From: Bill Rooney

To: "Avant, Bob"; "Schmitt, Brian C."; "McCutchen, Bill"; "Helms, Adam"; "Mullet, John E."; "jmgould@tamu.edu"

Cc: "Baltensperger, David"; "Schuerman, Peter L."; "Simpson, Shay"

Subject: RE: CONFIDENTIAL: Shell Meeting
Date: Wednesday, August 12, 2009 9:14:00 AM

Attachments: Hodnett et al 2009 Sorghum-Saccharum - 8-9-09.doc

# Bob et al.,

Shell has pretty much already seen the Bio level presentation back in late April (about the same time of Bio).

I plan to use the recommendations provided by Brian Schmitt; that we use the manuscript that was attached to the IP claims made in the documentation that was submitted in late July. While the IP claims in that IP document went beyond the manuscript, my discussion will be limited to the information that is provided in the manuscript, which will be submitted for publication in the very near future.

As a reference, the manuscript is attached. It has not yet been submitted for publication so I would appreciate it if you would not distribute beyond this group.

regards,

bill

Dr. William L. Rooney Professor, Sorghum Breeding and Genetics Chair, Plant Release Committee Texas A&M University College Station, Texas 77843-2474 979 845 2151

----Original Message-----

From: Avant, Bob [mailto:bavant@tamu.edu] Sent: Wednesday, August 12, 2009 9:00 AM

To: Schmitt, Brian C.; McCutchen, Bill; Helms, Adam; wlr@tamu.edu; Mullet, John E.;

jmgould@tamu.edu; rjessup@tamu.edu

Cc: Baltensperger, David; Schuerman, Peter L.; Simpson, Shay

Subject: Re: CONFIDENTIAL: Shell Meeting

I continue to feel that Bill Rooney discuss at the level discussed at Bio in Atlanta Bob Avant

512/422-6171 (Cell)

From: Schmitt, Brian C.

**To**: McCutchen, Bill; Avant, Bob; Helms, Adam; 'wlr@tamu.edu' <wlr@tamu.edu>; Mullet, John E.; 'jmgould@tamu.edu' <jmgould@tamu.edu>; 'rjessup@tamu.edu' <rjessup@tamu.edu>

Cc: Baltensperger, David; Schuerman, Peter L.; Simpson, Shay

Sent: Wed Aug 12 08:57:42 2009

Subject: RE: CONFIDENTIAL: Shell Meeting

Bill:

I agree that the meeting should proceed. However, I also feel that there are some

components of the WH patent application that should not be disclosed. I have already discussed what these are with the PI's, particularly Bill Rooney and Dave Stelly.

If anyone else has any questions on what these items are, feel free to contact me. I think the discussion about what should and should not be discussed should not be in email or in writing.

Regards,

**Brian Schmitt** 

Brian Schmitt, J.D.
Intellectual Property Manager
Registered Patent Attorney
Office of Technology Commercialization
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1700 Research Parkway, Suite 250
College Station, TX 77845
979-862-6832
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From: McCutchen, Bill

Sent: Tuesday, August 11, 2009 7:58 PM

To: Avant, Bob; Helms, Adam; 'wlr@tamu.edu'; Mullet, John E.; 'jmgould@tamu.edu';

'rjessup@tamu.edu'

Cc: Baltensperger, David; Schuerman, Peter L.; Schmitt, Brian C.; Simpson, Shay

Subject: Re: CONFIDENTIAL: Shell Meeting

I think we should definitely meet with Shell.

We should also discuss in some depth on WH across sorghum, cane, miscanthus - and WH's potential. After filing (and reading) recent patent applications, we are "IP protected". We should provide this exciting information, I would just advise not sharing any future RD Specifics that may hold IP. Might be important to provide the breadth and diversity (accessions) that we have for cane, sorghum, etc.

Bill

From: Avant. Bob

To: Helms, Adam; 'wlr@tamu.edu' <wlr@tamu.edu>; Mullet, John E.;

'jmgould@tamu.edu' <jmgould@tamu.edu>; 'rjessup@tamu.edu' <rjessup@tamu.edu> Cc: Baltensperger, David; McCutchen, Bill; Schuerman, Peter L.; Schmitt, Brian C.;

Simpson, Shay

**Sent**: Tue Aug 11 18:57:18 2009

Subject: Re: CONFIDENTIAL: Shell Meeting

I agree that we should meet. Just exercise care. I think we should limit WH to what Bill Rooney presented in Atlanta. For the rest give our generic presentations and don't volunteer IP ideas.

