% to 3% slopes	WD	1	0.48
1997 Tully, New York	32	4, 5	Palmyra gravelly loam, 0% to 3% slopes	WD	1	0.48
Existing willow biomass trials with new cultivars						
2005 Tully, New York	18	2, 3	Palmyra gravelly loam, 0% to 3% slopes	WD	1	0.48
2005 Belleville, New York	18	2, 3	Galway silt loam, 3% to 8% slopes	WD	2	0.39
2006 Constableville, New York	30	1, 2	Empeyville loam, 3% to 8% slopes, stony	MWD	2	0.24
2007 Middlebury, Vermont	30	1, 2	Vergennes clay, 2% to 6% slopes	PD	4	0.49
2006 Waseca, Minnesota	24	1, 2	Nicollet clay loam, 1% to 3% slopes	PD	2	0.80
2008 Big Flats, New York	6	1, 2	Unadilla silt loam, 0% to 3% slopes	WD	1	0.56
2008 Fredonia, New York	28	1, 2	Lordstown channery silt loam, 5% to 15% slopes	WD	3	0.34
2008 Escanaba, Michigan	26	1, 2	Onaway-Ossineke fine sandy loams, moraine, 1% to 6% slopes	MWD	2	0.35
New trials established in 2009						
2009 Sault Ste Marie (Brimley), Michigan	20	1	Rudyard silt loam, 0% to 3% slopes	MWD	2	0.32
2009 Skandia, Michigan	20	1	Munising fine sandy loam, 1% to 12% slopes, dissected	MWD	3	0.17
2009 Potsdam, New York	16	1	Adjidaumo silty clay	PD	4	0.40

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Trial name	Number of cultivars	Rotations harvested	Soil type[a]	Drainage[b]	Land capacity class[c]	NCCPI[d]
2009 Storrs, Connecticut	20	1	Woodbridge fine sandy loam, 3% to 8% slopes	WD	2	0.36
New trials established in 2010 and 2011						
2010 Savoy, Illinois	20	1	Catlin silt loam, 2% to 5% slopes	MWD	2	0.78
2010 West Point, Indiana	20	1	Troxel silty clay loam, 0% to 2% slopes	WD	1	0.94
2010 Onaway, Michigan	20	1	Detour flaggy loam, 0% to 3% slopes	SPD	2	0.26
2010 Lake City, Michigan	20	1	Emmet-Montcalm complex, 0% to 6% slopes	WD	2	0.44
2011 Albion, Michigan	20	1	Hillsdale sandy loam, 0% to 6% slopes	WD	3	0.61

a. *USDA National Resources Conservation Service (NRCS) Soil Survey Geographic database soil classification*
b. *USDA NRCS soil drainage classes: WD—well drained, MWD—moderately well drained, SPD—somewhat poorly drained, PD—poorly drained*
c. *Land Capacity Class rates land on a scale of 1 (few limitations to agriculture) to 8 (unsuitable for agriculture). Under good management, soils from class 1 to 4 are capable of producing common field crops and pasture without reducing the soil's long-term productivity. Soils 5 to 8 have limited value for commercial plant production but may be suitable for use as pasture, range, or forestland, and may also provide opportunities for recreation, wildlife, and water supply.*
d. NCCPI—National Commodity Crop Production Index. NCCPI is a model that interprets soil, landscape, and climate data to reflect soil's inherent capacity to produce dryland (nonirrigated) commodity crops.

Results and Outcomes

First-rotation yields were generated for a wide variety of cultivars across sites with a range of conditions across a broad geographical range. For trials planted after 2005, the yield of the top-producing, newer cultivars at the end of the first rotation ranged from 3.6 (Potsdam, New York) to 15.9 dry Mg ha^{-1} (Storrs, Connecticut). The mean across the sites was 10.6 $\pm$ 1.1 dry Mg ha^{-1} yr^{-1}. The current recommendation for large-scale plantings of willow biomass crops is that multiple cultivars should be planted at each site to minimize risk. Therefore, reporting yields of the top three or top five cultivars at each site is more representative. The yield of the top three cultivars across the sites ranged from 2.9 (Potsdam, New York) to 14.2 (at Middlebury, Vermont, and Storrs, Connecticut) dry Mg ha^{-1} yr^{-1}. The mean across the sites was 9.7 $\pm$ 0.9 dry Mg ha^{-1} yr^{-1}. The yield of the top five cultivars across the sites ranged from 2.7 (Potsdam, New York) to 13.9 (Middlebury, Vermont) dry Mg ha^{-1} yr^{-1} with a mean across all the sites of 9.2 $\pm$ 0.9 dry Mg ha^{-1} yr^{-1}.

Willow biomass crops are cultivated in a perennial system that is typically harvested multiple times on 3- or 4-year rotation cycles, and they have projected lifespans of over 25 years. However, data on the long-term production potential of willow biomass crops are very limited. The trials in this project have provided valuable results on the production of willow over multiple rotations. One trial planted in 1993 in Tully, New York, was maintained as part of this network and has now been harvested six times, providing the longest continuous set of yield data from a shrub willow trial in North America. While many of the cultivars planted in this trial have been replaced with more productive cultivars, one cultivar ('SV1') has been used for many years in both trials and large scale plantings of willow and is present in all the trials in this project. Over six rotations, the yield of 'SV1' ranged from 9.0 dry Mg ha^{-1} yr^{-1} in the first rotation to 15.3 dry Mg ha^{-1} yr^{-1} in the fourth rotation. In the sixth rotation, the yield decreased to 11.2 dry Mg ha^{-1} yr^{-1} but was still 26% greater than the first-rotation yield. Across all six rotations, the average annual yield was 12.4 dry Mg ha^{-1} yr^{-1}. A 12-year-old trial in Michigan compared poplar and willow hybrids under multiple 3-year harvest cycles and determined that while poplar initially thrived, it could not withstand repeated harvests as well as willow. These long-term data begin to provide verification that willow can be productive over multiple rotations and provide a basis for modeling these systems over 25 or more years.

This network of field trials with the large number of cultivars has provided essential information on potential yield increases associated with breeding and selection efforts. Yield increases associated with new cultivars have typically ranged from 15–25%, with some variation across sites. The broad range of sites included in this project has provided a valuable basis for understanding factors that influence willow production and genotype-by-environment interactions (Serapiglia et al. 2013). The factors that have greatest impact on yield can include temperature during the growing season, growing degree days, and regional pest pressure. Despite the heavy influence of site conditions on overall yield, some important patterns have emerged, including evidence that triploid hybrids, such as *Salix viminalis* x *S. miyabeana*, have demonstrated consistently greater yields compared to a range of other taxonomic groups. Since breeding and selection work is still at an early stage for willow, these results suggest that significant gains can still be realized by developing improved cultivars. Data from these sites, along with data from a number of other earlier trials, formed the data base for the development of models to predict the regional yields of willow that are used in the *2016 Billion-Ton Report* (DOE 2016).

Findings from a subset of trials in this network provided important data for a life-cycle analysis that was completed for willow biomass crops. This analysis included all activities beginning with site preparation, harvesting, and delivery of biomass to an end user. This analysis included seven 3-year harvest cycles. Uncertainty analysis was conducted on key variables including yield results from these trials. The GHG emissions from this study were negative for all scenarios (-125 to -48 CO_{2eq} per dry ton) (Caputo et al. 2014) when measurements of belowground biomass were included (Pacaldo et al. 2014). The net energy ratio of biomass delivered to an end user ranged from 18.3 to 43.4, meaning that for every unit of fossil energy invested in the production, harvest, and delivery of willow biomass, there are 18–43 units of stored energy in the willow chips delivered to the end user.

Wood samples collected at first- and second-rotation harvests from a number of these trials have been analyzed for specific gravity and biomass composition via high-resolution thermogravimetric analysis. There are significant differences in biomass composition by genotype, by site, and with significant genotype-by-environment interactions. There are significant positive correlations between yield and cellulose content, with negative correlations between yield and lignin content (Serapiglia et al. 2013; Fabio et al. in prep.). It is known that genotypes and environmental conditions can dramatically affect efficiency of sugar release and potential for conversion to biofuels (Serapiglia et al. 2013; Brereton et al. 2012). Data from these trials will vastly improve our understanding of how environmental factors influence biomass quality and conversion efficiency.

For perennial crops like willow, projection of yields over two or more decades is an important factor that influences key attributes, including the economic viability of these systems. The yield data collected across a range of sites and over multiple rotations as part of this project have provided a solid foundation for improving economic models of this system. Yield data from this network of trials were used to model returns from willow biomass crop systems using a cash flow model developed at the State University of New York College of Environmental Science and Forestry that was updated and improved in 2014 (Buchholz and Volk 2011, 2013; Heavey and Volk 2015). Yield data from this network of trials, and seven other additional trials outside of the network, were used to develop yield models using PRISM-ELM (Daly et al., 2018) across multiple regions of the United States (Figure 2). These yield results, along with production, management, and harvesting costs from EcoWillow 2.0 (http://www.esf.edu/willow/), will have been used in POLYSYS (De La Torre Ugarte and Ray, 2000) for the *2016 Billion-Ton Report* (DOE 2016).

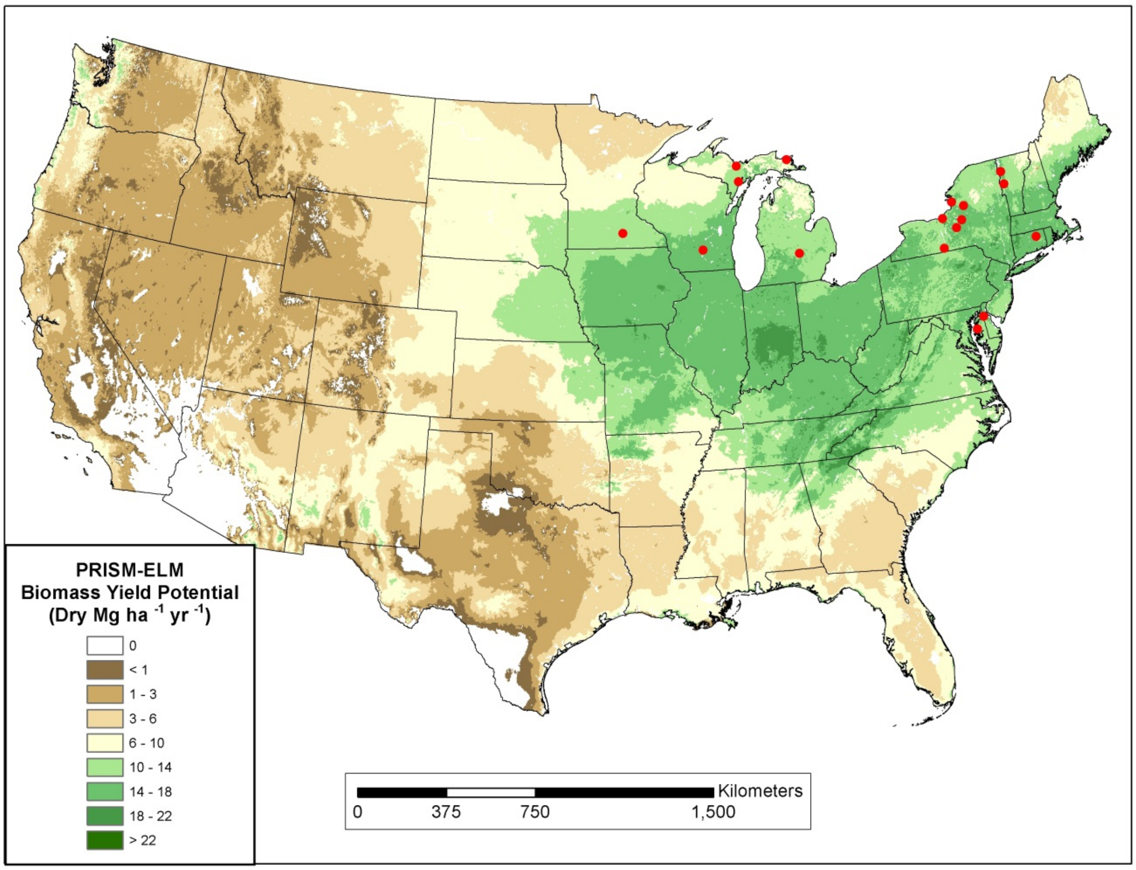

Figure 2. Potential willow biomass yields for the U.S. generated from the PRISM-ELM model and data from a network of 17 field trials (indicated by red dots on the map) (from Volk et al., 2017).

Two important barriers to the large-scale deployment of willow biomass crops include a stable and reliable market for the biomass and the overall economics of the system. As noted above, there is ongoing expansion of willow biomass production occurring in northern New York, with a commitment from ReEnergy to purchase all the willow biomass that is being grown in the area over an 11-year period. While the price that ReEnergy currently pays for wood chips would make it difficult to justify growing willow from a purely economic point of view, the support for landowners to plant willow biomass crops from the USDA Biomass Crop Assistance Program (BCAP) makes growing willow an economically viable option for the producer. The development of a long-term market and support to reduce upfront costs has made the expansion of willow in northern New York a reality. Another key barrier stems from the misperceptions about willow biomass that already exist among landowners and potential growers, as well as potential end users.

These misperceptions often stem from a common understanding that willow trees are a poor source of firewood. There is a perception that because willow trees tend to grow in wet areas that the wood is wetter and has less energy than other hardwoods. In reality, on a weight basis the moisture and energy content of freshly harvested willow is the same as other hardwoods like beech or maple (Eisenbies et al. 2014). The main difference between willow and other hardwoods is that willow wood density is 25%–35% lower than other hardwoods, which contributes to many of the perceptions about willow as a source of firewood. In terms of yield, willow can produce 5–10 times more biomass per hectare per year than surrounding natural forests, so willow stores more solar energy as biomass on each hectare of land. An additional misperception present among end users is that the particle size distribution of willow biomass is inconsistent and causes problems with feed systems at heating and power plants. This opinion is often based on some bad experiences that occurred years ago during initial trials with willow biomass that was produced using experimental harvesters. The chips from these machines were inconsistent and did cause problems in feed systems. These negative experiences have been widely discussed among plant operators over the years. However, recent developments in harvesting technology, by New Holland for example, have addressed this issue, and the material that is now being delivered to end users has a consistent quality. In the past few years, more than 4,000 Mg of willow biomass has been delivered to end users to generate heat and power. While there was initial skepticism about this fuel source among some plant operators, experience with many loads of willow biomass has shown them that willow is a good quality fuel that is easily mixed with other sources of woody biomass and used in their facilities without any serious problems.

This network of willow yield trials has provided locations where other studies are either underway or have been completed. Without this network of sites and the support to maintain these sites over an extended period of time, these studies would not have been possible. In New York, these related studies include assessments of belowground biomass, changes in soil carbon over time, characterizations of willow so the Revised Universal Soil Loss Equation can be used to estimate erosion potential under willow, measurements of sap flow in willow, assessments of fine root dynamics in willow, economic assessments of willow biomass crops, and examinations of genotype-by-environment interactions in willow and variations in willow compositions across a range of sites. In Connecticut, data generated from willow yield trials were used as the basis for developing other projects, including a study called “Agroforestry Riparian Project for Biofuel and Environmental Benefits.” This project is now using willow to restore a riparian buffer that impacts a number of ecosystem services including nitrogen and phosphorus runoff, erosion, and sedimentation. Yield trial data formed the foundation for the development of a genotype-by-environment trial of several willow varieties grown on different microsites with different soil attributes. The sites in Michigan were employed to conduct an investigation of the GHG and nitrogen impacts of changing land use from pastureland to short-rotation woody bioenergy crops

(Nikièma et al. 2012). Materials from these sites have also been supplied to a variety of other projects seeking to better understand variability of physical and chemical feedstock characteristics.

The network of trials has provided many unique opportunities to highlight willow biomass crops in different communities. Many of the sites were used for field days and extension and education activities, which has been important for the development and expansion of willow biomass crops. Two trials in northern New York in particular (Belleville and Constableville) were essential in the successful application for a USDA Biomass Crop Assistance project area. The trial at Belleville was established on school property, and the Future Farmers of America club at the school engaged in planting and monitoring the willow crop. The site was used for field days and for one of the first public demonstrations of a New Holland forage harvester being used to cut and chip willow stems. Similarly, the yield trial at the USDA NRCS Big Flats Plant Materials Center was highlighted at an annual field day event for several years, with over 100 participants each year. The data from these yield trials provided essential background information that was needed for this application and also provided key locations for landowners and potential growers to see willow biomass crops firsthand. Despite the fact that the sign-up period for this Biomass Crop Assistance Program project area was limited to about a six-week period, just over 473 ha were enrolled. Without the presence of these yield trials, this project would not have been successful, and this commercial expansion of willow biomass crops in the United States would not have been possible.

