Polyphenol oxidase (PPO) catalyses the undesirable darkening of wheat products such as Asian noodles. Genetic variation for PPO activity is present in bread wheat and low PPO activity is a current target of Australian wheat breeding programs. An improved understanding of the genetic control of PPO activity in wheat backgrounds that are relevant to yellow alkaline noodle (YAN) wheat production in northern NSW and QLD would expedite this process. QTL (Quantitative Trait Loci) mapping of polyphenol oxidase (PPO) activity was investigated in a doubled haploid (DH) population that was derived from a hybrid between a low PPO line DM5637*B8 and high PPO Australian wheat variety H45. These trials were conducted at The University of Sydney Plant Breeding Institute, Narrabri, NSW during the 2005 and 2006 growing seasons. In both seasons PPO activity was significantly (P<0.001) correlated with changes in brightness (∆L*) and yellowness (∆b*) of YAN. QTLs on chromosome 2A (QPPO.dmh45-2AL, Q∆L*.dmh45-2AL, Q∆b*.dmh45-2AL) were associated with the characters at a high level of significance (P>0.001) and had LRS values of 59.9, 47.7 and 53.8, respectively. In each case the identified QTL explained more than 50% of the phenotypic variation of the traits. A highly significant (P<0.001) association was identified between marker locus wPt-7024 (chromosome 2A), and PPO activity, ∆L* and ∆b*. This information may be applied to increase the efficiency of breeding efforts that aim to generate improved YAN wheat.

Keywords: polyphenol oxidase (PPO), doubled haploid (DH), QTL mapping, Asian noodle, YAN, colour

Introduction

Wheat flour noodles are an important part of the diet in many Asian countries. Noodles that are bright and have stable color are preferred by consumers. Those that are commonly sold fresh are sensitive to time dependent darkening which is undesirable (Baik et al., 1995; Hou and Kruk, 1998; Hatcher et al., 1999; Jimenez and Dubcovsky, 1999). Oxidative enzymes such as polyphenol oxidase (PPO), lipoxygenase (LOX), peroxidase and catalase cause time dependent darkening of wheat products (Feillet et al., 2000). PPO is a major contributor to browning of noodles, chapattis and other wheat products (Demek et al., 2001a). Polyphenol oxidase (PPO) catalyses the oxidation of phenolic compounds to quinones, which undergo further rearrangement, non enzymatic oxidation, and polymerization to produce dark or brown colored melanin (Whitaker and Lee, 1995; Demeke et al., 2001). Due to oxidation, this action causes discoloration of some wheat products including Asian noodles (Kruger et al., 1992; Baik et al., 1995, Fuerst et al., 2006), pasta (Simeone et al., 2002), pan breads and steam breads (Dexter et al., 1984). Cultivars with low PPO activity are
desirable for consumers and food manufacturers. The understanding of the influence of PPO activity on yellow alkaline noodle color is required by breeders that are developing wheat cultivars that may be used as an input of yellow alkaline noodle manufacture. The influence of PPO activity on YAN darkening is known but mapping approaches concerned with YAN color and color stability have rarely been used in Australian wheats.

Variation in PPO activity is reported between wheat cultivars in different locations (Baik et al., 1994; Park et al., 1997). The growing environment has an influence on PPO activity in wheat (Baik et al., 1994; Park 1997) and negatively impacts on the efficiency of selection for low PPO genotypes (Demeke et al., 2001). Furthermore, PPO activity is expressed in maternal tissue and it is difficult to select for low PPO activity early in the breeding cycle using standard substrate assays in segregating material. Because it might has the consequence of reducing population size. Identification of molecular markers that can select wheat lines with low PPO phenotypes will enhance the efficiency of plant breeding programmes that have low PPO activity as a target. Loci on the long arm of chromosomes 2AL (Raman et al., 2005; Sun et al., 2005; Zang et al., 2005; He et al., 2007) and 2DL (Zang et al., 2005; He et al., 2007) in hexaploid wheat have a major genetic effect on PPO activity in some genetic populations. Recently, He et al. (2007) found that a combination of markers (PPO33/PPO16) for chromosomes 2A and 2D loci was an efficient and reliable means of selecting low PPO genotypes. Hence, the present investigation aimed to identify the associations between PPO activity and yellow alkaline noodle color in a doubled haploid (DH) population derived from DM5637*B8 (low PPO line) x H45 (high PPO line). It also aimed to identify genomic regions that control PPO activity and color stability of YAN and molecular markers that will be useful to select wheat lines with high color stability of yellow alkaline noodles.

