

From: [Bill Rooney](#)
To: ["Bill McCutchen"](#)
Cc: ["Schmitt, Brian C."](#); ["Peter Schuerman"](#)
Subject: PRC recommendation to release Tx3361
Date: Saturday, August 29, 2009 12:10:00 PM
Attachments: [PRC Recommendation](#) [REDACTED]

Bill:

Now that the utility patents and other documentation has been filed, I am submitting [REDACTED] for official release.

I propose that we release the germplasm and register it in the Journal of Plant Registrations. To do so, seed will be deposited with NSSL and we will request that they hold any distributions for 20 years. This will last throughout the life of the patent. If you are in agreement, please sign and return to me. For your reference (and the lawyers and OTC), I've also attached a copy of the registration manuscript that we will submit once release is officially approved.

If you've got questions, please let me know.

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

TEXAS AGRILIFE RESEARCH
PLANT MATERIAL RELEASE NOTIFICATION
REPORT OF TECHNICAL COMMITTEE ON SEED RELEASE AND INCREASE

The attached proposal for plant release has been examined and reviewed by members of the TAES plant release committee. Based on this review, the following recommendations regarding release are made. Release procedures followed those given in the TAES Policy on the Management & Release of Plant Materials – 1995.

1. Species: **Sorghum** (*Sorghum bicolor* L. Moench)
2. Breeders: **L.C. Kuhlman and W.L. Rooney**
3. Type of Release: Cultivar Parental Line
 Germplasm Genetic Stock
4. Recommended for Release: **Yes**
 No
5. Designation to be applied upon release: XXXXXXXXXX
6. Distribution of Breeder's Seed: **Breeder**
7. Increase and maintenance of Foundation Seed Stocks: **n/a**
8. Responsibility for providing seed to state and federal seed laboratories: **Breeder**
9. Publicity (including Station Seed Leaflet): none
10. Other Recommendations:

11. Members of plant review committee considering release: Smith, Rudd, Peterson, Nelson, Starr, Rooney

Personnel to Receive Copies of this Recommendation and Release Proposal:

Director : **x** Breeder: **x** TFSS: OTC: **x** Unit:
SSPB: **n/a** NSSL: **responsibility of breeder** Other:

Forwarded by:

Approved as Recommended:

Approved with Changes (see below)



W.L. Rooney
Chair, Plant Review Committee

Date: 8/29/09

Changes in Release:

B.F. McCutchen
Associate Director, Texas Agrilife Research

Date:

ADDITIONAL INFORMATION (not for general distribution) –

This germplasm was considered by the PRC in June of 2007. It was recommended for release, but it was not released due to significant IP concerns regarding distribution. Since that time, we submitted a utility patent documented the use of Tx3361 to create wide hybrids with an array of different grass species. The plan is to officially release and register the germplasm. Upon registration, seed of the line will be deposited with the NSSL with instructions that it not be released for 20 years.

Release Proposal for Sorghum Germplasm with the *iap iap* Genotype

A single gene locus, designated as *Iap* (Inhibition of Alien Pollen), is one cause of reproductive isolation between cultivated sorghum (*Sorghum bicolor* L. Moench) and wild *Sorghum* species outside the *Eu-Sorghum* section. In the homozygous recessive condition, the *iap iap* genotype eliminates this reproductive isolation and allows hybrids to be recovered between *S. bicolor* and wild *Sorghum* relatives (Hodnett et al., 2005; Price et al., 2006). This unique genotype was first described in *S. bicolor* accession 'NR481' (Laurie and Bennett, 1989), but this accession has very undesirable agronomic characteristics such as tall height, pigmented testa, and extreme susceptibility to lodging. Its potential for use in an introgression program is limited as any wild species genetic variation recovered in introgression progeny will be in a poor genetic background.

An improved *S. bicolor* germplasm possessing the *iap iap* genotype would make the evaluation of recovered introgression progeny easier, and breeding with such progeny would not necessitate selecting against the deleterious agronomic characteristics. In the process of our introgression program, we have developed such a germplasm. Herein we propose the release of a sorghum germplasm with the *iap iap* genotype that has significantly improved agronomic performance as well as segregation of the *ms3* genetic male-sterility system.