The data from the network of trials, and especially the data from the trials with newer cultivars, have provided invaluable information for a commercial nursery partner in western New York—Double A Willow—to make decisions about which cultivars to plant in its nursery beds. Currently Double A Willow has about 67 ha of commercial nursery beds and is providing willow planting stock for various projects in the United States, with subcontracts to nurseries in Canada. The data from the yield trials are important for making decisions about what to plant in nursery trials because it takes several years before these nursery beds are productive.

Key outputs

Peer-Reviewed Publications

Caputo, J., S. Balogh, T. A. Volk, L. Johnson, M. Puettman, B. R. Lippke, and E. Oneil. 2013. “Incorporating Uncertainty into a Life-Cycle Analysis (LCA) Model of Short-Rotation Willow Biomass (*Salix* spp.) Crops.” *Bioenergy Research* 7 (1): 48–59. doi:10.1007/s12155-013-9347-y.

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Where do we go from here?

Maintaining a subset of these trials is important for monitoring some of the new cultivars over multiple rotations to provide data on their performance. In addition, findings from some trials with cultivars from breeding efforts that have been conducted over the past few years are beneficial for continuing to improve the genetic material that is available for future deployment. As willow crops are deployed on a commercial scale, it becomes especially valuable to conduct focused monitoring across large fields. This would provide valuable data on the economics, production, and sustainability of willow biomass crops at a much larger scale and provide an opportunity to optimize various parts of the system.

At the beginning of this project, willow biomass crops were limited to a small network of yield trials and a few scattered larger-scale demonstration plantings. This project has supported an important expansion of the network of yield trials and has enabled researchers in a number of regions to leverage this support for other projects and initiatives. As noted above, this network of trials has provided key data for the expansion of willow biomass production in northern New York.

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National Resources Assessment
PRISM Environmental Limitation Model (PRISM-ELM)

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Summary

As part of the Sun Grant program, a Regional Feedstock Partnership was developed to assess biomass feedstock potentials, conduct field trials, and estimate the nation's bioenergy production potential. Many new crops identified as potential feedstocks have little production history in the United States. A modeling and mapping system called PRISM-ELM (PRISM Environmental Limitation Model) (Daly et al., 2018) was developed to help answer the basic question: In the absence of detailed, quantitative information on the environmental limitations of a given feedstock, what is the spatial distribution of the major climate and soil constraints that limit biomass production for better characterized crop species? PRISM-ELM employs a limiting factor approach, where the final result is the lowest relative yield resulting from submodels that simulate water balance; winter low temperature response; summer high temperature response; and soil pH, salinity, and drainage. PRISM-ELM relative yield (0-100%) grids are transformed into actual yield potential maps through national linear regressions relating relative yield to reported yield data from field trials. An important component of the modeling process is close interaction between Sun Grant agronomists conducting the field trials and the PRISM-ELM modeling group. The model was parameterized and validated using grain yield data for winter wheat and maize. Mean absolute errors between predictions and observations for winter wheat and maize were 16 and 17 percent, respectively. The unexplained variability in yield may have been caused by differences in the environmental limitations of grain production versus biomass, and the many economic and management decisions not simulated in PRISM-ELM. Model settings for these well-known crops were used as anchor points for the parameterization and mapping of the production potential of herbaceous feedstocks: upland and lowland switchgrass, energycane, giant miscanthus, biomass sorghum, and conservation reserve program grasses; and woody feedstocks: willow, poplar, and southern pine. This set of maps, modeled in a consistent manner, and with expert input and data from Partnership agronomists, form the basis for economic and comparative analyses in the *2016 Billion-Ton Report* (U.D. DOE, 2016).

Introduction

As part of the Sun Grant program, a Regional Feedstock Partnership was developed to assess biomass feedstock potentials, conduct field trials of the more promising potential energy crops, and estimate the nation's bioenergy crop production potential through the findings collected from this research and other data. To organize and display research trial data and map crop model predictions, a team was assembled from several university and federal GIS (Geographic Information System) centers. The PRISM Climate Group at Oregon State University (http://prism.oregonstate.edu/) participated in this effort.

Biofuel crops have become a subject of national focus, with several new crops identified as potential feedstocks. Traditional crops such as wheat and maize provide residues that can serve as biofuel feedstocks, and have long production histories and rich knowledge bases with regard to physiology, production, and spatial distribution. However, many new crops identified as potential lignocellulosic feedstocks have little production history in the United States. It is not surprising, then, that planners tasked with assessing farming, transportation, processing needs, and infrastructure for new crops are asking the basic question: Where can these new crops be raised successfully and what kind of production can be expected within a given geographic region?

Estimating crop yield potential and mapping on a landscape scale differs from crop simulation modeling that is focused on specific site predictions. Landscape-level potential yield maps of crops being considered for meeting biofuel feedstock needs will allow for comparison of crops on a biomass production basis and economic analyses when conversion efficiencies are applied to raw yield estimates.

The focus of this mapping work was on nationally important feedstocks. Herbaceous feedstocks include upland and lowland switchgrass (*Panicum virgatum* L.), miscanthus (*Miscanthus* X *giganteus*), energycane (*Saccharum officinarum L.* X *Saccharum spontaneam L.*), biomass sorghum (*Sorghum bicolor*), and mixed Conservation Reserve Program (CRP) grasses. Woody feedstocks include willow (*Salix* spp.), poplar (*Populus* spp.), and southern pine (*Pinus taeda*).

Objectives

A major objective of the Sun Grant geographic information system component is to gain an understanding of the spatial distribution of current and potential biofuel/bioenergy crop resources across the country. To that end, specific objectives of this work were to: (1) develop a spatial model (PRISM-ELM) that estimates the production potential of biofuel feedstocks across the conterminous United States based on climate and soil conditions; (2) calibrate and validate the model with yield data from well-known crops and use these model settings as anchor points for the feedstock analysis; (3) establish a mapping approach for biomass feedstocks that emphasizes close interaction between the modeling and agronomic components of the Feedstock Partnership; and (4) produce long-term production potential maps to be used in economic and comparative analyses.

Methods

Model Description and Inputs

For a detailed peer review of PRISM-ELM, see Daly et al. (2018).

Attempts to estimate the potential spatial distribution and yield of new bioenergy crops have taken two main approaches: (1) empirical models based on field data and (2) application of mechanistic plant growth models (Jager et al. 2010; Nair et al. 2012). Commonly used empirical approaches involve statistical extrapolation of plot/field-level yield data to larger regions and climatic envelope modeling (e.g., Casler et al. 2007; Barney and DiTomaso 2010; Schmer et al. 2009; Araya et al. 2010; Jager et al. 2010; Wullschleger et al. 2010; Tulbure et al. 2011).

Plant growth models attempt to simulate the important physiological processes that affect growth, development, and yield. Most plant growth models simulate photosynthesis, carbon allocation, phenology, biomass production, and root/shoot partitioning. Examples of simulation models include EPIC (Williams et al. 1984; Brown et al. 2000), ALMANAC (Kiniry et al. 2008), and MISCANFOR (Hastings et al. 2009; Miguez et al. 2011).

Developing potential yield maps requires an understanding of how these plants respond to spatially variable growing conditions of climate and soils. Often modelers operate independently of agronomists, but an important component of this modeling process is the close interaction between agronomists conducting the yield trials for a given crop, and the modeling group.

In response to the need for an environmental assessment tool, a modeling and mapping system called PRISM-ELM (PRISM Environmental Limitation Model) was developed. PRISM-ELM draws from both statistical-empirical and crop growth modeling approaches, while keeping the modeling system very simple and universal so that assessments for a wide range of crops can be made in a consistent manner over large areas. PRISM-ELM stems from earlier work to estimate the suitability of U.S.-grown perennial grasses in China (Hannaway et al. 2005). The basic question PRISM-ELM was developed to answer is: In the absence of detailed, quantitative information on the climatic and soils tolerances of a given crop, what is the spatial distribution of the major environmental constraints that limit the biomass production of the crop? The interest here is only in general climatic and soil constraints on biomass production of any crop, rather than a detailed accounting of phenology, flowering, grain development, etc., of a specific species; this calls for a modeling system that is highly simplified and generalized. Environmental limitation maps estimated by PRISM-ELM are transformed into actual yield potential maps through linear statistical regressions which use the modeled environmental limitations and the yield data from field trials.

The centerpiece of PRISM-ELM is a semi-monthly Food and Agriculture–style water balance simulation (Figure 1), which tracks precipitation input, evapotranspiration, and soil moisture depletion (Allen et al. 1998). An estimate of monthly relative yield (0%–100%) is the product of the water stress coefficient and a temperature growth curve. In what is known as a "limiting factor" approach, the final relative yield is the lowest of the modeled yields resulting from the water balance simulation, plant injury curves for summer heat and winter cold, and growth constraints due to soil pH, drainage, and salinity.

Climate inputs of temperature and precipitation are provided to PRISM-ELM on a semi-monthly basis using 800-m resolution gridded daily data from the PRISM climate mapping system. PRISM datasets serve as the USDA's official 30-year "normal" digital climate maps (Daly et al. 2008; PRISM Climate Group 2015).

The water balance model uses PRISM precipitation (*P)* to determine total available water (*TAW*) in the soil profile (Halbleib et al. 2012) (Figure 1). Available soil water holding capacity (*AWC*) is estimated from the USDA NRCS U.S. General Soil Map Coverage (NRCS 2016), and the depth of the rooting zone (*Droot*) is defined by the user. PRISM monthly average temperature (*T*) is used to estimate potential evapotranspiration (*ETo*). Actual evapotranspiration (*ETa*) is a function of *ETo*, a water stress coefficient (*Ks*), the plant's water use efficiency (*Kc*, user-defined), and the root zone moisture depletion (*Dr*), which is the difference between the plant's moisture demand and the soil water supply. *ETa* in a given time interval reduces the next time interval's soil water supply, which is at least partially replenished by precipitation. At the end of each time interval, *Ks* is calculated as the difference between *TAW* and *Dr*. Relative yield for that interval is the product of *Ks* and a user-defined temperature growth response function, which defines the relationship between temperature and relative production for that crop.

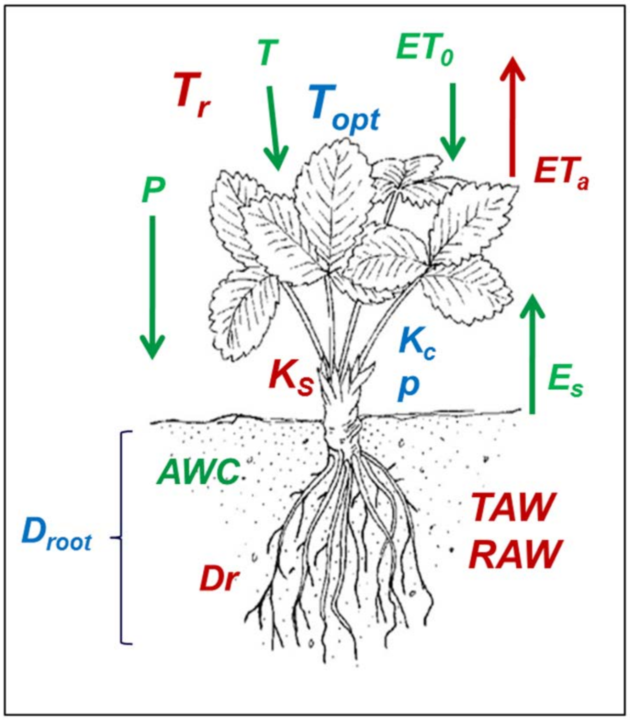

Figure 1. Schematic of the PRISM-ELM water balance model. Items in green are provided externally as spatially-varying (gridded) inputs. Blue items are crop-specific scalars provided as external inputs. Red items are calculated internally by the model and are spatially varying.

The output of the water balance model is a relative yield estimate ranging from 0 to 100% for each month (shown, for example, as RY_m in Figure 2). The user specifies a potential growth period, which is the range of months in which production is likely to occur across the modeling region. In the example in Figure 2, the potential growth period is March–October. The user also specifies the number of sequential months within the potential growth period over which maximum production is likely to occur. Relative yield values are averaged over these months to obtain a final water balance yield. For example, if the period of significant biomass accumulation is typically 3 months, the user would input N = 3, as shown in Figure 2. This maximum growth period is allowed to “float,” meaning the model will use the 3-month sequence with the highest average relative yield as the final water balance yield, to accommodate varying growing season timing under differing climates.

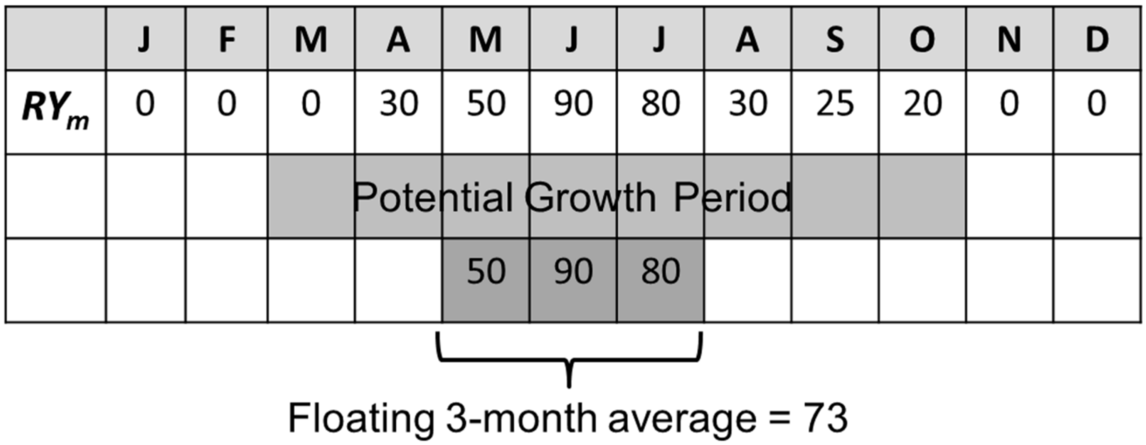

Figure 2. Example of the method used to calculate final water balance yield. This example assumes the period of significant biomass accumulation is typically 3 months; therefore, the model will use the 3-month sequence with the highest average relative yield as the final water balance yield.

The winter temperature constraint simulates a perennial crop's ability to tolerate and survive winter low temperatures. A two-tailed temperature response function relates the PRISM average January minimum temperature to expected damage or mortality on the cold tail and, if needed, loss of production due to inability to meet winter chilling requirements on the warm tail. The summer temperature constraint simulates a crop's ability to tolerate and survive average summer high temperatures. A single-tailed temperature response function relates the PRISM average July maximum temperature to expected damage or mortality and resultant loss of production.

The soil constraint function for soil pH uses a two-tailed curve that can be broadened or narrowed based on expected plant response to pH, and to accommodate application of amendments such as lime to raise the pH of acidic soils. The soil constraint function for salinity uses a one-tailed curve that represents growth reduction due to increasing soil salinity. The soil constraint function for drainage is based on the seven soil drainage classes as defined by the NRCS U.S. General Soil Map Coverage, ranging from very poorly drained to excessively drained. The expected plant response for each drainage class can be set individually, ranging from 0 (full constraint) to 100 (no constraint). Drainage class responses can be modified to account for field tiling and surface drainage to improve poorly drained soils.

The final relative yield is calculated as the *lowest* yield resulting from any of the constraint functions: water balance, winter low temperature, summer high temperature, soil pH, soil drainage, and soil salinity. Model output is in the form of a regularly spaced grid at a native 800-m resolution with an estimate of relative yield from 0 to 100%.

A land use grid can be applied to the relative yield map to mask out land use types that are not classified as agricultural, such as forests, deserts, parks, cities, roads, etc. A useful source of land use coverage is the NASS National Cultivated Data Layer (https://www.nass.usda.gov/Research_and_Science/Cropland/Release/), which is derived from annual remote sensing of agricultural crop lands for the continental USA.