**Bob Avant** 

512/422-6171 (Cell)

From: Helms, Adam

To: Bill Rooney <wlr@tamu.edu>; Mullet, John E.; J. Michael Gould

<jmgould@tamu.edu>; Russell Jessup <rjessup@tamu.edu>

Cc: Avant, Bob; Baltensperger, David; McCutchen, Bill; Schuerman, Peter L.; Schmitt,

Brian C. <Brian.Schmitt@tamu.edu>; Simpson, Shay

Sent: Tue Aug 11 17:49:36 2009 Subject: CONFIDENTIAL: Shell Meeting

I just spoke with Corey Radtke from Shell and he is under the impression that the NDA will not clear their legal counsel by the meeting this Thursday. Corey still believes a lot can be accomplished through discussion without the agreement in place.

Additionally, Dr. McCutchen would still like for us to have the meeting and give Shell a

field tour of the WH material.

Brian - would you be able to join us for this meeting Thursday and keep us within the IP safety arena? I have attached the agenda. Please let us know.

Thanks,

Adam

Adam Helms
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# Elimination of a reproductive barrier facilitates intergeneric hybridization of $Sorghum\ bicolor\ and\ Saccharum$

George L. Hodnett<sup>1</sup>, Anna L. Hale<sup>2</sup>, Dan J. Packer<sup>1</sup>, David M. Stelly<sup>1</sup>, Jorge da Silva<sup>3</sup> and William L. Rooney<sup>1</sup>

**Comment [reviewer1]:** who are these people? What was their contribution?

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<sup>&</sup>lt;sup>2</sup> United States Department of Agriculture – Agricultural Research Service - Sugarcane Research Unit, Houma, LA.

<sup>&</sup>lt;sup>3</sup>Texas AgriLife Research and Extension Center, Weslaco, TX.

## Abstract:

Growing interest in bioenergy production has increased efforts to breed for greater biomass through intra- and inter-generic hybridization. Consequently, increasing emphasis in both sorghum (Sorghum bicolor) and sugarcane (Saccharum spp.) breeding and genetic research is being placed on enhancing both the quantity and quality of biomass and ensuring the sustainability of the crop by minimizing input requirements through breeding for biotic and abiotic stress tolerance. The ability to consistently hybridize these species would facilitate the transport of complementary traits that increase adaptability, yields, and sustainability, from one crop to another. Previous efforts to hybridize these crops have had limited success, but the discovery of a specific mutant in sorghum has eliminated at least one prezygotic barrier to fertilization. Techniques used to produce a significant amount of seed from crosses between sorghum and sugarcane are described. Using these methods, our programs have grown 1,371 intergeneric hybrids. Seed set in the intergeneric crosses was affected by sugarcane pollinators, implying that breeding and selection of sugarcane pollen parents could further enhance successful hybridization. The Sorghum x Saccharum hybrids described in this paper are being used for introgression of traits into both species. Unlike previous attempts to hybridize these two genera, sufficient quantities of seedlings were produced to impose selection criteria with the goal of developing a new intergeneric cultivar with potential to be used for sugar or as a biomass feedstock. The long-term objective is to combine desirable traits of both sorghum and sugarcane.

# INTRODUCTION

Both sorghum (*Sorghum bicolor*) and sugarcane (*Saccharum sp.*) have been identified as potentially dedicated bioenergy crops. Consequently, increasing emphasis in both sorghum and sugarcane breeding and genetic research is being placed on enhancing both the quantity and quality of biomass and ensuring the sustainability of the crop through breeding for biotic and abiotic stress tolerance. An ideal bioenergy crop has numerous characteristics including high biomass, low input requirements, sustainability, and stress tolerance (Perlack et al., 2005). Biomass feedstocks have been explored in the past as a source of renewable energy, and today there are increasing numbers of studies assessing their strengths and weaknesses (Lipinsky, 1978; Clark et al., 1981; Goldemberg, 2007; Burner et al., 2009). Many of these strengths may be enhanced and the weaknesses could be overcome, if afforded the opportunity, by genes introgressed from other crops.

Crop improvement through breeding relies on genetic variation within the species. When this variation does not exist, or is limited, breeders turn toward wide hybridization or transgenic approaches to exploit genes from other sources. Transgenic approaches are effective for traits influenced by only a few genes and typically target a very specific trait. In addition, regulatory approval is cost prohibitive and public perception is sometimes a problem. For traits that are quantitatively inherited, introgression provides the most logical and effective approach to gene transfer, assuming that interspecific or intergeneric hybridization can be achieved. The probability of successfully hybridizing different crop species increases when the species are more closely related.

