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New Approaches to Selecting Resistance or Tolerance to SDS and Fusarium Root Rot

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Abstract

Fusarial rots are a significant problem worldwide affecting roots (and sometimes fruits) of most major crops including soybean, maize and wheat. Cultivar variation in partial resistance or tolerance is widespread and significant. Different cultivars of the soybean [*Glycine max* (L.) Merr.] have both resistance/tolerance to the leaf scorch known as Sudden Death Syndrome (SDS) and to the infection and root rot by the causal organism, *Fusarium virguliforme* (ex. *F. solani* f. sp glycines) hence the syndrome is composed of two diseases (1-3). Thirteen loci have been identified from analysis of 7 different crosses (2). Using new strains and new methods resistance loci in 'Hartwig' and 'Forrest', resistant cultivars clearly showed two loci underlie root resistance (lower LG G and D2) and four to eleven loci underlie leaf scorch resistance, depending on the cross made(eg, C2, F, I and upper G in ExF). Transcript abundance analysis of roots in response to *F. virguliforme* shows an orthologous set of transcripts accumulate during infection of resistant soybean cultivars and *Arabidopsis thaliana* that include the pathways leading to phenylpropanoid metabolism and its control, guanyl cylase a common second messenger and several transcription factors. Guanyl cyclase is also implicated in resistance in maize. In root disease resistance the genes implicated were known to be stress related. Therefore, *A. thaliana* is partially resistant and can be used to test both transgenes and mutants in candidate genes. Trangenics show fine maps to BACs have isolated some genes. For example, by fine mapping in NILs candidate genes underlying the controlling loci programming root resistance was a multi-stress resistance protein (lower G; *Rfs1*). For leaf scorch (*Rfs4*) an ascorbate peroxidase (C2) has been targeted. Also, *Rfs2*, a receptor like kinase (G) has been used to generate stable transgenic soybeans. Identification of the genes and loci conferring SDS resistance has provided options to breed improved cultivars with resistance to SDS.

Introduction

Among the top four loss causing diseases of soybean [*Glycine max* (L.) Merr.], worldwide were the root rot and leaf scorch called Sudden Death Syndrome (SDS; Wrather et al. 1996; 2003). Over a five year period 1999-2004 average losses around 1% or 0.9 million megagrams per harvest, worth $190 million a year, were reported. The syndrome was accurately predicted to intensify and spread over the next 20 years (Scherm and Yang, 1996). Improved genetic resistance in germplasm releases will be key to containing soybean losses to SDS (Gibson et al., 1994; Kazi et al., 2007).

SDS was shown to be caused by the blue-pigmented soil borne fungus *Fusarium virguliforme* (Aoki et al., 2003; ex. *Fusarium solani* (Mart.) Sacc. f. sp. glycines; Fsg; Roy, 1997). *F. virguliforme* is a member of an evolutionary group known as the "*F. solani* complex" that colonize a wide variety of habitats and hosts (Gray et al., 1999; O'Donnell, 2000). They are serious pathogens of many crops. Analysis in North America showed that only *F. virguliforme* prompted the symptoms of SDS on soybean but in south America two separate species, *F. tucumaniae* and *F. virguliforme*, were both responsible for SDS (Aoki et al., 2003;

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Covert et al., 2007).

The genetics of resistance to SDS is complex. Stephens et al. (1993) reported that a single dominant gene, *Rfs* controls SDS resistance in 'Ripley' soybean in greenhouse conditions. In contrast, the 'Essex' by 'Forrest' (ExF) population (Hnetkovsky et al., 1996; Chang et al., 1996; Kassem et al., 2006) showed that the SDS resistance was conditioned by several quantitative trait loci (QTL). By 2007, more than twenty detections of QTL for resistance to SDS have been reported among eight different recombinant inbred line (RIL) populations (Table 1). By assigning QTL detected in overlapping intervals to a single locus, the loci may be assigned to as few as 11-12 qRfs loci on nine linkage groups (LGs) including A2, C2, D2, F, G, I, J, L and N. The map of ExF showed three (Kassem et al., 2006, 2007) or four QTL (Iqbal et al., 2001) that mapped to linkage group G and one on each of linkage groups C2, F, J, I, L and N (qRfs1 to qRfs 9).

Some QTL for resistance to SDS have been confirmed (suffixed cqRfs-) by mapping to a similar location in separate populations and/or near isogeneic lines (NILs) derived from RILs segregating across regions encompassing QTL.

The confirmed QTL include C2 (Njiti et al., 1998; 2002), one on D2 (Farias-Neto et al., 2007), three all on G (Njiti et al. 2002; Prabhu et al., 1999), J (Sanitchon et al., 2004; Kassem et al., 2006, 2007) and N (Njiti et al., 2002; Hashmi, 2004). The ExF QTL on F and I (Iqbal et al., 2001; Kassem et al., 2007) were not yet confirmed by association in a second population by late 2007. Similarly not confirmed to date were the QTL found on A2 in 'Ripley' by 'Spencer' (Hashmi, 2004; Farias- Neto et al., 2007); L in 'Minsoy' x 'Noir 1' (Njiti and Lightfoot, 2006) and H in ExF grown in Argentina (unpublished).