The relative yield map is transformed into an actual yield map by developing statistical relationships between relative and actual yield using available field data. The transformation can be as simple as setting 100% relative yield to a maximum expected biomass yield and scaling the map accordingly, or as complex as using *in situ* yield reports to develop spatially varying relationships across the country.

Calibration and Validation

PRISM-ELM was initially calibrated and validated for two well-known crops for which substantial research and yield data were available (and have also been identified as sources of biomass for bioenergy conversion): winter wheat, an annual, overwintering crop with a C3 (cool-season) photosynthetic pathway; and maize, an annual crop with a C4 (warm-season) photosynthetic pathway. The process of setting of PRISM-ELM input parameters drew on three main types of information in an iterative fashion: (1) published literature on the quantitative tolerances of winter wheat and maize to environmental conditions; (2) the degree of adherence of resulting PRISM-ELM relative yield maps to known spatial patterns and limits of crop production; and (3) quantitative relationships with yield data from the USDA Risk Management Agency (RMA). Producers participating in the federal crop insurance program are required to report acreage planted and harvested yield, and these data are aggregated by RMA to produce county-level statistics. Counties that had at least thirty reports with a non-irrigated practice code for at least ten years during the period 2000-2015 were included in the analysis.

The RMA yield dataset was randomly divided into a training half and an evaluation half (fifty percent data withholding). The training half was used in the initial parameterization of PRISM-ELM and model performance assessed. Using the same parameter settings, PRISM-ELM was then applied to the evaluation half of the data, and finally to the entire dataset, with model performance assessed at each step.

To gain an understanding of the uncertainties associated with yield data, county-level yield data for winter wheat and maize were also obtained from the National Agricultural Statistics Service (NASS) of the USDA (https://quickstats.nass.usda.gov/). NASS yield data are collected through voluntary surveys, rather than being programmatically mandated. NASS data were available through 2012 at the time of the analysis, so data for the years 2000-2012 were used. Counties reporting with a non-irrigated practice code in at least eight years during 2000-2012 were selected for use; it was not possible to adhere to the ten-year rule as was done for the RMA data, because very few counties reported that frequently, especially for maize. The thirty-records-per-county criteria could not be implemented because NASS does not provide the number of records per county.

To compare the PRISM-ELM grid cell and RMA and NASS county-level yields, all PRISM-ELM grid cells within each county that were designated as cultivated were averaged to produce county-level values. The 2013 USDA NASS National Cultivated Layer was used to identify areas under cultivation.

Modeling Assumptions

One of the first tasks in the modeling process for bioenergy feedstocks was to identify what the yield potential maps would represent, and list the associated assumptions. The consensus among the participants in the Feedstock Partnership was that the PRISM-ELM estimated yield maps would show the long-term potential yield for a given crop at the field level, assuming best management practices. Specifically, the yield potential maps would represent:

- Long-term average yields based on climatological data for the period 1981-2010, not accounting for occasional damaging events, such as hail, flooding and wind
- Dryland conditions (non-irrigated)
- Yields from the best local cultivar available at the time of the yield trials
- Once-per-year harvest frequency (no summer fallow practices); estimated annual increment for woody perennials based on total biomass accumulation across multiple years.
- Field-scale yields, as opposed to test plot-scale yields (accounts for “yield gap”)
- Yields of fully established crops, if perennials; establishment years not included
- Best-practice fertilizer application using a mass balance approach for local soil type
- Best-practice pesticide application, typically minimal inputs

Sun Grant yield trials were conducted in a coordinated fashion, and site specific information for soils, crop management, fertility, and harvest methods were recorded for each location. This provided the opportunity to control for these variations across sites by selecting trials that were most consistent with mapping assumptions. Given that management practices greatly influence yields, controlling for these practices allowed the modeling work to focus on how climate and soil constraints influence yield distribution patterns.

Feedstock Modeling Approach

An overview of the modeling process for mapping bioenergy feedstock resources is shown in Figure 3. PRISM-ELM was provided with gridded climate and soils data, and a control file with crop-specific parameters. PRISM-ELM produced an initial grid of relative yield from 0-100%, where 100% represents no climate or soil constraints on growth and zero represents a full limitation. For a given crop, yield data from field trials conducted by Sun Grant agronomists were examined at face-to-face meetings with the modeling group. During this meeting, each yield data point was evaluated for adherence to the modeling assumptions presented above. The initial PRISM-ELM relative yield grid was used to provide a framework for evaluating the yield data. The goal of each meeting was to come to an agreement on which yield data points would be included in a national regression function relating PRISM-ELM relative yield to actual yield. This nationwide regression function allowed the PRISM-ELM relative yield grid to be transformed into an actual yield grid. The process of adjusting PRISM-ELM crop parameters and comparing the relative yield map to the observed data was done iteratively until a final solution was reached. The goal was to achieve the best agreement possible between PRISM-ELM and yield data, but within the constraints of model parameter values that were consistent with the type of crop being mapped.

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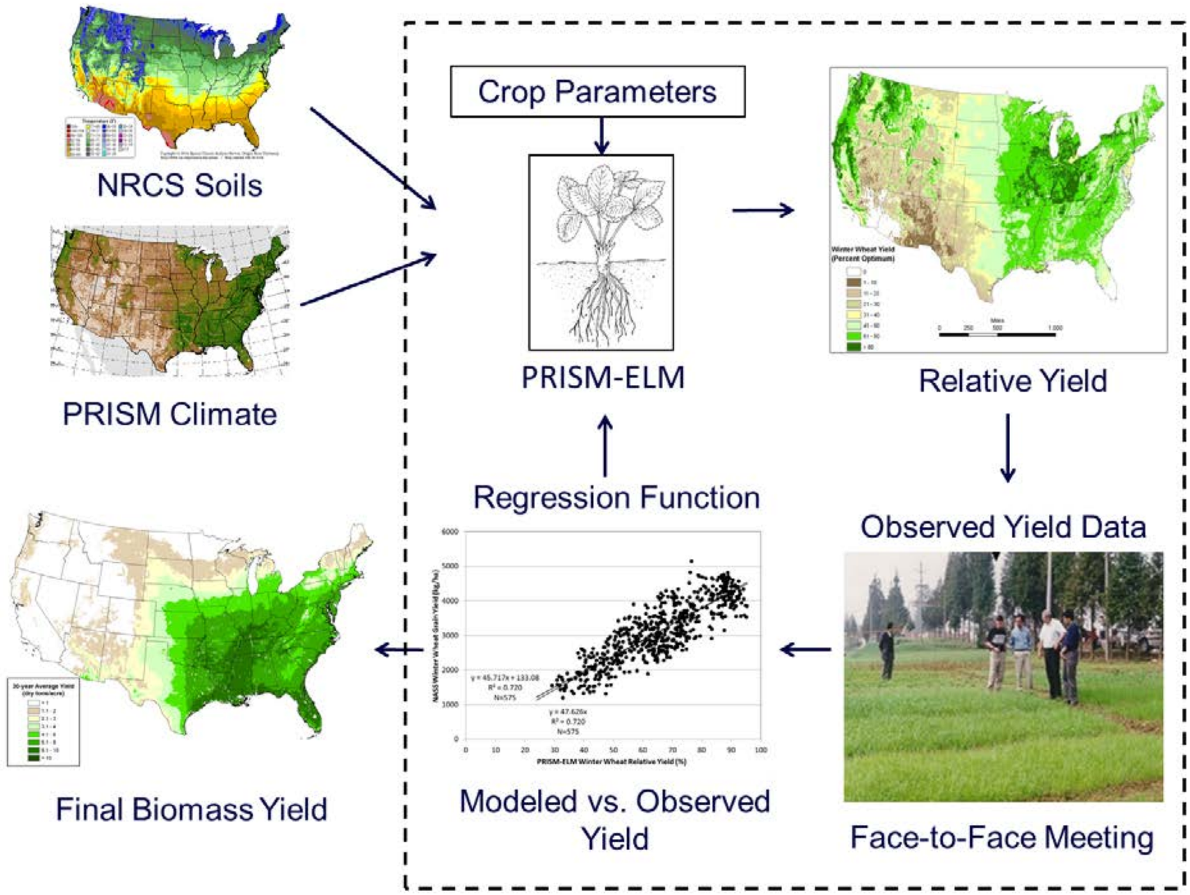

Figure 3. Schematic of the PRISM-ELM workflow for mapping bioenergy feedstock resources. Inputs to PRISM-ELM were gridded climate and soils data, and a preliminary parameter file for the crop to be modeled. An initial relative yield map was produced, and during a face-to-face meeting with agronomists, the relative yield map was evaluated against observed yield data to help screen for data outliers and adjust model parameters. Once a consensus was reached on model parameters and yield data to be used, a final regression function was developed and applied to the relative yield map to produce a final biomass yield map.

Results and Outcomes

Yield Data Characteristics

The spatial patterns of RMA and NASS yield data for non-irrigated winter wheat and maize are shown in Figures 4 and 5, respectively. RMA reports are more numerous than those from NASS, likely because the RMA requires yield reporting as a condition of participation in the federal crop insurance program, while NASS relies on voluntary survey responses. There were very few NASS maize reports in the non-irrigated category. It appears that the irrigation practice was unknown or not specified, because the NASS all-practice category had many more entries.

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However, the all-practices data contained many irrigated reports, which violated our assumption of dryland farm practice, and hence could not be used in the analysis.

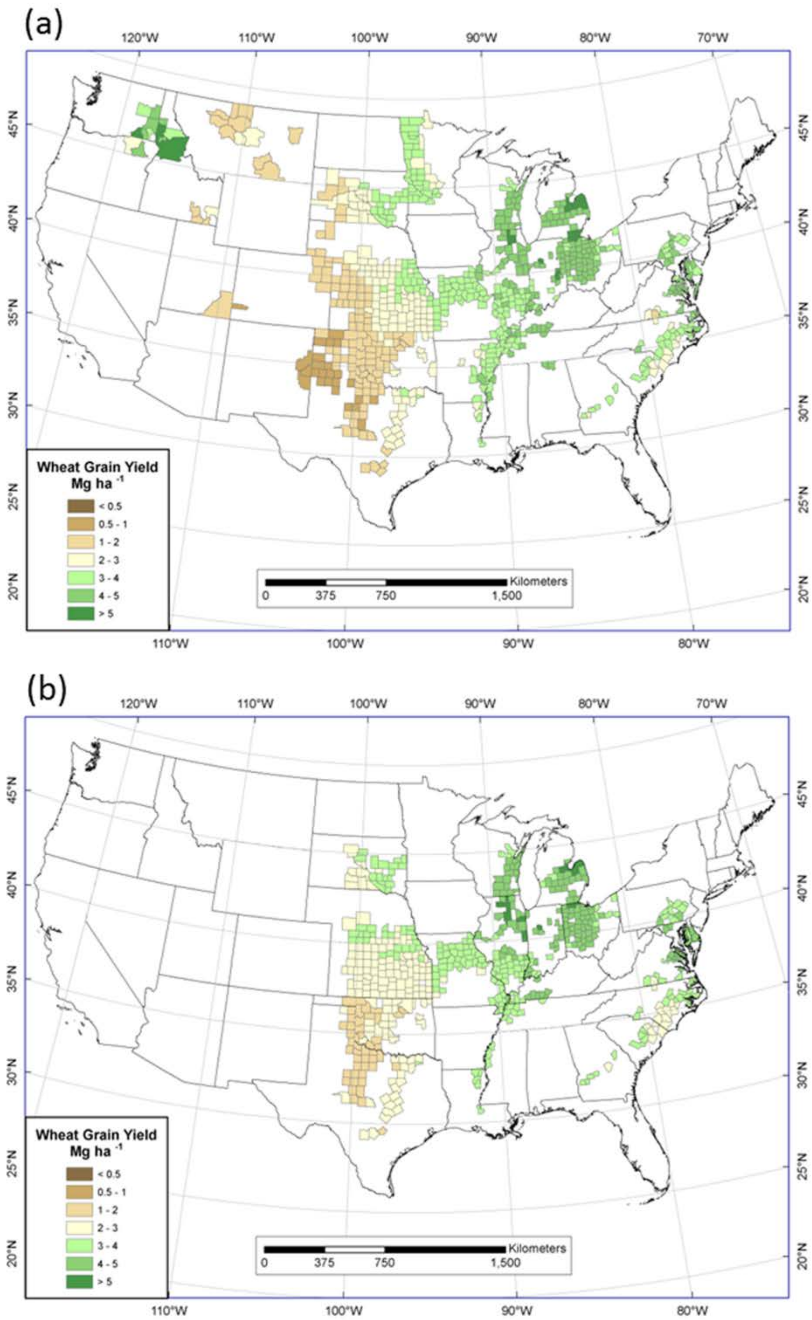

Figure 4. Spatial patterns of county-level yield data for non-irrigated winter wheat grain as reported by (a) RMA and (b) NASS. See text for county inclusion criteria.

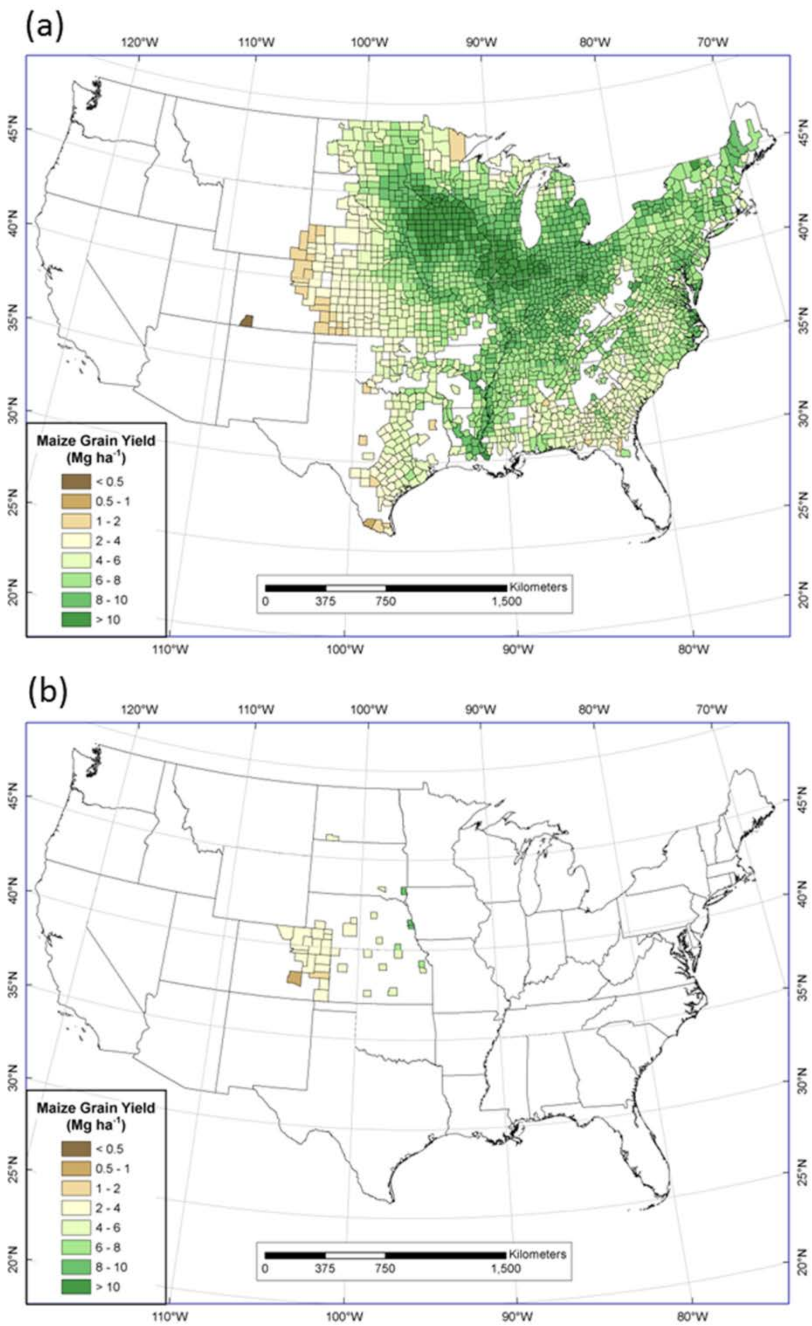

Figure 5. Spatial patterns of county-level yield data for non-irrigated maize grain as reported by (a) RMA and (b) NASS. See text for county inclusion criteria.

