Association mapping of semolina yield in diverse durum wheat germplasm

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INTRODUCTION

High semolina yield is important to the economic returns of the durum (*Triticum turgidum* L. ssp. *durum* (Desf.) Husn.) milling industry. Chaurand et al. (1999) found that genetic differences in semolina yield exceeded those due to environment, and suggested that breeding for semolina yield and other milling properties was worthwhile.

Semolina yield is difficult to select in breeding programs because of the high labour requirement for milling of samples. Various methods have been proposed to predict semolina yield, including micro-milling procedures, near-infrared reflectance spectroscopy (Ripetti-Ballester et al. 2000), the single kernel characterization system (Sissons et al. 2000), and image analysis (Novaro et al. 2001). These techniques show varying degrees of promise, but all still require labour to process large numbers of breeding samples.

Another approach to selection for semolina yield would be discovery and validation of DNA markers that could be applied to breeding material. Several reports for common wheat (*T. aestivum* L.) flour yield support this approach (eg. Parker et al 1999, Breseghello and Sorrells 2006). Many of the reported QTL from these studies reside on the A and B genomes, so similar research in durum wheat is warranted. There are no reports in the literature of QTL for durum semolina yield.

The objective of this work was to use association mapping of a diverse set of durum genotypes to identify putative QTL for semolina yield in durum wheat.

MATERIALS AND METHODS

One hundred diverse durum genotypes from Argentina, Australia, Canada, France, Germany, Italy, Iran, Mexico, Morocco, New Zealand, Russia, Spain and the United States were grown in field trials arranged in a 10 X 10 lattice designs with two replications. The trials were grown under rain-fed conditions at Regina, Saskatchewan and under irrigation at Vauxhall, Alberta in 2001 and 2002. Test weight and kernel weight were determined on all plots. Semolina yield was measured by milling 2 kg samples on an Allis-Chalmers laboratory mill as described by Dexter et al. (1990). Eighty-one of these genotypes overlapped with another study of linkage disequilibrium (Somers et al. 2007). The genotypic data from that study, comprising 245 microsatellite markers, were used in conjunction with the semolina yield data for association mapping using the 81 lines in common.

A pairwise genetic similarity matrix was calculated based on Rogers Euclidean distance (Rogers 1972) to determine population structure for association mapping. A Bayesian clustering approach was also used to infer the number of sub-populations (K) and to assign individuals to sub-populations based on membership proportion in each sub-population (Q-matrix) with the software STRUCTURE v.2 (Pritchard et al. 2000).

Marker-trait associations were tested with semolina yield least squares means for each environment in a linear mixed model with the program TASSEL 2.0.1 (Yu et al. 2006) using the Q matrix estimated for K=5 as a covariate. Rare alleles (frequency <5%) were either combined into a single genotypic class if their combined frequency was greater than 5%, or scored as missing data. This left 210 informative microsatellite markers to be used for the analysis. Significance of associations between loci and semolina yield was based on an F-test, at a significance level of $P \leq 0.01$, corrected for by performing 10,000 permutations. QTL were considered significant if probability of at least some of the chromosome region was <0.01 and not above 0.05, and at least 3 of the trial environments were involved.

RESULTS AND DISCUSSION

There was good genetic variation for semolina yield, ranging from approximately 61.5 to 70.5%. Marker associations for semolina yield were identified on chromosomes 2A, 2B, 4A, 5B, 6A and 7A (Table 1, Fig. 1). More tentative indications were observed on 1A, 3A, 3B, 5B, and 7B.

Test weight and kernel weight are known to be positively associated with semolina yield (Marshall et al.

1986; Dexter et al. 1987), so we checked to see if any of the observed semolina QTL coincided with test weight or kernel weight QTL. The 6A region *barc3*, a weak

Table1. Chromosome regions markers, and probabilities associated with semolina yield of durum wheat grown at Regina (Rg), SK and Vauxhall (Vh), AB in 2001 and 2002.

		Rg 01	Rg 02	Vh 01	Vh 02
1A	*wmc59*	.006	.011	.023	.016
2A	*wmc522*	.078	.039	.001	.025
2B	*gwm429*	.003	.012	.007	.0001
3A	*wmc559*	.048	0.10	0.003	.047
3A	*cfa2076*	.006	.014	.147	.024
3B	*gwm493*	.003	.030	.031	.095
	wmc43	.039	.009	.014	0.284
	gwm340	.011	.031	.049	.010
4A	*wmc313*	.058	.004	.016	.002
5B	*wmc415*	.017	.027	.069	.080
	wmc508	.013	.004	.006	.015
6A	*barc3*	.007	.014	.001	.042
7A	*wmc790*	.003	.004	.0001	.007
7A	*wmc809*	.0008	.002	.0001	.002
7B	*gwm333*	.09	.09	.003	.362

semolina yield QTL, was flanked with a moderate test weight QTL. The 2A region *wmc522-gwm95* coincided with a strong kernel weight QTL. The 7A region *wmc809* coincided with a moderate kernel weight and moderate to strong test weight QTL. In the latter case, the QTL associations for kernel and test weight were not as strong (ie. less significant) as the observed semolina yield QTL association, suggesting the possibility of additive effects of this locus on semolina yield. After discounting loci that are possibly indirectly linked to semolina yield through test weight or kernel weight, there were still strong, consistent associations with semolina yield on chromosomes 2B, 4A, 5B and 7A, and indications on 1A, 3A and 3B. The number of chromosome regions associated with semolina yield strongly suggests quantitative inheritance of the trait.

