CAAP Canadian Agricultural Adaptation Program

Final Report

Enhancing the commercial viability of switchgrass on marginal farmland through plant breeding Project Title

6604

Project Number

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> January 10, 2014 Final Report Submission Date

The final report forwarded to the CDAQ in hard copy and in Word copy must include:

- Let the deliverables described in Appendix C of the financial contribution agreement;
- Let the supporting documents, numbered and written in the Financing Plan and Expenditure Reconciliation document;
- □ the copies of the dissemination documents mentioning the CAAP's contribution according to the program's exposure rules.





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1. OBJECTIVES

1.1. General Objective

To enhance the commercial viability of switchgrass in Québec by establishing a plant breeding program to improve the agronomic performance and diversity of switchgrass cultivars available to Québec farmers for utilization on marginal farmland.

1.2. Specific Objectives

Objective 1: To develop six new breeding selections of switchgrass with improved adaptation and yield on marginal soils and/or short season production areas in Québec.

Objective 2: To improve seedling emergence and first year establishment in switchgrass through selection for increased seed size and single tiller production.

Objective 3: To make these new selections available for replicated cultivar trials in Québec and multiply breeding seed through a Québec switchgrass seed grower.

Objective 4: To disseminate project progress and results.

2. RESULTS AND ANALYSIS

2.1. Results Achieved and Analysis

The project has largely been implemented as planned, and we have accomplished all proposed milestones. The main deliverable of developing 6 advanced populations of switchgrass was primarily achieved. Establishment and breeding practices have been vastly improved upon and advanced populations of switchgrass were made available to CEROM after the first and second third breeding cycles. Dissemination activities have been undertaken at Ferme Norac, through conferences, workshop events and the REAP-Canada web site.

One of the key realizations made during this project was that fairly rapid progress can be made using a simplified breeding program. We have made incremental improvements on all aspects of our breeding program, which translates into considerable progress towards developing agronomically superior short season switchgrass cultivars for Eastern Canada. We have effectively reduced a typical 3-4 year Recurrent Restricted Phenotypic Selection (RRPS) program (Burton, 1972) to a 1-2 year cycle time. This was made possible by integrating several selection techniques in the breeding program and modifying greenhouse and field nursery practices to accelerate growth and enhance selection pressure. Subjecting the populations to selection at

the seed, seedling and mature plant stage appeared to be an effective approach. We believe the resulting germplasm available at the end of the 2013 field season are improved populations with reduced dormancy, increased seed size, reduced tillering, increased height, and increased weight of reproductive tillers. These results are discussed in detail below and supporting photos provided.

Activities	Expected date	Actual date	Purposes	Notes
Develop 4 improved populations of switchgrass	May-October 2012/2013	May-October 2012/2013	To increase biomass productivity and adaptability to marginal areas	Completed
Develop 2 synthetic populations of switchgrass	October 2013	October 2013	Create germplasm for both cool regions and wet soils	Completed but success of crossing needs to be determined
Improve seedling emergence/establishment	May 2011;2012;2013	May 2011;2012;2013	Further increase seed size and emergence	Completed
Seed multiplication, conditioning and distribution	October 2011/October 2012	October 2011/October 2012	Further testing by CEROM and use by Ferme Norac	First and second year seeds were distributed and third year seeds have been harvested Ongoing
Performance evaluation data	October 2011; 2012	October 2011; 2012	Establishment, yield, and growth evaluation data summarized from current and planned variety trials	Completed
Development of an improved Breeding program	October 2013	October 2013	Further optimize breeding techniques for improved adaptation and seedling vigor	Completed
Project communications	Fall / Winter 2011; 2012; 2013	Fall / Winter 2011; 2012; 2013	Enhance awareness and knowledge	A diversity of activities were completed; including field days, conferences & website dissemination

Table 2.1 Completed Activities

Deliverable 1 – Seed from improved populations

At the outset of the project the breeding plots of Cave-in-Rock II (CIR II), Cave-in-Rock Early maturity (CIR-EM), Tecumseh II and Blue Jacket II were well established. They had previously been field transplanted in the spring of 2009 and grew vigorously in both 2009 and in the spring/summer of 2010. This allowed these populations to undergo 2 full cycles of selection by the end of the project; creating Cave-in-Rock IV, Cave-in-Rock III EM, Tecumseh IV and Blue Jacket IV. We harvested seed from each of these populations in October 2013 and it will be processed and made available to the RPBQ for cultivar variety trials for next field season.

The two poly-cross nurseries were evaluated and harvested for seed in October 2013. The Cave-in-Rock IV x High Tide II (CIR IV x HT II) nursery yielded some of the tallest plants overall (See Figure 2.1), but there were some concerns about the success of crossings. The parents of the CIR IV (see Table 2.3) mature somewhat faster (approximately 5-10 days) than the parents of the High Tide II. The seed obtained from this breeding cycle will need to be evaluated for evidence of a successful cross. Overall we believe this cross met our original goals and was successful. As for the Blue Jacket III x Cave-in-Rock EM II (Blue Jacket III x CIR EM II) polycross nursery, there were more serious challenges experienced. There was excessive competition from the rows of CIR EM II switchgrass, which may have been due to the more extensive surface rooting, spring vigor and taller height. The rows of the CIR EM II plants provided too much competition for the alternate row plantings of Blue Jacket III plants. This caused them to lodge and decay in the bottom of the canopy. We have learned that making successful field crossings requires careful consideration of flowering periods as well as the level of competition between different populations. The row widths at 55cm appeared too narrow resulting in excessive below-ground competition and above-ground shading. Nonetheless we were able to source improved germplasm from the CIR EM II plants in this nursery. We will continue to improve upon the crossing methods, possibly using larger spacing between rows in the crossing nurseries to slightly reduce the between-cultivar competition. In addition to the competition concerns, the flowering period of the two cultivars may not have sufficiently overlapped. The parent material of the CIR EM II and the Blue Jacket III materials were estimated to have maturities of 147 days and 136 days respectively (see Table 2.3).



