2004

Seed quality QTL in a prominent soybean population

D. L. Hyten Jr.
University of Tennessee - Knoxville, david.hyten@unl.edu

V. R. Pantalone
University of Tennessee - Knoxville, vpantalo@utk.edu

C. E. Sams
University of Tennessee - Knoxville, carlsams@tennessee.edu

A. M. Saxon
University of Tennessee - Knoxville

D. Landau-Ellis
University of Tennessee - Knoxville

See next page for additional authors

Published in *Theoretical and Applied Genetics* 109 (2004), pp. 552–561; doi: 10.1007/s00122-004-1661-5

Submitted December 12, 2003; accepted March 8, 2004; published online June 24, 2004.

Seed quality QTL in a prominent soybean population

D. L. Hyten,[1] V. R. Pantalone,[1] C. E. Sams,[1] A. M. Saxton,[2] D. Landau-Ellis,[1]

T. R. Stefaniak,[3] and M. E. Schmidt[4]

1. Department of Plant Sciences, The University of Tennessee, 252 Ellington Plant Science Building, 2431 Joe Johnson Drive, Knoxville, TN, 37996, USA
2. Department of Animal Science, The University of Tennessee, Knoxville, TN, 37996, USA
3. Department of Agronomy, University of Kentucky, Lexington, KY, 40546, USA
4. Department of Plant, Soil, and General Agriculture, Southern Illinois University, Carbondale, IL, 62901, USA

Corresponding author – D. L. Hyten, email hytend@ba.ars.usda.gov. Present address: Soybean Genomics and Improvement Laboratory, USDA-ARSPSI, Building 006, 10300 Baltimore Avenue, Beltsville, MD, 20705, USA

Abstract
Soybean [*Glycine max* (L.) Merr.] is a versatile crop due to its multitude of uses as a high protein meal and vegetable oil. Soybean seed traits such as seed protein and oil concentration and seed size are important quantitative traits. The objective of this study was to identify representative protein, oil, and seed size quantitative trait loci (QTL) in soybean. A recombinant inbred line (RIL) population consisting of 131 F_6-derived lines was created from two prominent ancestors of North American soybeans ('Essex' and 'Williams') and the RILs were grown in six environments. One hundred simple sequence repeat (SSR) markers spaced throughout the genome were mapped in this population. There were a total of four protein, six oil, and seven seed size QTL found in this population. The QTL found in this study may assist breeders in marker-assisted selection (MAS) to retain current positive QTL in modern soybeans while simultaneously pyramiding additional QTL from new germplasm.

Introduction

Soybean is grown worldwide, with the United States, Brazil, China, Argentina, and India being the major producers (Liu 1997). The major use of soybean is for soybean meal, a

primary component that provides protein in livestock feed. An important by-product of meal production is soybean oil, extracted from the crushed meal. Soybean oil is a vegetable oil used worldwide in cooking, frying fats and in spreadable margarines. Seed size is important in soybean products used for direct human consumption such as tofu, miso, and texturized soy vegetable protein.

An increase in seed protein concentration in commonly grown cultivars is needed to keep soybean competitive as a livestock feed. The typical negative correlation between protein and oil has caused difficulties in increasing the protein level while keeping the seed oil concentration at current levels. Protein, oil, and seed size are quantitative traits controlled by multiple genes having small or large effects. There have been many studies that have used molecular markers to map quantitative trait loci (QTL), the regions of the chromosome responsible for quantitative traits such as protein, oil, and seed size (Diers et al. 1992; Sebolt et al. 2000; Csanadi et al. 2001). Previous studies have utilized QTL mapping populations with large parental differences for the trait under study. Other QTL mapping populations have used soybean plant introductions or exotic germplasm, such as *Glycine soja* (Sieb. and Zucc.) as one of the parents in order to find new QTL not present in common cultivars, which can then be used to enhance current lines for the trait (Sebolt et al. 2000; Diers et al. 1992).

Currently, SoyBase (1995) contains 180 QTL for seed oil and protein concentration and seed size that have been mapped in many different populations and environments. Breeders and other researchers wishing to explore the utilization of these QTL may be hindered by the fact that some QTL have been detected in very few populations, too few environments and/or through only one statistical method. Information as to which QTL in SoyBase (1995) are verified, which QTL have significant QTL × environmental interactions, and which positive QTL alleles are already present in current breeding populations could be helpful for planning future research.

The two ancestral cultivars 'Essex' (Smith and Camper 1973) and 'Williams' (Bernard and Lindahl 1972) have contributed to the genetic background of many northern and southern USA cultivars and elite breeding lines (Sneller 1994; Gizlice et al. 1996). The diversity created by the Essex × Williams cross is known for producing the very popular cultivar Asgrow 'A3127' which served as a genetic bridge between the northern USA germplasm pool and the southern USA germplasm pool (Sneller 1994). Williams and A3127 have been the most commonly used parents in northern germplasm in the history of US soybean breeding (Sneller 1994). The objective of this study is to discover QTL segregating between Essex and Williams in six different environments and to determine which allele was inherited by A3127. This should identify genomic regions that have been detected for similar QTL in different populations in different environments. In addition, discovery of QTL in this population and verification of the allele inherited by A3127 should determine which QTL alleles are present in current breeding germplasm.

Materials and methods

One hundred and thirty-one F_6 recombinant inbred lines (RIL) were created at the University of Tennessee Knoxville Experimental Station (KES), Knoxville, Tenn., USA, from the

cross between the soybean cultivars Essex and Williams. The $F_{6,8}$ RIL were planted in a randomized complete block design with three replications in each of three environments [KES-C field and KES-S field, and the West Tennessee Experiment Station (WTES) at Jackson, Tenn., USA] during the year 2000. Two 6.1-m rows per plot were planted at KES-S and WTES. One 6.1-m row per plot was planted at KES-C. In 2001, the 131 RIL as $F_{6,9}$ lines were divided into three maturity groups (early, medium, and late) based on their maturity observed at KES-C in the previous year. Overall there were 49 early, 37 medium, and 47 late, with 5 days separating the early and medium groups, 3 days separating the medium and late group, and two lines overlapping in the medium and late maturity groups. Within each maturity group, a randomized complete block design with three replications each were planted at KES-V and Harrisburg, Ill., USA, and two replications were planted in four 6.1-m row plots at WTES, all during the year 2001. In each environment, the three maturity blocks were randomized within replications.

Seed protein and oil concentrations were determined at the USDA Northern Regional Research Center at Peoria, Ill., USA. Measurements were taken on a moisture-free basis using near-infrared reflectance spectroscopy on 21–25 g of whole beans from each plot. Seed size was determined by weighing 100 seeds from each plot.

DNA was isolated by taking a single leaf from five plants and combining them into one sample. The Qiagen Plant Easy DNA Extraction Kit (Qiagen, Hilden, Germany) was used to obtain purified DNA.

Essex and Williams were tested with 568 simple sequence repeat (SSR) markers previously placed on the integrated soybean genetic linkage map (Cregan et al. 1999). The sequence information for the soybean SSR primers and current integrated soybean genetic linkage map are publicly available from the USDA internet site http://bldg6.arsusda.gov/~pooley/soy/cregan/soy_map1. html. One hundred polymorphic markers were chosen to achieve a reasonable coverage of all 20 molecular linkage groups.