PROPOSED NAMES AND SEED HANDLING

This line was selected, evaluated, and increased in the TAES sorghum breeding program managed by Dr. William L. Rooney at College Station, TX. Using the numbering system of the TAES sorghum improvement program, this genetic stock will be designated as [REDACTED] upon release. After release, the line will be registered in *Crop Science* and seed sent for storage at the National Seed Storage Laboratory in Fort Collins, CO. Seed of this line will be maintained and distributed upon request by personnel in the Department of Soil and Crop Sciences, Texas A&M University, College Station, TX 77843-2474.

BREEDING HISTORY AND METHODOLOGY

This line was developed from a cross between genetic male-sterile [REDACTED], an unreleased derivative of [REDACTED] containing the *ms3* allele for genetic male-sterility, and NR481, an unreleased line homozygous for the *iap* allele (Laurie and Bennett, 1989). The hybrid was backcrossed once to the [REDACTED] parent. Fertile [REDACTED] progeny were self pollinated and selected for 3-dwarf height, white pericarp, no awns, absence of pigmented testa, and reduced lodging in College Station, TX 2005. [REDACTED] progeny were grown in a greenhouse, hand emasculated and tested for maize pollen tube growth (Laurie and Bennett, 1989). Genotypes at the *Iap* locus are based on qualitatively measuring maize pollen tube growth to the base of the style in sorghum pistils 24 hours after pollination. Individuals that show maize pollen tube growth to the base of the style are considered *iap iap* (Figure 1). Selected *iap iap* individuals were self pollinated and progeny rows were grown the following season in College Station, TX. Lines were evaluated for lodging, height, awns, and segregation of the *ms3* allele. Selected male-

fertile and sterile plants [REDACTED]) within *ms3* segregating rows were sib-mated. Individual sib crosses were grown in Weslaco, TX and evaluated for stable backcross segregation of *ms3*, lodging, height, maturity, and maize pollen tube growth was used to confirm their *Iap* locus genotype (Table 1). The selected line was bulk sib-mated between male-sterile and fertile plants to produce breeder's seed of the proposed genetic stock. The selected line is a maintainer of sterility in the A1 cytoplasmic male sterility system.

The observed expression of the *iap iap* genotype, maize pollen tube growth to the base of the style, was at a lower frequency than previously reported (Laurie and Bennett, 1989) and is likely environmentally influenced. [REDACTED] has expression similar to [REDACTED] in all tested environments. This genetic stock can be used as a female parent to obtain interspecific crosses with exotic sorghum species and possibly species beyond the genus. Any recovered introgression will be in a more favorable genetic background for further evaluation and breeding.