Figure 2.1. A 3 metre tall CIR III Plant in a second year CIR III x HTII polycross nursery at Ferme Norac

Deliverable 2 – Improved seeds for Cultivar trials

As of November 2013 we have produced seed from 7 advanced populations of switchgrass with improved seed size, reduced tillering and increased height (Cave-in-Rock IV, Cave-in-Rock Early Maturity III, Tecumseh IV, Blue Jacket IV, High Tide III and synthetics of Cave-in-Rock IV x High Tide II and Cave-in-Rock Early Maturity II x Blue Jacket III). These adapted germplasm populations will be made available to researchers of RPBQ for replicated cultivar trials.



Figure 2.2. Summer and Cave-in-Rock seed from their original seed lots and second cycle Bluejacket breeder seed.

Increased seed size – Seed size has been increased from that of the original foundation seed populations. After three cycles of selection, we have seen a 19% increase in seed size from Sunburst (parent population) to Blue Jacket III and also a 19% increase from Cave-in-Rock

(parent population) to Cave-in-Rock III. This seed size difference represents the difference between standard seed lots of the material from field plantings and seed derived from that of space planted nurseries - high graded for large seed. A 22% increase in seed size between the seeds of Cave-in-Rock and Cave-in-Rock Early II and a 30% increase from Summer to Tecumseh III were also identified. There was also a 30% increase between the seeds Cave-in-Rock and the seeds selected in the Cave-in-Rock x High Tide crossing nursery and a 10% increase from the High Tide to the High Tide III (See Table 2.2). Please note these breeder seed populations were derived from space planted nurseries and the "original" seed was from field plantings. Through replicated field plot studies in the future, we will be able to determine if seed size is changed in the subsequent populations.

Increased seed size may partially account for the improved seedling emergence we have seen in the new materials. Breeding for increased seed size is extremely important for emergence, especially on heavy clay soils. For example, the small seeded Tecumseh can often suffer from stand failures on heavy clay soils because of the smaller seed size. On average the breeding cycle initiated in the spring of 2013 benefited from seed that was approximately 18% higher in 100g seed weights than commercially-sourced varieties. In general the Cave-in-Rock and Blue Jacket selections have a large seed size for upland switchgrass relative to other cultivars. Tecumseh and High Tide are in the lower end of the range reported for upland varieties (Elbersen 2001), and thus require further seed size improvement. Overall it appears increasing seed size to 0.200g per 100 seeds can have appreciable benefits to improving stand establishment. Switchgrass seed lots of 0.200 g per 100 seed were found to have improved germination, days to emergence, and a higher emergence percentage than seed of approximately 0.100g per 100 seed (Kneebone and Cremer 1955).

Cultivar	100 seed wt (g)	% increase
	(9)	
Sunburst	0.197	
Blue Jacket III	0.242	18.6
Cave-in-Rock	0.187	
Cave-in-Rock III	0.230	18.7
Cave-in-Rock	0.187	
Cave-in-Rock Early II	0.241	22.4
Summer	0.108	
Tecumseh III	0.154	29.6
Cave-in-Rock	0.187	
Cave-in-Rock III x High Tide II	0.266	29.7
High Tide	0.120	
High Tide III	0.134	10.4

Table 2.2. 100-Seed weights. The **original seed** weights (Sunburst, CIR, Summer, High Tide) represent the average between the seed lots sourced to establish the original breeding material, and those measured in Elbersen (2001). The **breeder seed (**Blue Jacket III, CIR III, CIR III EM, CIR III x HT II, HT III) represents the heaviest 2% of the seed harvested in the Fall of 2012 from spaced-plant nurseries.

Deliverable 3—Seed production fields

Seed production fields were harvested and seeded at Ferme Norac for Tecumseh II and Cave-in-Rock II switchgrass. The area harvested in fall 2011 was approximately .2 ha of Cave-in-Rock II switchgrass and .3 ha of Tecumseh II switchgrass. These seeds were subsequently reseeded at Ferme Norac in the spring of 2012 and 2013 to bring a final acreage of 0.75 ha of Tecumseh II and 1.5 ha of Cave-in-Rock II and Tecumseh II switchgrass. These fields were harvested in the fall of 2013 and are expected to produce approximately 150-200kg/ha of seed for further multiplication. The main problem experienced with the seed field establishment was annual grass pressure from crab grass and giant foxtail. It is now recognized that a comprehensive strategy to manage weeds is required in seed fields. This should include effective crop rotations prior to seeding switchgrass to reduce weed pressure, and seeding at a minimum of 5 kg/ha to ensure an adequate plant stand. Our observation was that there was overall less weed pressure in the CIR II than the Tecumseh II. This may have been result of the taller height and wider leaves of the CIR II and the more decumbent nature in the lower leaf canopy, which tends to make CIR more effective at weed suppression. Tecumseh II stands very erect and tends to let more light into the lower part of the canopy.

