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Construction of a genetic map in a sorghum recombinant inbred line using probes from different sources and its comparison with other sorghum maps

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Abstract. A genetic map was established using 120 F_5 sorghum recombinant inbred lines (RILs) developed from a cross between 2 Australian elite sorghum inbred lines, QL39 and QL41. A variety of DNA probes, including sorghum genomic DNA, maize genomic DNA and cDNA, sugarcane genomic DNA and cDNA, and cereal anchor probes, were screened to identify DNA polymorphism between the parental lines. Using 5 restriction enzymes, probe polymorphism levels were low (26·5%). A total of 155 restriction fragment length polymorphism (RFLP) loci and 8 simple sequence repeat (SSR) loci were mapped onto 21 linkage groups, covering a map distance of approximately 1400 cM. Genes for 3 simply inherited traits, awns (AW), mesocarp thickness (*Z*), and organophosphate insecticide (OPR) reaction, were also mapped. The relationships between this map and other published sorghum maps were reviewed and a comparison of major sorghum RFLP maps attempted. This comparison is expected to enhance the effectiveness of existing mapping information and will facilitate efforts to map agronomically important traits in sorghum.

Additional keywords: genome mapping, RFLP, SSR, RILs.

Introduction

Construction of a linkage map is the most fundamental step required for a detailed genetic study and marker-assisted breeding approach in any crop (Tanksley *et al*. 1989). Sorghum genome mapping based on DNA markers began in the early 1990s, and since then, several genetic maps of sorghum have been produced. The first maps of sorghum were based largely on DNA probes previously mapped in the maize genome (Hulbert *et al*. 1990; Binelli *et al*. 1992; Whitkus *et al.* 1992; Berhan *et al*. 1993; Pereira *et al*. 1994), and of these, only the map of Pereira *et al*. (1994) is complete, with 10 linkage groups. More recently, 3 maps were produced using mainly sorghum DNA probes (Chittenden *et al*. 1994; Ragab *et al*. 1994; Xu *et al*. 1994), and another recently published sorghum map was based on both maize and sugarcane probes (Dufour *et al*. 1997). The map of Chittenden *et al*. (1994) also contains 10 linkage groups. All of these maps were developed using restriction fragment length polymorphism (RFLP) markers and most of the mapping populations were F_2 with the exception of the map of Dufour *et al*. (1997) in which 2 recombinant inbred line (RIL) populations were used for the construction of a composite sorghum map. Although the above maps are the only major published maps of sorghum, some of the laboratories in which these early maps were constructed, as well as other laboratories, have continued to add additional markers to their maps, to increase these map coverages, to reduce the number of linkage groups, and to convert to RIL populations, as reported at recent meetings such as the Plant and Animal Genome Conference (Kong *et al*. 1997; Toure *et al*. 1997).

In this paper, we report the construction of a sorghum map using a RIL population and a wide range of probe sources. Most of the loci in the present map have been mapped onto these published maps. In addition, we report the relationship between our sorghum map and the 4 previously published sorghum maps, as revealed by comparisons based on common loci.

Material and methods

Plant material

Random F_5 RILs (120) were derived from an F_2 population from the cross QL39×QL41. Both parental lines were developed by the Queensland Department of Primary Industry (Henzell 1992; Henzell *et al*. 1994).

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Field trials and phenotypic data collection

All RILs were planted at the Queensland Department of Primary Industries Gatton Research Station in 1994–95 as single-row plots (5·5 m by 0·7 m) with 2 replications. Phenotypic data for awns and mesocarp thickness were collected before harvesting. Leaf burn data were obtained a few days after spraying with Lorsban R.

Awn

QL39 is awnless and QL41 has long awns. The RILs varied for presence and absence of awns. The length of awns varied, however, from weak awns to long awns. RILs with any visible awns were recorded as awned.

Mesocarp thickness

The gene *Z* affects the thickness of the pericarp, its main influence being on the mesocarp. QL39 has no mesocarp, whereas the mesocarp is present in QL41. The RILs varied for mesocarp thickness from absent to thick. This was visually scored as absent (*Z*–) when the grain had a translucent appearance, and present (*zz*) when the grain had a dull appearance.

Organophosphate insecticide reaction

Sorghum lines vary in reaction to insecticide. Those showing no leaf necrosis are resistant and those with any necrosis are susceptible. Susceptibility is dominant to resistance (Schertz and Stephens 1966). QL39 is resistant and QL41 is susceptible. The RILs showing no leaf necrosis were rated as resistant and those with any necrosis were rated as susceptible.