ACKNOWLEDGEMENTS

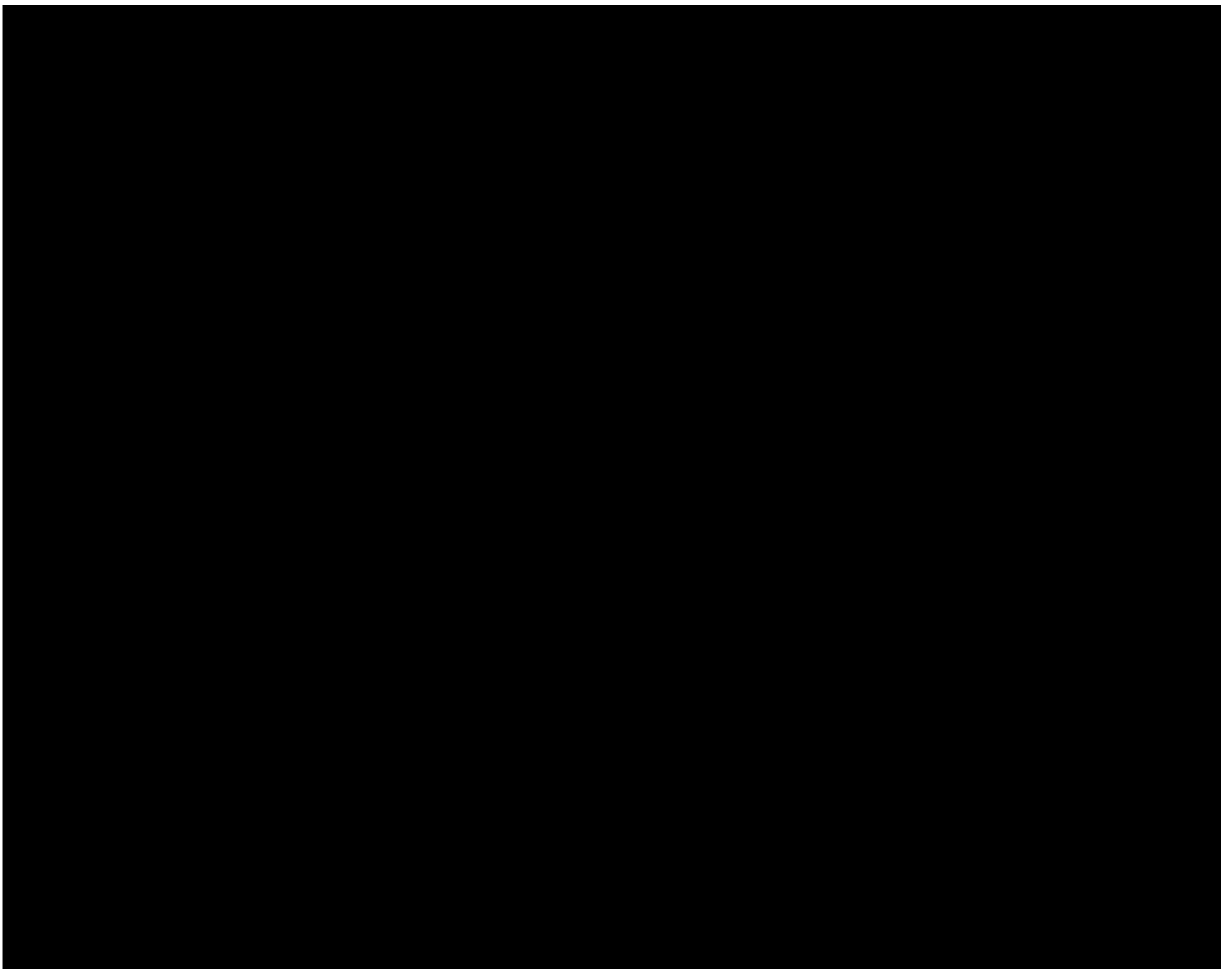
Financial support from the National Research Initiative of the USDA Cooperative State Research, Education and Extension Service, grant number # [REDACTED], the Texas Agricultural Experiment Station, and the USDA Sorghum Germplasm Committee.