Polymerase chain reaction (PCR) reactions were performed in a 384-well MBS Hybaid thermocycler (Hybaid Franklin, Mass., USA). The 10 μl PCR mix consisted of 40 ng template DNA, 1× PCR Buffer, 0.2 mM dNTP mixture (Pharmacia, Piscataway, N.J., USA), 1.0 μM forward and reverse primer, and 0.5 units of Klentaq (Ab Peptides, St. Louis, Mo., USA). The amplification conditions consisted of 94°C for 5 min followed by 35 cycles of 94°C denaturation for 25 s, 48°C annealing for 30 s, and 72°C extension for 30 s, ending with one cycle of 72°C for 5 min. PCR products were separated using 6% non-denaturing polyacrylamide gel electrophoresis with ethidium bromide staining for visualization (Wang et al. 2003). The RIL were scored based on visualization of the DNA bands. Because we utilized an F_6-derived RIL population, heterozygotes were dropped from the data set to allow only additive effects to be estimated.

One hundred and thirty-one lines were mapped with 100 SSR markers and three phenotypic markers (*W1*, *T*, and *Dt1*). MAPMAKER/EXP (Lander et al. 1987; Lincoln et al. 1992) was used to estimate genetic distances between SSR markers. A minimum likelihood of odds (LOD) ≥3.0 and a maximum distance ≤50 centimorgan (cM) were used to test linkages among markers.

QTL analysis was performed using the phenotypic trait values and SSR RIL scores on 131 RIL planted in the field environments. The soybean composite integrated map was

used as the genetic map because the map distances are based on five populations (Song et al. 2004) and should have less variability associated with the map distances. Simple linear regression and composite interval mapping (CIM) were performed using QTL Cartographer (Basten et al. 1994, 2001). CIM was used to identify and map significant QTL where two or more markers where linked and simple linear regression was utilized for QTL detection when a marker was not linked to another marker. One thousand permutation tests were performed on each trait to establish empirical LOD thresholds at the 5% alpha level (Churchill and Doerge 1994). Broad-sense heritability estimates were computed as

$$h^2 = \sigma^2_g/[\sigma^2_g + (\sigma^2_{ge}/e) = (\sigma^2/re)]$$

where h^2 represents heritability, σ^2_g the genotypic variance, σ^2_{ge} the genotype × environment variance, σ^2 the error variance, r the number of replications, and e the number of environments (Nyquist 1991).

Results

A total of 100 SSR markers plus three phenotypic markers were mapped in the Essex × Williams RIL population. The resulting genetic linkage map covered 869.4 cM, which is about 34% of the currently known recombination distance of 2,524 cM, averaged from the integrated soybean genetic linkage map (Song et al. 2004). The Essex × Williams genetic map did not show any major discrepancies with the integrated soybean genetic linkage map. SSR markers on linkage groups A2, B1, and B2 were not polymorphic except for a few unlinked SSR markers, so CIM was not performed on these linkage groups.

The average seed oil concentration for Essex and Williams averaged over six environments only differed by 9.5 g/kg while the range of the F_6 RIL population is 25.4 g/kg with the mean of the population falling between the two parents (table 1). The population would have needed to produce individuals with <189.4 g/kg or >219.3 g/kg oil to display significant transgressive segregation ($P \leq 0.05$). A negative correlation between seed protein concentration and seed oil concentration was significant in the RIL population ($r = -0.60$, $P < 0.0001$).

Table 1. Mean, standard deviation, range, parental means, and heritability for soybean seed traits in an Essex × Williams F_6 RIL population grown in six environments

Seed traits	Mean	Standard deviation	Range	Essex	Williams	h^2
Seed size (mg/seed)	131.0	15.0	99–166	120.0	154.0	0.95
Oil concentration (g/kg)	203.5	5.1	189.8–215.2	199.6	209.1	0.84
Protein concentration (g/kg)	422.7	9.7	399.7–450.9	429.5	424.7	0.91

There are a total of six seed oil concentration QTL that were discovered through composite interval mapping in the RIL population (table 2). The oil QTL are located on linkage groups C2, D1a, D2, L, and M. Only two of the oil QTL (Oil-5 and Oil-6) were significant when the trait values from the six environments were averaged and CIM used for QTL

analysis. Oil-2, Oil-3, and Oil-4 gave significant LOD scores in one or two environments while Oil-1 was significant in five environments. Oil-1 was the only QTL in this study with a significantly positive effect in one environment and a negative effect in other environments.

Table 2. Position and effects of QTL found significant at the 5% alpha level associated with soybean seed oil concentration (*Oil*), seed protein concentration (*Pro*), and seed size (*SS*) in an Essex × Williams F_6 RIL population, based on QTL Cartographer analysis. The linkage group of each QTL is given based on the integrated soybean genetic linkage map.

E Essex allele, *W* Williams allele, *H* heterozygous. The QTL position is given as the estimated position on the integrated soybean genetic map. *KES-C/00* Knoxville Experimental Station (C1-field) in 2000, *WTES/00* West Tennessee Experiment Station in 2000, *KES-S/00* Knoxville Experiment Station (S-field) in 2000, *KES-V/01* Knoxville Experiment Station (V-field) in 2001, *Harrisburg/01* Harrisburg in 2001, *WTES/01* West Tennessee Experiment Station in 2001. Combined CIM was performed on average trait value from six environments. The effect is given as the average change (g/kg) resulting from substituting one allele of Williams with one allele of Essex.

QTL	LG	SSR interval (allele A3127 inherited)	QTL position (cM)	95% Confidence interval of QTL position	Environment	r^2	LOD	Effect
Oil-1	C2	Satt277(H)-Satt460(W)	112.2	107.6–113.9	WTES/00	8.8	3.7	−2.6
			109.6	<107.6–112.2	KES-S/00	31.6	12.0	4.9
			117.8	107.6–121.3	KES-C/00	20.0	7.1	3.7
			112.2	107.6–113.9	Harrisburg/01	8.7	2.9	−1.7
			112.2	107.6–113.9	WTES/01	13.3	5.1	−2.7
Oil-2	D1a	Satt184(W)-Satt179(E)	26.0	21.5–46.0	WTES/00	8.1	3.3	−2.4
Oil-3	D2	Satt458(E)-Satt154(E)	24.5	<24.5–34.5	WTES/00	7.8	3.0	−2.3
Oil-4	L	Satt166(W)-Dt1(W)	66.5	66.2–72.5	KES-S/00	8.0	3.4	−2.3
			66.5	66.2–76.5	KES-V/01	9.9	3.3	−2
Oil-5	L	Satt229(W)-Satt373(W)	93.9	91.1–101.9	Combined	8.3	3.3	−1.5
			93.9	91.1–101.9	KES-S/00	6.0	2.9	−2.1
			93.9	91.1–101.9	KES-C/00	9.6	3.8	−2.4
Oil-6	M	Satt540(E)-Satt463(E)	39.9	<35.9–49.9	Combined	11.6	3.6	−1.7
			35.9	<35.9–43.9	WTES/00	7.4	3.2	−2.3
			41.9	<35.9–49.9	KES-V/01	10.7	2.9	−2.1
			37.9	<35.9–45.9	WTES/01	16.9	5.6	−2.7
Pro-1	C2	Satt277(H)-Satt202(W)	119.8	117.8–121.3	Combined	27.6	9.8	−5.3
			121.3	119.8–125.3	WTES/00	14.5	5.7	−4.5
			111.6	107.6–121.3	KES-S/00	27.6	10.1	−7.2
			119.8	116.0–121.3	KES-C/00	33.5	10.6	−8.5
			109.6	107.6–116.0	KES-V/01	20.8	8.4	−5.3