Probe sources

A total of 627 DNA clones from 7 sources were used as probes in the mapping study. The sources, codes, and number of different RFLP probes are summarised in Table 1. For most of the clones, a single insert of an expected size was amplified after polymerase chain reaction (PCR), but multiple inserts were amplified for some clones. These unexpected extra fragments were excised from the gels separately and used as anonymous probes in this study.

Single sequence repeat (SSR) primers

Seventeen SSR primer pairs were identified by Brown *et al.* (1996) as detecting polymorphisms between sorghum lines. These 17 primer pairs were then selected for genome mapping in this study. The primers were synthesised by GIBCO/BRL based on the sequence information provided by Brown *et al.* (1996). A list of the primers and their codes is given in Table 2.

RFLP and PCR techniques

Leaf tissue was collected from the field for DNA extraction. The detailed RFLP procedures were conducted as previously described in Tao *et al.* (1996).

All PCR was performed on a thermocycler in 25 μL volumes containing 10 ng sorghum genomic DNA, 10 pmol (50 ng) of each primer, 5 mM $MgCl_2$, 40 μM each dNTP, 1×PCR buffer (50 mM KCl, 10 mM Tris–HCl pH (8·0), 10 mM NaCl, 0·01 mM EDTA, 0·5 mM DTT, 0·1% TRITON X-100), and 1·3 units of Taq DNA Polymerase (Biotech). The samples were subjected to 40 cycles of denaturation for 1 min, annealing for 1 min, and amplification for 1·5 min. Denaturation was performed at 94°C, anealing at 65°C for the first 5 cycles, 60°C for the next 5 cycles, and 55°C for the remaining 30 cycles. Extension was performed at 72°C. DNA fragments produced by these amplifications were separated by electrophoresis in 2% agarose gels and stained with ethidium bromide.

Linkage analysis

Linkage analysis was performed by means of MAPMAKER/EXP version 3·0 (Lander *et al.* 1987). The 'group' command with a minimum LOD score of 3·0 and maximum distance of 30% recombination was used. Multipoint linkage analysis on loci within groups was subsequently performed. For each group, a 3-point linkage analysis was conducted followed by the 'order' command. After all scores were checked, the 'framework' command for each declared linkage group was used to construct the final map.

Results

Construction of a genetic linkage map for sorghum

Five restriction enzymes, *Dra*I, *Eco*RI, *Eco*RV, *Hin*dIII, and *Xba*I, were used to identify the probes which produced RFLPs between the parental lines of the mapping population. After autoradiography, it was found that signals detected from hybridisations using all heterologous genomic and most cDNA probes were as strong as those detected from the sorghum genomic DNA probes. Some of the sugarcane and maize cDNA probes gave weaker but still scorable signals. A total of 166 polymorphic probes were identified after parental screening of all 627 clones. The percentage of polymorphic probes varied with different probe sources, ranging from 17% (maize cDNA clones) to 32% (sorghum genomic DNA clones).

Table 1. Probe sources, number of markers produced, and percentage of polymorphic of probes surveyed in this study

Source	Code	Number of probes/markers			Percent polymorphic
		Screened probes	Polymorphic probes	Markers	
Sorghum gDNA	TXS	145	46	52	32
	PSB	80	25	27	31
Sugarcane gDNA	SSCIR	82	21	24	26
Sugarcane cDNA	TS	45	9	10	20
Maize gDNA	UMC/BNL	59	10	10	17
Maize cDNA	ISU	66	19	21	29
Cereals	RZ/CDO/BCD	150	36	38	24
Total/average		627	166	182	26·5

The genomic DNA clones in general gave slightly higher percentages of polymorphic probes than the cDNA clones. The average percentage of polymorphic probes is 26·5% (Table 1).

PCR products were obtained from all 17 SSR primer pairs used for parental screening under the same optimised PCR condition. After separation by electrophoresis on 2% agarose gels, polymorphisms were identified from the DNA fragments amplified from 10 SSR primer pairs (Table 2).