Some cultivars of soybean have a dual resistance to SDS leaf scorch and root infection by the causal organism, *F. virguliforme* that was consistent in both field and greenhouse (Njiti et al., 1997, 2001, 2003; Hartman et al., 1997). Among dually resistant lines are Forrest, 'Hartwig', 'Jack', 'Ripley' and several commercial lines. Most of the dually resistant lines are also resistant to *Heterodera glycines* HG Type 0 (race 3) of the soybean cyst nematode (SCN). Subsequently, linkage and pleiotropy with loci underlying resistance to SDS have been detected at the SCN resistance locus *rhg1* but not *Rhg4* (Meksem et al., 1999; Triwitayakorn et al., 2005; Ruben et al., 2006).

In contrast, cultivars that show root susceptibility to *F. virguliforme* combined with SDS leaf scorch resistance (like 'Pyramid', 'Fayette' and 'LS92-1920') have been associated with resistance to *Heterodera glycines* HG Type 1.3.6.7 (race 14) of SCN (Gibson et al., 1994) across a wide germplasm collection. Consequently, repulsion linkage and/or pleiotropy is expected with loci underlying resistance to SDS at loci that underlie resistance to Hg Type 1.3.6.7 (Webb et al., 1995; Schuster et al., 2001; Concibido et al., 2004).

Preliminary separation of loci underlying root and leaf resistance used near isogeneic lines (NILs) to show a single root resistance locus in 'Forrest' (cqRfs1, requested to be renamed cqSDS-003) was about 10 cM from cqRfs2/*rhg1* gene cluster that separately conferred partial resistance to SCN and SDS leaf scorch (Njiti et al., 1998; Meksem et al., 1999; Triwitaya-

Table 1. Catalog of QTL underlying resistance to SDS through mid 2009. Black boxes indicate loci were detected in that study most were confirmed in another population or by NILs.

SGC Anotation	Linkage group	Map Position cM	Essex x Forrest in USA	Pyramid x Dougals	Flyer x Hartwig	Ripley x Spencer	Asgrow x Cordell	GC87018-12-2B-1xGC89045-13	Minsoy x Noir 1	Essex x Forrest in Argentina	Loci (QTL= q)	Marker	Secondary Marker	Map Position cM
	B1	84	■								*qRfs13*	Satt583	Satt415	82
	C1	91				■			■		*Dt3/qRfs9*	Sat_311	Satt578	65
	C2	146	■	■	■					■	*Rfs4*	Satt371	Satt307	
cqSDS001	D2	97		■	■	■	■				*qRfs7*	Sat_001	Satt226	
	F	42	■						■		*qRfs12*	Satt160	Satt510	85
cqSDS002	G	0	■								*qRfs3*	Satt163		
		3	■	■	■						*Rfs2*	Satt309		
		9	■								*Rfs1*	Satt570		
		38	■		■						*Rfs*	Sat_403	Satt130	
		140	■	■							*Dt2/qRfs11*	Satt138		
	H									■	*Rfs12*	Satt353		
	I	46	■		■		■				*qRfs5*	Satt354	Satt299	100
	J	42	■					■			*qRfs10*	Satt183	Satt249	
	L	78				■			■		*Dt1/(qRfs8)*	Sat_99	Satt226	
	N	45		■	■						*qRfs6*	Satt80		

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korn et al., 2005; Supplementary Table 1). The other loci on G (cqRfs2; or cqSDS-002) the locus *Rhg4* on LG A2 and the locus on C2 (cqRfs4; or cqSDS-004) were shown to have no effect on root infection (Njiti et al., 1998; Triwitayakorn et al., 2005).

The cultivar Hartwig was resistant to both leaf scorch and root rot (Wrather et al., 1995; Njiti et al., 1997, 2001; Mueller et al., 2003) and HG Type 1.3.6.7 (race 14) of SCN. Therefore, Hartwig might contain superior alleles underlying a combined SCN and SDS resistance. Cultivar Flyer was susceptible to SCN and both leaf scorch and root rot of SDS (Njiti et al., 1997; 2001). Recombinant inbred lines were developed from the cross of Flyer by Hartwig (FxH), released (Kazi et al., 2007) and used for preliminary QTL detection (Prabhu et al., 1999). A locus for resistance to root infection (Rfs1) was detected on LG G in the same interval as *rhg1* but not *Rhg4* in ExF (Prabhu et al., 1999; Table 1).