Materials and Methods

Plant Materials

A doubled haploid population (n = 187) that was generated from a hybrid between DM5637*B8 (AUS1408/Sunco), a line with low PPO activity, sprouting tolerance and low blackpoint incidence (Howes pers. comm.) and Australian cultivar H45 (Ciano67/2*Olympic/3/WW80/3*Anza/Kalyonson a/Bluebird) was used in this study. The doubled haploid population was developed using the wheat x maize system (Ahmed pers. comm.; Johnson et al., 2005). For the PPO experiments, plants were sown in 3m rows and a sub set of 48 lines and parents were grown in 2 m x 6 m plots for the yellow alkaline noodles (YAN) testing. A RCBD design with 2 replications was employed for both experiments. Trials were conducted at the Plant Breeding Institute, Narrabri during the 2005 and 2006 growing seasons.

PPO Assay

The grain PPO activity was determined by using tyrosine as substrate for the assay procedure as described by Bernier and Howes (1994) and McCaig et al. (1999) except that the incubation period was increased from 2.5 to 3.5 h and the optical density was determined at 450 nm by a Bench Mark Plus-Microplate Spectrophotometer (BIORAD laboratories 2000, Alfred Nobel Drive, Hercules, CA 94547). Twenty whole seeds for each line were incubated in 0.01 M disodium tyrosine solution with 0.2% Tween 20 at 37°C for 3.5 h, and, after leaving the seeds, the absorbance of the solution was measured at 450 nm.

Yellow Alkaline Noodles (YAN)

The yellow alkaline noodle evaluation experiments for the crop grown in 2005 and 2006 were conducted in Western Wheat Quality Laboratory (WWQL), Washington State University, USA and the Plant Breeding Institute, University of Sydney (PBIN), Narrabri, NSW, Australia, respectively.

Flour Milling

Grains of the lines were milled using a Quadrumat Junior Mill (Brabender® OHG DUISBURG, Germany). The flour extraction rate was adjusted to 60 percent. Moisture and protein contents were measured using Near Infrared Reflectance (NIR; Perten Instruments, Type 9100/01, Germany). The samples were conditioned to 15% moisture overnight by adding the
appropriate amount of water before milling. Flour and bran yield was recorded for each sample.

Yellow Alkaline Noodle (YAN) Sheet Preparation

At WWQL, the noodle sheet was produced as described by Morris et al. (2000). The stock solution was prepared by mixing 200 g NaCl with 50 g of Na₂CO₃ in 1000 mL of H₂O in a volumetric flask. Five mL of this stock solution containing 1 g of NaCl and 0.25 g of Na₂CO₃ was added to 50 g of flour in this test. At PBIN, the stock solution was prepared according to the method of Mares and Campbell (2001) by mixing 25 g NaCl, 15 g KCO₃ and 10 g Na₂CO₃ with 875 mL water. The yellow alkaline noodle sheets were prepared by adding 10 g of flour with 3.7 mL of alkaline noodles solution.

Yellow Alkaline Noodle (YAN) Sheet and Flour Color Measurement

Wheat noodle sheet color (CIE L*, a*, b* denotes lightness which is white-black, red-green, and yellow-blue scales, respectively) were measured with a chromameter (Model-300, Minolta Camera Co., Ltd., Osaka Japan) with a 50 mm diameter measuring tube using a white tile background. The average of three readings, that were taken after moving the head each time, was employed in analysis. Four color readings at 0, 2, 24 and 48 h (2 h reading was not taken at WWQL) were taken. Noodles sheets were stored in plastic bags at 21°C between color readings. The change in color values (CIE ΔL*, Δa*, and Δb*) was calculated by subtracting readings at 2, 24 and 48 h from those immediately taken after noodle sheets were made (time zero). Values from duplicate noodle sheets were used in all of the analyses.

DNA Isolation

Genomic DNA extractions were performed using a modified CTAB protocol according to Doyle et al. (1990). 2.5μg of restriction quality DNA of each DH genotype and the parents of the DM5637*B8 x H45 population were prepared. Genomic DNA was extracted from fresh leaf tissue (0.5 to 1g) from a bulk of four plants. Leaves from three weeks old plants were taken. Leaves that were dry, disease free and free from sticking soil particles were selected for DNA extraction. CTAB extraction buffer was prepared with 2% (w/v) CTAB (sigma), 1.4 M NaCl, 0.2% mM EDTA, 100 mM Tris-HCl, (pH 8.0).