Sorghum is considered one of the closest relatives of the Saccharum complex, having diverged from a common ancestor as little as five million years ago (Al-Janabi et al.,

1994). Guimaraes et al. (1997) illustrated this relationship by showing colinearity of 190 RFLP probes on genetic maps of *Sorghum* and *S. officinarum*. This close relationship has been recognized for some time as *Saccharum* x *Sorghum* crosses have been reported with limited success (Venkatraman and Thomas, 1932; Bourne, 1935; Moriya, 1940; De Wet et al., 1976). Bourne (1935) also attempted the cross with sorghum as the female parent but was not successful. Only recently, Nair (1999) reported on the production of progeny from a *Sorghum* x *Saccharum* hybridization, but the frequency of viable progeny was low. From 3,670 well-pollinated florets only five seedlings were recovered. While there is obvious interest in creating and utilizing these hybrids between the two species, progress could be hastened by increased seed set and the ability to make selections among the resulting progeny.

The primary barrier to interspecific and intergeneric hybridization in sorghum is prezygotic; pollen tubes of alien species cease growth in pistils of sorghum before reaching the egg (Hodnett et al., 2005). Laurie and Bennett (1989) identified a sorghum gene, *Iap* (*Inhibition of Alien Pollen*), that permitted maize pollen tube growth to continue through the ovary to the micropyle when the sorghum female was homozygous for *iap*, but the recovery of sorghum-maize hybrids was not reported. Price et al. (2006) discovered that the same *iap* mutant removes the reproductive isolation between sorghum and several closely related wild taxa (*S. angustum*, *S. macrospermum* and *S. nitidum*) allowing the relatively easy production of new interspecific hybrids. Following this work, Kuhlman et al. (2008) documented the backcrossing of the previously described *S. macrospermum* hybrid to cultivated sorghum through the derivation of stable inbred lines with confirmed introgression from *S. macrospermum*. This introgression proves that

**Comment [reviewer2]:** Did they identify a gene, trait, or a marker? Bill likes to point out to me that there is no gene. It's a patented trait.

large segments of chromosomes can be moved across Poaceae species, which can facilitate the intergeneric transfer of important and quantitatively inherited traits.

Given the potential benefits to sugarcane and sorghum crops and the renewed interest in both crops as bioenergy feedstocks, there is a logical interest in hybridization to combine their desirable characteristics. These characteristics include, but are not limited to, drought tolerance and wide adaptation from sorghum along with sugar concentration and perennial growth habit from sugarcane. Another potential benefit of wide hybridization between the species is the possibility of introducing the seed production capacity of sorghum into sugarcane and, in the long-term, developing a sugarcane variety that can be planted from true botanical seed as opposed to the current labor-intensive whole-stalk or billet planting methods. The objective of this study was to determine if sorghum germplasm possessing the *iap* mutant can be used to increase the frequency of sorghum/sugarcane hybrids and to assess the relative effect of sugarcane pollinators on seed set and progeny viability.

# MATERIALS AND METHODS

Production of Sorghum/Sugarcane Hybrids: Seed of Tx3361, a line homozygous for iap and segregating for male sterility (Kuhlman et al. in review), was planted in pots in the greenhouse from mid-July through mid-September to ensure flower synchronization between the sorghum and sugarcane plants. At the onset of anthesis, male sterile plants of Tx3361 were identified based on anther phenotype and isolated from unknown pollen by covering with a paper bag. Sorghum/sugarcane pollinations were made at the USDA-ARS Sugarcane Research Unit in Houma, Louisiana between late September and early

November of 2007 and 2008. Additional pollinations were made in College Station, Tx in January and February of 2009.

A total of 67 basic and commercial sugarcane breeding lines were used as male parents.