			116.0	112.2–121.3	Harrisburg/01	20.0	8.1	−4.9
Pro-2	F	Satt335(W)-Satt144(E)	89.7	79.7–99.7	Combined	18.1	4.4	4.1
			93.7	83.7–112.1	WTES/00	17.3	3.5	4.7
			87.7	79.7–97.7	KES-S/00	20.1	5.0	5.7
			85.7	71.7–36.3	Harrisburg/01	23.3	4.6	4.9
Pro-3	K	Satt539(W)-Satt102(W)	15.8	5.8–25.8	Combined	24.4	4.3	5.4
			21.8	7.8–36.3	KES-V/01	13.4	2.7	3.7
Pro-4	M	Satt540(E)-Satt463(E)	41.9	<35.9–50.1	WTES/01	13.3	3.0	3.7
SS-1	C2	Satt277(H)-Satt460(W)	114.0	107.6–119.8	Combined	7.7	4.8	−4.5
			112.2	<107.6–121.3	WTES/00	6.2	2.9	−3.8
			112.2	109.6–117.8	KES-V/01	13.8	6.7	−9.1
			112.2	107.6–114.0	Harrisburg/01	12.9	7.3	−7.4
SS-2	D1a	Satt179(E)-Satt071(E)	58.0	42.0–88.2	Combined	13.9	4.0	−5.5
			62.0	48.0–82.2	Harrisburg/01	14.9	5.2	−7.1
SS-3	F	Satt114(E)-Satt335(W)	69.7	51.2–77.7	Combined	9.8	4.7	4.6
			67.7	51.2–71.7	KES-S/00	12.6	5.5	5.5
			63.7	53.2–69.7	KES-C/00	9.0	5.1	4.3
			67.7	49.2–75.7	Harrisburg/01	10.3	5.0	5.8
SS-4	G	Satt394(E)-Satt340(E)	43.4	33.1–47.4	Combined	4.4	2.9	−3.2
			43.4	33.1–48.6	KES-S/00	4.8	2.9	−3.5
			47.4	39.1–56.5	KES-C/00	5.3	2.7	−3.3
SS-5	I	Satt292(W)-Satt148(E)	82.8	74.1–90.8	KES-V/01	6.4	3.0	5.6
SS-6	K	Satt518(E)-Satt273(E)	46.6	40.9–52.6	KES-S/00	5.0	3.3	−3.6
SS-7	L	Satt156(E)-Dt1(W)	66.5	62.2–70.5	Combined	28.2	14.5	−8.2
			68.5	66.5–72.5	WTES/00	26.3	9.0	−7.2
			66.2	62.2–68.5	KES-S/00	42.4	18.4	−10.3
			68.5	60.2–72.5	KES-C/00	31.5	12.9	−8.1
			68.5	62.2–72.6	KES-V/01	43.0	12.0	−14.5
			68.5	62.2–74.5	Harrisburg/01	23.7	9.1	−9.4
			64.2	60.2–72.5	WTES/01	26.1	8.0	−6.4

Currently there are 53 seed oil concentration QTL reported in SoyBase (1995). Forty of the 53 oil QTL were scanned by markers in this study which were within 20 cM of the reported position on the QTL reported in SoyBase (1995) (table 3). Many of the oil QTL in SoyBase (1995) were discovered through simple linear regression methods and have not

been detected through interval mapping. This study reports only significant QTL found through CIM, but to create a complete comparison with previously reported QTL, the markers not significantly associated with an interval via CIM but found to be significantly associated with the trait through simple linear regression are included in tables 3, 4, and 5.

Table 3. QTL locations, effects, significance, and mapping parents associated with soybean seed oil concentration in this study and in SoyBase. The linkage group assignation of each QTL is based on the integrated soybean genetic linkage map.

QTL	LG	Marker	Map position	r^2	LOD	*P*-value	Parent 1	Parent 2	References
Oil 3-3	A1	A329_2	30.3	5	—	0.001	Minsoy	Noir 1	Mansur et al. (1996)
Un-assigned	A1	Satt591	31.1	—	—	0.006	Essex	Williams	This study
Oil 4-1	A1	K400_1	53.4	14	—	0.02	A87-296011	CX1039-99	Brummer et al. (1997)
Oil 4-2	A1	A975_1	75.4	11	—	0.009	C1763	CX1159-49-1	Brummer et al. (1997)
Oil 8-1	A1	Satt174	88.6	10	4	—	Minsoy	Archer	Orf et al. (1999)
Oil 4-3	A1	A104_1	92.3	19	—	0.003	M82-806	HHP	Brummer et al. (1997)
Oil 3-2	A1	T155_1	93.6	7	—	0.001	Minsoy	Noir 1	Mansur et al. (1996)
Oil 10-1	A1	T155_1	93.6	13	3	—	Minsoy	Noir 1	Orf et al. (1999)
Oil 13-1	A1	B170_1	94.9	4	5	—	Minsoy	Noir 1	Specht et al. (2001)
Oil 1-1[a]	A2	T153_1, A111_1	50.4	36	6	—	Minsoy	Noir 1	Mansur et al. (1993)
Oil 4-4[a]	A2	A505_1	132.3	9	—	0.03	C1763	CX1039-99	Brummer et al. (1997)
Oil 4-5[a]	B1	A109_1	29.2	31	—	0.0001	C1763	CX1159-49-1	Brummer et al. (1997)
Oil 2-6[a]	B2	A242_1	33.1	39	—	0.0001	A81356022	PI468916	Diers et al. (1992)
Oil 14-1[a]	B2	Satt020	72.1	3	3	—	Ma. Belle	Proto	Csanadi et al. (2001)
Oil 8-2[a]	C1	SOYGPATR	10.3	11	3	—	Minsoy	Archer	Orf et al. (1999)
Oil 9-1[a]	C1	SOYGPATR	10.3	7	3	—	Noir 1	Archer	Orf et al. (1999)
Oil 6-1	C1	A063_1	90.7	13	—	0.05	PI97100	Coker237	Lee et al. (1996)
Oil 9-2[a]	C2	Satt432	38.0	11	3	—	Noir 1	Archer	Orf et al. (1999)
Oil 4-6	C2	L148_1	97.2	9	—	0.03	A87-296011	CX1039-99	Brummer et al. (1997)
Un-assigned	C2	Satt557	112.2	16b	6b	—	Essex	Williams	This study
Un-assigned	D1a	Satt184	26.0	8	3	—	Essex	Williams	This study
Oil 13-2	D1a	Satt468	69.9	9	4	—	Minsoy	Noir 1	Specht et al. (2001)

