Flour yield and water absorption in wheat – a pedigree mapping approach

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ABSTRACT

Flour yield and flour water absorption are two of the most important quality traits for milling wheats. A number of studies have tried to locate QTL for these traits, using doubled haploid mapping populations. Results from these studies suggest that the traits are determined by genes in different parts of the genome in different populations, and that large genotype by environment interactions occur. A pedigree mapping approach may be more amenable for genetic analysis of these traits. This approach uses knowledge accumulated for related breeding lines from an improvement program, and incorporates data from a number of years and trial sites. Using this method, we aim to identify more robust QTL for flour yield within the breeding populations and compared the identified marker-trait associations with those found in other studies. The often reported negative relationship between flour yield and flour water absorption has made it difficult to breed for both traits simultaneously. It has been shown that lines do exist which combine both high milling yield and high water absorption, but these lines are rare. Because phenotypic screening for quality traits generally does not commence until late in the selection process, there is a high probability of lines with high flour yield and high flour water absorption being discarded. If markers could be applied early in the selection process to enrich breeding populations for these desirable genes, step-wise progress could be made for important traits.

INTRODUCTION

Genetic studies of end-use quality traits in wheat suggest that the genetic determination of the fine differences between a good and an excellent quality genotype in a breeding program is often gene-pool specific. Many of these quality traits are also strongly influenced by environmental effects and by genotype by environment interaction. The pedigree mapping approach allows us to study genetic relationships within breeding populations, to define these gene-pools, and identify regions of the genome identical-by-descent, and associate that information with a pool of data on quality parameters collected over a number of sites and years. The aim is to define robust, useful marker-trait associations for these often subtle quality improvements, for specific sets of germplasm, and for specific donors of these elite quality traits within breeding programs.

Most studies which have identified QTL (quantitative trait loci) for quality traits are based on bi-parental doubled haploid (DH) populations. The majority of wheat germplasm grown in northern Australia has a significant genetic contribution from the ancestral varieties Cook or Hartog.

Recent studies using germplasm related to Cook or Hartog have identified a number of QTL for flour milling yield. Schmidt et al (2004 and pers. comm.) studied a Kukri/Janz DH population. Janz is a Cook-derived variety, bred by QDPI&F. They identified significant QTL for milling yield on 1B, 2A, 2D, 4B, 4D, 3B, 5A, 7D. The 1B locus is associated with the Bx7 over-expression, derived from Kukri. The 4B locus was closely associated with the Rht1 dwarfing locus.

Lehmensiek et al (2006) identified QTL for flour yield in three populations, Sunco/Tasman, CD87/Katepwa and Cranbrook/Halberd. Sunco is closely related to Cook. In the Sunco/Tasman population, milling yield QTL were identified on chromosomes 2BS, 4B, 5AL and 6BL. In this population, the only QTL contributed by Sunco was on chromosome 2B, associated with a large introgression from *Triticum. timopheevii*. The QTL identified near the Rht1 locus on 4B was the only QTL found in more than one population (Sunco/Tasman and CD87/Katepwa). The allele contributing to improved flour yield was from Tasman, which carries the *rht1* tall allele. QTL associated with protein content and hardness were also identified as likely to influence flour milling yield.

Another study used a Lang/QT8766 DH population (Lehmensiek et al pers com). Lang is related to Cook and QT8766 is related to Hartog. In this study a strong negative relationship was found between flour milling yield and water absorption. Some lines were identified however with both good milling yield and water absorption (Kelly et al pers com). A negative relationship was found between flour milling yield and grain protein. A highly significant positive relationship was found between flour milling yield and both test weight and plant height. QTL associated with flour milling yield were found on 4BS and 4DS from QT8766 and Lang respectively for all 3 year by sites. A QTL on 3DL from QT8766 was detected in 2 year/sites and QTL were detected in single year/sites on 2AS and 6AS from

QT8766 and on 2BS from Lang. A single QTL for water absorption was detected in one year/site on 4DS from QT8733. This locus appears to be in repulsion with that from Lang associated with flour yield.