Deliverable 4 - Performance Analyses

Yield performance measures, heights and seed weights were assessed at two sites for the populations of Cave in Rock II, Blue Jacket II, Tecumseh II and Cave in Rock early. Variables examined during the season included size, yield and phenological stages of growth, see Appendix II. The results demonstrated that the improved selections produced significantly higher yields than the original parent cultivars of Sunburst and CIR. There was an average of 11.9% yield gain between the cycle 2 Blue Jacket population and its parent cultivar, and a 15.5% yield gain in the cycle 2 CIR population. We also saw that the dynamics of the production of tillers differs between cultivars. At the beginning of the season the plant produces many tillers, but those that are small and shaded die during the season, and the plant eventually converges to an optimal number of tillers or a 'tiller equilibrium'. Therefore, selecting for reduced tillering at the seedling stage should prove to be a worthwhile practice towards decreasing tiller mortality.

Using the refined breeding system, we were able to improve establishment and first year growth enough so that selections were able to be made and plants produced significant amounts of seed in the first year. This was not possible prior to the implementation of the improved production methods developed in the greenhouse and transplant nurseries.

The phenologies of the original and subsequent breeding material were also monitored throughout the 2011 and 2012 seasons to determine the average days to maturity for each population. Monitoring the maturities ensures the populations chosen have a maturity rating that is suitable to short season zones in Québec and allows a more accurate determination of

which materials can be successfully cross-bred. In order to do field crossings it is imperative to have synchronous flowering periods between the populations to ensure optimal crossover time during anthesis. It was found that the average days to maturity for each of the breeding populations was usually within 3-5 days of the original parent population. Blue Jacket is the earliest maturing population, Summer is about 7 days later, and Cave-in-Rock is approximately 7 days after Summer. High Tide is the latest maturing material at about 12 days later than Cave-in-Rock (See Table 2.3). However, there was quite a bit of variability in rates of maturity between plants in each plot and less frequent sampling was taken in 2012 (3 assessments) than in 2011 (6 assessments). This may have led to a less precise measurement, as the Cave in rock II was identified to be appreciably later in the 2012 season.

The appreciable difference in maturity (~11 days) between the Blue Jacket II and Cave in Rock E populations may partially account for the difficulty experienced within the Cave-in-Rock E II x Blue Jacket III crossing nursery.

Table 2.3. Average days to maturity. The original material represents the populations sourced establish the original breeding material. The days to maturity of the Breeding material is the average over 2 sites after 2 cycles of selection at each site.

Average Days to maturity		Average Day	s to maturity
Original	Material	Breeding Material	
Sunburst	133	Blue Jacket II	136
Summer	140	Tecumseh II	142
Cave-in-Rock	146	Cave-in-Rock II	153
Cave-in-Rock	146	Cave-in-Rock Early	147
High Tide	158*	High Tide	N/A

*estimated from Cortese & Bonos, 2013



Figure 2.3. Left: Sunburst population (Parent material of Blue Jacket); Right: Blue Jacket II

Deliverable 5 - Development of an Improved Breeding Program

The objective of the new breeding program was to reduce cycle time and improve seedling traits in switchgrass. The overall breeding techniques were refined as the program evolved and the team gained further field experience. The main successes have been in incrementally strengthening the first year establishment in the field and optimizing processes. Using the selection methods described in Smart et al. (2003; 2004) along with those developed throughout the project, proved to be valuable breeding techniques. The resulting germplasm demonstrated improved first year growth and were better adapted to the environmental conditions in eastern Canada. By the end of this project we had successfully hastened emergence and improved establishment so much so that we were able to develop populations that produced a significant amount seed in the first year of establishment.

A detailed description of the breeding system used with the modifications to date is fully outlined in Appendix I.

Deliverable 6- Communication and Project Dissemination

The project has used a diversity of strategies to disseminate results including conferences, field visits and trade shows to expose the switchgrass breeding work in Quebec, Ontario the United States and Europe. The widest exposures to audiences have been through conference and trade show events.

REAP hosted a trade show booth and annual general meeting at the Guelph Organic Conference in February 2012, 2013, and will again be present in February 2014. This event attracts a diversity of interested parties in warm season grass production and utilization. It also allows us to network with a diversity of interested parties including farmers, scientists and business developers.

We also exposed the research at technical meetings on biomass and switchgrass production. This included a presentation at the 2013 Switchgrass II conference in Madison, Wisconsin on September 10-12, where Roger Samson was an invited to give an oral presentation on switchgrass breeding in Canada. This conference brought together switchgrass scientists and other prairie grasses specialists to discuss prairie grass research and opportunities for collaboration within the field. This event is the largest North American event for switchgrass breeders and is held on a biennial basis. It also provided Roger Samson an opportunity to spend several days with Dr. Annie Claessen from Agriculture Canada to discuss future collaboration opportunities on switchgrass breeding in Eastern Canada.

Oral presentations on the yield performance and morphological aspects of the breeding lines developed by REAP-Canada were presented by research associate Erik Delaquis. This occurred in Quebec City at the 81st Association Francophone pour le Savoir (ACFAS) conference in May

2013, and at the 21st European Biomass Conference and Exhibition in Copenhagen, Denmark in June 2013. This latter event is one of the world's premier events in the biomass sector. Roger Samson gave an oral presentation to the Ontario Biomass Producers Group (OBPG) in Guelph Ontario on February 4, 2013 on the REAP-Canada switchgrass genetic improvement program and highlighted the new germplasm and breeding techniques that were under development.