Flour yield QTL have also been reported on chromosomes 1A, 2A, 6A and 7A of hexaploid wheat, although only *barc*107 on 6A (Kuchel et al. 2006) appears to be close to that found here (http://wheat.pw.usda.gov/cgi-bin/graingenes/quickquery.cgi?query=nearbyloci&arg1=Xbarc107-6A&arg2=10). One would not expect close agreement of QTL for semolina yield and flour yield given the differences in the milling procedures.

Further work is required to resolve which of the chromosome locations and QTL clusters are worth pursuing for potential marker-assisted selection. To that end, doubled haploid lines have been generated from an inter-cross of a high and a low semolina yield line from this study, and will be genotyped and screened for semolina yield to further investigate the genome regions identified here. Additional validation will be carried out by haplotype analysis of durum lines in registration trials, which are subjected to rigorous milling assessment.

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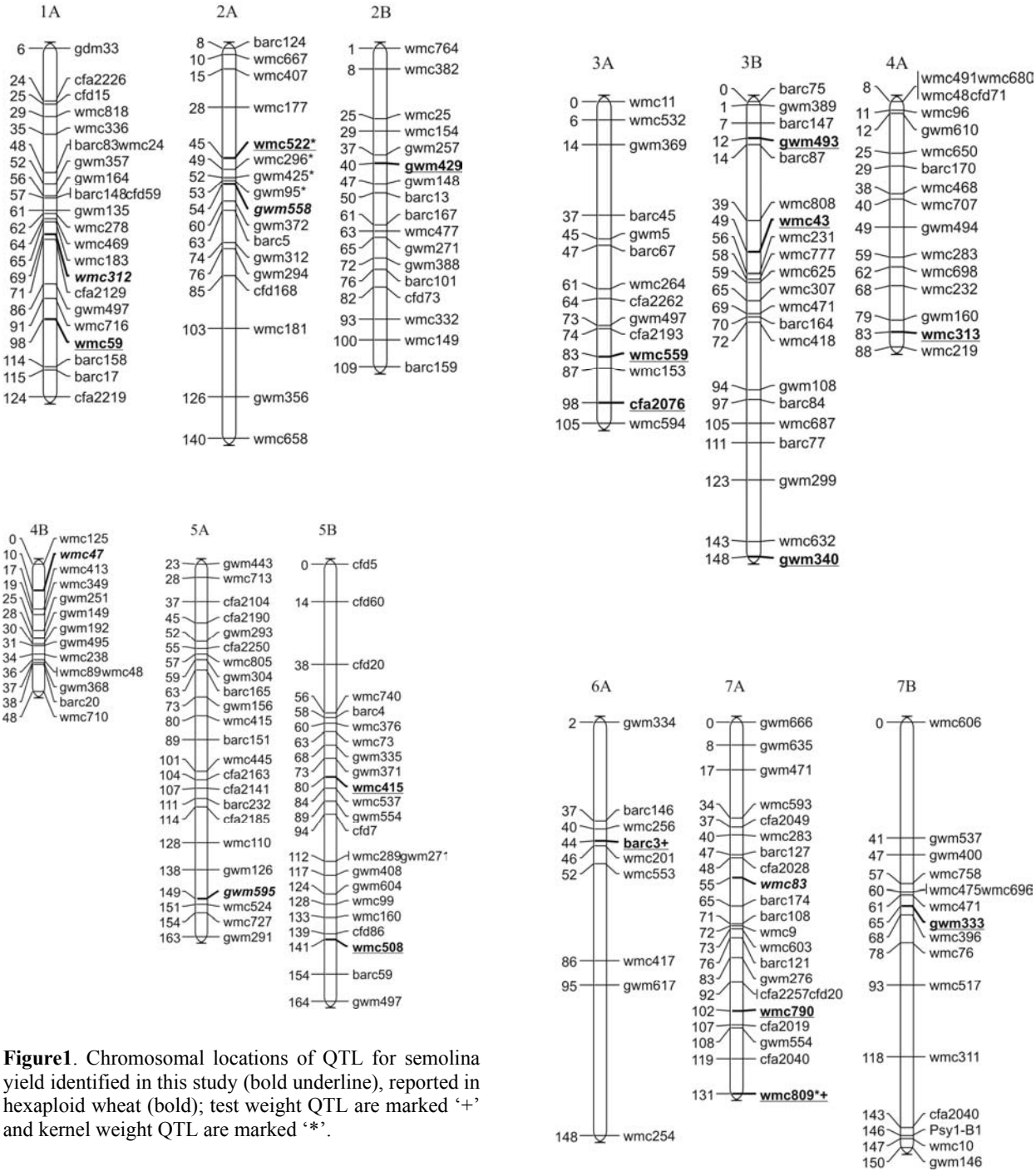

Figure1. Chromosomal locations of QTL for semolina yield identified in this study (bold underline), reported in hexaploid wheat (bold); test weight QTL are marked '+' and kernel weight QTL are marked '*'.