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Given the differing methods of data collection by NASS and RMA, and inconsistencies in county inclusion criteria, it is not surprising that RMA and NASS yield data differed somewhat (Figure 6). Agreement was good for winter wheat yields above about 3 Mg/ha, but below this level, RMA reported yields were lower than those from NASS (Figure 6a). These discrepancies were located primarily in western Kansas, Oklahoma, and Texas (Figure 4). Maize yield in the few counties reported by NASS agreed well with those reported by RMA, but RMA yields were slightly lower than NASS yields below about 4 Mg/ha (Figure 6b), again in the western plains states.

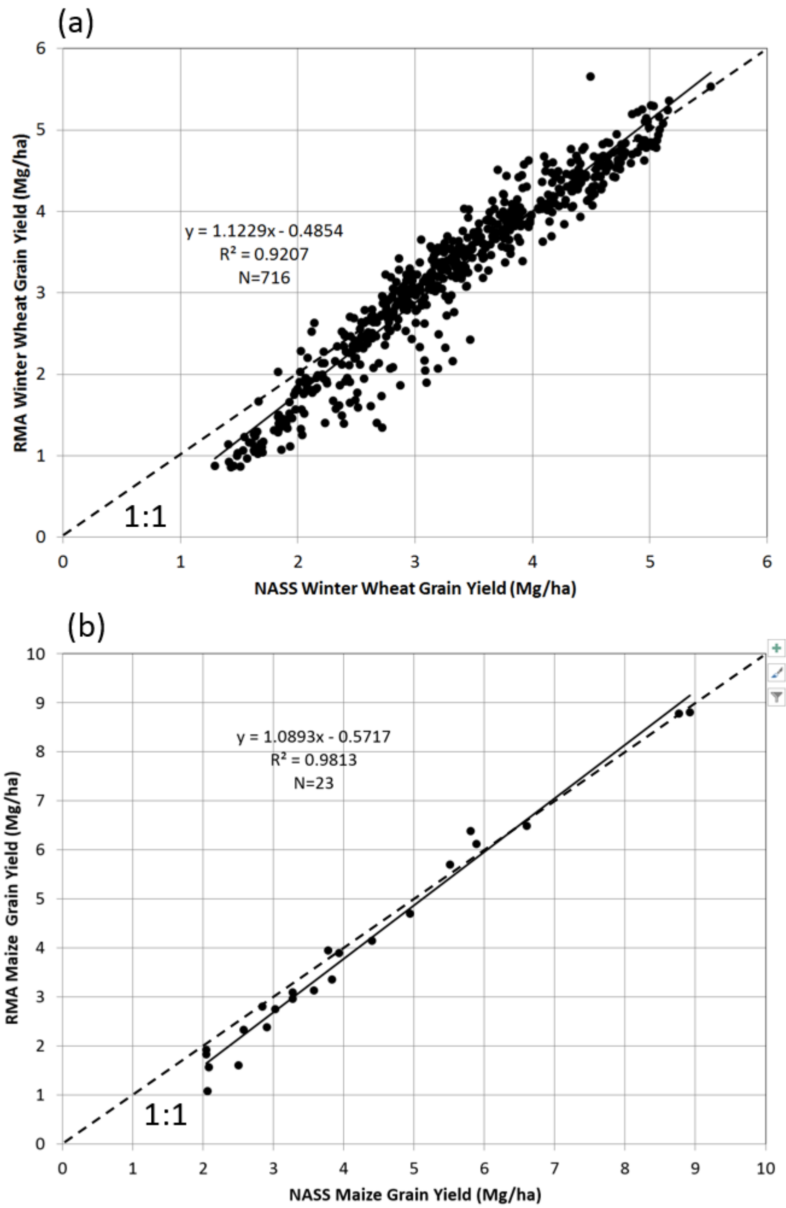

Figure 6. Comparison of county-level yield data for (a) winter wheat and (b) maize. Data points shown are from counties where both RMA and NASS reporting inclusion criteria were met. See text for details. (Adapted from Daly et al., 2018)

Model Results and Performance

PRISM-ELM relative yield maps for winter wheat and maize are shown in Figure 7. Wheat and maize relative yields were highest in the Midwest and eastern US, where temperature and precipitation conditions are generally adequate during their respective growing seasons. Wheat's cooler temperature optimum places its peak production period primarily in the spring compared to maize's summer production period. As a result, wheat production is less limited than maize on the West Coast, where seasonal precipitation is greater in spring than in summer.

The patterns of PRISM-ELM relative yield match those of the reported yields reasonably well where there are data (Figures 4 and 5). Both indicate maximum maize yields in the Iowa-Illinois-Indiana corridor, and relatively high yields in the northeast. Modeled and reported yields decrease along the east-to-west precipitation gradient in the plains in a similar fashion. Winter wheat data coverage is not as extensive, but maximum yields are also reproduced in the Midwest and east, as is a similar gradient in the plains. PRISM-ELM's relatively high yields in the Pacific Northwest are also corroborated by RMA data.

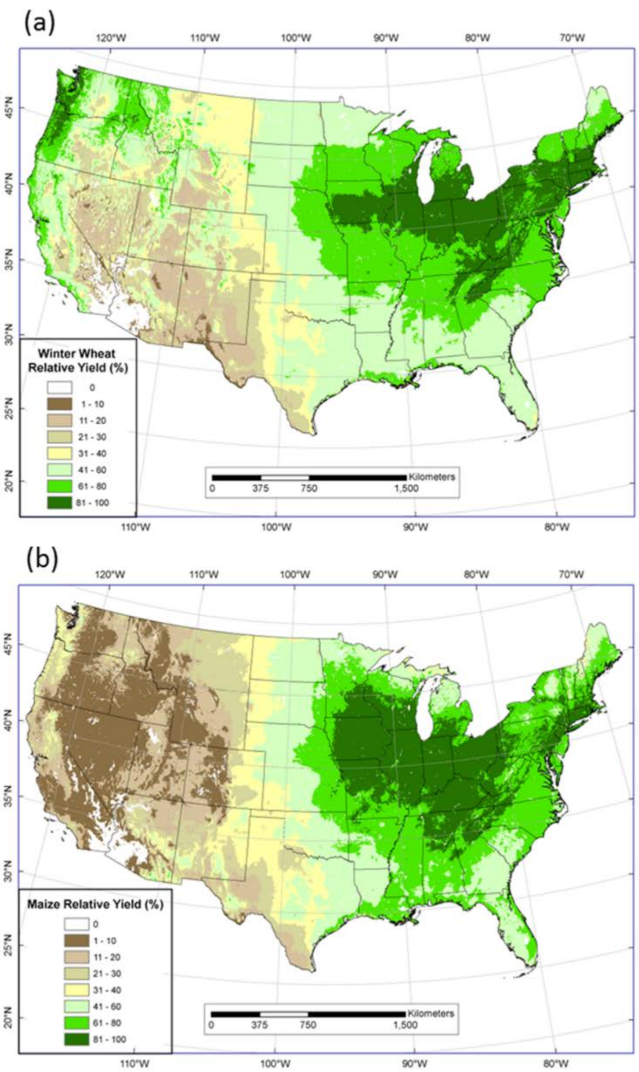

Figure 7. PRISM-ELM relative yield maps for (a) winter wheat and (b) maize.

The statistical relationship between PRISM-ELM relative yield and RMA average winter wheat and maize yields was fairly consistent for the training, evaluation, and full RMA datasets (Table 1). Winter wheat correlation coefficients (for a least-squares regression forced through zero) declined from 0.75 for the training dataset to 0.71 for the evaluation dataset; the mean absolute error (MAE), a measure of difference between two continuous variables, changed little, averaging about 16 percent. Correlation coefficients for maize were lower, ranging from 0.59 for the training dataset to 0.46 for the evaluation dataset. However, percent MAEs did not differ substantially from those of winter wheat, averaging about 17 percent.

Table 1. PRISM-ELM performance statistics for RMA winter wheat and maize yield. Comparisons were made after the PRISM-ELM relative yields were transformed into actual yields using national regression equations forced through zero. The dataset was divided into training and evaluation halves, with performance statistics calculated for each, as well as for the entire dataset. Model parameter settings were the same for both the training and evaluation datasets.

	Regression Equation	R^2	Bias (Mg ha^{-1} / %)	MAE (Mg ha^{-1} / %)
Winter Wheat				
RMA Training Dataset (N=358)	y=0.0501x	0.75	0.10 / 3.3	0.49 / 15.8
RMA Evaluation Dataset (N=358)	y=0.0508x	0.71	0.04 / 1.4	0.49 / 15.9
RMA Full Dataset (N=716)	y=0.0505x	0.73	0.07 / 2.3	0.49 / 15.6
Maize				
RMA Training Dataset (N=1042)	y = 0.0992x	0.59	0.03 / 0.4	1.2 / 16.5
RMA Evaluation Dataset (N=1043)	y = 0.0929x	0.46	-0.05 / -0.9	1.10 / 17.2
RMA Full Dataset (N=2085)	y = 0.0964x	0.55	0.0 / 0.0	1.18 / 17.1

Scatterplots between PRISM-ELM relative yield and RMA and NASS reported yields for winter wheat were similar (Figure 8). One notable difference is that RMA yields fell below the linear regression line at lower reported yields (Figure 8a), while NASS yields did not; this is similar to the discrepancy between RMA and NASS yields in the western plains (Figure 6a). For maize, there was significant scatter in the relationship between PRISM-ELM relative yield and RMA reported yield (Figure 9). The relationship with the few NASS data available was much stronger, but the y-intercept was greater than zero. This is likely because the NASS data encompassed a strong east-to-west precipitation gradient across the central plains, which was captured by the PRISM-ELM water balance. As the RMA yield data show, there were factors affecting maize yield in the eastern US that were probably local (e.g., soil conditions) or non-environmental in nature. An unknown amount of limited irrigation could also have affected yields.

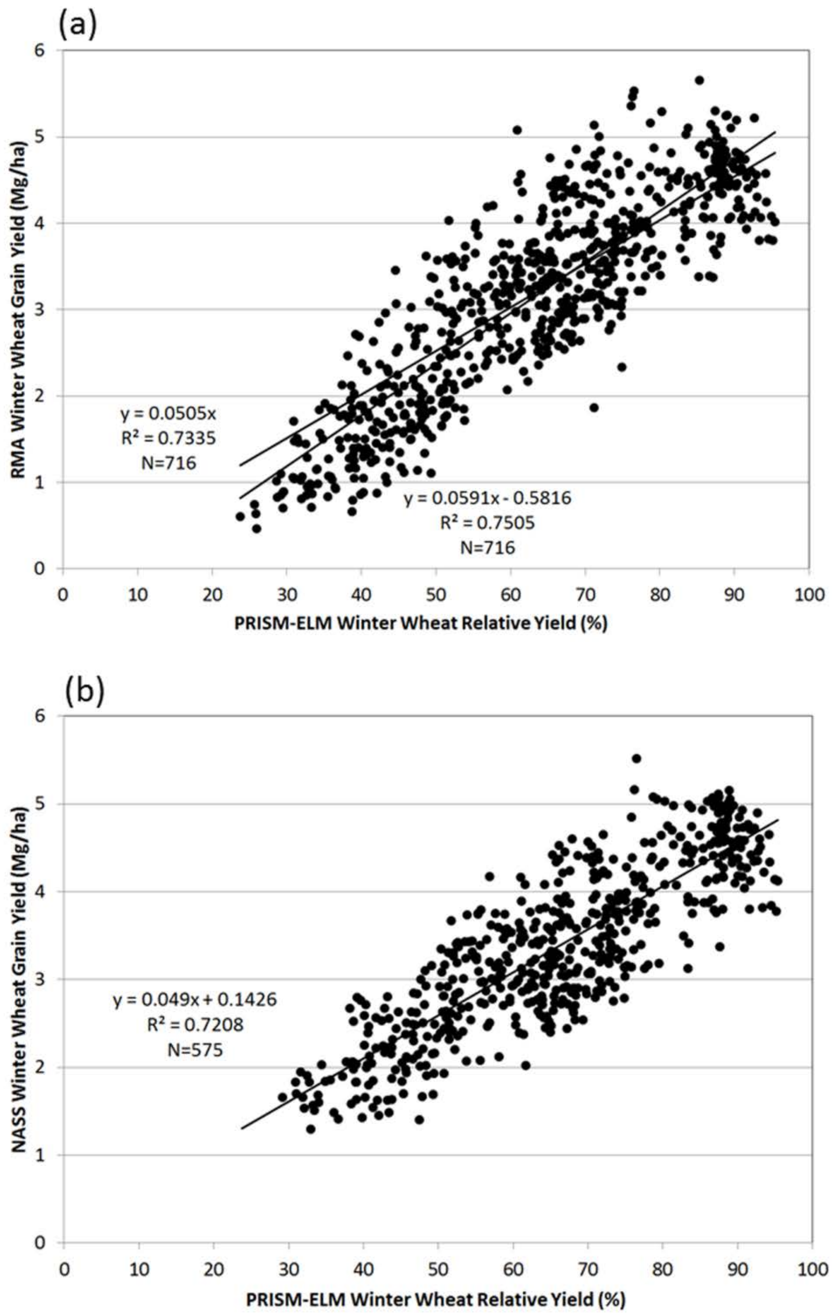

Figure 8. Scatterplots and least-squares linear regressions between county-level PRISM-ELM relative yield for winter wheat and reported yields from (a) RMA and (b) NASS. Also shown is the regression function with RMA data having a zero y-intercept; this function was used to transform relative yield to actual yield.

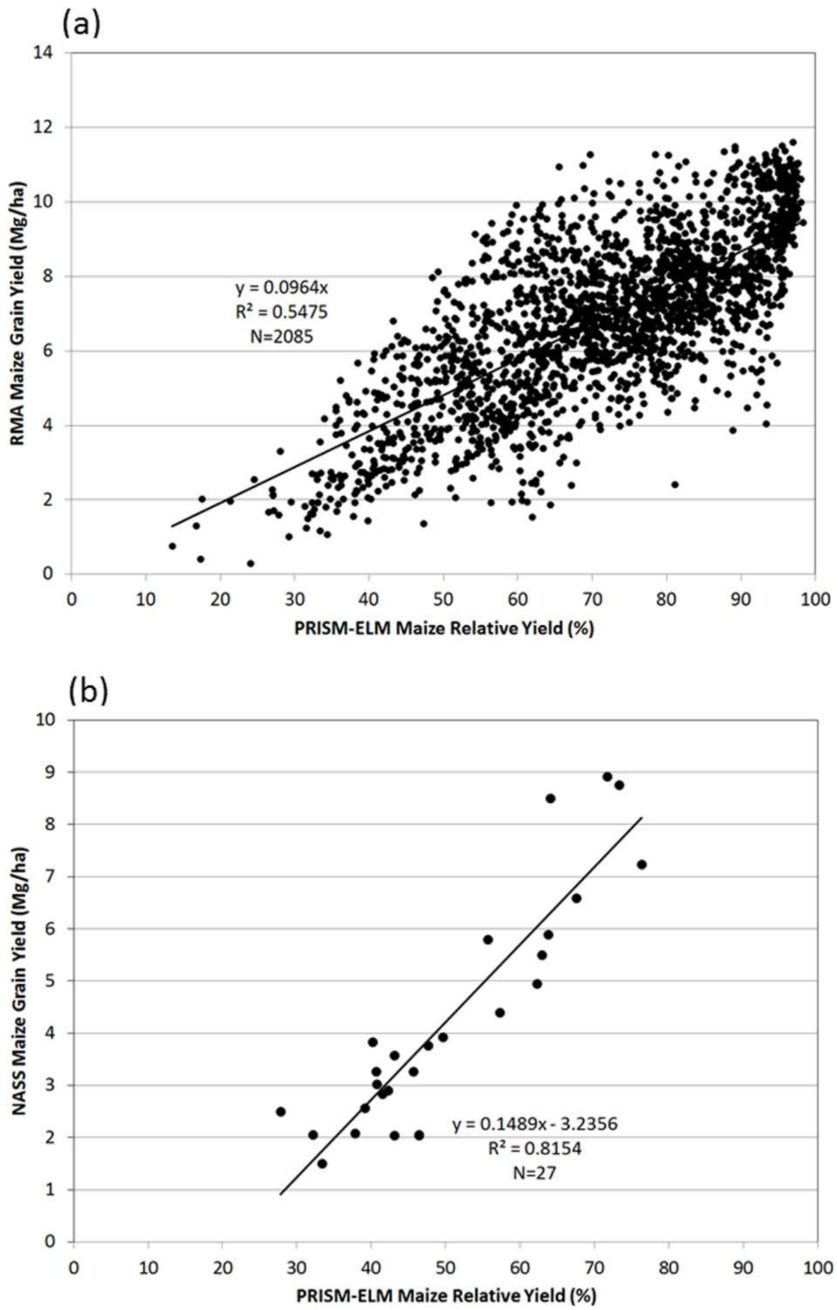

Figure 9. Comparison of county-level PRISM-ELM relative yield for maize with reported yields from (a) RMA and (b) NASS. The regression function with RMA data had a zero y-intercept, and was used to transform relative yield to actual yield.