The mechanisms underlying resistance to root infection by *F. virguliforme* appear to include the increases in the abundance of transcripts encoded by stress- and defense-related genes. The response, over time, prevents the inhibition of cellular transcription found in susceptible roots (Iqbal et al., 2005). In turn, the *F. virguliforme* genome encodes several pathogenicity factors found in other Martiella species (Meksem and Covert, unpublished, 2007). These general plant pathogen responses might underlie the association between resistance to SCN and SDS. However, other mechanisms of resistance do operate. For example, since the pathogen is active in lignin degradation (Lozovaya et al., 2005), plant processes related to isoflavonoid production (Iqbal et al., 2003; Lozovaya et al., 2004), lignin deposition or modification (Triwitayakorn et al., 2005) might help prevent infection.

Mechanisms for leaf scorch development were expected to include infection rate and pathogen load (Njiti et al., 1997, 1998). However, there is evidence that genotypes with root resistance in the absence of sufficient leaf scorch resistance show higher amounts of scorch (Triwitayakorn et al., 2005; Lightfoot, unpublished). Involved in the leaf scorch are at least 4 different toxins (Baker and Nemec, 1994; Jin et al., 1996; Ji et al., 2006; Bhattacharryya et al., unpublished, 2007; Li et al., unpublished, 2007). Production, excretion, translocation, uptake and metabolism of the toxins are all stages at which plant genetic diversity might act. SCN infestion might indirectly alter toxin responses by weakening the plants or altering translocation.

Microarray analysis in both soybean and *Arabidopsis thaliana* suggests the mechanisms of resistance include the production of isoflavones and resistance to toxins that disrupt transcription (Iqbal et al., 2002; 2005; Yuan et al., 2008).

Materials and Methods

The genetic material used in this study consisted of; 92 $F_{5:14}$ RILS derived from the cross of 'Essex' x 'Forrest' (Lightfoot et al., 2005); NILs of ExF 11, 34, 60 and 77, ; 92 FxH F5:11-derived recombinant inbred line (RIL) mapping population (Yuan et al., 2002; Kazi et al., 2007); 92 $F_{5:14}$ RILs derived from the cross of 'Pyramid' x 'Douglas'; 200 $F_{5:14}$ RILs derived from the cross of 'Minsoy' x 'Noir'; *Arabidopsis thaliana* cv Columbia; and *Nicotiana tabacum* SR1. *Fusarium virguliforme* Mont-1 was used for inoculated experiments, natural infestations were used for field experiments.

In greenhouse experiments methods followed (Njiti et al., 2001). Plants were grown with a 14 h photoperiod and the air temperature ranged from 20±2°C at night to 27±2°C during the day in the green house. Parents and non-inoculated control plants were included in the experiments. RILs were sown in sterilized 1:1 (v/v) of sand and soil inoculated with *F. virguliforme* virulent strain Mont-1 in four-inch plastic cups and kept saturated at the lower 5 cm of the pot with water.

Sudden death syndrome DS was rated determined on the basis of the degree of leaf damage (chlorosis/necrosis) on each plant, and was rated on a scale of 1 to 9 (1 = 0–10%/1–5%, 2= 10–20%/6–10%, 3 =20–40%/10–20%, 4 =40–60%/20–40%, 5=60%/_40% of leaf surface chlorosis/necrosis, respectively, 6= up to 33% premature defoliation, 7 = up to 66% premature defoliation 8=66% premature defoliation, and 9 =premature death of plant). DI was on a percentage scale 1-100 (field only). SDS was rated at R5R6 and R7 in the field with regression to a R6 score for each line. In the greenhouse DS was at 21and/or 28 days after inoculation.

Roots were harvested and the green fluorescence was measured in the roots using Fluorometer of BIOTEK SYNERGY 2. Sample preparation for fluorometer readings took about 100 mg of root tissue ground in liquid nitrogen. The frozen slurry was mixed with 100 µl water in a 1.5 ml eppendorf tube by vortexing and was used to record fluorometer reading. The wave lengths used to record the fluorescence were; excitation at 485+20 nM; emission at 528+20 Nm; sensitivity 50; reading from the top 50% of the plate.

Results and Discussion

'Pyramid' by 'Douglas'

Four quantitative trait loci (QTLs) for resistance to SDS derived their beneficial alleles from 'Pyramid'. Three from the resistant parent were identified on linkage group D2 by OPZ19 (567 bp allele, P=0.0001, R^2=22.0%). G by BARC-Satt163 (261-bp allele, P=0.0005, R^2=16.0%) and linkage group N by BARC-Satt080 (230-bp allele, P=0.0009, R^2=15.6%). Beneficial alleles of three of the QTL were previously identified in 'Forrest', or 'Hartwig'. A QTL for resistance to SDS on linkage group C2 identified by BARC-Satt307 (292-bp allele, P=0.0008, R^2=13.6%) derived the beneficial allele from 'Douglas'. A beneficial allele of this QTL was previously identified in 'Essex'. Recombinant inbred lines that carry the beneficial alleles at all four QTL for resistance to SDS were significantly ($P<0.05$) more resistant than other recombinant inbred lines . Among these recombinant inbred lines resistance to SDS was environmentally stable. Therefore, gene pyramiding will be an effective method for developing cultivars with stable resistance to SDS. Crosses among the best lines of three populations were made in 2004 and selections are beginning.