Roger Samson also presented a poster on the switchgrass breeding research at the OMAFRA Ontario Bioeconomy Workshop in Guelph, Ontario on November 26, 2013. This event provided an opportunity to network with leading Canadian research scientists working on biomass production and utilization. As well, it provided an opportunity to discuss market opportunities with various individuals and institutions present, including Mushrooms Canada, pellet producers and biocomposite producers.

Due to the concern of contamination of our switchgrass breeding nurseries by the large nearby field switchgrass plantations, we reduced planned dissemination activities at Ferme Norac in 2013 and instead included the sites at McGill in a number of field tours. A RPBQ field day was held in May of 2013 at the Ste. Anne-de-Bellevue research site was visited by MAPAQ, CEROM, Ag-Canada, McGill University and REAP-Canada. On several other occasions, the research sites at Ferme Norac in Salaberry-de-Valleyfield, QC and those in Ste-Anne-de Bellevue, QC were visited by commercial organizations including Ernst Conservation Seeds, the Ontario Biomass Producers Group, La Coop Fédérée, and Lafarge Cement.

In October, 2013 we invited research scientists from Ag-Canada in Quebec City to assess our materials at the end of the field season. Collaborative opportunities were discussed extensively, resulting in a proposal being submitted for a science cluster under Ag-Canada's AgriInnovation program to develop a joint breeding program on switchgrass as an agri-fibre feedstock.

Finally we recognized that there were serious limitations in using the western Quebec sites to enable farmers to learn more about switchgrass as an alternative crop for marginal lands in Quebec. In 2013 we decided to partner with La Coop Fédérée to plant and showcase several of the new switchgrass cultivars we have under development at their demonstration site near Mont-saint Hilaire, QC. This is a great exposure opportunity for the improved switchgrass germplasm as it is in a more centrally located demonstration site for existing and new potential switchgrass producers. It is more accessible for field exposures than the site at Ferme Norac located in the corn belt of Quebec near the Ontario border. We also exposed farmers to the project through Canada's outdoor farm show in Woodstock Ontario in 2012 and 2013. REAP President Don Nott and REAP-Vice-president Urs Eggimann hosted a trade show booth on behalf of REAP-Canada and handed out literature related to switchgrass development in eastern Canada.

The REAP Canada web site also provided wide visibility to highlight our work on genetic improvement of grasses. All the aforementioned conference and workshop presentations are

now available on the REAP Canada web site as well as several blogs. The numerous youth interns (23 in total) that worked on the warm season grass improvement program also wrote about the activity in their personal blogs to give other youth an exposure to the grass improvement effort.

SEE TABLE 2.3 FOR FURTHER DETAILS

2.2. Dissemination of Results

TABLE 2.3 DISSEMINATION OF RESULTS

APPENDIX A's planned activities	Completed Activities	Description (theme,title, location, etc.)	Completion date	# of people reached	Exposure given to the CAAP (logo, mention)
Conferences /Speaking Engagements 31 st & 32 nd Annual Guelph Organic Conference & Expo	Trade show booth	Organic & sustainable Agriculture Expo, Showcase REAP's switchgrass breeding and other activities; Guelph, ON	Feb 2012; 2013	300	Logo ; Mention
81st Congress of the ACFAS,	Performance de 11 sélections de panic érigée adaptée au sud du Québec.	Showcasing Francophone & international research; University Laval, Quebec City, Quebec	May 6-10, 2013	20-30	Mention
21 st European Biomass Conference and Exhibition	Presentation: Assessing cultivars of switchgrass in a Northern Environment	International scientific conference on biomass research, development, and policy. One of the world's leading events in the biomass sector; Copenhagen, Denmark	June 3-7, 2013	200 -250	Mention

APPENDIX A's planned activities	Completed Activities	Description (theme,title, location, etc.)	Completion date	# of people reached	Exposure given to the CAAP (logo, mention)
Switchgrass II Conference	Presentation: Development of improved switchgrass selections in a northern environment	A conference for switchgrass scientists and other prairie grasses specialists to discuss prairie grass research; Madison, Wisconsin	Sept 10-12, 2013	80	Logo; Mention
OMAFRA Ontario Bioeconomy Day	Poster presentation on Breeding Switchgrass in Eastern Canada	Ontario's annual meeting to highlight biomass production and bioconversion economy research	November 26, 2013	200	Logo; Mention
Field days	7 field visits	CEROM site visit with provincial grass specialists to discuss improved cultivars	May, Nov 2013	15	Mention
		RBPQ; CEROM; Ag-Canada; MAPAQ; McGill Lodds Agronomy Research farm Ste- Anne de Bellevue,	May 2013 Nov 2012	15-20	Mention
		Outdoor Farm Show Woodstock, ON	Sept 2012 Sept 2013	300	Logo; Mention

APPENDIX A's planned activities	Completed Activities	Description (theme,title, location, etc.)	Completion date	# of people reached	Exposure given to the CAAP (logo, mention)
		Coop Fédérée Research and farm exposure;	Nov. 2012	Potentially 100/ year	
		Ontario Biomass Producers Cooperative	Nov 2013	2	Mention
		LaFarge Cement & Ag-Canada	May 2013	10	Mention
Websites & Blog Material	Several news blogs and material posted on REAP blog, website and REAP intern blog site.	Dissemination events, updates on the project, intern's perspectives on switchgrass breeding.	Ongoing	300/day	Mention; Logo
Demonstration Days	Seed planted at La Coop Fédérée Demonstration centre - Le Domaine de Rouville	Seed of REAP's advanced populations of switchgrass was planted for permanent exhibition by La Coop Fédérée for increased exposure during their demonstration and field days In the region of Mont-Saint Hilaire	June 2013; ongoing	200 annually	Mention

3. CONCLUSION

The breeding cycles of the germplasm were incrementally improved each year and we gained experience in refining techniques to improve seed separation, early seedling growth and field transplanting, maintenance, genetic selection and cross breeding. Overall the project appears to have developed several highly promising germplasms which can be readily scaled up. As well, we have developed grass populations with better adaptation to marginal conditions that will be invaluable in future breeding efforts to develop these materials into commercially viable strains. Through our close collaboration with Ferme Norac, we have developed improved seed strains that will strengthen the plant material resource base for Eastern Canadian farmers to enable the expansion of warm season grasses especially in the provinces on Quebec and Ontario.