PRISM-ELM actual yield maps were produced using the linear least-squares relationship between actual yield and RMA yield, forced through zero (Figure 10). The spatial patterns are the same as the relative yield maps (Figure 7), given that the regression function served only to scale the data to different units. Modeled winter wheat grain yields were roughly half those of maize in the eastern US, but exceeded those of maize in the western US because of the

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availability of spring moisture and moderate climatic conditions prevalent in some western wheat growing regions.

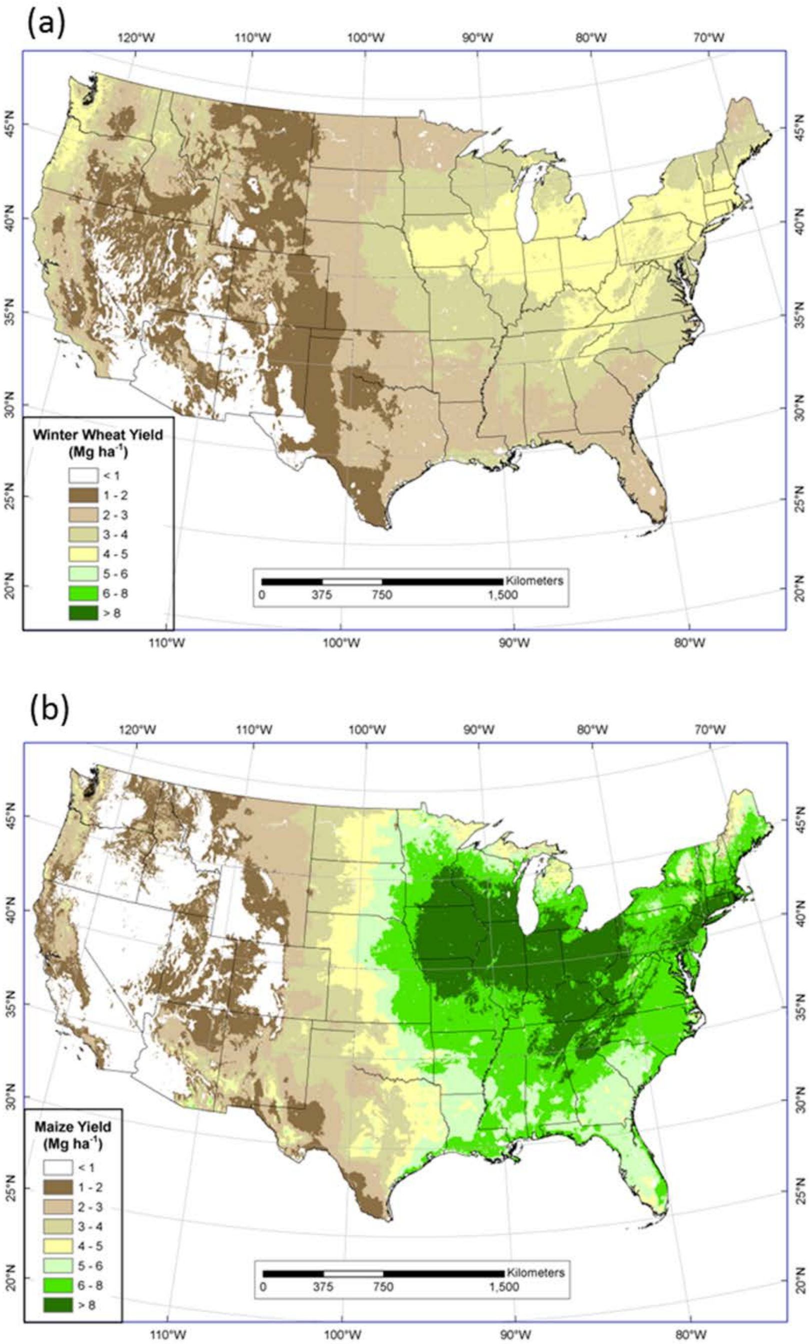

Figure 10. PRISM-ELM 1981-2010 estimated average yields, transformed from relative yield to actual grain yield for: (a) winter wheat, using the equation y=0.0505x (see Figure 8a); and (b) maize, using the equation y=0.0964x (see Figure 9a).

Difference maps between actual PRISM-ELM estimates and RMA and NASS reported winter wheat grain yields show reasonably good agreement (Figures 11 and 12). Overall, the strong east-to-west gradient of decreasing yield with decreasing precipitation across the plains was captured well by PRISM-ELM. RMA winter wheat yields were over-predicted in the western plains, but not NASS yields, which were over-predicted in the eastern plains. The pattern of western plains over-predictions was repeated for RMA maize yields. There were few NASS maize yield values available for comparison, but this discrepancy in the pattern of differences is consistent with lower RMA yields compared to NASS in lower yielding areas, which are located primarily in the western plains (Figure 6). In the eastern United States, where precipitation is generally sufficient, differences may have been caused by local soil conditions, management decisions (e.g., variety selection and fertilizer application), or other economic influences.

Comparisons between PRISM-ELM relative yields and reported grain yields are useful for model validation, but possess uncertainties. PRISM-ELM was driven by 1981–2010 mean climatic conditions, while the yield data represented averages over shorter periods. In addition, grain yield is likely sensitive to different environmental limitations, and their timing and magnitude, than biomass yield. Finally, reported yield data represent a sampling of production outcomes that reflects a myriad of interrelated management decisions and economic forces that can mask the environmental limitations modeled in PRISM-ELM.

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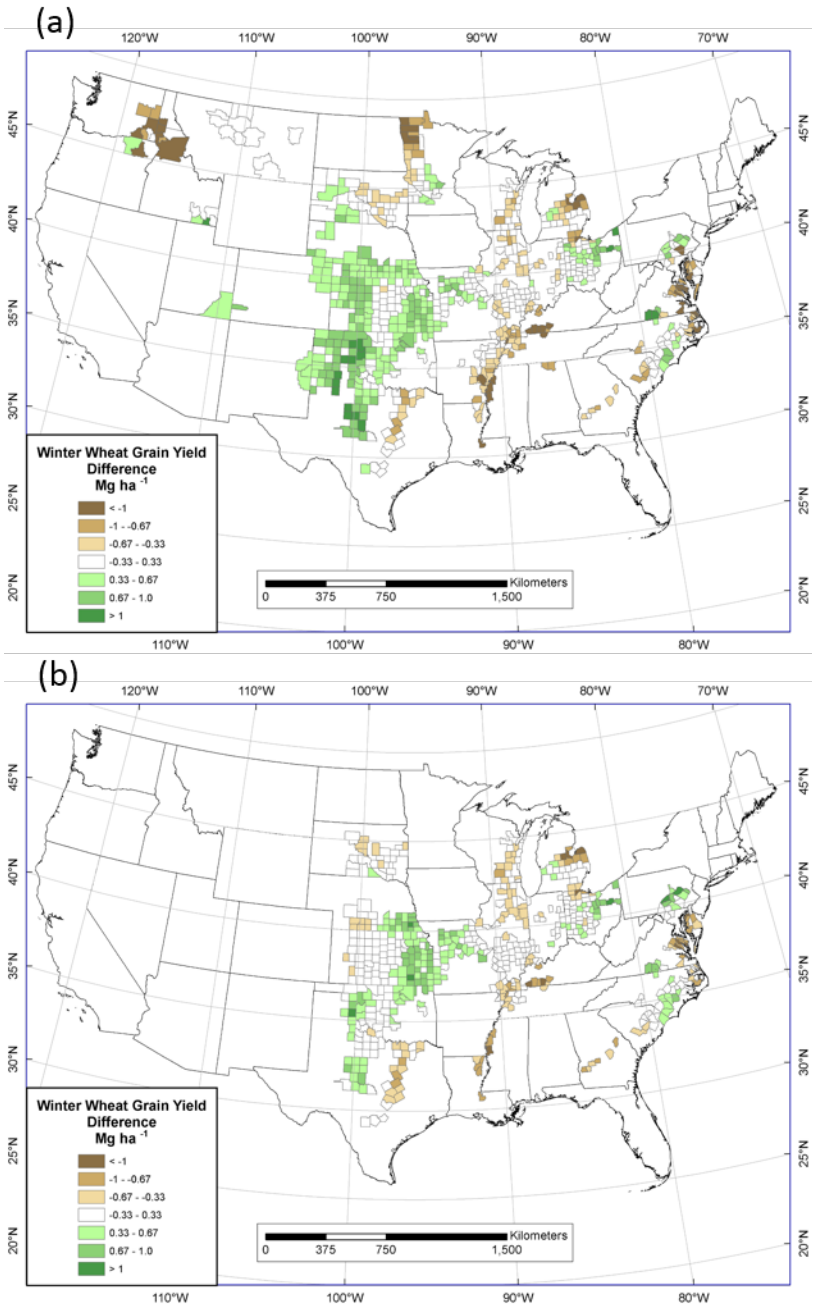

Figure 11. Differences (PRISM-reported) between PRISM-ELM 1981-2010 estimated average yields and county-level winter wheat yields reported by (a) RMA and (b) NASS.

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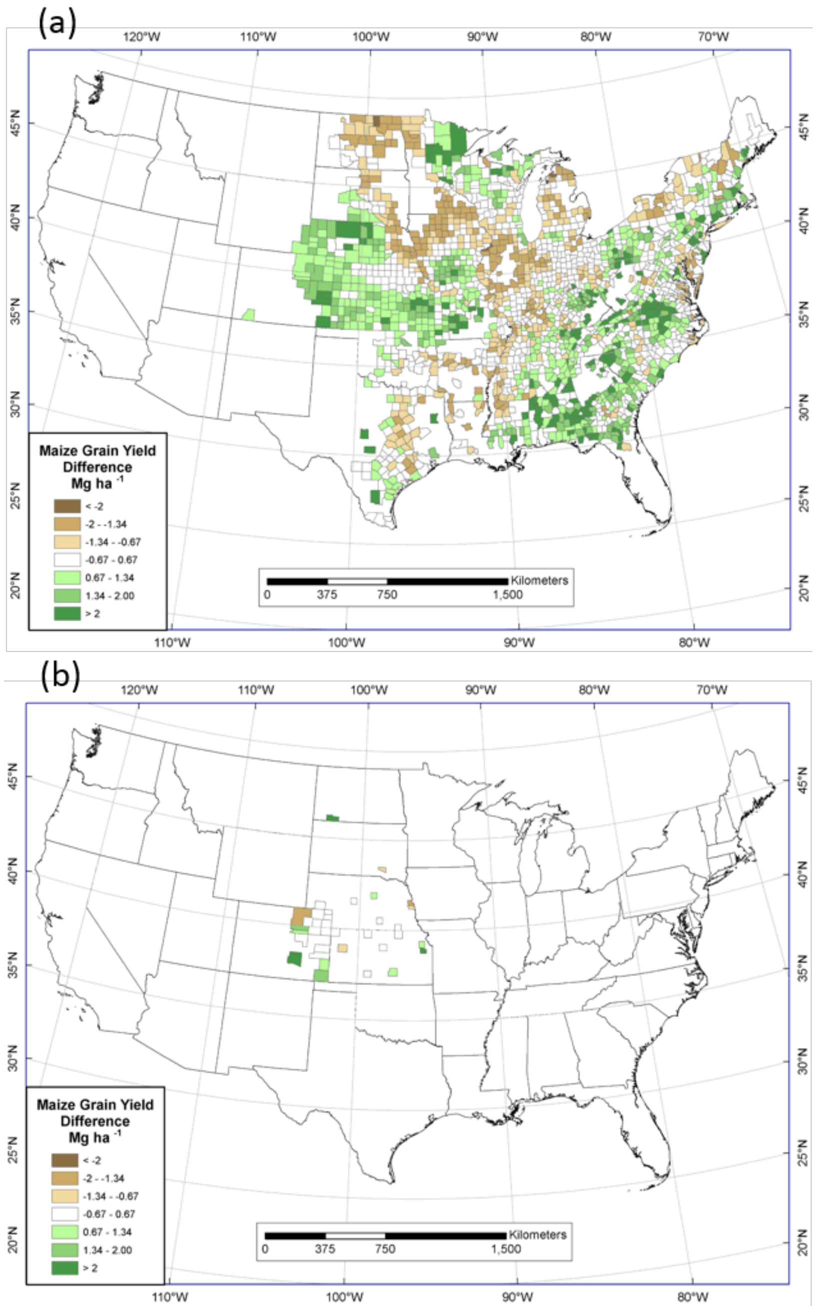

Figure 12. Differences (PRISM-reported) between PRISM-ELM 1981-2010 estimated average yields and county-level maize yields reported by (a) RMA and (b) NASS.

<u>Feedstock Resource Mapping</u>

PRISM-ELM was parameterized for six herbaceous (Lee et al., 2018) and three woody (Volk et al., 2018) biomass feedstocks considered nationally important for the Sun Grant program. There is very limited quantitative tolerance information for these feedstocks. As a result, model parameters were developed based on a combination of information sources. With the help of Sun Grant agronomists, the feedstocks were arrayed in order of perceived optimum temperature response (*OptT*), from cool to warm, anchored by wheat and maize with their fixed parameter sets (Table 2). Although there were no anchor species available for the woody feedstocks, these were also arrayed in the same manner (Table 3). Settings for other parameters, such as water use efficiency and heat and cold tolerances, were based on the strength of PRISM-ELM relative yield relationships with field data, and constrained by known patterns of growth and survival as perceived by the feedstock experts. The parameter settings should be viewed as initial values which will likely be refined as more field trial data are collected.

Table 2. Herbaceous feedstock species ordered by PRISM-ELM optimum growth temperature (*OptT*).

Species	***OptT* (°C)**
CRP	17
Winter Wheat*	18
Giant Miscanthus	20
Upland Switchgrass	21
Maize*	21.5
Lowland Switchgrass	24
Energycane	26
Biomass Sorghum	26

**"Anchor" species used to guide the parameterization of the biomass feedstock species.*

Table 3. Woody feedstock species ordered by PRISM-ELM optimum growth temperature (*OptT*).

Species	***OptT* (°C)**
Willow	20
Poplar	21
Southern Pine	22

A linear regression function relating PRISM-ELM relative yield to observed yield data was developed for each feedstock (Table 4). These functions were used to transform the PRISM-ELM relative yield grids into actual yield grids. The exception was southern pine (*Pinus taeda*), for which no yield data were available in this study. Based on expert information from the Sun Grant program, a PRISM-ELM relative yield of 100% was assigned an actual yield of 15.69 Mg/ha-yr (7 dry tons/acre-year) for southern pine (Tim Rials, personal communication), and all other values scaled accordingly.

Table 4. Linear regression functions relating observed yield (in Mg ha^{-1}) to PRISM-ELM relative yield for each feedstock species, where x is the PRISM-ELM relative yield and y is the estimated actual yield. The y-intercept of the regression function was forced through zero.

Species	Regression Equation	R^2	N
CRP	y=0.0426x	0.84	6
Giant Miscanthus	y=0.2485x	0.55	17
Upland Switchgrass	y=0.1169x	0.45	24
Lowland Switchgrass	y=0.2192x	0.69	22
Energycane	y=0.2295x	0.85	6
Biomass Sorghum	y=0.2926x	0.77	7
Willow	y=0.1964x	0.52	17
Poplar	y=0.1601x	0.60	17
Southern Pine	y=0.1569x	--	0

Using the equations in Tables 3 and 4, maps of 1981–2010 average potential biomass production were produced for energycane, upland and lowland switchgrass, biomass sorghum, CRP grasses, miscanthus, willow, poplar, and southern pine (Appendix A) (Daly et al., 2018; Lee et al., 2018; Volk et al., 2018). These maps provide a first look at the distribution of potential biomass production for these nationally important bioenergy feedstock species, using a common modeling and data collection framework, and close collaboration with Partnership agronomists. The maps were used as the basis for the economic and comparative analyses in the *2016 Billion-Ton Report*. In addition, the yield potential maps will be indispensable for organizations exploring biomass feedstock supplies in different regions of the country, addressing issues of water use and land-use change, and determining the impact of climate change in the future.