In 2007 pollinations made in Houma were completed by dusting the sorghum panicle with freshly collected sugarcane pollen and by rubbing the sorghum panicle through the sugarcane tassel. Male parents included one released commercial sugarcane variety, one released energy-cane variety, three commercial breeding lines, four S. spontaneum accessions, and one Erianthus accession. Also included were six clones resulting from wide hybridization (basic breeding lines). These included clones resulting from the following crosses: one S. spontaneum x sugarcane (F<sub>1</sub>); one S. officinarum x sugarcane  $(F_1)$ ; one spont complex (S. spontaneum x S. spontaneum); two  $F_1$  x sugarcane (BC<sub>1</sub>); one BC<sub>1</sub> x sugarcane (BC<sub>2</sub>). In addition, one cross was made using multiple male parents (a polycross). In 2008, crosses were made by tapping tassels of a single sugarcane parent over the top of one to three sorghum panicles. To improve pollen load on the panicle, this was followed by rubbing the sorghum panicles into the sugarcane tassels. For a single cross, pollinations were repeated for 3-4 consecutive days during sugarcane anthesis. Males included five commercially released sugarcane varieties, 24 sugarcane breeding lines, two Erianthus accessions, one S. spontaneum accession, and 13 basic breeding lines. The basic breeding lines included 12 F<sub>1</sub> hybrids between S. spontaneum and sugarcane and one BC<sub>2</sub>. One polycross was also included in 2008. Pollinated sorghum plants were returned to College Station for seed development and maturation. The sorghum x sugarcane crosses made in College Station were completed using five commercial sugarcane breeding lines from the Texas AgriLife sugarcane breeding program in Weslaco, TX. Each sorghum panicle was pollinated only one time using the techniques developed in Houma in 2008.

Seed Harvest and Germination: Seed was allowed to develop and mature for 46, 41, and 27 days post pollination in 2007, 2008, and 2009, respectively. Seed from 2007 was stored from 30 to 90 d prior to germination. A high frequency of vivipary was observed in 2007 resulting in a loss of hybrids. To eliminate this problem in 2008 and 2009, seeds were not stored but were immediately germinated. Prior to germination seeds were surface sterilized by soaking them in a liquid suspension of Captan and Apron (Syngenta) for at least half an hour and then immersing them in a 30% solution of Chlorox TM (Proctor and Gamble, Oakland, CA) bleach for 20 minutes. Following surface sterilization, seeds were rinsed in sterile water and placed embryo side up in a petri dish containing a culture medium of Murashige-Skoog basal salts and vitamins (Murashige and Skoog, 1962) supplemented with 10 mg L<sup>-1</sup> glycine, 10 mg L<sup>-1</sup> L-arginine-HCl, 10 mg L-1 L-tyrosine, 100 mg L-1 inositol, and 30 g L-1 sucrose, solidified with 0.7% agar (plant tissue culture grade, Phytotechnology Laboratories, Shawnee Mission, KS) (Sharma, 1999). Petri dishes were maintained between 27 and 30 C under Gro-Lux<sup>TM</sup> flourescent lights (Sylvania, Danvers, MA) set to 14 hours per day. All seeds that showed good root and shoot development were placed in 4" pots. Once established, they were transferred to the greenhouse.

Confirmation of Intergeneric Hybrid Plants: Intergeneric hybrids were initially classified by morphology. As they developed, all hybrids exhibited numerous characteristics of sugarcane (e.g. height, tillering, and maturity) that the maternal parent did not possess. Plants assumed to be hybrids based on morphology were confirmed

Comment [reviewer3]: Add information on Texas males

**Comment [g4R3]:** I am assuming these are commercial breeding lines until we find out otherwise

Comment [reviewer5]: Not sure about Crop Science format, but you may need a location listed for Syngenta. using somatic chromosome numbers. Chromosome spreads were prepared from root tips using a method described by Jewell and Islam-Faridi (1994) with the following modifications. Young actively growing root tips were pretreated with a saturated aqueous solution of α-bromonaphthalene for 2.75 h at room temperature and fixed overnight in 95% ethanol/glacial acetic acid (3:1 v/v). Following fixation, root tips were rinsed several times with distilled water, hydrolyzed for 10 min in 0.2 M HCl and again rinsed in distilled water for 10 min. Cell walls were digested for 35 to 60 minutes at 37 C with an aqueous solution of 5% cellulase (Onozuka R-10, Yakult Honsha Co. Ltd., Tokyo) and 1.0% pectolyase Y-23 (Seishin Corporation, Tokyo) at pH 4.5 and subsequently rinsed three times with distilled water. Meristems were placed on a clean glass slide in an ethanol/glacial acetic acid (3:1) solution, macerated and spread with fine-tipped forceps, air-dried at room temperature for 2 d, and stained with Azure Blue. Root tip spreads were examined using a Zeiss Universal II microscope (Carl Zeiss Inc., Gottingen, Germany) with 63X and 100X apochromat objectives. Images were captured with an Optronics VI-470 system (Optronics Inc., Goleta, CA) and digitally stored and processed with Optimas (v. 6.1) image analysis software (Optimas Corp., Bothell, WA).