Un-assigned	D1a	Satt147	108.0	—	—	0.004	Essex	Williams	This study
Un-assigned	D2	Satt458	24.5	8	3	—	Essex	Williams	This study
Oil 5-5	D2	K258_2	73.5	9	—	0.05	Young	PI416937	Lee et al. (1996)
Unas-signed	D2	Satt082	87.0	—	—	0.034	Essex	Williams	This study
Oil 5-4	D2	CR142_1	—	13	—	0.05	Young	PI416937	Lee et al. (1996)
Oil 5-6	D2	CR326_1	—	9	—	0.05	Young	PI416937	Lee et al. (1996)
Oil 2-5[a]	E	SAC7_1	6.3	43	—	0.0001	A81356022	PI468916	Diers et al. (1992)
Oil 2-3[a]	E	Pb	13.6	27	—	0.0001	A81356022	PI468916	Diers et al. (1992)
Oil 2-8	E	K229_1	28.3	22	—	0.001	A81356022	PI468916	Diers et al. (1992)
Oil 2-4	E	A454_1	30.9	23	—	0.0008	A81356022	PI468916	Diers et al. (1992)
Oil 2-9	E	A203_1	34.6	18	—	0.006	A81356022	PI468916	Diers et al. (1992)
Unas-signed	E	Satt268	44.0	—	—	0.009	Essex	Williams	This study
Oil 5-1	E	A069_2	—	7	—	0.05	Young	PI416937	Lee et al. (1996)
Oil 13-3	F	Satt510	71.4	6	3	—	Minsoy	Noir 1	Specht et al. (2001)
Unas-signed	F	Satt335	77.7	—	—	0.026	Essex	Williams	This study
Oil 4-7	G	A584_1	65.6	19	—	0.009	C1763	CX1039-99	Brummer et al. (1997)
Oil 4-8	G	A816_1	67.5	11	—	0.007	A87-296011	CX1039-99	Brummer et al. (1997)
Oil 4-9	G	A890_1	67.7	15	—	0.004	M81-382	PI423949	Brummer et al. (1997)
Oil 6-4	G	L002_2	97.7	14	—	0.05	PI97100	Coker237	Lee et al. (1996)
Oil 6-2	G	L154_1	99.3	17	—	0.05	PI97100	Coker237	Lee et al. (1996)
Oil 4-10	H	A069_1	33.2	18	—	0.0003	C1763	CX1159-49-1	Brummer et al. (1997)
Unas-signed	H	Satt192	44.0	—	—	0.033	Essex	Williams	This study
Oil 7-1[a]	H	B072_1	124.0	21	—	0.002	Peking	Essex	Qui et al. (1999)
Oil 6-5	H	A566_2	—	10	—	0.05	PI97100	Coker237	Lee et al. (1996)
Oil 14-3	I	Satt562	22.8	6	—	0.01	Ma. Belle	Proto	Csanadi et al. (2001)
Oil 11-1	I	A144_1	32.4	39	4	—	A81356022	PI468916	Sebolt et al. (2000)
Oil 12-1	I	A144_1	32.4	15	—	0.0008	Parker	PI468916	Sebolt et al. (2000)
Oil 13-4	I	BLT002_1	35.2	10	5	—	Minsoy	Noir 1	Specht et al. (2001)

Oil 2-1	I	K011_1	38.1	27	—	0.0002	A81356022	PI468916	Diers et al. (1992)
Oil 2-2	I	A407_1	39.4	28	—	0.0005	A81356022	PI468916	Diers et al. (1992)
Oil 13-5	I	L026_2	119.1	5	3	—	Minsoy	Noir 1	Specht et al. (2001)
Oil 5-2[a]	J	B122_1	54.8	7	—	0.05	Young	PI416937	Lee et al. (1996)
Oil 1-2	K	BC1, A315_1	28.7	24	3	—	Minsoy	Noir 1	Mansur et al. (1993)
Oil 4-11	K	K387_1	98.9	16	—	0.002	C1763	CX1039-99	Brummer et al. (1997)
Oil 14-2[a]	K	Satt196	104.8	7	—	0.01	Ma. Belle	Proto	Csanadi et al. (2001)
Un-assigned	L	Satt523	27.9	—	—	0.042	Essex	Williams	This study
Oil 2-7	L	A023_1	36.7	32	—	0.0001	A81356022	PI468916	Diers et al. (1992)
Oil 5-3	L	A023_1	36.7	7	—	0.05	Young	PI416937	Lee et al. (1996)
Un-assigned	L	Satt166	66.5	9[b]	3[b]	—	Essex	Williams	This study
Oil 3-1	L	Satt006	92.0	9	—	0.001	Minsoy	Noir 1	Mansur et al. (1996)
Un-assigned	L	Satt229	93.9	8	3	—	Essex	Williams	This study
Oil 9-3	L	A489_1	95.4	19	6	—	Noir 1	Archer	Orf et al. (1999)
Un-assigned	M	Satt540	39.9	12	4	—	Essex	Williams	This study
Oil 6-3	—	A235_4	—	15	—	0.05	PI97100	Coker237	Lee et al. (1996)

a. QTL that had no SSR markers tested within 20 cM in the Essex × Williams population.
b. Average of significant environments from table 2.

The Essex × Williams RIL population has several oil QTL detected close to previously identified oil QTL (table 3). Only linkage groups G, I, and K contained previously reported QTL that were not detected in any environment in this study. Linkage group A1 did have a QTL detected through simple linear regression at 31 cM, but there are several previously reported QTL found over 20 cM away throughout the chromosome. The oil QTL from this study on linkage group M has not been detected in any previous study.

The average seed protein concentration of Essex and Williams was very similar with Essex's average equal to 429.5 g/kg and Williams' average equal to 424.7 g/kg. The range of the F_6 RIL population was 399.7–450.9 g/kg with the mean of the population falling slightly below the two parents (table 1). The RIL population demonstrated significant negative and positive transgressive segregation for protein concentration.

There are a total of four seed protein concentration QTL that were discovered through CIM in the RIL population (table 2). Three of the protein QTL were significant in the combined environments while Pro-4 was only significant in one environment. The protein allele present on C2 in Williams conferring higher protein concentration is closely linked to

an early maturity allele in Williams presumed to be *E1* (data not shown). The other three protein QTL detected received the beneficial allele from the Essex parent.

Currently there are 61 seed protein concentration QTL reported in SoyBase (1995). Fifty of the 61 protein QTL were in regions of the genome that were within 20 cM of markers used in this study (table 4). Few of the QTL reported in SoyBase (1995) were found in close proximity to a QTL found in the current study. Only QTL previously reported in SoyBase (1995) on linkage groups C2, F, K, and M were significant with markers linked closely to significant QTL intervals found in the Essex × Williams population.

Table 4. QTL locations, effects, significance, and mapping parents associated with soybean seed protein concentration in this study and in SoyBase. The linkage group assignation of each QTL is based on the integrated soybean genetic linkage map.