Key outputs

Peer reviewed Publications

Daly, C., M. D. Halblei, D. B. Hannaway, and L. M. Eaton. 2018. "Environmental limitation mapping of potential biomass resources across the conterminous United States." *GCB Bioenergy* doi: 10.1111/gcbb.12496.

Conference Proceedings

Halbleib, M., C. Daly, and D. Hannaway. 2012. "Nationwide Crop Suitability Modeling of Biomass Feedstocks." In *Proceedings of the 2012 Sun Grant National Conference: Science for Biomass Feedstock Production and Utilization*, New Orleans, LA, October 2–5. https://ag.tennessee.edu/sungrant/Documents/2012%20National%20Conference/ConferenceProceedings/Volume%202/Vol2.pdf.

Workshops

Daly, C., M. Halbleib, and L. Eaton. 2013. “Nationwide Bio-Fuel Resource Mapping: Energycane Biomass Feedstocks.” Organized and conducted workshop for the U.S. Department of Energy/Department of Agriculture/Department of Transportation Sun Grant Initiative, Jackson, MS, May 7–8.

———. 2013. “Nationwide Bio-Fuel Resource Mapping: Switchgrass Biomass Feedstocks.” Organized, conducted, and hosted workshop for the U.S. Department of Energy/Department of Agriculture/Department of Transportation Sun Grant Initiative, Corvallis, OR, May 29–30.

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Presentations and Panels

Daly, C., and M. Halbleib. 2009a. “Western Region Sun Grant GIS Team Status Report.” Presented at the Sun Grant Regional Biomass Feedstock Partnership Workshop, Washington, D.C., March 9.

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———.2009c. “Using Map Server Technology and Environmental Datasets for Feedstock Development and Assessment.” Presented at the Sun Grant Initiative Energy Conference, Washington, D.C., March 12.

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Daly, C., M. Halbleib, M. Doggett, and D. Hannaway. 2011. “Nationwide Biomass Modeling of Bio-Energy Feedstocks.” Presented at the Sun Grant Feedstock Partnership annual meeting, Knoxville, TN, February 15–16.

Daly, C., M. Halbleib. 2011a. “Biomass Mapping of Bio-Energy Feedstocks.” Presented at the U.S. Navy Green Fleet workshop, Honolulu, HI, March 7.

———.2011b. “Nationwide Bio-Fuel Resource Mapping: Estimating the Potential Distribution and Yield of Biomass Crops.” Presented at Texas A&M University Biomass Group, June 10.

Halbleib, M., and C. Daly. 2011. “Nationwide Biomass Mapping of Bio-Energy Feedstocks” Presented at Interagency Biofuels Infrastructure Workshop, Washington, DC, June 13-14.

Halbleib, M., C. Daly, M. Doggett, and D. Hannaway. 2012. “Modeling of Bio-Energy Feedstock Biomass in the US.” Presented at the Sun Grant Feedstock Partnership Annual Meeting, Indianapolis, IN, March 14–15.

Halbleib, M., C. Daly, M. Doggett, and D. Hannaway. 2013 “Nationwide Bio-Fuel Resource Mapping: Estimating the Potential Distribution and Yield of Biomass Crops” Presented at 2013 DOE Regional Feedstock Partnership Annual Meeting, Tunica, MS, February 14-15.

Daly, C., and M. Halbleib. 2014a. “Potential Yield Mapping of Bioenergy Crops.” Presented at breakout session and panel discussion, “Potential Yield, Composition, and Supply of Dedicated Energy Crops: Results and Outcomes of the Sun Grant Regional Feedstock Partnership,” at the BIO International Bioenergy Congress, Philadelphia, PA, May 12–14. https://www.bio.org/sites/default/files/WorldCongress/Chris%20Daly.pdf

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M. Halbleib., and C. Daly. 2015. “Biomass Feedstock Regional Partnership - Geographic Information System Yield Mapping”. U.S. Department of Energy Bioenergy Technologies Office: Project Peer Review. Alexandria, VA, March 23-27.

Daly, C. 2016 “Biomass Productivity Modeling and National Yield Potential”. U.S. Department of Energy Bioenergy Technologies Office: Project Peer Review. Washington, DC, July 14.

Where do we go from here?

The yield potential maps developed through this work are a first look at the potential long-term yield distribution of important feedstocks, and as such represent a strong basis for future expansion and refinement. Several topics need to be addressed in future work. These include: (1) estimating the expected temporal variability in yields; (2) improving soil characterizations in the model; and (3) updating the maps with more recent Partnership yield data using newly developed improved cultivars. The potential biomass maps produced by PRISM-ELM represent estimates of average yields expected each year over a 30-year period (1981–2010). The logical next step is to apply the model on a year-by-year basis over those 30 (and more recent) years to obtain a distribution of potential yields that can be used to develop risk assessments. This type of analysis would help answer questions about the long-term stability of expected feedstock yields over time, given the historical variability in weather conditions. An example shown in Figure 13 is a cumulative yield distribution function derived from annual PRISM-ELM winter wheat yields, which estimates the probability of attaining a given winter wheat grain yield in Wheatland County, Montana.

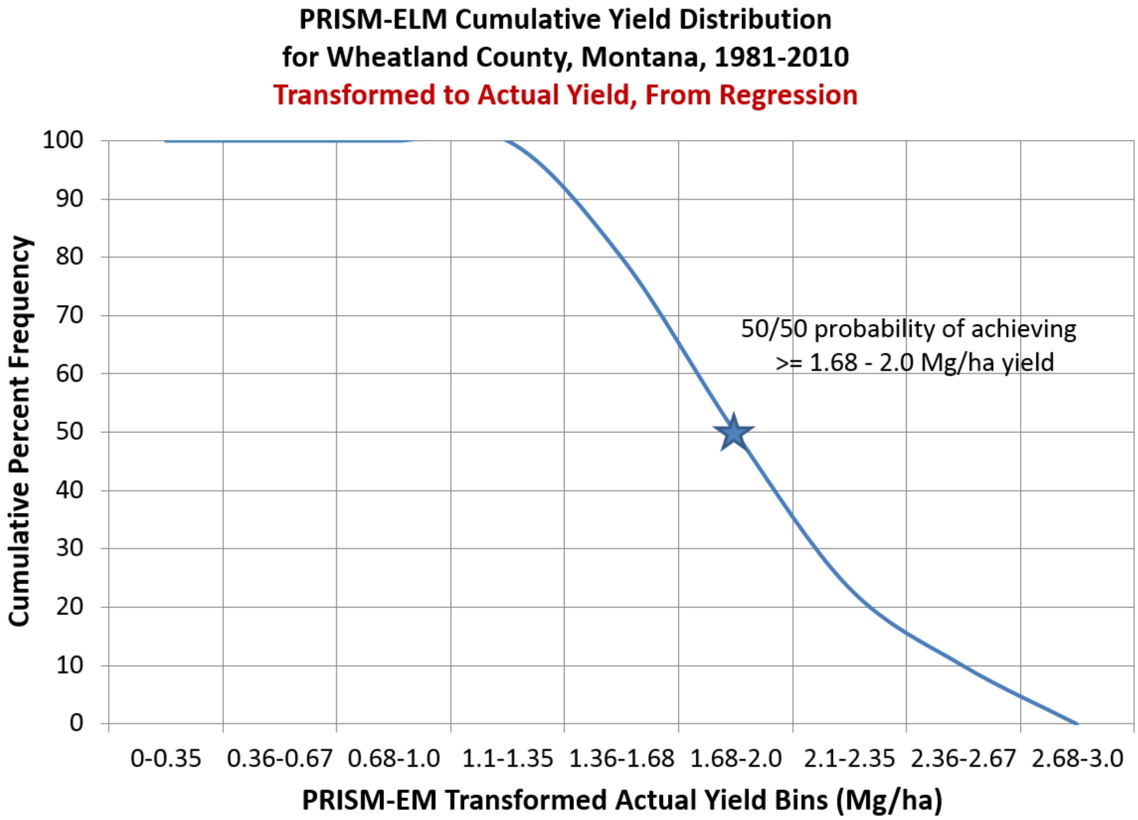

Figure 13. Cumulative yield distribution of PRISM-ELM estimated yields for winter wheat in Wheatland County, Montana, for the years 1981–2010. The probability curve shows the percent chance of attaining a given yield, with the 50th percentile marked with a star.

PRISM-ELM modeling efforts currently use soils data derived from the U.S. General Soils Maps; representative values are extracted from this map and averaged to an 800-meter grid cell. It is now possible to use recently-completed, higher-resolution soils data from the USDA NRCS to improve soil characterizations. In addition, given that the soils data are provided in polygon format rather than grid cells, implementing a modeling approach that is based on polygons rather than grid cells would increase the effective resolution of the modeling results, especially at field trial locations. PRISM-ELM also could be re-cast to run in “point” mode for the field trial locations, and have soil characteristics specified based on data collected by agronomists conducting the field trials. The results would be more representative of the fine-scale conditions at the field level, and may provide improved and more representative correlations between observed and modeled yields. A key soil characteristic that is not yet included in the PRISM-ELM modeling framework is a metric for fertility and overall soil health. Soil organic matter (SOM), is a good choice, as it plays key roles in overall soil health by increasing carbon content, acting as a buffer for soil pH acidification, and contributing to soil structure and water holding capacity.

The PRISM-ELM yield potential maps used all available data at the time of their development. However, additional years of yield data have been collected since the maps were created. It is the desire of both the modelers and agronomists that the yield maps be refined to incorporate new data and knowledge gained over the most recent years. In some cases, up to three additional years of Partnership data may be available for this purpose. In addition, it would be worthwhile to forecast potential future yields using assumptions from various climate change models

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Appendix A

PRISM-ELM maps of 1981-2010 average potential biomass production for nationally important herbaceous and woody feedstocks.

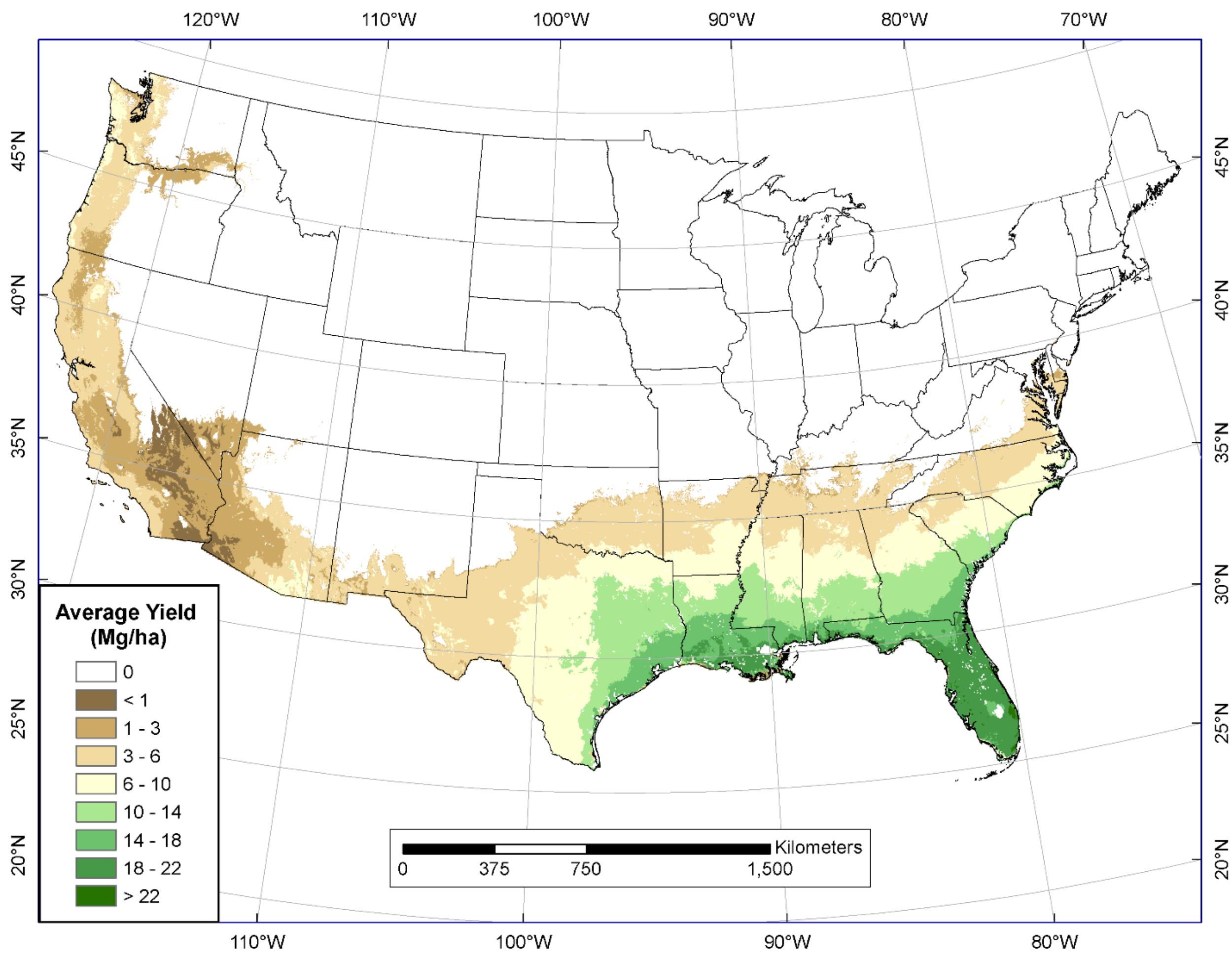

Figure A1. PRISM-ELM 1981-2010 estimated average biomass yield for energycane.

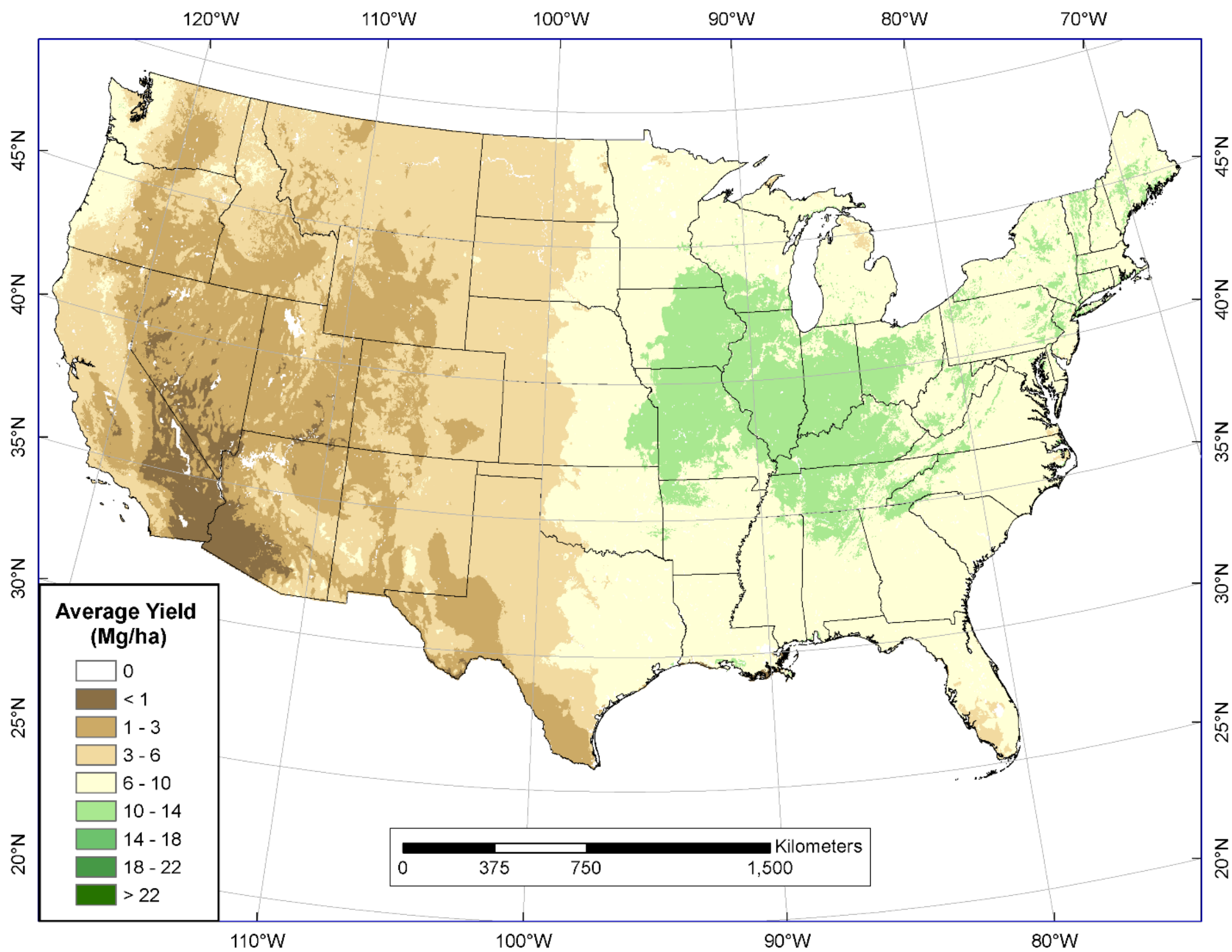

Figure A2. PRISM-ELM 1981-2010 estimated average biomass yield for upland switchgrass.