Effect of Sugarcane Pollinator on Hybrid Seed Set: For each cross made in Houma in 2008, the sugarcane parent, date of pollination, location of pollination, pollen rating, florets/panicle, seeds/panicle and seedlings produced were recorded. Pollen rating was a subjective measurement determined at the time hybrid seed was harvested by observing the amount of pollen present on stigmas of the sorghum panicle. The amount of pollen present on the stigmas was observed under a dissecting microscope and scored as 1, 2, or

3 with 1 being the least and 3 being the most. For each cross made in College Station in 2009 the sugarcane parent, seeds/panicle and seedlings produced were recorded.

To determine relative effect of location, date of pollination and sugarcane pollinator on seed set and pollen rating, was used. Only sugarcane males that had been used in at least three pollinations were included in the analysis. All effects were considered fixed and only interactions involving the pollinator were included the analysis of variance.

## **RESULTS**

2007 Hybrid Seed Production, Confirmation and Growth: In the fall of 2007, a total of 24 pollinations were made using 17 different pollinators (Table 1). Based on stigma reaction, it was apparent by two to three days post pollination that fertilization had occurred. Seed development was slower and the size was smaller when compared to intraspecific hybridization of sorghum. Embryo loss during seed development, and vivipary after development, became evident when the seed was prepared for germination. Further analysis revealed that these were common problems with 39% of the seed having no embryo, and 32% being viviparous. Seedlings were confirmed as intergeneric hybrids through chromosome counts, and represented a wide range of phenotypes, ranging from very poor in growth to highly vigorous.

From these pollinations, 23 of the most vigorous hybrids were transplanted to pots and placed in the greenhouse. Somatic chromosome counts for these hybrids ranged from (Fig. 1). These hybrids displayed a wide range of phenotypes but all had numerous long narrow leaves like sugarcane and most tillered profusely. Two hybrids,

vigorous growth than the others. In seven months, stalks of hybrid were 2.7 m in height and those of were 3.1 m (Fig. 1). Unlike both of these hybrids were photoperiod sensitive like sugarcane, and flowered from mid December through January in College Station whereas flowers in approximately 65 d regardless of planting date. The panicles on these two hybrids, which were slightly more compact than those of sugarcane (Fig. 1), were male sterile, and attempted backcrossings with the sorghum parent did not produce seed. In August, several stalks of each hybrid were cut to test for the ability to vegetatively propagate and to assess the accumulation of soluble sugars and their distribution. Vegetative propagation through nodal cuttings was successful and internode Brix values ranged from 8.5 to 19% with concentrations

2008/2009 Hybrid Seed Production and Enhancement of Process: In 2008 a total of 155 sorghum panicles (totaling 74,300 florets) were pollinated. From these pollinations, 10,347 seed were recovered, resulting in an average seed set of 14%. Percent seed set was not measured in the 2009 pollinations, but it appeared similar to that observed in 2008. Seed was harvested 40 d and 28 d post pollination in 2008 and 2009, respectively. Germination rates for the 2008 seed still suffered some from vivipary. In addition it was discovered that many of the embryos could not grow through the seed coat, which further limited germination rates in this year. In 2009 an additional decrease in maturation time further reduced vivipary, and excising the pericarp prior to planting removed the seed coat barrier. These minor modifications significantly improved germination rates from 2.5% in 2007, to 5.7% in 2008, and to a much improved 33% in 2009.

increasing with internode maturity as is seen in sugarcane (Whittaker and Botha, 1997).



From the combined 2008/2009 pollinations, a total of 1348 seedlings were transplanted to the greenhouse. The phenotypic variation present in these hybrids was extensive, but all were morphologically more like sugarcane than sorghum. These hybrids are expected to follow growth and development patterns observed in the limited set of hybrids evaluated from the 2007 crosses.

Effect of Pollinator Parent on Seed Set and Germination: Analysis of variance detected a significant effect of pollinator parent on seed set (Table 2), indicating that the source of sugarcane pollen is critical in the success of the production of intergeneric hybrids with had good seed set when pollinated with sugarcane clones which had seed set rates of respectively. These pollinators are of particular interest for the production of intergeneric hybrids, while other clones with poor seed set percentages (i.e. <10%) should be avoided (Table 3).