In this study we aimed to identify some regions of the genome associated with flour milling yield in a set of germplasm with the ancestral wheat variety Cook in its pedigree.

MATERIALS AND METHODS

Data for flour milling yield from 27 designed experiments in 2000 to 2004 from the QDPI&F wheat breeding program was used. Flour yield (FY) was assessed as the percentage of total grain weight accounted for by the combination of all flour fractions. Grain samples were conditioned to 15% moisture content using AACC Method 26-10A (2000) prior to milling through a Buhler, MLU-202, pneumatic mill by AACC Method 26-21A (2000). Flour water absorption was measured using a 50g Farinograph (Brabender® OHG Duisburg, Germany) according to RACI Method 06-02 (2003). Data for analysis were analysed summaries (that is, genotype means and statistical weights) of flour yield adjusted for both field and laboratory trend from single analyses of these two phase (that is, field and laboratory) experiments.

106 SSR markers were selected for maximum genetic coverage of chromosomes 2B, 2D, 3B, 3D, 4A, 4B, 4D and 7A and expected polymorphism. DNA was extracted from a pool of up to 10 plants. Some of these primer pairs also produced peaks associated with other chromosomes. Assignment of fragments to loci was based on reported band sizing and genetic location. Approximate locations reported in Table 1 were drawn from composite maps (Rudi Appels pers comm.).

There were 48 genotypes for which data for both flour yield and markers were available. Genotype means and statistical weights from the individual analyses of raw data were used in a combined analysis over all trials, initially in a succession of single marker, or allele by allele analyses. Reported here are markers for which at least one allele showed a greater than 5% probability of being associated with a difference in flour yield. All represented alleles were scored as either present "1" or absent "0", alleles were excluded if there was less than 10 observations in either class. Association between the quality traits and marker scores was determined by a sequential F-test for the fitted (fixed) allele effect.

RESULTS AND DISCUSSION

This study identified 70 markers from 13 chromosomes for which at least one allele was associated with significant differences in flour milling yield. In some cases groups of markers are probably associated with individual QTL, in others they are spread across entire chromosomes. Some of the regions identified as associated with flour milling yield are in common with those identified in bi-parental populations involving related genetic material and others are unique.

Table 1. 70 significant associations were detected between markers and milling yield on 13 chromosomes. Genetic distances are normalised cM values produced as part of developing a composite map.

Chr	Marker	Location (Morgans)	Chr	Marker	Location (Morgans)
2A	Gdm005.1	0.29	4A	Wmc161.2	1.05
2B	Gwm614.1	0.03	4A	Gwm350.1	1.08
2B	Gwm257.1	0.47	4A	Wmc262.1	1.14
2B	Gwm429.1	0.60	4A	Wmc232.1	1.19
2B	Gwm374.1	0.74	4A	Gwm160.1	1.29
2B	Gwm120.1	0.89	4B	Wmc048.3	0.30
2B	Gdm093.1	1.19	4B	Gwm113.1	0.31
2B	Wmc317.1	1.25	4B	Gwm495.1	0.33
2D	Wmc111.1	0.13	4B	Gwm513.1	0.33
2D	Wmc025.1	0.26	4B	Gwm192.3	0.36
2D	Wmc112.1	0.27	4B	Gwm368.1	0.36
2D	Gwm484.1	0.31	4B	Wmc149.1	0.39
2D	Gwm102.1	0.44	4B	Gwm251.1	0.45
2D	Wmc018.1	0.46	4D	Wmc285.1	0.00
2D	Wmc190.1	0.47	4D	Gwm608.2	0.41
2D	Gwm539.1	0.71	4D	Wmc331.1	0.60
2D	Wmc041.1	0.98	4D	Gdm145.1	0.71
2D	Gwm349.1	1.04	5D	Wmc161.1	1.16
2D	Gwm301.2	1.07	6A	Wmc243.1	1.02
2D	Gwm301.1	1.17	7A	Gwm233.1	0.03
3B	Gwm389.1	0.10	7A	Gwm350.3	0.24
3B	Gwm533.1	0.14	7A	Wmc479.1	0.29
3B	Wmc085.1	0.35	7A	Wmc283.2	0.63
3B	Gwm566.1	0.43	7A	Wmc052.2	0.66
3B	Wmc326.1	0.74	7A	Wmc083.2	0.66
3B	Gwm114.2	0.89	7A	Gwm282.1	1.70
3B	Gwm340.1	0.98	7A	Gwm332.1	1.71
3B	Gwm547.1	1.00	7A	Wmc273.3	2.06
3D	Gwm161.1	0.25	7B	Gwm573.2	0.51
3D	Wmc529.1	0.53	7B	Wmc273.1	0.89
3D	Gdm088.1	0.67	7D	Gwm635.1	0.19
3D	Gwm383.1	0.82	7D	Gwm350.2	0.28
3D	Gwm114.1	1.17	7D	Gdm145.2	0.40
4A	Wmc048.1	0.54	7D	Wmc405.2	0.97
4A	Gwm637.1	0.94	7D	Wmc273.2	2.29