Table 2. A list of sorghum SSR primers surveyed and their codes in this study

Asterisk indicates that primer produced polymorphic markers in the mapping population. For sequence information refer to Brown *et al.* (1996)

Primer ID	Code	Primer ID	Code
*SBKAFGKI	M-KA	Sb 5–206	MB5–206
ZMADH2N	M-H2N	Sb 5–214	MB5–214
*Sb 1–1	MB1–1	Sb 5–236	MB5–236
*Sb 1–10	MB1–10	Sb 6–36	MB6–36
*Sb 4–15	MB4–15	*Sb 6–57	MB6–57
*Sb 4–22	MB4–22	*Sb 6–84	MB6–84
*Sb 4–32	MB 4–32	*Sb 6–325	MB6–325
Sb 4–121	MB4–121	Sb 6–342	MB6–342
*Sb 5–85	MB5–85		

A total of 194 loci were produced from the 166 polymorphic RFLP probes, 10 SSR primers, and 2 anonymous probes after progeny screening. Of these loci, 90% (174 out 194) were characterised by co-dominant alleles. The remaining 20 loci behaved as dominant alleles. Most of the polymorphic RFLP probes produced only one single segregating fragment which was then mapped as a single locus onto the map. However, 12 probes produced 2 loci. These included 5 TXS probes, 2 ISU probes, and 1 TS, RZ, and SCSIR probe.

A total of 21 linkage groups were identified and named LG1–LG21 in order of decreasing genetic distance. Six small linkage groups only contained 2 loci (LG16–LG21), and one group had 3 loci (LG15). The number of loci in the remaining 14 groups ranged from 4 (LG10) to 23 (LG1) and the genetic distances varied from 28·0 cM (LG14) to 230·3 cM (LG1). The total estimated map size was 1400 cM and the average distance per locus was about 10 cM.

Eight SSR loci were located on 6 linkage groups. Two were on LG1 and LG8, respectively, and one each on LG3, LG5, LG12, and LG19 (Fig. 1). This result indicates that these SSR markers were distributed throughout the sorghum genome.

Genes for 2 morphological traits, mesocarp thickness (Z) and awnless (awn), were mapped onto linkage groups LG6 and LG14, respectively. The gene for organophosphate insecticide reaction (OPR) was mapped onto linkage group LG7 of the current map (Fig. 1). The location of the gene at a distance of 13·6 cM from marker TXS713 indicates that it is the same gene mapped by Toure *et al.* (1997).

Allele frequencies and ratios of genotypic classes at most of the loci fitted the expected segregation patterns. Segregation distortion was found for 31 loci. Loci with distorted segregation were not distributed at random throughout the genome. Most of these loci fell into 3 clusters on 3 linkage groups: LG2, LG6, and LG8. The aberrant loci in LG2 showed distorted segregation in favour of QL39 as did the loci in LG6. In contrast, the loci located in LG8 were distorted in favour of the other parent, QL41 (Fig. 1).

Comparisons between the present map and other sorghum maps

In this study, subsets of probes representing some of the mapped loci were used to compare the current map with the 4 major previously published sorghum maps (Table 3).

Comparison between present map and map of Chittenden et al. *(1994)*

Nineteen loci were common between the current map and the map of Chittenden *et al.* (1994). The 19 loci were distributed onto 9 of the 10 linkage groups of the map of Chittenden *et al.* (1994). Six of the 9 linkage groups contained multiple DNA markers, providing good evidence for linkage group alignment.

Four PSB loci and BNL5·09 of LG3 (present map) were colinear with LGC (Chittenden *et al.* 1994), and colinearity was also observed between 4 PSB loci of LG2 and LGF. Closely linked loci UMC4 and PSB266 in LG5 were also closely linked to each other in LGB, as was the case for linked loci PSB115 and PSB632 in LG1 and in LGI.

Comparison between present map and map of Xu et al. *(1994)*

Thirty-two common loci were found between the current map and the map of Xu *et al.* (1994). The 32 loci were distributed onto 12 of the 14 linkage groups of Xu *et al.* (1994). Of these, 8 contained multiple markers, and the remaining 4 linkage groups contained single common markers. Good agreements occurred between LG1, LG2, LG4, and LG12 of this map and LGB, LGI, LGC, and LGF of their map. LG3 covered the loci from both LGA and LGJ of the map of Xu *et al.* (1994).