Short-term benefits

A major benefit to the agricultural sector is the development and release of high yielding, switchgrass germplasm adapted to eastern Canada. The seed multiplication nurseries and fields of promising accessions established at Ferme Norac are a positive step towards growing the warm season grass seed production industry in Quebec. The description of a low-labour breeding technique for C4 grasses and the data on the switchgrass accessions produced can be used to further develop materials that have improved seedling vigor for short season zones and imperfectly drained soils.

Medium-term benefits

The medium term benefits of the research include the expansion of the switchgrass seed production industry in both Québec and Ontario. The development of improved germplasm will hasten the increase in switchgrass plantings in Eastern Canada and provide biomass for further expanding and diversifying markets. The research will help develop switchgrass as an economically viable alternative to high-input crops on marginal land. This land could include imperfectly or coarsely drained soils or production areas in a low heat unit range (i.e. <2500 CHU areas). Ultimately the introduction of switchgrass in these areas will help improve soil fertility and reduce farmers' risks for crop production in marginal areas. The communication and dissemination activities will result in an increased utilization and awareness of switchgrass for planting on marginal land. This leads to increased commercialization efforts for the crop as a renewable energy source, and for biofibre and agrifood industries. The breeding approach developed here could also be adapted to include other native grass crops to further diversify the perennial grasses grown in Canada for biofibre and bioenergy applications.

Long-term benefits

In the long-term this grass improvement program will increase farm receipts in some of the most disadvantaged farming areas in eastern Canada. The development of higher yielding germplasm with improved establishment will increase profitability of these crops for farmers and enhance adoption. This will contribute to acreage expansion, which will help further

increase the confidence that the crop can be scaled up to meet the growing need for raw material resources of biomass conversion facilities. As well, we believe this research has identified the need for further development of switchgrass as a new fibre crop for eastern Canada. Increasing the biomass quality for fibre markets by increasing fibre strength and cellulose content will expand the commercial value of the material for these new emerging market applications.

Recommendations

We believe the project has demonstrated that genetic improvement of switchgrass using an integration of several selection methods at the seed, seedling and mature plant stage is the most logical step forward to help domesticate switchgrass as a field crop for eastern Canada. Fairly rapid progress can be made using the simplified breeding program developed here. Seed size can be effectively increased from cycle to cycle and rapid selection for early emergence reduces seed dormancy and overall emergence. Selection for single tillers increases seedling height and from the literature appears to contribute to increases in height and biomass of mature plants. A main priority is to reduce tiller mortality in growing swards by selecting for reduced tiller production in the seedling stage. Selecting at the seedling stage for the fastest emerging plants helps reduce dormancy and the efforts made in the greenhouse ensures plants are well established before field planting. We believe that further refinement of these techniques and developing a focused effort on improving the fibre quality of switchgrass for biomaterial applications will further help accelerate the business opportunities for switchgrass for producers and entrepreneurs looking to utilize the biomaterial. We also believe that other warm season grass species should be developed in eastern Canada (such as big bluestem) that may have improved fibre quality and will reduce the biological risks of growing switchgrass monocultures on a large area.

Project sustainability

We regularly collaborate with the Ontario Biomass Producers Cooperative, switchgrass farmers in Quebec and Ontario, CEROM, la Coop Fédérée and end users of switchgrass to keep abreast of the breeding priorities of farmers growing the crop and the end use markets. The fact that the project links directly with switchgrass seed producers to multiply these new cultivars ensures that farmers will have a sustainable supply of seed in the future through commercial sources.

The breeding effort in this project has allowed us to more fully understanding the morphological traits of switchgrass and ways to further improve biomass quality. By producing a better final product for bioconversion, we will improve the sustainability of enterprises introducing these new biomaterials into the marketplace by improving consumer acceptability. As well it is by advancing populations of grasses with better adaptation to marginal conditions that we will grow the land base available to grow biomaterials while minimizing interference with conventional food production systems.

Overall it can be projected that in Eastern North America, the development of appreciable acreage of switchgrass could be realized to supply the diversity of existing and emerging markets that come on stream. There is also a very large opportunity for switchgrass to be used as a straw substitute and wood shavings substitute for livestock bedding. Switchgrass is normally drier than cereal straw, and the increased cellulose content is highly desirable as improves absorbency. The mushroom trade is also a major user of straw in eastern North America and currently lacks fibre. Switchgrass is of interest to mushroom producers because of its long straw and structural strength that helps support an aerobic composting process. As well, it will help mushroom producers with a more stable feedstock price due to the reliability of production. Using improved breeding practices to develop market-specific switchgrass with higher yields to ensure consistent production will broadly increase switchgrass acceptance in the biomaterial and agri-food industries. These improvements will help accelerate planting and reduce farmers risk as it will increase the acceptance value of the material to stimulate a broad market development opportunity.