'Flyer' by 'Hartwig'

Three QTL found in earlier studies were confirmed; one contributing resistance to leaf scorch on LG C2 (Satt277; P=0.004, R^2=15%) and two on LG G underlying root infection at R8 (Satt038; P=0.0001, R^2=28.1%; Satt115; P=0.003, R^2=12.9%). Satt038 was linked to *rhg1* underlying resistance to SCN Hg Type 0. Two QTL were discovered, the first was on linkage group (LG) G underlying resistance to SDS leaf scorch measured by disease index (Satt130; P=0.003, R^2=13%). The second QTL was on LG D2 underlying resistance to root infection at R6 (Satt574; P=0.001, R^2=10%). The QTL was in an interval previously associated with resistance to SCN Hg Type 1.3.6.7 and showed repulsion linkage that may explain the relative susceptibility to SDS of some SCN resistant cultivars. The loci and markers will provide tagged alleles with which to improve the breeding of cultivars combining resistances to SDS leaf scorch, root infection and SCN HG Type 1.3.6.7.

'Essex' by 'Forrest'

Three parameters were previously used to observe resistance to soybean sudden death syndrome (SDS) which includes Mean Disease Severity (MNDS), Mean Disease Incidence (MNDI), and Mean Disease Index (MNDX) (Kassem et al., 2006, 2007). Six QTL have previously been reported on linkage groups F (Satt160-Satt252), C2 (Satt489-Satt286), N (Satt080-Satt387), and three on G (OIO3-Satt122; Satt309-Satt214; Sct010-OPE02). BES derived SSRs confirmed a QTL for MNDS from QTL Cartographer (version 2.0). This QTL was found on linkage group F identified within the marker interval of SIUC_B15L05 BARC-Satt160 (Table 1). Beneficial alleles from 'Forrest' contribute to the reduction in MNDS by a mean difference of 0.04 between 'Essex' (1.50) and Forrest (1.46). MNDS LOD scores were graphed across the linkage group F by composite interval mapping in ExF RIL population. The QTL was reported significant as it was detected above a LOD score of 2.5 (Figure 1). The new marker SattB15L05 delimited the QTL to one side. BES derived SSRs confirmed a QTL for MNDX on linkage group F identified within the marker interval of SIUC_B15L05 BARC-Satt160 (Table 1). Beneficial alleles from 'Forrest' contribute to the reduction in MNDX by a mean difference of 0.7 between 'Essex' (9.6) and 'Forrest' (8.9). MNDX LOD scores were graphed across the linkage group F by composite interval mapping in ExF RIL population. The QTL was reported weakly-significant as it was detected slightly below a LOD score of 2.5.

Two new QTL on LG B1 was discovered by CIM re-analysis of the data. The first QTL was associated with Satt583-Satt415, a 3.2 cM interval, LOD 3.2 14.4, that explained 37% of the trait. The QTL was named qRfs13. The second was on LG G between Satt324-Satt594, LOD 3.0 that explained up to 35% of the trait variation. This locus was detected previously in 'Minsoy' x 'Noir' (qRfs11; Table 1) and was close to a determinacy locus *Dt2*. The new QTL will be tested to confirm in the NIL lines 57, 60 and 73 that segregate for SDS response but not at any of the 6 QTL detected previously.

Root Fluorescence in 'Essex' by 'Forrest'

There was a weak but significant correlation between root fluorescence and root resistance (r=0.08 and P=0.005). Non-infected roots showed major QTL on near Satt129 (D1a, 0.0032, R^2=11%), near Sat_001 (D2, 0.0072, R^2=10%) near Satt303 (G, 0.0318, R^2=7%), and near Satt 579 (D1b, 0.0582, R^2=6%).

Figure 1. Root rot scores from 1-9 does not correlate with leaf symptoms severity DS across a variety of RILs and NILs.

Table 2. Composite interval map reanalysis of the SDS data using genome sequence validated markers.