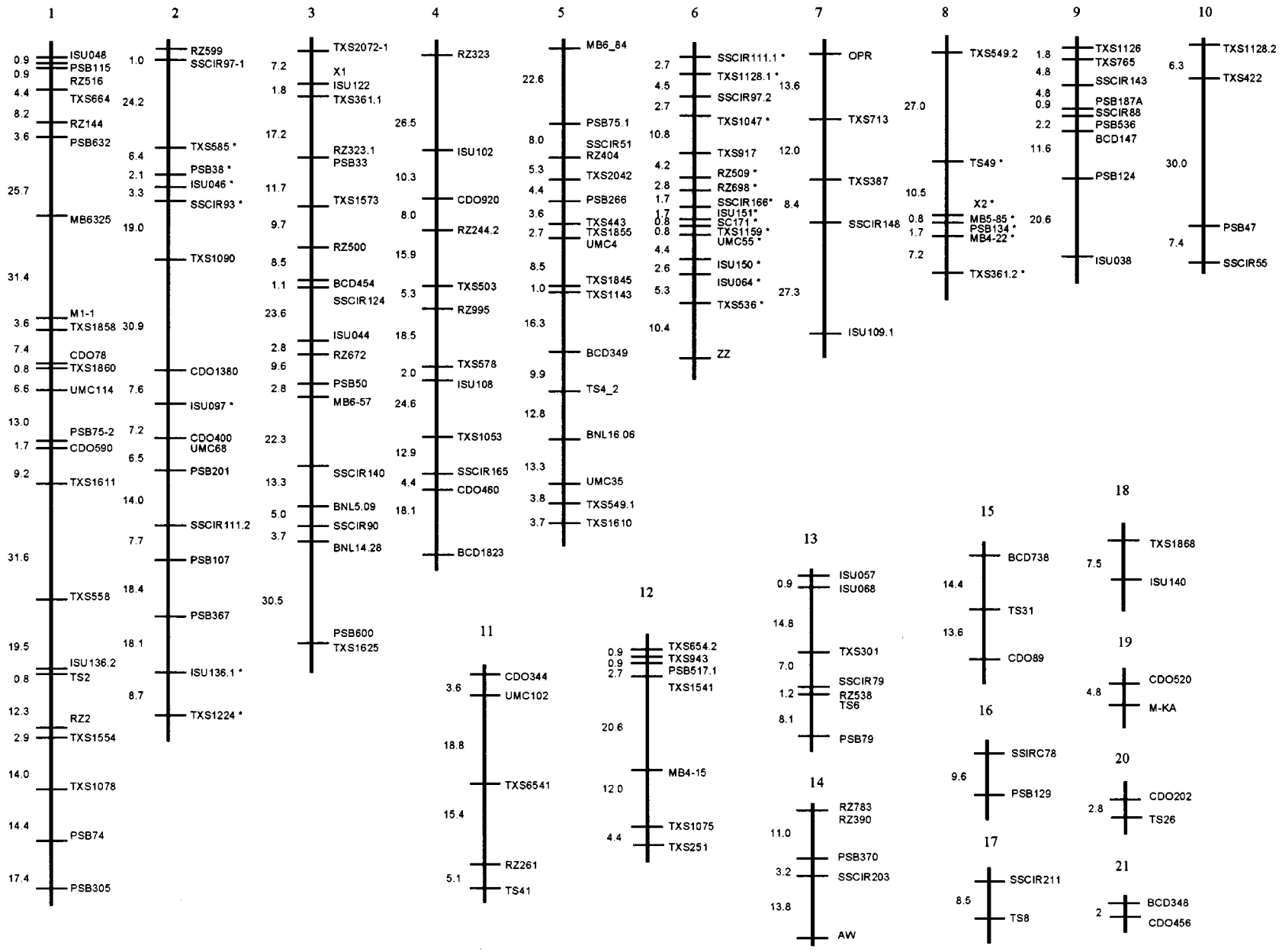

Fig. 1. Sorghum genetic map of the recombinant inbred population derived from a cross QL39×QL41. Linkage groups at minimum LOD 3·0, maximum distance 30·0 cM. Distorted loci are marked with *.

Comparison between present map and map of Pereria et al. (1994)

Nineteen loci are common between the current map and the map of Pereria *et al.* (1994). The 19 loci were distributed onto 7 of the 10 linkage groups of Pereria *et al.* (1994), of which 5 contained multiple markers. Both LG3 and LGC share the same synteny of 3 loci, ISU122, ISU044, and BNL5·09. But loci ISU64 and ISU150 in LGC and loci UMC55 and ISU151 in LGF of the map of Pereria *et al.* (1994) were mapped to the same linkage group, LG6, of this map.

Comparison between present map and map of Dufour et al. (1997)

Only 10 common loci were found between the current map and the map of Dufour *et al.* (1997). The 10 loci were distributed onto 5 of the 12 linkage groups of the map of Dufour *et al.* (1997) and 2 of the 5 contained multiple markers. Four loci in LG6 occurred in the same order in LGF of the map of Dufour *et al.* (1997). Two closely linked maize RFLP loci, BNL14·28 and BNL5·09, gave the same synteny in both LG3 and LGC.