Project Follow-up

A proposal was submitted to the Ag-Canada AgriInnovation Program to continue the progress made with our switchgrass populations during this project. The next step is to further adapt our germplasm to the needs of the emerging markets. Breeding efforts on the advanced switchgrass germplasm developed in this project will continue with a focus on enhancing the fibre quality and yield of the populations. This will include efforts to increase the stem to leaf ratio of switchgrass and increase the percentage of reproductive tillers versus vegetative tillers. Aside from livestock feed uses, using the breeding program to develop a more fibrous switchgrass biomaterial will broadly improve the overall market acceptability and value of switchgrass for both biomaterial and the existing agri-food industry uses. Future market development for energy markets such as fuel pellets and cellulosic ethanol will benefit from an increased fibre content of the grass as well. It will improve the energy value of the feedstock by reducing ash content and increasing cellulose content. We believe that the introduction of improved cultivars will help switchgrass emerge as the first new large acreage crop in eastern Canada in the next 10 years.

4. SUMMARY OF PROJECT ACHIEVEMENTS

This project responds to the need to develop new high yielding biomass crops to enable farmers to diversify their crop production on marginal farmlands in eastern Canada. Farmers like growing switchgrass as a biomass crop but are requesting new genetic materials that are easier to establish, less prone to lodging, higher yielding and are better adapted to the cooler zones of eastern Canada. We optimized a breeding program for the region, as there were no existing breeding programs aimed at developing upland switchgrass in the eastern great lakes region. The main activity completed was the development of five advanced selections of switchgrass and two synthetic populations with improved agronomic traits. Seed of promising new selections was field multiplied by Ferme Norac, a commercial switchgrass seed grower, in Valleyfield Quebec. The project worked to increase seed size in switchgrass in order to improve germination, and to increase emergence to improve first year establishment. The breeding program also selected for reduced tillering in the grasses to prevent lodging, to prevent biomass from decaying in the bottom of the crop canopy as a means to increase yield and to increase height and weight of reproductive tillers.

The project identified and refined methods to more effectively enable switchgrass selection at the seedling and mature plant stage. The integrated breeding approach developed helps control the cost of operating breeding programs and to improve the efficiency of the selection process while reducing the plant breeding cycle from 3-4 years to 1-2 years.

The main goals of the native grass genetic improvement program are 1) to work with switchgrass seed producers to mass multiply the most advanced selections to enable farmers in eastern Canada to have access to improved switchgrass germplasms; 2) to help expand the commercial viability and impact of the crop in eastern Canada; 3) to continue genetically improving switchgrass and big bluestem in eastern Canada for bioindustrial and agrifood markets and 3) to continue refining low-cost breeding methods for native grasses and to improve the efficiency of selection for yield and agronomic traits while keeping the breeding cycle relatively short.

Further regional testing of these improved germplasms will be required through the RPBQ in Quebec and through the University of Guelph in Ontario to establish their suitability to the various zones in Ontario and Quebec. We anticipate that the acreage of warm season grasses in eastern Canada can expand by approximately 30% per year in the future through a concentrated effort to genetically improve the yield of grasses and to develop plant materials with improved quality traits for biomaterial and agri-food markets.



Figure 2.3 On-farm cooperator Normand Caron and Erik Delaquis in a breeding plot

5. FINANCING PLAN AND EXPENDITURE RECONCILIATION

Fill and forward the Financing Plan and Expenditure Reconciliation *(linked to Appendix B of the financial contribution agreement), a copy of which you have received electronically in MS Excel format.*

You must join all invoice copies related to the budgetary items. Contributions from the applicant and partners must also be justified. **No payment will be made before the paid supporting documents are submitted**.

Refer to the *instructions* available *in the first sheet of the* Excel worksheet entitled *Financing Plan and Expenditure Reconciliation*.

Any project can be submitted to an audit.

In accordance with the contribution agreement, you must keep the CDAQ informed of changes to the project and the financing plan.

Last update of the form by CDAQ : March 17, 2010

[References]

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<u>Methods – Switchgrass Breeding</u>

The 2 year breeding cycle system used previously appeared vulnerable to suboptimal weather conditions, which can cause delays in the breeding cycle and forcing a 3 year cycle. Both the field transplanting system and second year plots had modest results in 2011 because of less than optimal weather conditions in early 2011 (cool spring followed by a dry early summer). As a result, we decided efforts were required to further improve first year growth to make second year plants less vulnerable to drought. The modified system incorporates both greenhouse and field selection, creating a highly useful and efficient approach to quickly upgrade important traits in populations. This will lead to more rapid development of desired agronomic traits, requiring less time and labour by enabling fewer field plants to manage.

1. Seed Selection

Seeds are harvested from the fall populations and threshed and conditioned in the spring. Only the largest seed of each population is retained for the next breeding cycle by passing the seed collected through the air separator, which discards the lightest seeds and retains the heaviest. A large seed separation device was also developed that more effectively separates the heavy and light seed. This has given us greater air control regulation and speeds up seed processing in both the breeder seed lots and the seed increase plots at Ferme Norac.