Pollen rating in sugarcane (Table 2) is influenced by genotype and environment, with the date of pollination having a significant effect on pollen shed (Moore and Nuss, 1987). In our study, clones with a low mean pollen rating consistently produced crosses with low seed set, but using clones with a high mean pollen rating did not necessarily produce high seed set. Six of the top seven sugarcane pollinators (defined by seed set percentage) had an average or above average mean pollen rating, while mean pollen ratings in males producing below average seed set varied (Table 3). These results imply that males must not only produce high pollen ratings but that they must also have favorable genetic and/or genomic compatibility with Tx3361.

Analysis of variance of the 2009 data indicated that once the seed was set, neither pollination environment nor sugarcane pollinator influenced percent germination. Based on the current methods of managing seed production and germination, it is reasonable to expect between 25-40% of seed to be viable regardless of which pollinator is used and where the pollination is made.

## DISCUSSION

An average seed set of 53% when using sugarcane pollinator was unexpectedly high for an intergeneric cross, considering attempts by previous researchers resulted in no more than a few plants (Nair, 1999). The high rate of seed production is attributed to the elimination of pre-fertilization barriers through the use of as well as compatibility of this line with particular sugarcane pollinators. Once produced, management of the hybrid seed prior to germination was critical to maximize production. Marked increases in viable seedlings were observed in each successive crossing year as problems affecting germination were identified. These increases resulted from the elimination of vivipary and physical barriers through early harvest and the removal of the pericarp.

Eliminating hybridization barriers and improving the germination rate has substantially increased the capacity to generate hybrids when compared to previous work. Nair (1999) "thoroughly pollinated" 3,670 florets and produced five seedlings for a success rate of 0.13%. In 2008, 16,813 florets were pollinated using males with a high pollen rating. Of these pollinations, 162 plants were produced for a success rate of 1%. Assuming that "thoroughly pollinated" is equivalent to a high pollen rating, this

results reported by Nair (1999). As modifications were made to the seed treatment, an additional 6-fold increase in recoverable progeny was achieved in 2009. Thus, the combined increases resulted in approximately a 40-fold increase in recovered progeny when compared to the previous report.

A limited number of male parents were screened in the current study. It is logical to assume that further screening will uncover additional compatible sugarcane pollinators that will expand production of intergeneric hybrids by increasing seed set and by improving seed quality. Therefore continued screening of *Saccharum* pollinators will be necessary to identify the best males for intergeneric hybrid production. The capacity to produce large-scale quantities of intergeneric *Sorghum/Saccharum* hybrids opens a wide range of possibilities for genetic improvement of sugar and bioenergy crops. While successful hybridization between sorghum and sugarcane, *S. spontaneum*, and early generation *Saccharum* hybrids, are described in this study, there is a need to determine the range of germplasm that can be hybridized using the developed lines and techniques. It may be possible to hybridize sorghum with other grasses of the Poaceae (e.g. *Miscanthus*, *Erianthus*, etc.) to facilitate introgression of positive traits among the genera/species.

The genetic and phenotypic variation present among the newly developed Sorghum/Saccharum hybrids presents significant opportunities. Given the amount of variation present and the large numbers of hybrids produced, segregation is expected to allow for the selection of elite hybrids. Even in 2007, the lowest of the three reported years for seedling production, there was enough variation among the 23 viable seedlings to select <u>two</u> that were visually superior to the others based on agronomic type. Further characterization of these <u>two</u> selected seedlings, as well as characterization of <u>of future</u> <u>selections is</u> necessary to determine unique strengths and weaknesses of the hybrids.

Selected hybrids can be used to introgress large genomic regions that control valuable quantitative traits from one species to the other. For example, the potential to transfer drought tolerance from sorghum to sugarcane or to introgress enhanced sugar production from sugarcane into sorghum could significantly influence energy and sugar production throughout the world. Because the initial F<sub>1</sub> hybrids appear to be steriledo not produce seed when crossed with sorghum, cytological manipulations will likely be needed, but established procedures provide approaches to mitigate this obstacle (Kuhlman et al., 2008).

If the hybrids possess unique and desirable agronomic characteristics and they perform well in agronomic trials, there is the potential to develop a new intergeneric hybrid crop. For example, a "sorcane" hybrid with high sugar accumulating capacity and enhanced water-use efficiency may be valuable as either a seed or vegetatively propagated crop. Additional research and development on sorghum seed parents and sugarcane pollinators must be completed to maximize seed production and development to make seed propagation a viable option. However, the germplasm and techniques described will produce seed quantities suitable for introgression, selection, and testing purposes.

Comment

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Kulman et al., in review

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