Discussion

Level of polymorphism

Only 26·5% of the probes surveyed showed polymorphism between the 2 parental lines after digestion with 5 restriction enzymes. This percentage of polymorphic probes is much lower than in other previously reported mapping studies; 50·8% from the same 5 restriction enzymes in the study by Xu *et al.* (1994), 95% from only 3 enzymes in the study by Chittenden *et al.* (1994), and 37% from only 2 enzymes in the study by Pereira *et al.* (1994). The diversity in sorghum, in general, is fairly low compared with other major crops (Tao *et al.* 1993), and selection for agronomically important traits in sorghum breeding can further decrease the level of polymorphism between elite inbreds (Jordan *et al.* 1996), as used as parents of the present mapping population. The low level of DNA polymorphism for the population used in the present study made it much more difficult to obtain sufficient polymorphic probes to construct a map. Consequently, a greater effort has been expended in identifying polymorphic probes from a variety of sources.

Genetic distance of the linkage map

Fewer loci (166) have been mapped in the present map than in the maps reported by Pereira *et al.* (1994), Chittenden *et al.* (1994), and Xu *et al.* (1994), in which 201, 276, and 190 loci were mapped, respectively. The total distance of the present map (about 1400 Cm) is also smaller compared with the total distance of these maps of 1530 cM, 1445 cM, and 1851 cM, respectively.

Table 3. Relationships between the current recombinant inbred line map and the four previously published sorghum maps

Numbers of common loci between different linkage groups are in parentheses

Present (map)	Chittenden *et al.* (1994)	Xu *et al.* (1994)	Pereira *et al.* (1994)	Dufour *et al.* (1997)
1	I(2)	B(5)	H(3)	H(1)
2	F(4)	I(3)	D(2)	–
3	C(4)	A(2), J(1)	C(3)	C(2)
4	–	C(3)	D(1), G(1)	–
5	B(2)	G(4), C(1)	–	F(4)
6	–	H(2), C(1)	F(2), C(2)	F(1)
7	–	A(1), N(1)	–	–
8	G(1)	–	–	–
9	H(1), J(1)	D(2)	A(1)	A(1)
10	E(1)	C(1)	–	–
11	–	–	I(1)	I(1)
12	–	F(3)	–	–
13	A(1)	D(1)	G(2)	–
14	A(1)	–	–	–
15	–	–	–	–
16	A(1)	–	–	–
17	–	–	–	–
18	–	E(1)	E(1)	–
19	–	–	–	–
20	–	–	–	–
21	–	–	–	–
Total	19	32	19	10

More than 10 linkage groups have been obtained in the present map and 33 loci remained unlinked, indicating the presence of gaps between linked loci in the current map. A major contributing factor is that the 2 parents of the mapping population share common ancestors. The coefficient of parentage between the 2 parents, based on pedigree information, is 0·12 (data not shown). Selection for agronomically important traits during line development means that the true coefficient of parentage may in fact be greater than the theoretical estimate. If a chromosomal segment was inherited in both parents from a common ancestor, no polymorphism could be detected in this region, resulting in a gap in the map.

Distortion

Aberrant segregation has been repeatedly observed in the constructions of genome maps for different plant species, particularly for the populations derived from distant crosses. Distorted segregation ratios have been also reported in other sorghum mapping studies (Chittenden *et al.* 1994; Xu *et al.* 1994; Dufour *et al.* 1997). The frequency of aberrant loci is higher in this study and the clusters of distorted loci are different from the ones found in other studies. One of the major reasons for aberrant segregation may be the elimination of gametes or zygotes, which is controlled by a lethal factor located in the neighbouring region of the marker. In rice, it was suggested that the presence of a lethal factor located on the rice chromosome III caused partial gametic selection in both the male and female side (Causse *et al.* 1994). The action of the lethal factor was found to be effective on both male and female gametes with a fertilisation ability 41·5% of that of the normal gametes (Cheng *et al.* 1996). The population used in this mapping study is known to be segregating for a gene(s) causing partial female sterility (data not shown). The increase of segregation distortion in RILs has possibly resulted from the cumulative effect of selection against alleles of one of the parents during propagation of the RILs.

SSR markers

PCR primers for the amplification of DNA fragments containing SSRs from sorghum were successfully developed through 3 different approaches by Brown *et al.* (1996) and it was also reported that sorghum fragments can be amplified using maize SSR primers (Brown *et al.* 1996). A total of 13 sorghum SSR loci were also characterised by Taramino *et al.* (1997) and 7 of these SSR markers were mapped, using an existing sorghum RFLP map (Taramino *et al.* 1997). SSR markers have been incorporated into an existing RFLP based on the map of Xu *et al.* (1994) (Kong *et al.* 1997).