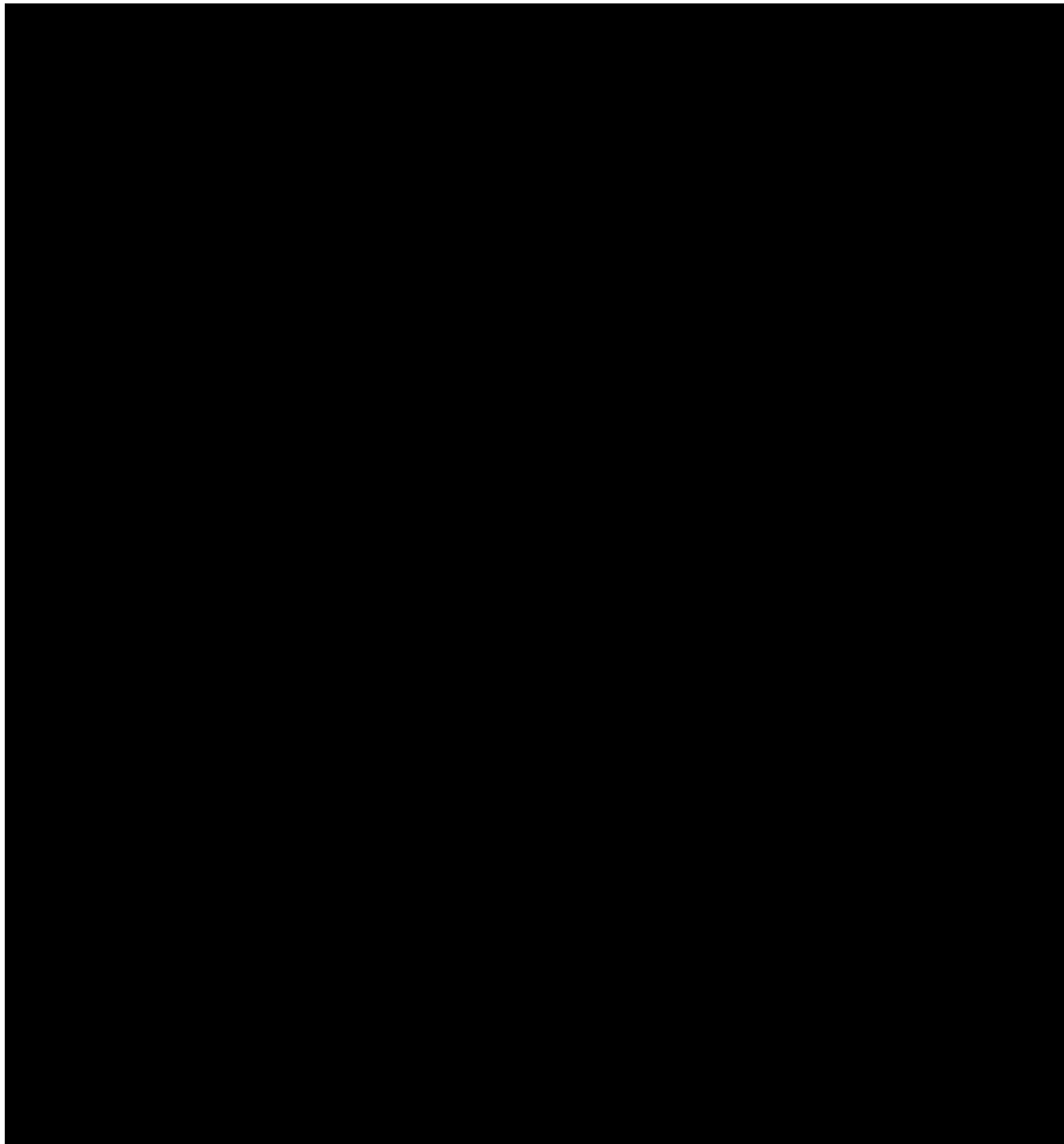
SCIENTISTS and TECHNICAL SUPPORT CONTRIBUTING TO THIS RELEASE

L.C. Kuhlman, Graduate Research Assistant, Tom Slick Graduate Fellow, TAMU, Soil and Crop Sciences, College Station, TX
W.L. Rooney, Professor, TAES, TAMU, Soil and Crop Sciences, College Station, TX
H.J. Price (Deceased), Professor, TAMU, Soil and Crop Sciences, College Station, TX
S.D. Collins, Research Associate, TAES, TAMU, Soil and Crop Sciences, College Station, TX

REFERENCES

- Hodnett, G.L., B.L. Burson, W.L. Rooney, S.L. Dillon, and H.J. Price. 2005. Pollen-pistil interactions result in reproductive isolation between *Sorghum bicolor* and divergent *Sorghum* species. *Crop Sci.* 45:1403-1409.
- Price H.J., G.L. Hodnett, B.L. Burson. S.L. Dillon, D.L. Stelly, and W.L. Rooney. 2006. Genotype dependent interspecific hybridization of *Sorghum bicolor*. *Crop Sci.* 46:2617-2622.
- Laurie, D.A., and M.D. Bennett. 1989. Genetic variation in *Sorghum* for the inhibition of maize pollen tube growth. *Ann. Bot.* 64:675-681.