QTL	LG	Marker	Map position	r^2	LOD	*P*-value	Parent 1	Parent 2	References
Prot 2-3	A1	A329_2	30.3	5	—	0.001	Minsoy	Noir 1	Mansur et al. (1996)
Prot 2-1	A1	T155_1	93.6	9	—	0.001	Minsoy	Noir 1	Mansur et al. (1996)
Prot 9-1	A1	T155_1	93.6	15	4	—	Minsoy	Noir 1	Orf et al. (1999)
Prot 12-1	A1	B170_1	94.9	5	5	—	Minsoy	Noir 1	Specht et al. (2001)
Prot 3-1[a]	A2	A505_1	132.3	11	—	0.01	C1763	CX1039-99	Brummer et al. (1997)
Prot 14-1[a]	A2	Ti	—	2	—	0.05	M91-212006	SZG9652	Vollmann et al. (2002)
Prot 3-2[a]	B1	A109_1	29.2	8	—	0.008	McCall	PI445815	Brummer et al. (1997)
Prot 4-11[a]	B2	A352_1	29.2	10	—	0.05	Young	PI416937	Lee et al. (1996)
Prot 1-6[a]	B2	A242_1	33.1	19	—	0.004	A81356022	PI468916	Diers et al. (1992)
Prot 4-10[a]	B2	B142_1	43.6	10	—	0.05	Young	PI416937	Lee et al. (1996)
Prot 9-2[a]	C1	SOYGPATR	10.3	12	4	—	Minsoy	Noir 1	Orf et al. (1999)
Prot 4-4[a]	C1	A463_1	21.0	7	—	0.05	Young	PI416937	Lee et al. (1996)
Prot 12-2[a]	C1	K001_1	33.3	7	4	—	Minsoy	Noir 1	Specht et al. (2001)
Prot 7-2	C1	Satt578	65.1	12	6	—	Minsoy	Archer	Orf et al. (1999)
Prot 3-3	C1	A063_1	90.7	12	—	0.007	M84-492	Sturdy	Brummer et al. (1997)
Prot 4-3	C1	A338_2	—	10	—	0.05	Young	PI416937	Lee et al. (1996)
Prot 4-2	C1	EV3_1	—	11	—	0.05	Young	PI416937	Lee et al. (1996)
Prot 4-1	C1	GC197_1	—	13	—	0.05	Young	PI416937	Lee et al. (1996)
Un-assigned	C2	Satt460	119.8	28	10	—	Essex	Williams	This study

Prot 13-2	C2	Sct_028	122.0	7	3	—	Ma. Belle	Proto	Csanadi et al. (2001)
Prot 3-4	D1a	A398_1	6.4	28	—	0.009	LN83-2356	PI360843	Brummer et al. (1997)
Prot 3-5	D1a	A691_1	40.8	10	—	0.02	C1763	CX1039-99	Brummer et al. (1997)
Prot 13-1	D1a	Satt077	77.5	5	—	0.01	Ma. Belle	Proto	Csanadi et al. (2001)
Un-assigned	D1b	Satt459	118.6	—	—	0.021	Essex	Williams	This study
Prot 1-5[a]	E	SAC7_1	6.3	24	—	0.003	A81356022	PI468916	Diers et al. (1992)
Prot 3-6	E	B174_1	30.9	11	—	0.02	McCall	PI445815	Brummer et al. (1997)
Prot 5-1	E	A454_1	30.9	9	—	0.05	PI97100	Coker237	Lee et al. (1996)
Prot 4-5	E	C	—	7	—	0.05	Young	PI416937	Lee et al. (1996)
Prot 4-6	E	CR167_1	—	7	—	0.05	Young	PI416937	Lee et al. (1996)
Prot 4-13	E	CR274_1	—	8	—	0.05	Young	PI416937	Lee et al. (1996)
Prot 3-7	F	K002_1	47.6	9	—	0.03	A87-296011	CX1039-99	Brummer et al. (1997)
Un-assigned	F	Satt335	89.7	18	4	—	Essex	Williams	This study
Prot 6-2	F	B148_1	105.8	17	—	0.0001	Peking	Essex	Qui et al. (1999)
Prot 3-8	G	A816_1	67.5	12	—	0.005	A87-296011	CX1039-99	Brummer et al. (1997)
Prot 3-9	G	A890_1	67.7	16	—	0.003	C1763	CX1159-49-1	Brummer et al. (1997)
Prot 1-8	G	A245_2	90.0	12	—	0.01	A81356022	PI468916	Diers et al. (1992)
Prot 3-10	G	A235_1	97.2	8	—	0.04	C1763	CX1039-99	Brummer et al. (1997)
Prot 3-11	H	A069_1	33.2	7	—	0.05	C1763	CX1159-49-1	Brummer et al. (1997)
Prot 6-1[a]	H	B072_1	124.0	32	—	0.002	Peking	Essex	Qui et al. (1999)
Prot 5-2	H	A566_2	—	14	—	0.05	PI97100	Coker237	Lee et al. (1996)
Prot 1-4	I	A688_1	32.4	25	—	0.001	A81356022	PI468916	Diers et al. (1992)
Prot 1-3	I	A144_1	32.4	24	—	0.0007	A81356022	PI468916	Diers et al. (1992)
Prot 3-12	I	A144_1	32.4	28	—	0.0002	M82-806	HHP	Brummer et al. (1997)
Prot 11-1	I	A144_1	32.4	44	—	0.0001	Parker	PI468916	Sebolt et al. (2000)
Prot 10-1	I	Satt127	35.3	65	9	—	A81356022	PI468916	Sebolt et al. (2000)
Prot 1-1	I	K011_1	38.1	42	—	0.0001	A81356022	PI468916	Diers et al. (1992)
Prot 1-2	I	A407_1	39.4	39	—	0.0001	A81356022	PI468916	Diers et al. (1992)

Prot 4-7	J	B166_1	27.7	8	—	0.05	Young	PI416937	Lee et al. (1996)
Un-assigned	K	Satt539	15.8	24	4	—	Essex	Williams	This study
Prot 5-4	K	R051_2	31.8	10	—	0.05	PI97100	Coker237	Lee et al. (1996)
Prot 12-3	K	Satt178	40.9	3	3	—	Minsoy	Noir 1	Specht et al. (2001)
Unas-signed	K	Satt260	80.1	—	—	0.002	Essex	Williams	This study
Prot 13-4	K	Satt196	104.8	5	—	0.01	Ma. Belle	Proto	Csanadi et al. (2001)
Prot 5-3	K	A065_3	—	11	—	0.05	PI97100	Coker237	Lee et al. (1996)
Prot 5-5	K	Q043_1	—	10	—	0.05	PI97100	Coker237	Lee et al. (1996)
Prot 1-7	L	A023_1	36.7	16	—	0.01	A81356022	PI468916	Diers et al. (1992)
Prot 8-1	L	Satt166	66.5	11	3	—	Noir 1	Archer	Orf et al. (1999)
Prot 2-2	L	Satt006	92.0	8	—	0.001	Minsoy	Noir 1	Mansur et al. (1996)
Prot 12-4	M	Satt567	33.5	27	13	—	Minsoy	Noir 1	Specht et al. (2001)
Prot 13-3	M	Satt567	33.5	7	—	0.01	Ma. Belle	Proto	Csanadi et al. (2001)
Prot 7-1	M	R079_1	39.0	6	3	—	Minsoy	Archer	Orf et al. (1999)
Unas-signed	M	Satt540	41.9	13	3	—	Essex	Williams	This study
Prot 4-8	N	A071_2	30.3	11	—	0.05	Young	PI416937	Lee et al. (1996)
Prot 4-9	N	GC34_2	—	8	—	0.05	Young	PI416937	Lee et al. (1996)
Prot 12-5	O	Satt478	71.1	6	3	—	Minsoy	Noir	1
Prot 4-12	—	Ga1_25	—	14	—	0.05	Young	PI416937	Lee et al. (1996)
Prot 5-6	—	A132_4	—	13	—	0.05	PI97100	Coker237	Lee et al. (1996)

a. QTL that had no SSR markers tested within 20 cM in the Essex × Williams population