In this study, a single optimised PCR condition was successfully applied for all 17 primers tested. The level of polymorphism of these primers, 60% (10 out 17), was higher than obtained for the RFLP probes. The mapped SSR loci were distributed throughout the genome. All of these factors are encouraging for the extensive use of SSR markers in sorghum mapping. More SSR loci can be easily mapped as more SSR primers become available in the future.

Loci for simply inherited traits

Genetic studies of morphological traits in sorghum began early this century and many genes for morphological traits have been identified; however, the precise location of these genes in a complete genome map is still unclear (Doggett 1988). Use of morphological traits as genetic markers in sorghum breeding programs has been limited as only few morphological markers are available and the chance that a morphological marker is linked to an agronomically important trait is very low. This situation would be changed if a morphological trait was mapped onto a complete genetic linkage map. Then, it could be used as an easily recognised landmark for a whole linkage group rather than just a single marker. For example, from the result of this study, mesocarp thickness and awns can be used as a landmark for linkage groups LG6 and LG14, respectively. This gives morphological markers much more power when they are used in a breeding program, particularly if any useful major gene or QTL alleles are mapped onto these 2 linkage groups. In addition, the gene for organophosphate insecticide reaction was also located onto LG7 of the current map. The marker linked to this gene can then be used to select for resistance to the insecticide without needing to spray (and kill) the plants.

Comparison of sorghum maps

Few common loci can be found in the maps of Chittenden *et al.* (1994), Pereira *et al.* (1994); Xu *et al.* (1994) and Dufour *et al.* (1997). For the present study, probes were chosen from the same sources as used for these maps. Common loci were thus mapped onto both the current map and one or more of the other maps, enabling comparisons to be made between these maps for the first time.

As demonstrated in Table 3, the current LG1 corresponds to LGI of the map of Chittenden *et al.* (1994), LGB of the map of Xu *et al.* (1994), and LGH of the

maps of both Pereira *et al.* (1994) and Dufour *et al.* (1997). The current LG2 corresponds to LGF of the map of Chittenden *et al.* (1994), LGI of the map of Xu *et al.* (1994), and LGD of the maps of Pereira *et al.* (1994). LG5 of the current map corresponds to LGB of the map of Chittenden *et al.* (1994), LGG of the map of Xu *et al.* (1994), and LGF of the maps of Dufour *et al.* (1997.

It was noted that linkage rearrangements occurred for some of the published maps. Loci in LGA and LGJ of Xu *et al.* (1994) were mapped onto the same LG3 of the current map, and corresponded to LGC of the maps of Pereira *et al.* (1994), Dufour *et al.* (1997), and Chittenden *et al.* (1994). The same linkage rearrangements were also observed by other groups with the same probes (G. Hart, pers. comm.). Similarly, 2 closely linked loci in LGC and another 2 closely linked loci in LGF of the map of Pereira *et al.* (1994) were joined together in LG6 of the current map, indicating that some linkage groups may still be questionable.

Information from comparisons between the current map and other maps also facilitated the joining of some linkage groups in this study. LG5 and LG6 are very likely to be part of the same linkage group as both linkage groups contain loci of the same LGF of the map of Dufour *et al.* (1997). Comparisons between the current map and rice and sugarcane maps provided additional evidence for this (data not shown). Another possible combination could be between LG4, LG13, LG14, and LG16 of the current map. Both groups LG4 and LG13 have loci of the same linkage group LGG of the map of Pereira *et al.* (1994) and LG13, LG14, and LG16 each have a single locus in common with LGA of the map of Chittenden *et al.* (1994). Strong evidence also comes from a comparison between the current map and the rice map (data not shown). Ultimately, it should be possible to join some linkage groups of the current map to give a final number of 10 linkage groups.

Complete alignment of all of the sorghum maps is still far from satisfactory as only half of the linkage groups can be unambiguously aligned at this stage. Constructing a bridging map for several existing maps was more difficult than expected. Lack of sufficient common loci with which to compare and frequent inconsistency in probe location (data not shown) were major problems for this study.

Future applications

A genetic map was established using 120 F_5 sorghum RILs developed from a cross between 2 Australian elite sorghum inbred lines. Recombinant inbred lines offer several advantages over other populations used for genetic mapping. The most important benefit from RILs is that RILs can be distributed, replicated, and evaluated from experiments in different environments, which is essential for the analysis of quantitative traits (Burr *et al.* 1988; Zamir *et al.* 1993; Goldman *et al.* 1995; Paran *et al.* 1995). The particular population used in this study was chosen because the parents are segregating for 2 important traits, stay-green and midge resistance, which we are currently attempting to map. In addition, both parents show good agronomic performance and have been widely used as parents of commercial hybrids and breeding lines in Australia. Any markers developed from this cross could directly benefit Australian sorghum production.