For example, markers from nearly the entire length of chromosome 2B were associated with significant

differences in flour milling yield in our set of data. This is likely associated with a large, interstitial translocation present in Cook and many of it's derivatives between chromosome 2B and *T. timopheevii*. This genetic region was also identified by Lehmensiek et al (2006) and Lehmensiek et al (pers com) as associated with improved milling yield, and was contributed from Cook and its relative Lang respectively in these studies.

The region on chromosome 4B identified as associated with flour yield in this study is relatively tightly flanked by markers within about 15cM, and may represent a single QTL. This chromosome was identified as associated with milling yield by Schmidt et al (2004). This region was also found to be associated with milling yield by Lehmensiek et al (2006) but in this case the positive allele was contributed by Tasman rather than the Cook related parent Sunco. This region was the only one found to be associated with flour milling yield in more than one population in their study. In a second study, 4B QTL for flour milling yield were identified in all 3 year/sites by Lehmensiek et al (pers com), with the high milling yield allele contributed by QT8766, the Hartog-derived parent. This part of chromosome 4B carries the Rht1 gene for plant height, but whether this linkage is due to a pleitrophic effect of this gene, or genetic linkage is unclear.

Other chromosomes identified as associated with flour milling yield in Kukri/Janz, namely 2A, 2D, 3B, 4D and 7D also showed associations in this study. A QTL on 2AS was found by Lehmensiek et al (pers comm.) at a single year/site with the desirable allele contributed by QT8733. A QTL on 4DS was also identified in all 3 site/years with the high milling yield allele contributed by Lang. This QTL appeared to be linked in repulsion to one contributing high water absorption. Chromosome 4D also carries the Rht2 dwarfing gene. Its location is distal but linked to the large region identified in this study as associated with flour milling yield.

QTL on 6AS and 2AS were found by Lehmensiek et al (pers comm.) at a single year/site with the desirable alleles contributed by QT8733. This studied identified a single marker-trait association on 6AL. A gliadin gene is located on 6AS. They also identified a QTL on 3DL in 2 year/sites.

The region on chromosome 4A identified here as associated with flour milling yield has not been identified in other studies. This region hosts major genes for resistance to pre-harvest sprouting (PHS) and has also been reported to be associated with later-maturity alpha amylase (LMA). QTL have also been reported on 7B for LMA (Carter et al 2002). The effect on milling yield of these genetic regions may be due to the effect of PHS or LMA rather than a QTL for milling yield *per se*.

Region found here to be associated with milling yield but not reported in other studies of related material were on 5D and 5A. 5D hosts the Pin genes associated with hardness, but the marker associated with flour milling yield in this study is genetically unlinked to these.

This is the first step in the identification of QTL for specific sets of wheat germplasm based on pedigree and source of alleles associated with improved quality. The next steps in this research will be: to repeat this analysis on a larger data set of both genotypes and markers; to develop methods to refine identification of significant regions; to identify associations with traits such as protein content and seed size and reassess the data with these traits as cofactors; to trace alleles identical by descent back through pedigrees to identify subsets of genetic material carrying the effective allele and assess epistatic effects; and to apply this methodology to water absorption and other important quality traits.

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