The average seed size over six environments was 120 mg/seed for Essex and 154 mg/seed for Williams (table 1). The seed sizes of the F_6 RILs ranged from 99 to 166 mg/seed but significant transgressive segregation was not detected ($P \leq 0.05$). The mean of the RIL population fell between the two parents and the heritability of the trait was very high. There were a total of five seed size QTL (SS-1, SS-2, SS-3, SS-4, and SS-7) that were significant across the six environments (table 2). Seed size QTL SS-5 and SS-6 were significant in only one environment. With the exception of SS-3 and SS-5, the alleles contributing to larger seed came from Williams, the parent with the large seed size. A major QTL for seed size was found on linkage group L which was in close proximity to the determinancy gene (*Dt1*). Forty-seven of the 66 seed size QTL reported in SoyBase (1995) were within 20 cM of a marker used in the analysis of the Essex × Williams RIL population. Only linkage

groups A1, C1, E, J, M, and O contained reported QTL that were not in close proximity to QTL found in this study (table 5). All of the QTL found in this study through CIM were in close proximity to QTL reported in other populations except for QTL SS-5 on linkage group I. This QTL is 38.8 cM from the nearest marker reported in SoyBase (1995) to be significantly associated with seed size.

Table 5. QTL locations, effects, significance, and mapping parents associated with soybean seed size in this study and in SoyBase. The linkage group assignation of each QTL is based on the integrated soybean genetic linkage map.

QTL	LG	Marker	Map position	r^2	LOD	*P*-value	Parent 1	Parent 2	References
Sd wt 6-4	A1	Satt449	27.8	6	3	—	Noir 1	Archer	Orf et al. (1999)
Sd wt 7-3	A1	Satt174	88.6	7	3	—	Minsoy	Noir 1	Orf et al. (1999)
Sd wt 10-1	A1	Satt200	92.9	3	3	—	Minsoy	Noir 1	Specht et al. (2001)
Sd wt 1-3	A1	T155_1	93.6	6	—	0.001	Minsoy	Noir 1	Mansur et al. (1996)
Sd wt 4-5[a]	A2	T153_1	50.4	7	—	0.007	V71-370	PI407162	Maughan et al. (1996)
Sd wt 6-2[a]	A2	Satt187	54.9	7	3	—	Noir 1	Archer	Orf et al. (1999)
Sd wt 7-1[a]	A2	Satt508	108.8	8	4	—	Minsoy	Noir 1	Orf et al. (1999)
Sd wt 10-2[a]	A2	Satt470	116.7	6	3	—	Minsoy	Noir 1	Specht et al. (2001)
Sd wt 1-1[a]	A2	K443_2	119.1	11	—	0.001	Minsoy	Noir 1	Mansur et al. (1996)
Sd wt 6-3[a]	B1	T028_1	5.1	8	4	—	Noir 1	Archer	Orf et al. (1999)
Sd wt 4-1[a]	B1	A118_1	58.9	14	—	0.001	V71-370	PI407162	Maughan et al. (1996)
Sd wt 11-1[a]	B1	A089_2	59.7	11	—	0.013	Pureunkong	Jinpumkong 2	Lee et al. (1996)
Sd wt 10-3[a]	B1	Sat_095	81.3	4	3	—	Minsoy	Noir 1	Specht et al. (2001)
Sd wt 10-4[a]	B2	BLT057_2	72.5	5	5	—	Minsoy	Noir 1	Specht et al. (2001)
Sd wt 3-8[a]	B2	Cr395_1	—	6	—	0.03	PI97100	Coker237	Mian et al. (1996)
Sd wt 2-1[a]	C1	A059_1	18.6	10	—	0.0001	Young	PI416937	Mian et al. (1996)
Sd wt 6-7[a]	C1	K001_1	33.3	9	5	—	Noir 1	Archer	Orf et al. (1999)
Sd wt 5-2	C1	L192_1	73.2	6	3	—	Minsoy	Archer	Orf et al. (1999)
Sd wt 7-5[a]	C2	L199_2	23.3	9	5	—	Minsoy	Noir 1	Orf et al. (1999)
Sd wt 10-5[a]	C2	L199_2	23.3	5	5	—	Minsoy	Noir 1	Specht et al. (2001)
Sd wt 1-2[a]	C2	A262_4	25.1	6	—	0.001	Minsoy	Noir 1	Mansur et al. (1996)

Sd wt 2-2[a]	C2	A635_1	95.6	6	—	0.01	Young	PI416937	Mian et al. (1996)
Sd wt 6-5[a]	C2	Satt277	107.6	7	3	—	Noir 1	Archer	Orf et al. (1999)
Unassigned	C2	Satt100	114.0	8	5	—	Essex	Williams	This study
Unassigned	D1a	Satt179	58.0	14	4	—	Essex	Williams	This study
Sd wt 7-4	D1a	Sat_036	75.3	6	3	—	Minsoy	Noir 1	Orf et al. (1999)
Unassigned	D1b	Satt459	118.6	—	—	0.01	Essex	Williams	This study
Sd wt 3-1	D2	A257_1	10.3	8	—	0.0087	PI97100	Coker237	Mian et al. (1996)
Unassigned	D2	Satt461	80.2	—	—	0.001	Essex	Williams	This study
Sd wt 2-3	E	BLT049_5	46.3	14	—	0.0001	Young	PI416937	Mian et al. (1996)
Sd wt 6-1	E	G214_26	47.6	6	3	—	Noir 1	Archer Orf et al. (1999)	Sd wt 6-1
Sd wt 11-2	E	A069_2	—	10	—	0.015	Pureunkong	Jinpumkong 2	Lee et al. (1996)
Sd wt 10-6	F	Satt343	3.0	3	3	—	Minsoy	Noir 1	Specht et al. (2001)
Sd wt 3-2	F	K002_1	47.6	7	—	0.0168	PI97100	Coker237	Mian et al. (1996)
Sd wt 10-7	F	K265_1	51.5	3	2.9	—	Minsoy	Noir 1	Specht et al. (2001)
Sd wt 2-4	F	BLT025_1	59.8	5	—	0.02	Young	PI416937	Mian et al. (1996)
Unassigned	F	Satt114	69.7	10	4.7	—	Essex	Williams	This study
Sd wt 6-6	F	L050_14	71.4	7	3	—	Noir 1	Archer	Orf et al. (1999)
Sd wt 11-3	F	CR321_2	—	13	—	0.005	Pureunkong	Jinpumkong 2	Lee et al. (1996)
Sd wt 7-2	G	Satt163	0.0	7	3	—	Minsoy	Noir 1	Orf et al. (1999)
Sd wt 10-8	G	Satt163	0.0	5	4	—	Minsoy	Noir 1	Specht et al. (2001)
Unassigned	G	Satt394	43.4	4	3	—	Essex	Williams	This study
Sd wt 4-2	G	A816_1	67.5	10	—	0.001	V71-370	PI407162	Maughan et al. (1996)
Sd wt 10-9	G	Satt517	69.9	4	3	—	Minsoy	Noir 1	Specht et al. (2001)
Sd wt 3-3	G	A235_1	97.2	10	—	0.004	PI97100	Coker237	Mian et al. (1996)
Sd wt 11-4	G	A235_1	97.2	8	—	0.035	Pureunkong	Jinpumkong 2	Lee et al. (1996)
Sd wt 2-5	G	B031_2	—	22	—	0.0001	Young	PI416937	Mian et al. (1996)
Sd wt 12-8	I	Satt562	22.8	12	—	0.01	Ma. Belle	Proto	Csanadi et al. (2001)
Sd wt 8-1	I	A144_1	32.4	29	3	—	A81356022	PI468916	Sebolt et al. (2000)
Sd wt 9-1	I	A515_1	44.0	4	—	0.04	Parker	PI468916	Sebolt et al. (2000)
Unassigned	I	Satt292	82.8	6	3	—	Essex	Williams	This study