Moreover, the relationships between different sorghum maps and colinearity and synteny between sorghum and other economically important and well-studied crops such as maize, rice, and sugarcane will be facilitated by this study (data not shown). By bringing all the information together, this map should help sorghum breeders to establish whether variation for a particular characteristic in this population is due to the same major gene or QTL allele of other mapping populations, either within sorghum or even in different species, using common loci (colinearity and synteny) between the maps as landmark loci for reference. In addition, any markers developed from this population can be easily tested and transferred to other populations within or between species.

Generally, the alignment of the different sorghum maps will serve sorghum researchers to use all of the mapping information more effectively. First of all, most genome maps available are far from saturated, which implies that uncertainties remain in terms of genetic distance, order of loci, and even linkage groups in the map. The map user cannot make their own judgement on the accuracy of available maps and the reliability of the linkage for the particular region(s) in which genes for agronomically important traits may reside until additional information from different mapping studies is available. From this viewpoint, using a bridging map covering different sources of mapping information, like the map presented in this paper, will help to avoid some confusion and assist in further applications of the maps. Secondly, it is difficult to get sufficient polymorphic markers from a single source based on one map to develop a useful map for gene tagging or QTL mapping. If a RIL population is used for particular traits of interest, lower levels of diversity make genotyping even more difficult. However, once an overall bridge map is established, probes from all sources become informative and can be pooled. The problem of limited choice for RFLP probes and other kinds of markers can then be solved.

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References

Berhan, A. M., Hulbert, S. H., Bulter, L. G., and Bennetzen, J. L. (1993). Structure and evolution of the genomes of S*orghum bicolor* and *Zea mays*. *Theoretical and Applied Genetics* **86**, 598–604.

Binelli, G., Gianfranceschi, L., Pe, M. E., Taramino, G., Busso, C., Stenhouse, J., and Ottaviano, E. (1992). Similarity of maize and sorghum genomes as revealed by maize RFLP probes. *Theoretical and Applied Genetics* **84**, 598–604.

Brown, S. M., Hopkins, M. S., Mitchell, S. E., Senior, M. L., Wang, T. Y., Duncan, R. R., Gonzalez-Candelas, F., and Kresovich, S. (1996). Multiple methods for the identification of polymorphic simple sequence repeats (SSRs) in sorghum [*Sorghum bicolor* (L.) Moench]. *Theoretical and Applied Genetics* **93**, 190–8.

Burr, B., Burr, F. A., Thompson, K. H., Albertson, M. C., and Stuber, C. W. (1988). Gene mapping with recombinant inbreds in maize. *Genetics* **118**, 518–26.

Causse, M., Fulton, T. M., Chap, Y. G., Ann, S., Chunwongse, J., Wu, K. S., Xiao, J. H., Ronald, P. C., Harrington, S. E., Second, G., McCouch, S. R., and Tanksley, S. D. (1994). Saturated molecular map of the rice genome based on an interspecific backcross population. *Genetics* **138**, 1251–74.

Cheng, R., Satio, A., Takano, Y., and Ukai, Y. (1996). Estimation of the position and effect of a lethal factor locus on a marker linkage map. *Theoretical and Applied Genetics* **93**, 494–502.

Chittenden, L. M., Schertz, K. F., Lin, Y. R., Wing, R. A., and Paterson, A. H. (1994). A detailed RFLP map of *Sorghum bicolor*×*S. propinquum*, suitable for high-density mapping, suggests ancestral duplication of *Sorghum* chromosomes or chromosomal segments. *Theoretical and Applied Genetics* **87**, 925–33.

Dufour, P., Deu, M., Grivet, L., Dhont, A., Paulet, F., Bouet, A., Lanaud, C., Glaszmann, J.C., and Hamon, P. (1997). Construction of a composite sorghum genome map and comparison with sugarcane, a related complex polyploid. *Theoretical and Applied Genetics* **94**, 409–18.

Doggett, H. (1988). 'Sorghum.' 2nd Edn. pp. 165–99. (John Wiley and Sons: New York.)

Goldman, I., Paran, I., and Zamir, D. (1995). Quantitative trait locus analysis of a recombinant inbred line population derived from a *Lycopersicon esculentum*×*L. cheesmanii* cross. *Theoretical and Applied Genetics* **90**, 925–32.