No.	Trait	LG	Marker/Interval	Position (cM)	LOD	Additive Effect	R^2
1	MNDI	B1	Satt583–Satt415	83	3.2		0.37
		J	Satt285a–Satt132	49	4.8		0.39
		G	Satt309a–Satt309b	12	6.3		0.18
		G	Satt324–Satt594	152	3.0		0.35
2	MNDS	G	Satt309a–Satt309b	12	4.0		0.12
		F	Sat_039–Satt160	16	4.8		0.20
3	MNDX	G	Satt309a–Satt309b	12	4.8		0.14
		F	Sat_039–Satt160	6	4.5		0.19

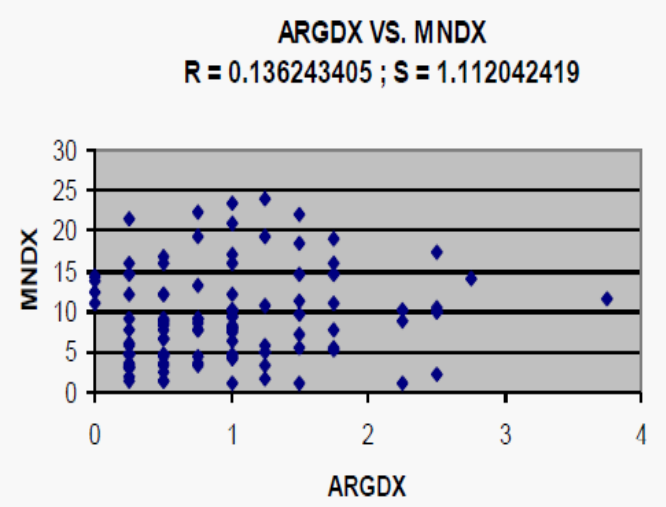

Figure 2. Correlation among SDS in Argentina and Mean Disease Index (MNDX) in USA. The values on the X and Y axes are those as expressed by ARGDX and MNDX respectively giving a distribution of points over 100 ExF RILs. It is a right-skewed (non-symmetric or non-normal) distribution as it is largely concentrated on the left side.

The QTL on G appeared to segregate in NIL population 34 and was encompassed by build 3 contig 763 and sequence scaffold 68 (see SoyGD) providing genomic resource for fine mapping and gene isolation. The QTL was distinct from the genes *Fr1*, *Fr2* and *Fr3* (blue fluorescence from UV light) and may be the previously detected but un-mapped *Fr4*, *Fr5* or a new gene *Fr6* (green fluorescence from blue light). Therefore, fluorescent root exudates, including phytoalexins, differ in 'Essex' and 'Forrest' and may underlie part of the resistance to root pathogens. The chemical nature of the fluorescence will be determined by GC-MS.

Marker Associations with SDS in Argentina (ARGDX)

Two markers SIUC_H32P20a (J-3; Q4S) SSR (LG D2) and SIUC_B27B08 SSR (Q3; Q4S) were weakly associated ($0.005<P<0.05$) with ARGDX (appendices). The linkage of B27B08 was not determined. Allelic mean difference was 0.51 between 'Essex' (0.86) and 'Forrest' (1.37) for marker H32P20a and 0.42 between 'Essex' (0.85) and 'Forrest' (1.27) for marker B27B08. The R^2 value was 0.13 with H32P20a and 0.07 with B27B08 (ctg 198 – FiS1i19 on this ctg). No QTL was detected with QTL Cartographer analysis for this trait. H32P20a is on B3 ctg 193 LG J near BSR®, plant height, leaf length. Correlations between SDS in Argentina and SDS in USA (MNDI, MNDS, and MNDX) were calculated (Figure 2). There was no linear relationship found between SDS in Argentina and MNDI, MNDS, and MNDX of SDS in USA. Correlation coefficient value was 0.16 (ARGDX vs. MNDI).

Arabidopsis thaliana Microarrays

The analyses of microarrays measuring TA in whole plants after *A. thaliana* cv 'Columbia' was challenged with fungal pathogen *F. virguliforme*. Infection caused significant variations in TAs. The number of increased transcripts was nearly four times more than decreased in abundance. A putative resistance pathway involved in responding to the pathogen infection in *A. thaliana* was identified and compared to that reported in soybean. The coordinated regulation of genes adjacent or clustered at certain genomic regions was detected. Microarray experiments allowed the identification of plant pathways likely to be involved in plant resistnce to fusarial pathogens. Dissection of the set functional orthologous genes between A.thaliana and *G. max* enabled a broad view of the functional relationships and molecular interactions among plant genes involved in *F. virguliforme* resistance.

Transgenic Plants

The analysis of transgenic tobacco expressing the *gdhA* gene showed resistance to root rot by *Fusarium*. This resistance is broad in nature, to all necrotrophic rotting fungi and seems to be related to alterations in special nitrogen metabolism that increases anti-fungal metabolites (Fakhoury and Lightfoot, 2010). In contrast the plants transgenic with the RLK at *rhg1* (*Rfs2*) showed both a significant increase in root resistance (3 IS units) and reduction in leaf scorch (4 DS units) compared to non-transgenic controls (Lightfoot, 2011; Srour et al., 2012). Arabidopsis plant transgenic with whole BACs (*Rfs1*) showed root resistance and leaf scorch resistance. In conclusion, the era of QTL confirmation by gene isolation has arrived (Ullah et al., 2012). Insights into the mechanisms of resistance should allow improved selection for resistance.