Registration of [REDACTED] Sorghum Germplasm

L.C. Kuhlman¹² and W.L. Rooney^{1*}

¹Dep. of Soil & Crop Sciences, Texas A&M University, College Station, Texas 77843-2474

² Current address: Pioneer HiBred International, _____, Lawrence, KS _____.

Registration by CSSA.

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*Corresponding author (wlr@tamu.edu)

Abstract

The sorghum [*Sorghum bicolor* (L.) Moench] germplasm [REDACTED] (Reg. No. GP-____) was developed and released by Texas Agrilife Research sorghum breeding program in August 2009. This germplasm is unique as it lacks a key factor that represses alien pollen growth on the stigma. The inability to repress alien pollen tube growth increases the frequency of interspecific and intergeneric fertilization in sorghum. The increase is influenced by both the specific species and particular cultivars that are being tested. This line allows sorghum improvement programs with a unique opportunity to create interspecific and intergeneric sorghum hybrids for use as introgression parents.

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23 A single gene locus, designated as *Iap* (Inhibition of Alien Pollen), is one cause of
24 reproductive isolation between cultivated sorghum (*Sorghum bicolor* L. Moench) and
25 wild *Sorghum* species outside the *Eu-Sorghum* section. In the homozygous recessive
26 condition, the *iap iap* genotype eliminates this reproductive isolation barrier and allows
27 hybrids to be recovered between *S. bicolor* and wild *Sorghum* relatives (Hodnett et al.,
28 2005; Price et al., 2006). This unique genotype was first described in *S. bicolor*
29 accession [REDACTED] (Laurie and Bennett, 1989), but this accession has very undesirable
30 agronomic characteristics such as tall height, pigmented testa, and extreme susceptibility
31 to lodging. Its potential for use in an introgression program is limited as any wild species
32 genetic variation recovered in introgression progeny will be in a poor genetic
33 background. The development of [REDACTED] provides the sorghum breeding community
34 with an agronomically improved *S. bicolor* germplasm possessing the *iap iap* genotype
35 and segregating for Ms3 genetic male sterility to facilitate hybridization.

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37 **Methods**

38 [REDACTED] was selected, evaluated, and increased in the Texas Agrilife Research sorghum
39 breeding program at College Station, TX. The germplasm was developed from a cross
40 between genetic male-sterile [REDACTED] an unreleased derivative of [REDACTED] containing the
41 *ms3* allele for genetic male-sterility, and NR481, an unreleased line homozygous for the
42 *iap* allele (Laurie and Bennett, 1989). The hybrid was backcrossed once to the
43 [REDACTED] parent. Fertile [REDACTED] progeny were self pollinated and selected for 3-dwarf
44 height, white pericarp, no awns, absence of pigmented testa, and reduced lodging in
45 College Station, TX 2005. BC₁F₂ progeny were grown in a greenhouse, hand

emasculated and tested for maize pollen tube growth (Laurie and Bennett, 1989). Genotypes at the *Iap* locus are based on qualitatively measuring maize pollen tube growth to the base of the style in sorghum pistils 24 hours after pollination. Individuals that show maize pollen tube growth to the base of the style are considered *iap iap* (Figure 1). Selected *iap iap* individuals were self pollinated and progeny rows were grown the following season in College Station, TX. Lines were evaluated for lodging, height, awns, and segregation of the *ms3* allele. Selected male-fertile and sterile plants (BC₁F₃) within *ms3* segregating rows were sib-mated. Individual sib crosses were grown in Weslaco, TX in the fall of 2006 and evaluated for stable backcross segregation of *ms3*, lodging, height, maturity, and maize pollen tube growth was used to confirm their *Iap* locus genotype (Table 1). The selected line was bulk sib-mated between male-sterile and fertile plants to produce breeder's seed of [REDACTED]