2. Greenhouse planting

The growing medium used was a half and half mixture of PRO-MIX[®] HP-CC[™] high-porosity peatbased soil and PRO-MIX[®] BX[™] general purpose peat-based soil. We found that this mixture provided the optimal seed bed and drainage and was ideal for promoting seedling growth. Each cell was filled to within about 1 cm of the rim with the potting mixture and packed. Approximately 10-15 of the selected seeds were broadcast in each cell of a 38 cell tray, with 26 trays in total (approximately 1000 plants). The tray was then topped with soil and compressed to create a firm seed bed. The trays were watered every 1-2 days, to keep the soil moist but not soaked to ensure an aerobic soil environment.

Population	Date planted in	Date emerged			
	greenhouse (d/m/y)	(d/m/y)			
Blue Jacket IV	04/04/2013	22/04/2013			
Cave-in-Rock IV	02/04/2013	16/04/2013			
Cave-in-Rock Early Maturity III	02/04/2013	16/04/2013			
Cave-in-Rock IV x High Tide II	03/04/2013	17/04/2013			
Tecumseh IV	02/04/2013	17/04/2013			

3. Seedling Selection

The seedlings emerge approximately 2 weeks after planting. At this stage, each cell is thinned to the single fastest growing seedling per pot. This occurs when seedlings are approximately 1 cm tall. This rapid selection for early emergence helps to improve crop establishment under field conditions and appears to be effective in reducing seed dormancy and increasing seedling vigor.



Seedling selection in the 2 weeks after planting –thinned to the single fastest emerging seedling per cell

4. Single-Tiller selection

Selecting seedlings for reduced tillering may translate into a higher yield per tiller and less tiller mortality in mature plants (Smart et al. 2003;2004). After 8 weeks each population undergoes selection for low tillering. The 200 plants with the least amount of tillers are selected from each population.

Formerly, the seedlings were transplanted directly into the field after selections were made at this stage. Transplanting these young plants caused appreciable stress to seedlings and made them vulnerable to subsequent wind damage. Furthermore, considerable labour was required to water the vulnerable young transplants.

5. Greenhouse Transplanting

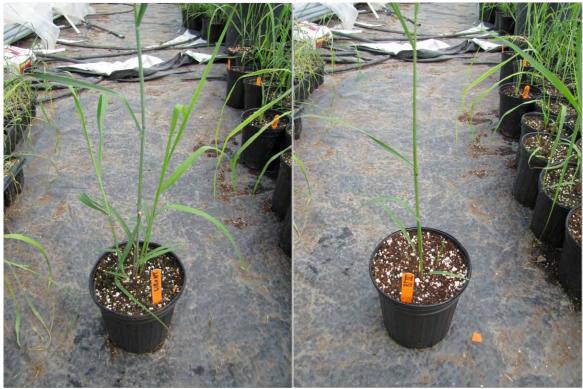
The 200 selected plants in each population are now transplanted into ¾ gallon pots and allowed to further mature in the greenhouse for an additional two weeks. In the previous year the ideal planting dates and duration of additional greenhouse growth had been determined for optimal growth. It was found that an extra two weeks' growth in the greenhouse not only reduces transplant shock, but improves overall first year establishment and growth, first year seed production potential, and allows for a reduced breeding cycle time. The improved establishment as a result of this additional step enables more vigorous and well-developed plants to be identified in the first or second year. In previous cycles where there were increased environmental stresses, such as a cold spring or dry weather, superior plants were not easily distinguished until the second and sometimes third year.



Switchgrass seedlings in the greenhouse - post transplanting into 3/4 gallon pots and ready to go to the field

6. Leaf Trimming

Due to observations of wind damage to young seedlings after field transplanting to the field, it was decided that the leaves should be trimmed to a shorter length before field transplanting. Therefore prior to removal from the greenhouse, the longest leaves of each plant are clipped, reducing the height of the plants to approximately 2 feet. This reduces damage caused by wind during establishment in the field and reduces moisture stress.



Left: Multiple tiller selection with one dominant centre tiller; Right: Selection with no emerging side tillers.

7. Spaced-Plant Nurseries & Location

The 200 selected plants are then transplanted in the spaced-plant nurseries. The ideal configuration was determined to be 8 rows of 25 plants, with 55cm between rows and 40 cm between plants. This spacing configuration enhances natural inter-plant competition, enabling superior plants to be easily identified. It also reduces labour requirements as there is less weeding required.

During the 2012 season, Ferme Norac seeded 15 hectares of Cave-in-Rock switchgrass approximately 150 meters southeast of the breeding nursery area. Extensive gene contamination can happen within 2 km of a pollen source of the same species, and viable grass pollen has been known to travel upwards of 10 km (Raynor et al. 1972; Watrud et al. 2004). Therefore, having the research plots located near local switchgrass fields is not recommended, and can potentially reverse any genetic gains made over the span of several years. In the final cycle, a new research site was found at McGill University's Agricultural campus, at the horticultural research farm. This was isolated from any outside sources of switchgrass grown at the Lods Agronomy Research Farm. It was decided that it was essential to plant the advanced populations in isolated nurseries at this location to remove any risk of pollen contamination.



Pépinière de sélection de Cave-in-Rock IV, trois semaines après la plantation.

Population	Date planted in nurseries
Cave-in-Rock Early Maturity III	June 19, 2013
Cave-in-Rock IV	June 20, 2013
Tecumseh IV	June 20, 2013
Cave-in-Rock IV x High Tide II	June 21, 2013
Blue Jacket IV	June 25, 2013

Table 3: Dates of transplanting to field nurseries



Cave-in-Rock IV breeding nursery, three weeks after planting.