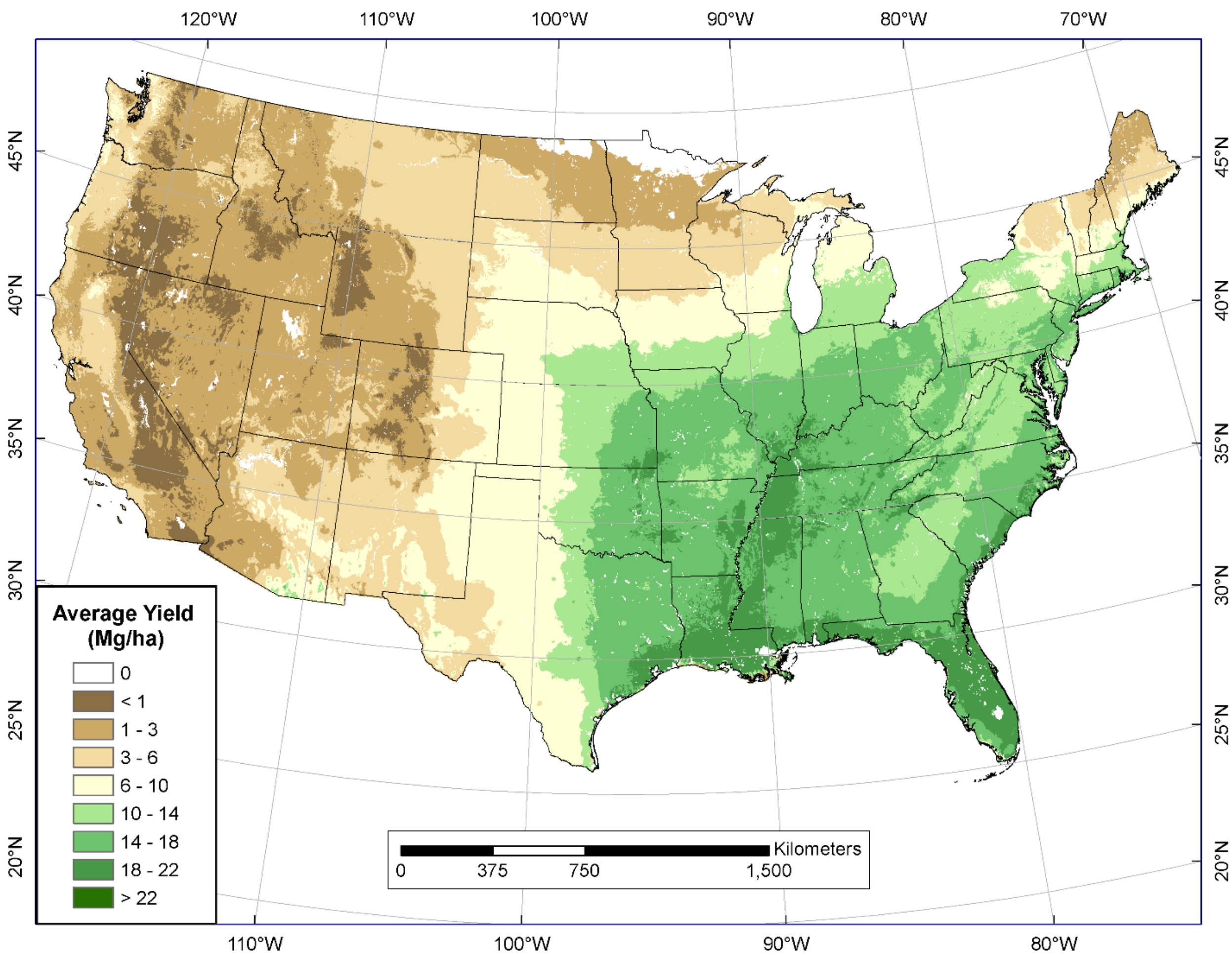

Figure A3. PRISM-ELM 1981-2010 estimated average biomass yield for lowland switchgrass.

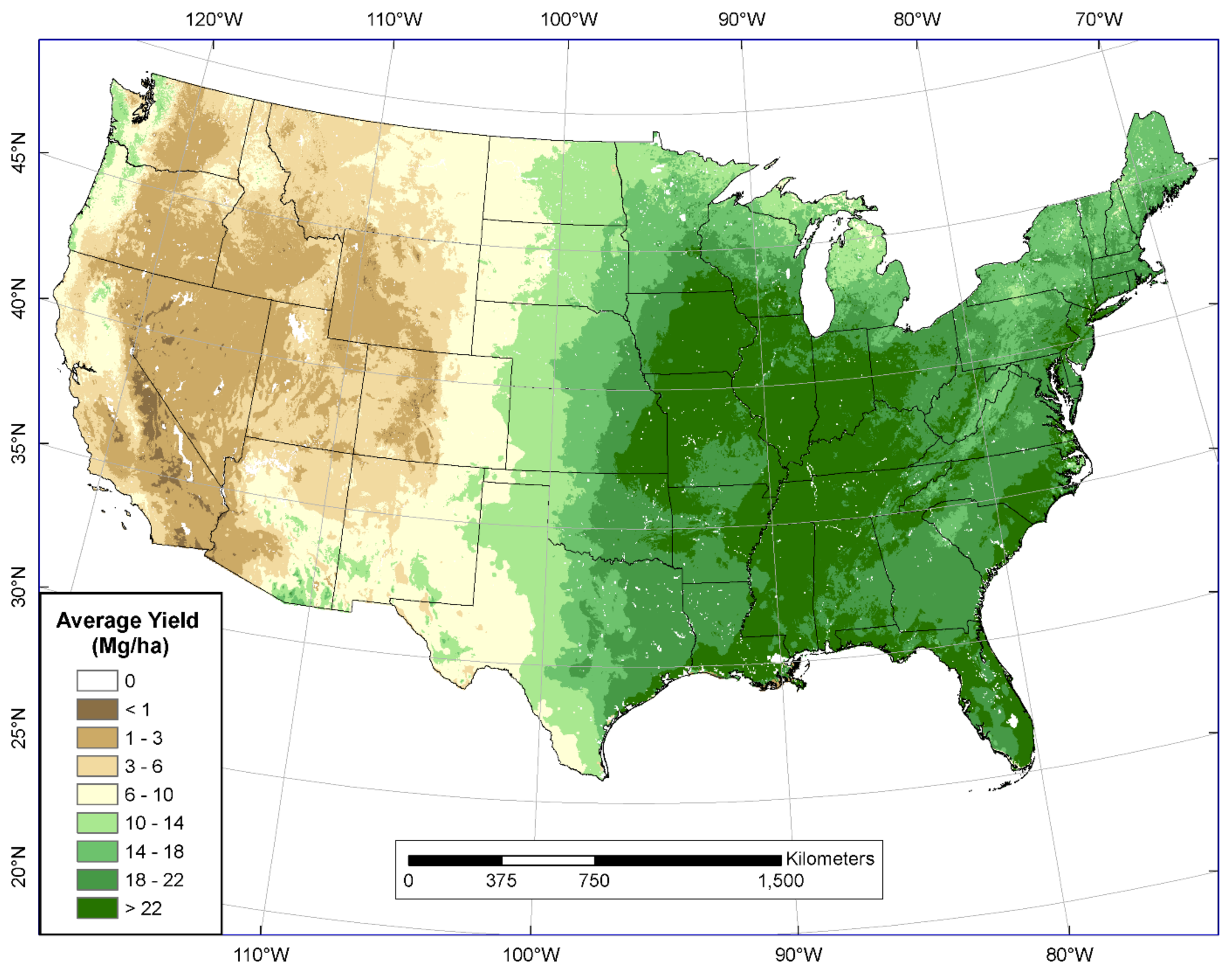

Figure A4. PRISM-ELM 1981-2010 estimated average biomass yield for biomass sorghum.

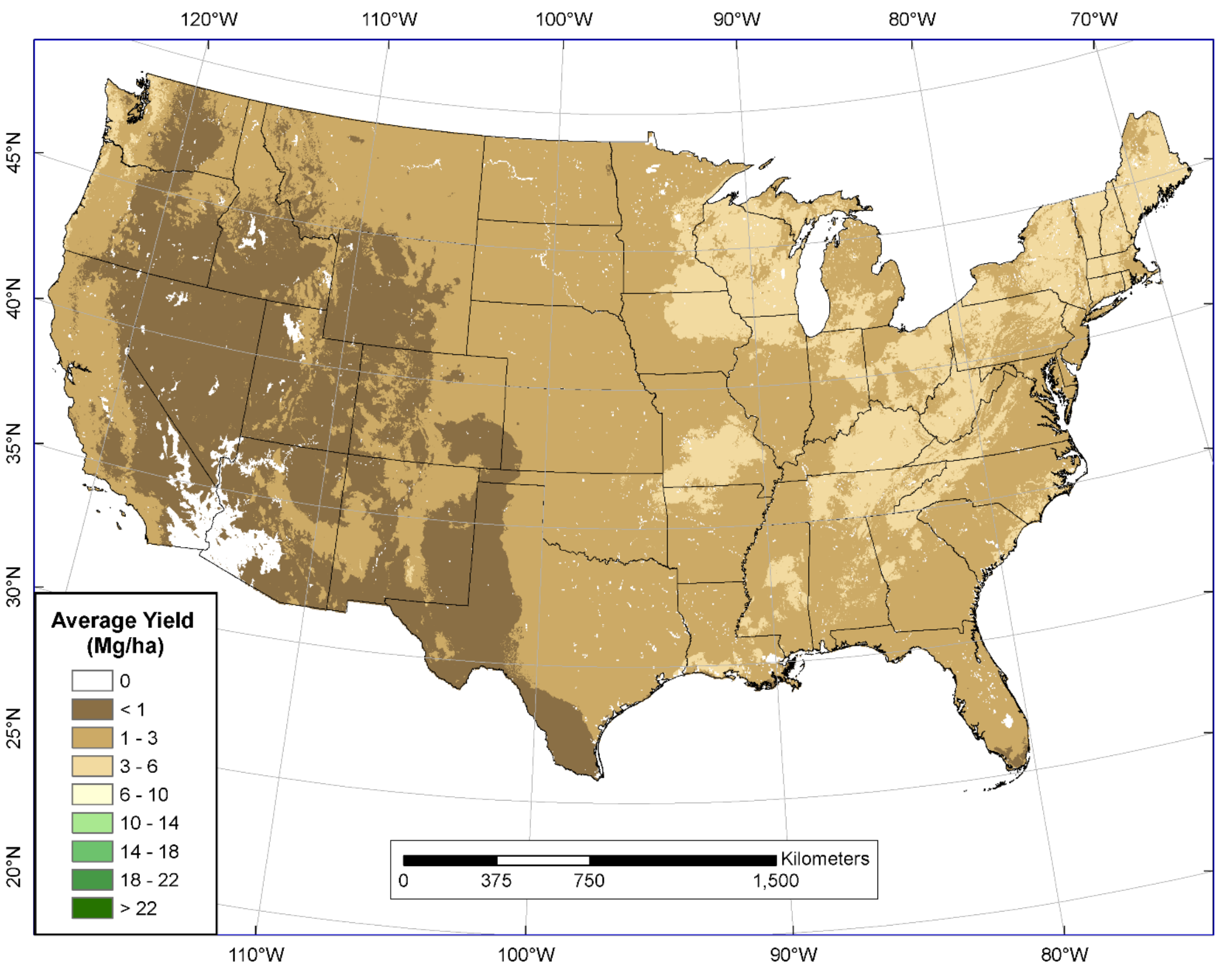

Figure A5. PRISM-ELM 1981-2010 estimated average biomass yield for CRP grasses.

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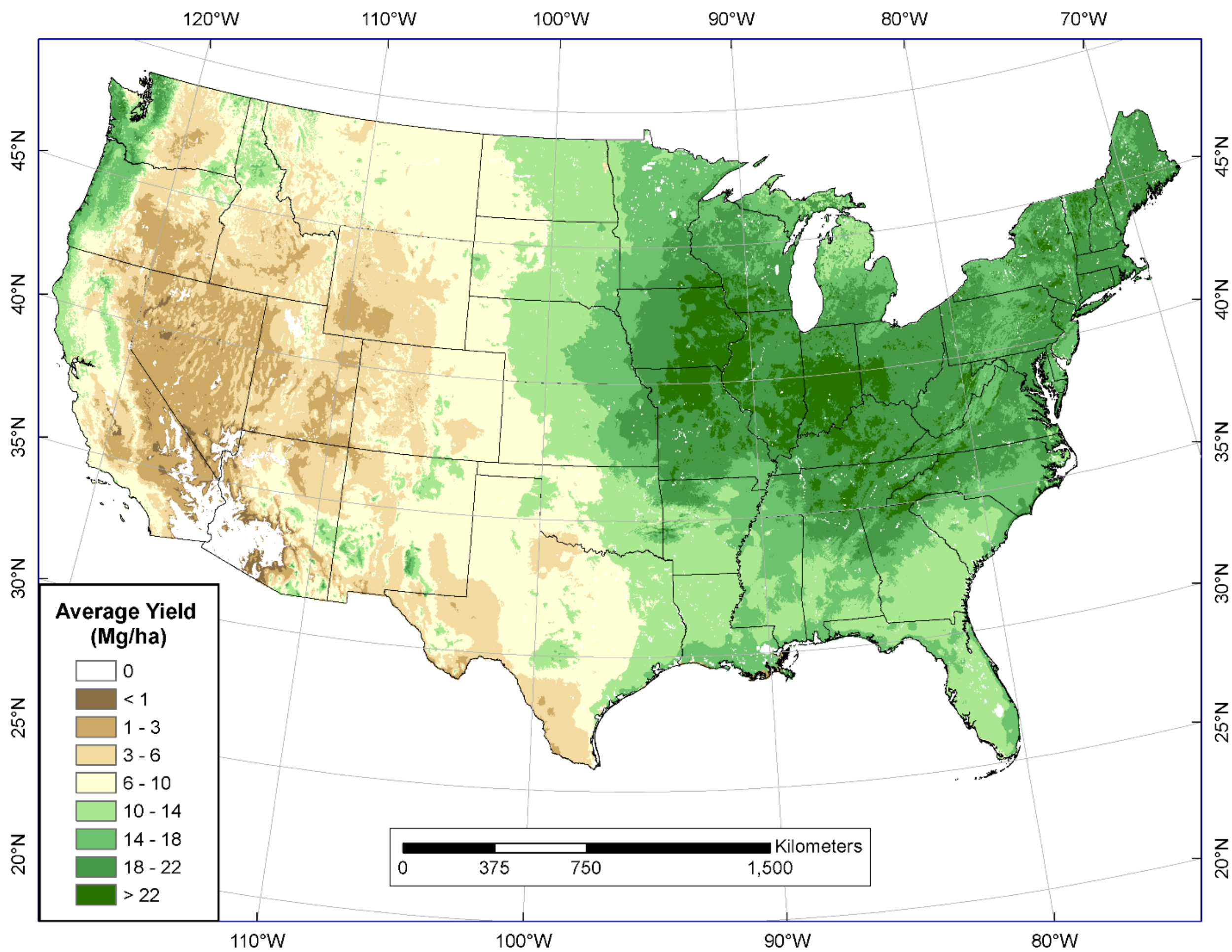

Figure A6. PRISM-ELM 1981-2010 estimated average biomass yield for miscanthus.