Characteristics

[REDACTED] is a maintainer of sterility in the A1 cytoplasmic male sterility system. It is genetically a three dwarf with height similar to most grain sorghum parental lines (Table 1). [REDACTED] is 10-12 d earlier flowering than BTx623 in all environments, but it is 4-5 d later than [REDACTED]. Lodging in [REDACTED] were similar and both had reduced lodging compared to [REDACTED] (Table 1). The penetrance of the *iap iap* genotype, based on maize pollen tube growth to the base of the style, was lower than previously reported by Laurie and Bennett (1989) and is likely environmentally influenced. [REDACTED] has expression similar to [REDACTED] in all tested environments. This genetic stock has been used as a female parent to obtain interspecific crosses with exotic sorghum species and

possibly species beyond the *Sorghum* genus (Kuhlman et al. 2008, 2009; Hodnett et al., 2005, 2009; Price et al., 2006).

Availability

Seed of [REDACTED] will be maintained by personnel in the Department of Soil and Crop Sciences, Texas A&M University, College Station, TX 77843-2474. Requests for [REDACTED] should be directed to the Office of Technology Commercialization, Texas A&M University, College Station, Texas 77843-2474.

Acknowledgements

Financial support from the National Research Initiative of the USDA Cooperative State Research, Education and Extension Service, grant number # [REDACTED], the Texas Agricultural Experiment Station, and the USDA Sorghum Germplasm Committee.

References

- Hodnett, G.L., B.L. Burson, W.L. Rooney, S.L. Dillon, and H.J. Price. 2005. Pollen-pistil interactions result in reproductive isolation between *Sorghum bicolor* and divergent *Sorghum* species. *Crop Sci.* 45:1403-1409.
- Hodnett, G.L., A.L. Hale, D.J. Packer, D.M. Stelly, J. da Silva and W.L. Rooney. 2009. Elimination of a reproductive barrier facilitates intergeneric hybridization of *Sorghum bicolor* and *Saccharum*. *Crop Science* (submitted).

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96 recovered germplasm. *Genome* (accepted).

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99 46:2617-2622.

100 Laurie, D.A., and M.D. Bennett. 1989. Genetic variation in *Sorghum* for the inhibition of
101 maize pollen tube growth. *Ann. Bot.* 64:675-681.

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Table 1. Agronomic trait means of the two parents and Tx3361 when evaluated in Wesalco, Texas in the fall of 2006.

	NR481	BTx623ms3	Tx3361	LSD _(.05)
Dwarf Loci ¹	$_ dw_2 _ _^\dagger$	$dw_1 Dw_2 dw_3 dw_4$	$dw_1 Dw_2 dw_3 dw_4$	
Pericarp Color	Red	White	White	
Awns	Yes	No	No	
Pigmented Testa	Yes	No	No	
<i>ms3</i> backcross segregation	No	Yes	Yes	
Maize PTG ²	22.5% ^{A 3}	0.0% ^B	15.3% ^A	11.0%
<i>Iap</i> Locus	<i>iap iap</i>	<i>Iap Iap</i>	<i>iap iap</i>	
Height (in.)	92 ^{A 3}	54 ^B	54 ^B	7.5
Exsertion (in.)	8.3 ^A	3.6 ^B	4.5 ^B	2.8
Lodging ⁴	5.7 ^A	0.6 ^B	1.8 ^B	1.5
Days to 50% Anthesis	49 ^C	65 ^A	53 ^B	3.5