References

Achenbach L, J Patrick, and L Gray (1996) Use of RAPD markers as a diagnostic tool for the identification of Fusarium solani isolates that cause soybean sudden death syndrome. Plant Dis 80: 1228-1232.

Afzal AJ and DA Lightfoot (2007) Soybean disease resistance protein RHG1-LRR domain expressed, purified and refolded from Escherichia coli inclusion bodies: preparation for a functional analysis. Protein Expr Purif 53:346-55.

Afzal J, A Wood, and DA Lightfoot (2008) Plant receptor-like serine threonine kinases: Roles in signaling and plant defense. Molecular Plant Microbe Interactions 21:507-517.

Anand SC (1992) Registration of "Hartwig" soybean. Crop Sci. 32:1060-1070.

Aoki T, K O'Donnell, Y Homma, and AR Lattanzi (2003) Sudden-death syndrome of soybean is caused by two morphologically and phylogenetically distinct species within the Fusarium solani species complex-F. virguliforme in North America and F. tucumaniae in South America. Mycologia 95: 660-684.

Baker RA, and S Nemec (1994) Soybean sudden death syndrome: isolation and identification of a new phytotoxin from cultures of the causal agent, Fusarium solani. abstract). Phytopathology 84:1144.

Chang SJC, TW Doubler, V Kilo, RJ Suttner, JH Klein, ME Schmidt, PT Gibson, and DA Lightfoot (1996) Two additional loci underlying durable field resistance to soybean sudden-death syndrome. SDS). Crop Sci 36:1624-1628.

Covert SF, T Aoki, K O'Donnell, D Starkey, A Holliday, DM Geiser, F Cheung, CD Town, A Strom, J Juba, M Scandiani, and XB Yang (2007) Sexual reproduction in the soybean sudden death syndrome pathogen Fusarium tucumaniae. Fungal Genetics and Biology 44: 799-807.

Farias-Neto AF, R Hashmi, ME Schmidt, SR Carlson, GL Hartman, S Li, RL Nelson, and BW Diers (2007) Mapping and confirmation of a sudden death syndrome resistance QTL on linkage group D2 from the soybean genotypes 'PI 567374' and 'Ripley'. Mol Breeding 20: 53-62.

Gibson PT, MA Shenaut, VN Njiti, RJ Suttner, and O Myers Jr (1994) Soybean varietal response to sudden death syndrome. p. 436–446. In D. Wilkinson. ed.. Proc. Twenty-fourth Soybean Seed Res. Conf., Chicago, IL. 6–7 Dec. 1994. Am Seed Trade Assoc Washington DC.

Gray LE, LA Achenbach, RJ Duff, and DA Lightfoot (1999) Pathogenicity of Fusarium solani f. sp. glycines isolates on soybean and green bean plants. J Phytopathol 147:281-284.

Hartman GL, YH Huang, RL Nelson, and GR Noel (1997) Germplasm evaluation of Glycine max for resistance to Fusarium solani, the causal organism of sudden death syndrome. Plant Dis 81:515-518.

Hashmi RY (2004) Inheritance of resistance to soybean sudden death syndrome. SDS. in 'Ripley' x 'Spencer' F5 derived lines. PhD Dissertation, Plant Biology, SIUC, Carbondale, USA.

Hnetkovsky N, SJC Chang, TW Doubler, PT Gibson, and DA Lightfoot (1996) Genetic mapping of loci underlying field resistance to soybean sudden death syndrome (SDS). Crop Sci 36:393-400.

Iqbal MJ, K Meksem, VN Njiti, MA Kassem, and DA Lightfoot (2001) Microsatellite markers identity three additional quantitative trait loci for resistance to soybean sudden-death syndrome (SDS) in Essex x Forrest RILs. Theor Appl Genet 102:187-192.

Iqbal MJ, S Yaegashi, R Ahsan, KL Shopinski, and DA Lightfoot (2005) Root response to Fusarium solani f. sp. glycines: temporal accumulation of transcripts in partially resistant and susceptible soybean. Theor Appl Genet 110:1429-1438.

Ji J, MP Scott, and MK Bhattacharyya (2006) Light is essential for degradation of ribulose-1,5-biphosphate carboxylase-oxygenase large subunit during sudden death syndrome development in soybean. Plant Biology 8, 597-605.

Jin H, GL Hartman, CD Nickell, and JM Widholm (1996) Characterization and purification of a phytotoxin produced by Fusarium solani, the causal agent of soybean sudden death syndrome. Phytopathology 86, 277–282.

Kassem MA, J Shultz, K Meksem, Y Cho, AJ Wood, MJ Iqbal, and DA Lightfoot (2006) An Updated 'Essex' by 'Forrest' Linkage Map and First Composite Interval Map of QTL Underlying Six Soybean Traits. Theor Appl Genet 113:1015-1026.