8. Field Selection & Seed Harvest

The nurseries are continually monitored, and towards the end of the season (September-October) the best plants are visually selected and identified. All breeding selections are made under a relatively low input system to minimize the influence of herbicide and traffic on individual plants. Newly seeded breeding plots are kept relatively weed-free by mechanical weeding with a wheel hoe and by hand-hoeing. No N-fertilizer is applied in order to apply selection pressure favoring plants with adaptability to low N soil environments. Previous research has shown that there is a positive response to selection in low input environments (Rose et al. 2007). All seed and biomass is hand harvested for analysis at the end of the season (September - October). Seed is harvested by selecting 10-30 seed heads per plant, depending on the amount of tillering within the whole population. All seed is harvested on those plants previously labelled as superior.



Cave-in-Rock IV spaced-plant nursery – First year material. The second plant to the left is exhibiting excellent growth form with tall height, upright leaf architecture high into the canopy, limited tillering, and the majority of tillers are reproductive.

Table 3: Fall Harvest dates		
Population	Date harvested in	
	field nurseries	
Tecumseh IV	October 7, 2013	
Blue Jacket IV	October 11, 2013	
Cave-in-Rock Early Maturity II	October 15, 2013	
Cave-in-Rock IV	October 17, 2013	
Cave-in-Rock IV x High Tide II	October 21 2013	

9. Transplanting to the Next Breeding Cycle

It was also decided to recycle the best plants (approximately 5%) from each population into the next cycle, this helps to maintain superior genetics within the subsequently developed populations.

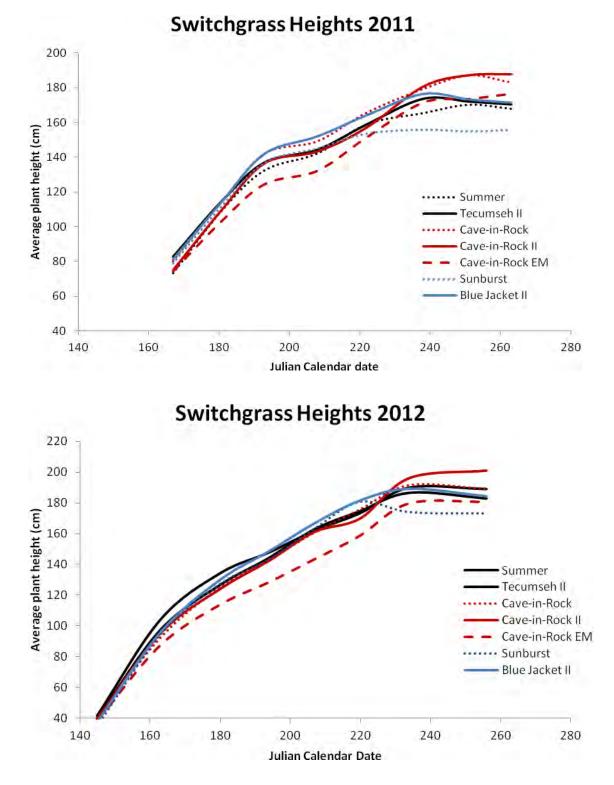
These plants are visually selected and labeled for transplanting into the next breeding cycle. It was found that in order reduce labour and transplant stress, it was better to only remove ¼ of the plant instead of moving the whole plant. The process of moving an entire plant proved labour intensive and significant transplant shock was experienced unless the plants were irrigated regularly. Both growth and seed production were greatly diminished unless the plants were constantly tended. Therefore, a corner of the selected mature plant is dug up and placed in a 2 US gallon pot. The plants are clipped to a height corresponding to at least 3 nodes on the stem (this was the height that resulted in the fastest re-growth). This plant is then allowed to grow in the greenhouse with regular watering for approximately 2 weeks, after which it can be transplanted into the next cycle with minimal transplant shock.

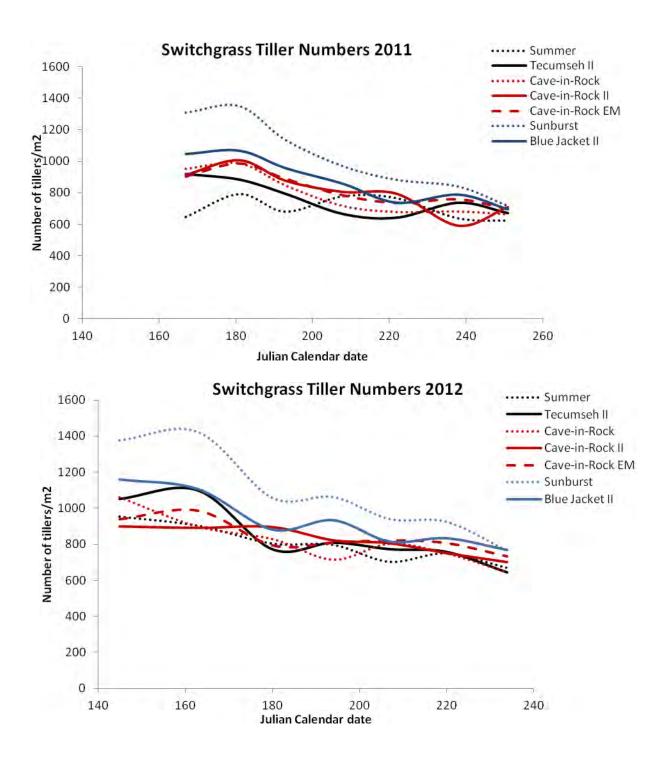


Blue Jacket III Spaced Plant Nursery - First year material

APPENDIX II







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