Sd wt 2-6	J	B166_1	27.7	8	—	0.006	Young	PI416937	Mian et al. (1996)
Sd wt 4-4	J	K384_1	28.2	11	—	0.001	V71-370	PI407162	Maughan et al. (1996)
Sd wt 10-10	K	Satt102	30.3	3	3	—	Minsoy	Noir 1	Specht et al. (2001)
Unassigned	K	Satt518	46.6	5	3	—	Essex	Williams	This study
Sd wt 3-4	K	A199_1	67.7	5	—	0.0501	PI97100	Coker237	Mian et al. (1996)
Sd wt 2-7	K	K003_1	84.5	8	—	0.004	Young	PI416937	Mian et al. (1996)
Unassigned	L	Satt523	27.9	—	—	0.043	Essex	Williams	This study
Sd wt 12-5	L	Satt313	34.5	5	—	0.01	Ma. Belle	Proto	Csanadi et al. (2001)
Sd wt 4-3	L	A023_1	36.7	9	—	0.002	V71-370	PI407162	Maughan et al. (1996)
Unassigned	L	Satt166	66.5	28	15	—	Essex	Williams	This study
Sd wt 5-1	L	Satt527	70.4	6	3	—	Minsoy	Archer	Orf et al. (1999)
Sd wt 7-7	L	Sat_099	78.2	6	3	—	Minsoy	Noir 1	Orf et al. (1999)
Sd wt 3-5	L	Dt1	89.1	10	—	0.0028	PI97100	Coker237	Mian et al. (1996)
Sd wt 12-3	L	Satt229	93.9	5	—	0.01	Ma. Belle	Proto	Csanadi et al. (2001)
Sd wt 4-6	L	K385_1	101.3	11	—	0.001	V71-370	PI407162	Maughan et al. (1996)
Sd wt 3-6	L	EV2_1	—	7	—	0.0256	PI97100	Coker237	Mian et al. (1996)
Sd wt 10-11[a]	M	Satt590	7.8	10	7	—	Minsoy	Noir 1	Specht et al. (2001)
Sd wt 7-6	M	Satt150	18.6	7	3	—	Minsoy	Noir 1	Orf et al. (1999)
Sd wt 12-4	M	Satt306	80.0	7	—	0.01	Ma. Belle	Proto	Csanadi et al. (2001)
Sd wt 12-1	M	Satt210	112.1	5	—	0.01	Pureunkong	Jinpumkong 2	Lee et al. (1996)
Sd wt 3-7	M	CR529_1	—	11	—	0.015	PI97100	Coker237	Mian et al. (1996)
Sd wt 12-6[a]	O	Satt358	5.4	8	—	0.01	Ma. Belle	Proto	Csanadi et al. (2001)
Sd wt 12-7[a]	O	Satt477	82.1	6	3	—	Ma. Belle	Proto	Csanadi et al. (2001)
Sd wt 12-2	O	Satt219	—	12	4	—	Ma. Belle	Proto	Csanadi et al. (2001)
Sd wt 3-9	—	CR235_1	—	10	—	0.0159	PI97100	Coker237	Mian et al. (1996)

a. QTL that had no SSR markers tested within 20 cM in the Essex × Williams population

All the SSR markers were used to assay the allelic status of cultivar A3127 to determine which of the QTL detected in this study were inherited by A3127. Table 2 reports these results with the allele for the respective SSR marker present in A3127 in the SSR interval column. Many of the QTL for the three traits contained crossovers between the two markers where the QTL was detected, so the inherited allele could not be determined. Nine of

the QTL detected in this study did not contain crossovers between the two markers that flank the putative QTL so the inherited allele could be inferred in A3127. QTL inherited by A3127 that negatively impacted oil, protein, and seed size were Oil-3, Oil-6, Pro-3, SS-2, SS-4, and SS-6. Conversely Oil-4, Oil- 5, and Pro-4 are QTL presumed to be inherited by A3127 that increase oil and protein concentration.

Discussion

The genetic map created by the Essex × Williams population only covered approximately 34% of the genome but the genome coverage was sufficient to span regions that contain 76% of the protein, oil, and seed size QTL reported in SoyBase (1995). Many of the QTL not scanned by this study were located on linkage groups A2, B1, B2, and C1 where there was a lack of polymorphic markers. In addition, numerous gaps of >20 cM are present in the Essex × Williams map which could lead to many important QTL not being detected. As new SSR markers and single nucleotide polymorphism markers become available it may be possible to scan these four linkage groups and the gaps for previously reported and new QTL. Alternatively, new polymorphic markers may not be discovered on linkage groups A2, B1, B2, and C1 because Essex and Williams may be genetically identical in these regions.

Seed protein concentration was the only seed quality trait to demonstrate transgressive segregation. Transgressive segregation for seed protein and oil concentration was expected in the F_6 RIL population because of the diversity of the two parents for these traits. There was a trend for the parent with the greater trait value to contribute more QTL alleles with positive effects. The protein QTL on linkage group C2 was a major QTL at which the Williams allele increased protein concentration while Pro-2, Pro-3, and Pro-4 were QTL at which the Essex allele increased protein concentration. The three QTL alleles that increased protein concentration came from Essex which had a mean protein concentration 4.8 g/kg greater than the mean for Williams. In only one case did the Williams QTL allele increase protein concentration.

Oil and seed size values from the RIL population numerically exceeded parental values but did not demonstrate significant transgressive segregation. Five of the six QTL alleles associated with increased oil concentration were inherited from Williams which has 9.5 g/kg higher seed oil concentration than Essex. The oil QTL Oil-1 demonstrated a large Oil × location interaction. At the Knoxville locations (which are in the eastern part of Tennessee) the Oil-1 Essex allele gave a positive additive effect whereas in Southern Illinois and western Tennessee the same allele was associated with significantly decreased oil concentration. Seed oil concentration had the lowest heritability of the three traits studied which most certainly was reduced by the Oil-1 QTL × location interaction. In the case of seed size the parent with the larger seed size, Williams, contributed five of the seven positive QTL alleles for seed size.