Kassem MA, K Meksem, AJ Wood, and DA Lightfoot (2007) Additional QTL for SDS and SCN resistances in a soybean recombinant inbred line population of 'Essex' by 'Forrest'. Reviews in Biology & Biotechnology 6 (1): 11-19.

Kazi S (2005) Identification of twenty three loci conditioning agronomic traits in recombinant inbred and near isogeneic soybean lines derived from Flyer by Hartwig. MS Thesis SIUC Carbondale IL, USA, pp 212

Kazi S, J Shultz, R Bashir, J Afzal, VN Njiti, and DA Lightfoot (2008) Separate loci underlie resistance to soybean sudden death syndrome in 'Hartwig' by 'Flyer'. Theor and App Genet 116: 967-977.

Kazi S, VN Njiti, TW Doubler, J Yuan, MJ Iqbal, S Cianzio, and DA Lightfoot (2007) Registration of the Flyer by Hartwig Recombinant Inbred Line Mapping Population. J Plant Registr 1: 175-178

Li S and GL Hartman (2003) Molecular detection of Fusarium solani f. sp. glycines in soybean roots and soil. Plant Pathol 52:74-78.

Lightfoot DA (2008) Soybean Genomics: Developments through the use of cultivar Forrest. International J of Plant Genomics 2008:1-22. doi:10.1155/2008/793158.

Lozovaya VV, AV Lygin, OV Zernova, S Li, GL Hartman, and JM Widholm (2004) Isoflavonoid accumulation in soybean hairy roots upon treatment with Fusarium solani. Plant Physiol Biochem. 42:671-679.

Lozovaya VV, AV Lygin, OV Zernova, S Li, JM Widholm, and GL Hartman (2005) Lignin degradation by Fusarium solani f. sp. glycines. Plant Disease 90: 77-82.

Meksem K, P Pantazopoulos, VN Njiti, DL Hyten, PR Arelli, and DA Lightfoot (2001) 'Forrest' resistance to the soybean cyst nematode is bigenic: saturation mapping of the Rhg1 and Rhg4 loci. Theor and App Genet 103:710-717.

Meksem K, TW Doubler, K Chancharoenchai, VN Njiti, SJC Chang, PR Arelli, PB Cregan, LE Gray, PT Gibson, and DA Lightfoot (1999) Clustering among loci underlying soybean resistance to Fusarium solani, SDS and SCN in near-isogenic lines. Theor Appl Genet 99:1131-1142.

Mueller DS, RL Nelson, GL Hartman, and WL Pederson (2003) Response of commercially developed soybean cultivars and ancestoral soybean lines to Fusarium solani f. sp. glycines, the causal organism of sudden death syndrome. Plant Dis 87:827-831.

Njiti VN and DA Lightfoot (2006) Genetic analysis infers Dt loci underlie resistance to SDS caused by Fusarium virguliforme in indeterminate soybeans. Can J Plant Science 41:83-89.

Njiti VN, JE Johnson, TA Torto, LE Gray, and DA Lightfoot (2001) Inoculum rate influences selection for field resistance to soybean sudden death syndrome in the greenhouse. Crop Sci 41: 1726-1731.

Njiti VN, K Meksem, MJ Iqbal, JE Johnson, MA Kassem, KF Zobrist, VY Kilo, and DA Lightfoot (2002) Common loci underlie field resistance to soybean sudden death syndrome in Forrest, Pyramid, Essex, and Douglas. Theor Appl Genet 104:294-300.

Njiti VN, L Gray, and DA Lightfoot (1997) Rate-reducing resistance to Fusarium solani f.sp. phaseoli [nee: glycines] underlies field resistance to soybean sudden-death syndrome. SDS). Crop Sci 37: 1–12

Njiti VN, MA Shenaut, RJ Sutter, ME Schmidt, and PT Gibson (1996) Soybean response to soybean sudden-death syndrome: inheritance influence by cyst nematode resistance in Pyramid x Douglas progenies. Crop Sci 36: 1165-1170

Njiti VN, O Myers Jr, D Schroeder, and DA Lightfoot (2003) Roundup ready soybean: Glyphosate effects on Fusarium solani root colonization and sudden death syndrome. Agronomy J 95: 1140-1145.

Njiti VN, TW Doubler, RJ Suttner, LE Gray, PT Gibson, and DA Lightfoot (1998) Resistance to soybean sudden death syndrome and root colonization by Fusarium solani f. sp. glycines in near-isogeneic lines. Crop Sci 38:472-477.

O'Donnell K (2000) Molecular phylogeny of the Nectrai hematococcca-Fusarium solani species complex. Mycologia 92:919-938.

Prabhu RR, VN Njiti, JE Johnson, ME Schmidt, RJ Klein, and DA Lightfoot (1999) Selecting soybean cultivars for dual resistance to cyst nematode sudden death syndrome with two DNA markers. Crop Sci 39:982-987.