Henzell, R. G. (1992). Grain sorghum breeding in Australia: current status and future prospects. *In* 'Proceedings of the Second Australian Sorghum Conference, Gatton, 4–6 February 1992'. (Eds M. A. Foale, R. G. Henzell, and P. N. Vance.) Ocassional Publication No. 68, pp. 70–82. (Australian Institute of Agricultural Science: Melbourne.)

Henzell, R. G., Franzmann, B. A., and Brengman, R. L. (1994). Sorghum midge resistance research in Australia. *International Sorghum and Millets Newsletter* **35**, 41–7.

Hulbert, S. H., Richter,T. E., Axtell, J. D., and Bennetzen, J. L. (1990). Genetic mapping and characterization of sorghum and related crops by means of maize probes. *Proceedings of the National Academy of Sciences USA* **87**, 4251–5.

Jordan, D. J., Tao, Y., Godwin, I. D., Cooper, M., and McIntyre, C. L. (1996). Reduced levels of genetic diversity in midge resistant sorghum hybrids. *In* 'Proceedings of the Third Australian Sorghum Conference'. pp. 383–90.

Kong, L., Dong, J., and Hart, G. E. (1997). Linkage mapping of simple-sequence repeat loci in *Sorghum bicolor* L. Moench. (Abstract.) *In* 'Plant & Animal Genome V, San Diego, CA. January 1997'. p. 100.

Lander, E. S., Green, P., Abrahamson, J., Barlow, A., Daly, M. J., Lincoln, S. E., and Newburg, L. (1987). MAPMAKER: An interactive computer package for constructing primary genetic linkage maps of experimental and natural populations. *Genomics* **1**, 174–81.

Paran, I., Goldman, I., Tanksley, S. D., and Zamir, D. (1995). Recombinant inbred lines for genetic mapping in tomato. *Theoretical and Applied Genetics* **90**, 542–8.

Pereira, M. G., Lee, M., Bramel-Cox, P., Woodman, W., Doebley, J., and Whitkus, R. (1994). Construction of an RFLP map in sorghum and comparative mapping in maize. *Genome* **37**, 236–43.

Ragab, R. A., Dronavelli, S., Saghai-Maroof, M. A., and Yu, Y. G. (1994). Construction of a sorghum RFLP linkage map using sorghum and maize DNA probes. *Genome* **37**, 590–4.

Schertz, K. F., and Stephens, J. C. (1966). Compilation of gene symbols, recommended revisions and summary of linkages for inherited characters of *Sorghum vulgare*. *In* 'Pers. Technical Monograph 3, April 1966'. (Texas A&M University: College Station, TX.)

Tanksley, S. D., Young, N. D., Paterson, A. H., and Bonierbale, M. W. (1989). RFLP mapping in plant breeding: new tool for an old science. *Biotechnology* **7**, 257–64.

Tao, Y. Z., Manners, J. M., Ludlow, M. M., and Henzell, R. G. (1993). DNA polymorphism in grain sorghum (S*orghum bicolor* (L.) Moench). *Theoretical and Applied Genetics* **86**, 679–88.

Tao, Y. Z., McIntyre, C. L., and Henzell, R. G. (1996). Applications of molecular markers to Australian sorghum breeding programs. I: Construction of a RFLP map using sorghum recombinant inbred lines. *In* 'Proceedings of the Third Australian Sorghum Conference'. pp. 443–50.

Taramino, G., Tarchini, R., Ferrario, S., Lee, M., and Pe, M. E. (1997). Characterization and mapping of simple sequence repeats (SSRs) in *Sorghum bicolor*. *Theoretical and Applied Genetics* **95**, 66–72.

Toure, A., Xu, W. W., Rosenow, D. T., Peterson, G. C., and Nguyen, H. T. (1997). Inheritance of insecticide phytotoxicity in sorghum. (Abstract.) *In* 'Plant & Animal Genome V, San Diego, CA., January 1997'. p. 103.

Whitkus, K., Doebley, J., and Lee, M. (1992). Comparative genome mapping of sorghum and maize. *Genetics* **132**, 1119–30.

Xu, G. W., Magill, C. W., Schertz, K. F., and Hart, G. E. (1994). An RFLP linkage map of S*orghum bicolor* (L.) Moench. *Theoretical and Applied Genetics* **89**, 139–45.

Zamir, D., Paran, I., Eshed, Y., Sarfatti, M., Ori, N., and Fluhr, R. (1993). Recombinant inbred populations for high-resolution mapping in tomato. *In* 'Molecular Biology of Tomato'. (Ed. J. Yoder.) pp. 49–58. (Technomic Publishing Co.: Lancaster, Basel.)

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