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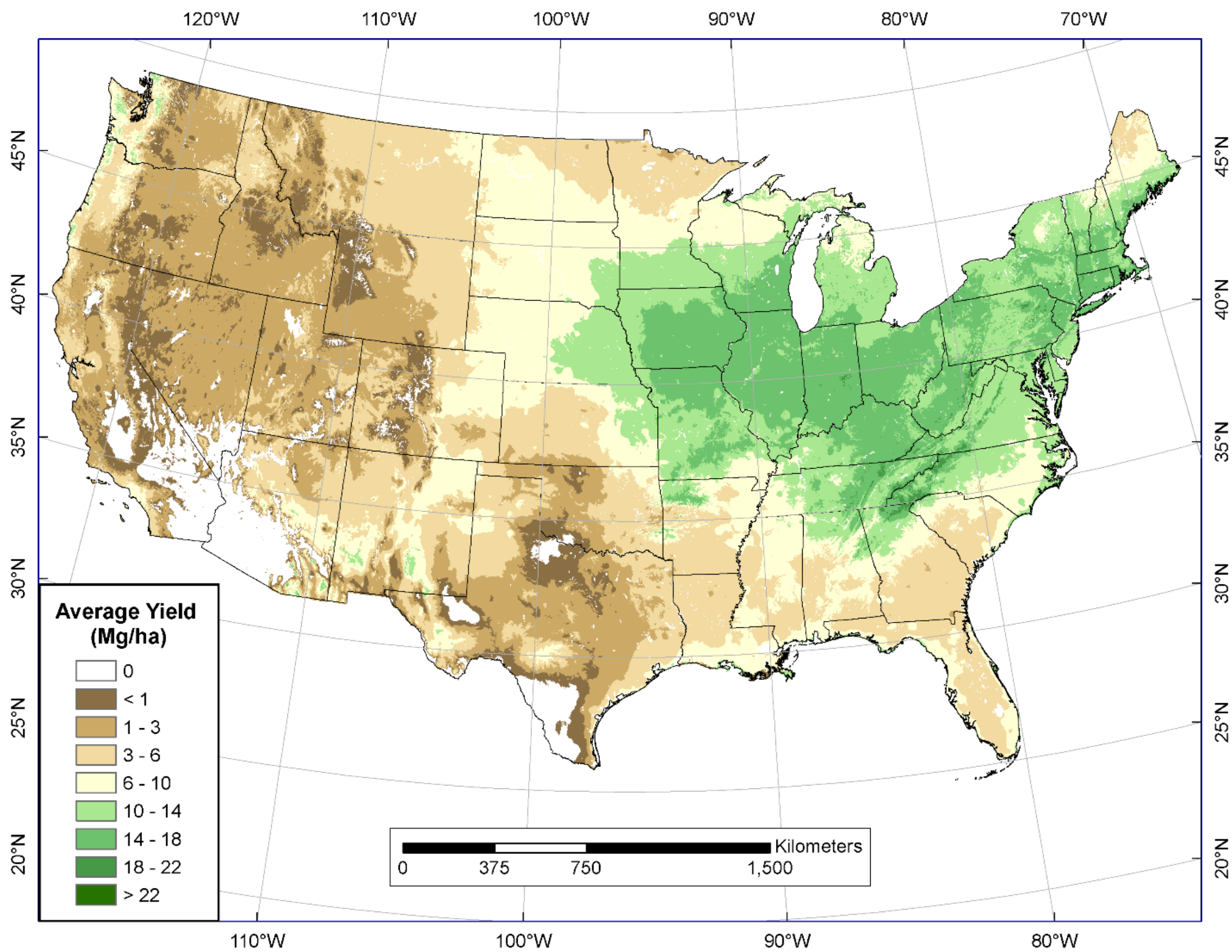

Figure A7. PRISM-ELM 1981-2010 estimated average biomass yield for willow.

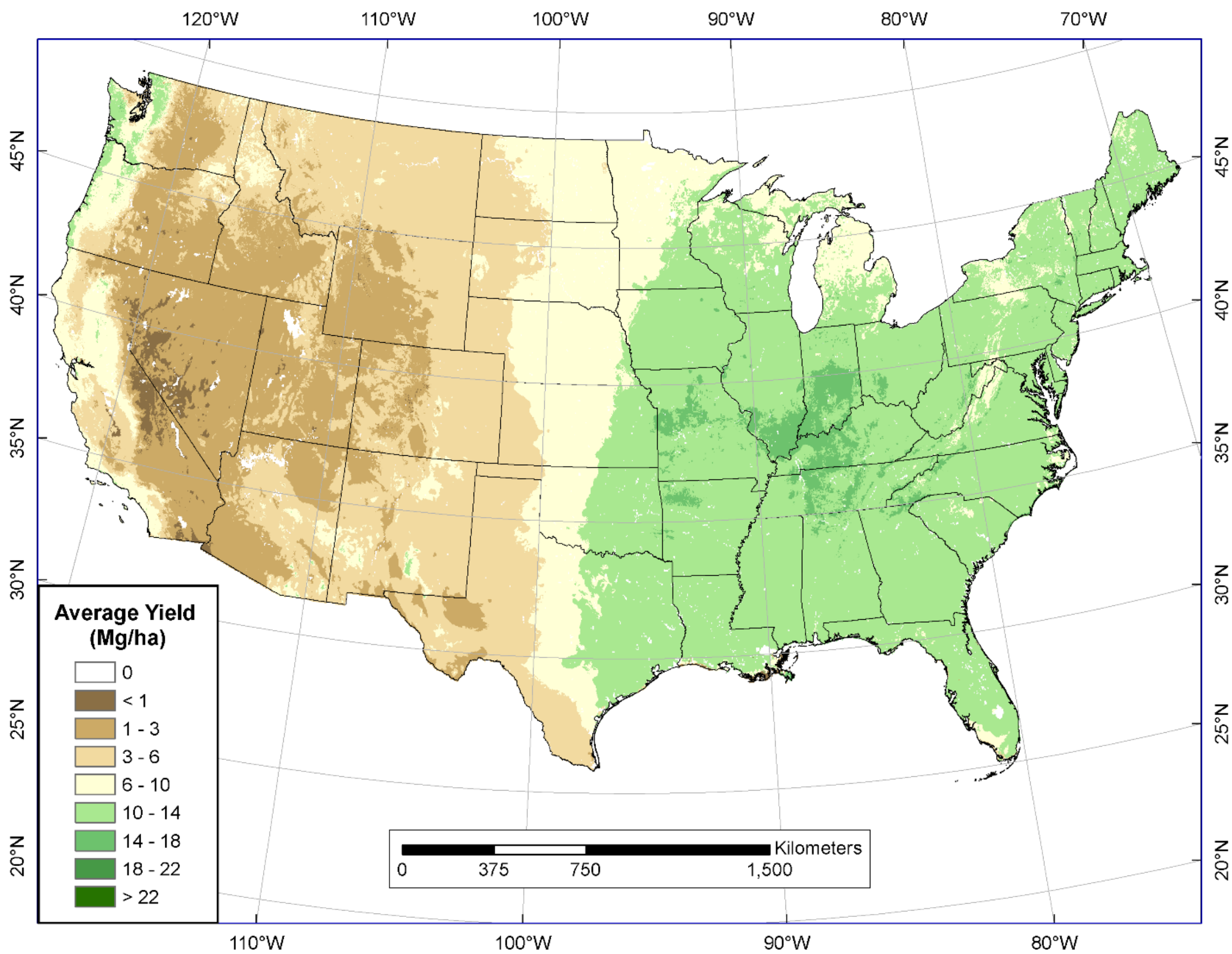

Figure A8. PRISM-ELM 1981-2010 estimated average biomass yield for poplar.

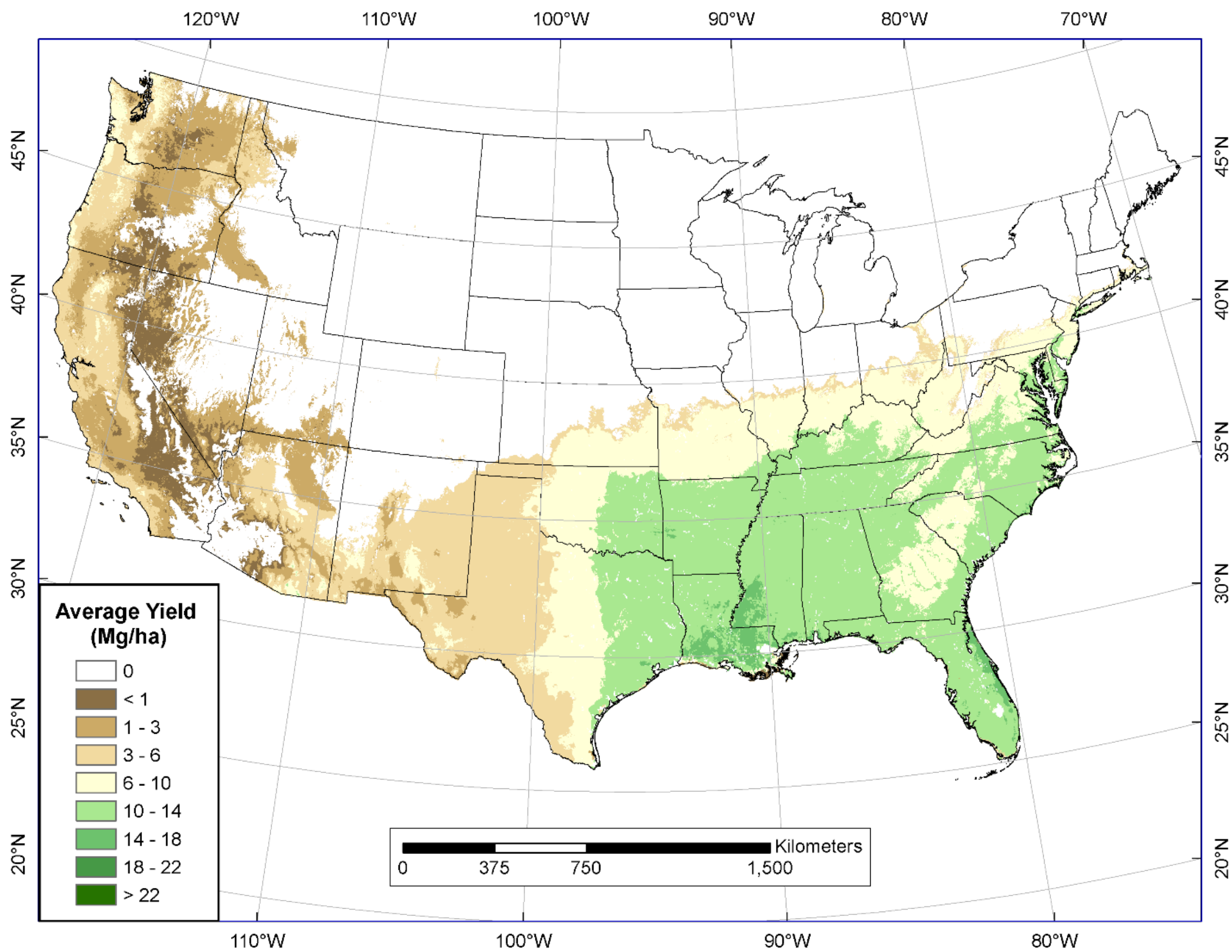

Figure A9. PRISM-ELM 1981-2010 estimated average biomass yield for southern pine.

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Outreach and Information Transfer

Submitted by: Timothy Rials and Jessica McCord, University of Tennessee

An important goal for the Partnership has been timely dissemination of information and insights generated by the research and development program, including conventional products like peer-reviewed publications and presentations at professional meetings. The trial network established by the Partnership also provided the perfect backdrop for numerous field days, tours, and other landowner education programs around the country (Figure 1). Examples of other valuable outreach events and tools are described below.

Figure 1. A field day at a poplar plantation in Tennessee (image courtesy of Jessica McCord, University of Tennessee).

The Sun Grant National Conference

The Sun Grant Initiative showcases regionally focused research that targets biomass, bioenergy, and bioproduct-related topics at national conferences across the United States. It has been important to document and highlight the activities and outcomes of the Partnership at various venues and in diverse media throughout the duration of the program. A key example includes hosting the Sun Grant National Conference in 2012 in New Orleans, LA. Several Partnership presentations were among the 123 papers presented at this conference. This conference led to a special issue of *Bioenergy Research* which included a paper from the Partnership (Figure 2).

Special Publications and Other Professional Presentations

The corn stover team also published a series of Partnership papers in *Bioenergy Research* and three papers have also been published as a set in *GCB Bioenergy*. Importantly, herbaceous yield data will be included for public access in that paper. In total, Partnership team members have published more than 130 peer-reviewed papers, four book chapters, more than 25 conference proceedings, developed extension/outreach materials, and given more than 200 presentations at other local, regional, and national events.

Figure 2. Partnership work was published in a number of journals, including two special issues of *Bioenergy Research*. The issue on the left (BioEnergy Research Volume 7, Issue 2) is a compilation of work done by the corn residue team and the issue on the right is from the 2012 Sun Grant National Conference (BioEnergy Research Volume 7, Issue 3).

BioWeb

In addition to field trials and crop modeling projects, educational and outreach work is underway to help agricultural producers, industry, and other stakeholders prepare for a future that includes converting biomass crops into energy and other products. One educational product from the Partnership is the BioWeb (bioweb.sungrant.org), which is an online resource for biomass and bioenergy information. The BioWeb has drawn from some of the country's top biomass authorities to provide a comprehensive analysis of the current state of biomass production, agronomy, harvest, collection, storage and preprocessing, as well as conversion technologies, and attempts to quantify impacts associated with biomass industry development, where possible. Content of the BioWeb is outlined in four major areas: 1) feedstocks, 2) biofuels, 3) biopower, and 4) bioproducts. In each of these areas, research coordinators, most from the Partnership, have assembled teams of research expertise that represents the spectrum of expertise in the biomass arena. The BioWeb was created primarily for the interested public and the academic specialist.

GeoSpatial Information Tools

Collaborations among regional partnerships are critical in developing sustainable biomass production and crop rotation strategies for both existing and new biomass resources. One DOE-sponsored Partnership activity was a regional and national biomass resource assessment. As part of this effort, the Southeastern Sun Grant Center worked with Oak Ridge National Laboratory to develop The Southeast Biomass Atlas (biomassatlas.org), which is a regionally focused, web-based atlas for policy and planning to support the expansion of biomass use for energy and biobased products.

The Southeast Biomass Atlas includes a spatial, county-level inventory of dedicated energy crops, with a scope of nine states and two territories (Figure 3). The atlas includes switchgrass, sorghum, willow, and other bioenergy crops. Using all available information on the existing and proposed facilities that use biomass for energy, advanced biofuels, and pellet production, this tool provides an estimate of current and future regional biomass demand. Environmental, socioeconomic, agricultural, geographic, and industry data, coupled with data from the U.S. Billion-Ton Update and estimated biomass supply through 2022, provide quick access to biomass availability information in a format relevant to policymakers, industry leaders, landowners, and resource providers.

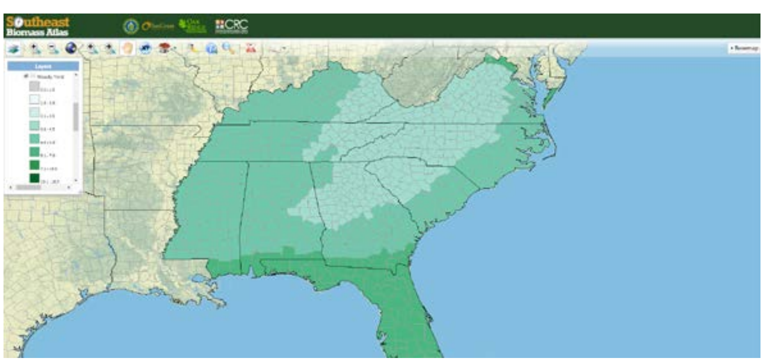

Figure 4. This map was generated using the Southeast Biomass Atlas and depicts availability of woody biomass in the Southeast through 2022 (information freely available at biomassatlas.org).