Many QTL that have been previously discovered and reported in SoyBase (1995) were in close proximity to QTL found in this study. As anticipated, a number of QTL catalogued in SoyBase (1995) were not detected in our study. We detected a number of QTL via simple linear regression that appeared to coincide with identical QTL catalogued in SoyBase

(1995) (Oil 3-3, Oil 5-5, Oil 2-9, Prot 7-1, Sd wt 3-1, Sd wt 12-5, etc.). However, further analysis with CIM indicated that those QTL did not reach the threshold for significance. A knowledge of the statistical rigor with which the QTL are detected may assist researchers in setting priorities for which QTL they want to study or manipulate. In addition, several QTL reported in SoyBase (1995) were not detected by our analysis methods in the current study. These QTL would seem to be fixed or not present in the Essex × Williams population. To determine if the fixed allele is the QTL allele with positive or negative effect additional crosses would need to be made with diverse parents. Once the QTL are mapped in these populations the fixed allele contribution from Essex and Williams to modern cultivars can be determined. With the knowledge of the QTL segregating in the Essex × Williams population along with the contribution of fixed QTL, researchers and breeders should have a more complete picture of which QTL are present in current breeding populations and which of these QTL from previous studies need to be introgressed.

The inheritance of alleles in A3127 also helps to complete the picture of which QTL are currently present in modern breeding populations. It is well known that A3127 was selected and released as a cultivar because of its outstanding yield potential. Selection for protein, oil, and seed size would have been incidental and related to increased seed yield. This leads to the suggestion that the QTL inherited by A3127 that increased protein, oil, and seed size such as Oil-4, Oil-5, and Pro-4 may not have a negative affect on yield. The favorable alleles for these three QTL are probably present in modern cultivars. It may be useful to fine map these QTL using alternative methods such as near isogenic lines, association analysis, or positional cloning so that the gene(s) contributing to these QTL may be discovered. These genes may lend insight on how to increase seed quality traits while maintaining yield. It was interesting that half of the QTL detected for the seed quality traits in this study contained crossovers in A3127 between the two markers that flanked the QTL. A better definition of the exact position of these QTL is needed in order to determine if these QTL are present in modern breeding populations.

Acknowledgments – The authors would like to thank James H. Klein III (Southern Illinois University at Carbondale, Ill., USA), Donna Thomas (USDA Northern Regional Research Center at Peoria, Ill., USA), and Gordon G. Percell (WTES) for their technical support and Dr. Perry B. Cregan (USDA-ARS, Beltsville, Md., USA) for his suggestions. This research was supported by the United Soybean Board project no. 1240 and by the Tennessee Soybean Promotion Board.

References

Basten CJ, Weir BS, Zeng Z-B (1994) Zmap-a QTL cartographer. In: Smith C, Gavora JS, Benkel B, Chesnais J, Fairfull W, Gibson JP, Kennedy BW, Burnside EB (eds) Proceedings of the 5th world congress on genetics applied to livestock production: computing strategies and software, vol 22. Guelph, Ontario, pp 65–66

Basten CJ, Weir BS, Zeng Z-B (2001) QTL Cartographer version 1.15. Department of Statistics, North Carolina State University, Raleigh

Bernard RL, Lindahl DA (1972) Registration of 'Williams' soybean. Crop Sci 12:716

Brummer EC, Graef GL, Orf J, Wilcox JR, Shoemaker RC (1997) Mapping QTL for seed protein and oil content in eight soybean populations. Crop Sci 37:370–378

Churchill G, Doerge R (1994) Empirical threshold values for quantitative trait mapping. Genetics 138:963–971

Cregan PB, Jarvik T, Bush AL, Shoemaker RC, Lark KG, Kahler AL, Kaya N, Van Toai TT, Lohnaes DG, Chung T, Specht JE (1999) An integrated genetic linkage map of the soybean genome. Crop Sci 39:1464–1490

Csanadi G, Vollmann J, Stift G, Lelley T (2001) Seed quality QTLs identified in a molecular map of early maturing soybean. Theor Appl Genet 103:912–919

Diers BW, Keim P, Fehr WR, Shoemaker RC (1992) RFLP analysis of soybean seed protein and oil content. Theor Appl Genet 83:608–612

Gizlice Z, Carter TE Jr, Gerig TM, Burton JW (1996) Genetic diversity patterns in North American public soybean cultivars based on coefficient of parentage. Crop Sci 36:753–765

Lander ES, Green P, Abrahamson J, Barlow A, Daly MJ, Lincoln SE, Newburg L (1987) MAPMAKER: an interactive computer package for constructing primary genetic linkage map of experimental and natural populations. Genomics 1:174–181

Lee SH, Bailey MA, Mian MAR, Carter TE, Shipe ER, Ashley DA, Parrott WA, Hussey RS, Boerma HR (1996) RFLP loci associated with soybean seed protein and oil content across populations and locations. Theor Appl Genet 93:649–657

Lincoln SE, Daly MJ, Lander E (1992) Constructing genetic maps with MAPMAKER/EXP 3.0, 3rd edn. Whitehead Institute Technical Report

Liu K (1997) Soybeans: chemistry, technology and utilization. Chapman & Hall, New York, p 279

Mansur LM, Orf JH, Lark KG (1993) Determining the linkage of qualitative trait loci to RFLP markers using extreme phenotypes of recombinant inbreds of soybean (Glycine max L. Merr.) Theor Appl Genet 86:914–918

Mansur LM, Orf JH, Chase K, Jarvik T, Cregan PB, Lark KG (1996) Genetic mapping of agronomic traits using recombinant inbred lines of soybean. Crop Sci 36:1327–1336

Maughan PJ, Saghai-Maroof MA, Buss GR (1996) Molecular-marker analysis of seed-weight: genomic locations, gene action, and evidence for orthologous evolution among three legume species. Theor Appl Genet 93:574–579

Mian AR, Bailey MA, Tamulonis JP, Shipe ER, Carter TE Jr, Parrott WA, Ashley DA, Hussey RS, Boerma HR (1996) Molecular markers associated with seed weight in two soybean populations. Theor Appl Genet 93:1011–1016

Nyquist WE (1991) Estimation of heritablility and prediction of selection response in plant populations. Crit Rev Plant Sci 10:235–322

Orf JH, Chase K, Jarvik T, Mansur LM, Cregan PB, Adler FR, Lark KG (1999) Genetics of soybean agronomic traits: I. comparison of three related recombinant inbred populations. Crop Sci 39:1642–1651

Qui BX, Arelli PR, Sleper DA (1999) RFLP markers associated with soybean cyst nematode resistance and seed composition in a 'Peking' × 'Essex' population. Theor Appl Genet 98:356–364

Sebolt AM, Shoemaker RC, Diers BW (2000) Analysis of a quantitative trait locus allele from wild soybean that increases seed protein concentration in soybean. Crop Sci 40:1438–1444

Smith TJ, Camper HM (1973) Registration of 'Essex' soybean. Crop Sci 13:495

Sneller CH (1994) Pedigree analysis of elite soybean lines. Crop Sci 34:1515–1522

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

SoyBase (1995) A genome database for Glycine. http://www. SoyBase.org. Verified 21 October 2003

Specht JE, Chase K, Macrander M, Graef BL, Chung J, Markwell JP, Germann M, Orf JH, Lark KG (2001) Soybean response to water: a QTL analysis of drought tolerance. Crop Sci 41:493– 509

Vollmann J, Schausberger H, Bistrich H, Lelley T (2002) The presence or absence of the soybean Kunitz trypsin inhibitor as a quantitative trait locus for seed protein content. Plant Breed 121:272–274

Wang D, Shi J, Carlson SR, Cregan PB, Ward RW, Diers BW (2003) A low-cost, high-throughput polyacrylamide gel electrophoresis system for genotyping with microsatellite DNA markers. Crop Sci 43:1828–1832