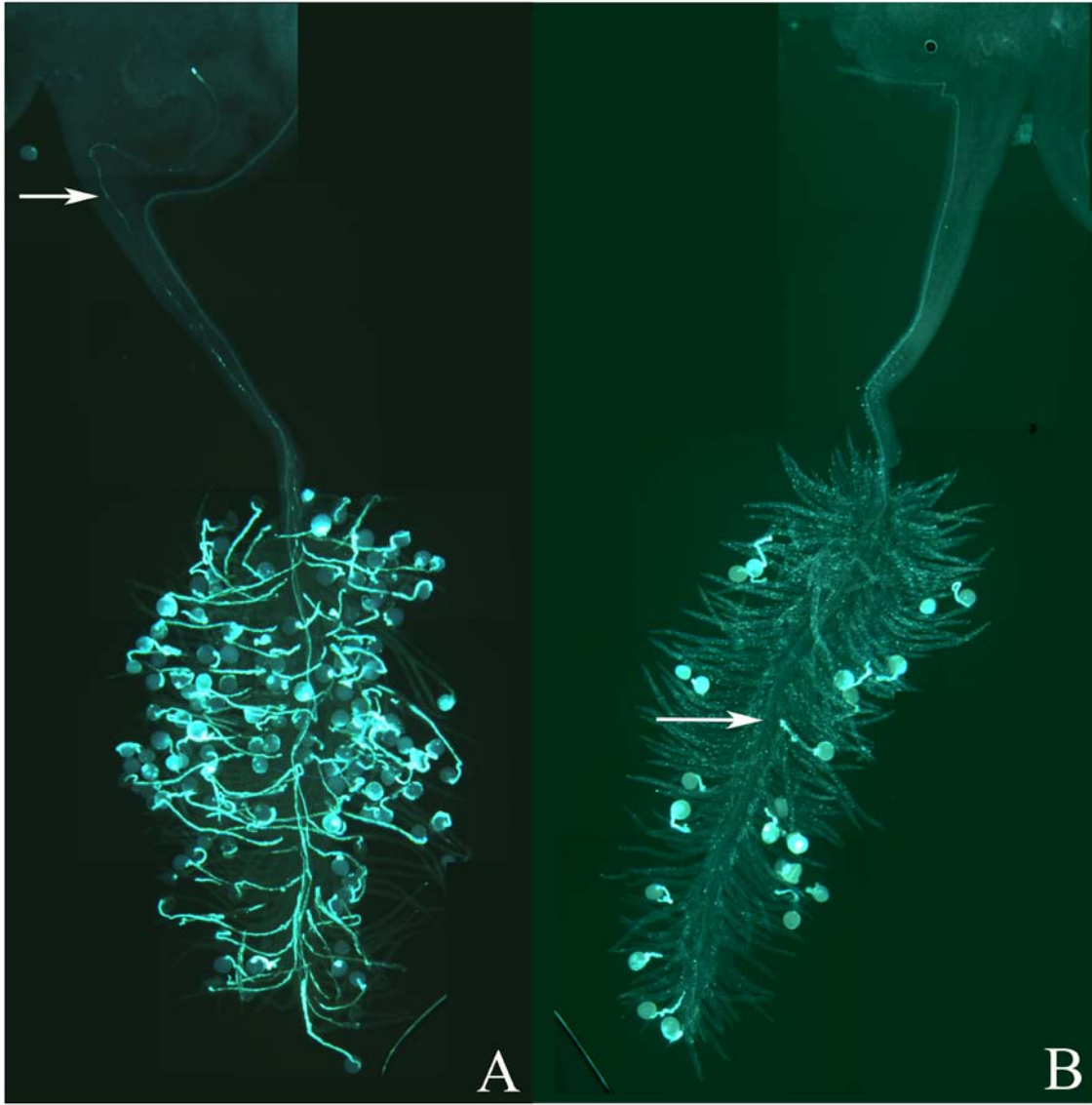
¹ Dwarf Loci: represents the homozygous allele at each dwarfing locus

[†] NR481' has 2 loci that are recessive but only the genotype at *Dw2* is known

² Frequency of sorghum pistils with maize pollen tube growth to the base of the style

³ Different letters within rows indicate significantly different means $\alpha = .05$

⁴ Lodging: 0 - 9 scale, 0 = 0-10%, 9 = 90-100% lodging



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107 Figure 1. (A) Tx3361 pistil showing maize pollen tube growth, arrow shows maize
108 pollen tube growing through the base of the style into the ovary and (B) BTx623 sorghum
109 pistil showing no maize pollen tube growth, arrow shows maize pollen tube failing to
110 enter the stigma axis.