Roy KW (1997) Fusarium solani on soybean roots: Nomenclature of the causal agent of sudden death syndrome and identity and relevance of F. solani form B. Plant Dis 81:259-266.

Sanithchon J, A Vanavichit, S Chanprame, T Toojinda, T Triwitayakorn, VN Njiti, and P Srinives (2004) Identification of simple sequence repeat markers linked to sudden death syndrome resistance in soybean. Science Asia 30:205-209.

Scherm H and XB Yang (1996) Development of sudden death syndrome of soybean in relation to soil temperature and soil water potential. Phytopathology 86:642-649.

Schuster I, RV Abdelnoor, SRR Marin, VP Carvalho, AS Kiihl, JFV Silva, CS Sedyama, EG Barros, and MA Moreira (2001) Identification of a new major QTL associated with resistance to the soybean cyst nematode. Heterodera glycines). Theor Appl Genet 102:91-96.

Shultz JL, CR Yesudas, S Yaegashi, JA Afzal, S Kazi, and DA Lightfoot (2006b) Three minimum tile paths from bacterial artificial chromosome libraries of the soybean. Glycine max cv. 'Forrest': Tools for structural and functional genomics. Plant Methods 2:9-18.

Shultz JL, D Kurunam, K Shopinski, MJ Iqbal, S Kazi, KF Zobrist, R Bashir, S Yaegashi, N Lavu, AJ Afzal, CR Yesudas, MA Kassem, C Wu, HB Zhang, CD Town, K Meksem, and DA Lightfoot (2006a) The Soybean Genome Database. SoyGD): A browser for display of duplicated, polyploid, regions and sequence tagged sites on the integrated physical and genetic maps of Glycine max. Nucleic Acids Res 34:D758-765.

Shultz JL, S Kazi, JA Afzal, R Bashir, and DA Lightfoot (2007) The development of BAC-end sequence-based microsatellite markers and placement in the physical and genetic maps of soybean. Theor Appl Genet 114:1081-1090.

Song QJ, LF Marek, RC Shoemaker, KG Lark, VC Concibido, X Delannay, JE Specht, and PB Cregan (2004) A new integrated genetic linkage map of the soybean. Theor Appl Genet 109: 122-128.

Srour A, AJ Afzal, N Saini, L Blahut-Beatty, N Hemmati, DH Simmonds, H El Shemy, CD Town, H Sharma, X Liu, W Li, and DA Lightfoot (2011) The receptor like kinase transgene from the Rhg1/Rfs2 locus caused pleiotropic resistances to soybean cyst nematode and sudden death syndrome. BMC Genomics (in review).

Stephens PA, CD Nickell, and FL Kolb (1993) Genetic analysis of resistance to Fusarium solani in soybean. Crop Sci 33:929-930.

Triwitayakorn K, VN Njiti, MJ Iqbal, S Yaegashi, CD Town, and DA Lightfoot (2005) Genomic analysis of a region encompassing QRfs1 and QRfs2: genes that underlie soybean resistance to sudden death syndrome. Genome 48: 125-138.

Ullah H, Jasim M and DA Lightfoot (2012) Using a minimum tile path for plant transformations encompassing the entire soybean genome Journal of Plant Genome Sciences. In Press.

Ullah H, M Jasim, and DA Lightfoot (2012) Using a minimum tile path for plant transformations encompassing the entire soybean genome. Journal of Plant Genome Sciences 1 (2): xx–xx. doi: 10.5147/jpgs.2011.0066. In Press.

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Wrather JA, SR Kendig, SC Anand, TL Niblack, and GS Smith (1995) Effects of tillage, cultivar, and planting date on percentage of soybean leaves with symptoms of sudden death syndrome. Plant Dis 79:560-562.

Wrather JA, SR Koenning, and TR Anderson (2003) Effect of diseases on soybean yields in the United States and Ontario. 1999 to 2002. Plant Health Progr. online doi 10.1049.

Wrather JA, TR Anderson, DM Arsyad, J Gai, DL Ploper, A Portapuglia, HH Ram, JT Yorinori (1996) Soybean disease loss estimates for the top ten producing countries during. Plant Disease 79:107-110.

Yuan J, M Zhu, DA Lightfoot, MJ Iqbal, JY Yang, and K Meksem (2008) In silico comparison of transcript abundances during Arabidopsis thaliana and Glycine max resistance to Fusarium virguliforme. BMC Genomics. 2008 2:S6.

Yuan J, VN Njiti, K Meksem, MJ Iqbal, K Triwitayakorn, MA Kassem, GT Davis, ME Schmidt, and DA Lightfoot (2002) Quantitative trait loci in two soybean recombinant inbred line populations segregating for yield and disease resistance. Crop Sci 42: 271-277.

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