DArT Marker Analyses

2.5 μg of restriction quality DNA of each DH genotype (n=92) and the parents of the population DM5637*B8 x H45 was sent to Triticarte, Canberra (Australia; http://www.triticarte.com.au) for whole genome profiling using Diversity Array Technology. The 92 lines that were the subject of this study were randomly selected from the population. The loci were scored as present (1) or absent (0) and the locus designations were as used by Triticarte Pty. Ltd. The P value reflects how well the two phases (present=1 vs absent=0) of the marker are separated in the sample and P is based on ANOVA which is an estimate of marker quality. Markers with P values 80, 77-80, and 75-77 are termed as extremely reliable, usually scored, and provide useful information, respectively (Wenzl et al., 2004; Huttner et al., 2006).

Linkage Map Construction and QTL Mapping

The initial linkage mapping was performed with Cartablanche software, version 1.5.0(111), Keygene Products B.V. Linkage groups were further reassessed and reconstructed with Map Manager (QTXb20). The assignment of markers in linkage groups for individual chromosome relied on the consensus map of the Triticarte Ltd. and ongoing construction of consensus map for the DArT assay (Akbari et al., 2006; Howes pers. comm.; http://www.triticarte.com.au)

Interval mapping was performed at P=0.01 for marker-trait association using Map Manager QTXb20 (Manly et al., 2001). Single marker regression and simple interval mapping tools were used for this purpose. The marker regression function (P=0.01) was performed to find single marker loci linked with the quantitative data. The analysis was carried out for each of the two individual environments/years and for the pooled environments. The LRS thresholds in the regression analyses P<0.01 and P<0.001 were used for declaring significant and highly significant levels, respectively. The Map Manager QTXb20 (Manly et al., 2001) and MapChart version 2.2 (Voorrips 2002) were used to generate QTL map and chromosome map figures for presentation.
Results

PPO Assay

The PPO activities determined for each of the lines were consistent across years and were significantly correlated ($r^2=0.38$, $P<0.001$; Figure 1). The low PPO parent, DM5637*B8 had consistently low PPO activities in 2005 and 2006 ($A_{450}$ of 0.229 and 0.274, respectively). The PPO activity of H45 was high in both seasons ($A_{450}$ of 0.400 and 0.559, respectively). The PPO activity ($A_{450}$) of lines ranged between 0.144 to 0.603 with a population mean of 0.314 in 2005. In 2006, the $A_{450}$ ranged from 0.164 to 0.698 and the mean for the population was 0.367. In both seasons the frequency distributions of PPO activity were near normal (data not shown). Transgressive segregation was evident in this population. However, sixty nine (37%) lines in the population possessed a significantly higher PPO activity than the low PPO parent DM5637*B8 across seasons (LSD$_{0.05}=0.137$).

Noodle Sheet Color Trait L*

The correlation between PPO activity ($A_{450}$) and YAN ($\Delta L^*$ [0-24h]) was significant $r^2=0.52$ and 0.78 ($P<0.001$) in 2005 and 2006, respectively (Figure 2). Results of both years revealed that YAN brightness ($L^*$) at 24 hours was clearly influenced by PPO activity (i.e. lower PPO activity = YAN brightness) of the genotypes.

Noodle Sheet Color Trait b*

The $\Delta b^*$ values were significantly correlated ($r^2=0.53$ and 0.67, $P<0.001$) with PPO activity in 2005 and 2006 (Figure 3). Results indicate that low PPO activity is associated with elevated $\Delta b^*$ in this population.

Figure 1 Relationship between PPO activity of lines from DM5637*B8 x H45 DH population in 2005 and 2006 seasons.

Figure 2 Relationship between PPO activities and YAN $\Delta L^*$ (0-24h) in a set of lines from the DM5637*B8 x H45 DH population in (a) 2005 and (b) 2006 (n=50).
Figure 3. Relationship between PPO activity and YAN ∆b* (0-24h) in a set of lines derived from the DM5637*B8 x H45 DH population in (a) 2005 and (b) 2006 (n=50).

DArT Marker Screening

The DNA of 92 lines and the two parents of the DM5637*B8 x H45 population were analysed by DArT. Three hundred and eighty four polymorphic DArT markers were obtained with an overall call rate of 94.28%. The overall marker P values were extremely reliable and ranged between 73.87 and 98.84 with a mean of 89.63. Only 17 markers had P values below 77. Of the 384 polymorphic markers that were identified 317 markers were assembled into 30 linkage groups using Cartablanche software, version 1.5.0(111), Keygene Products B.V. and QTXb20. The remaining markers were unassigned.

QTL Analyses

Polyphenol oxidase activity

Interval mapping analysis identified three genomic regions that controlled PPO activity in the 2005 trial (Table 1). A highly significant (P<0.001) QTL (QPPO.dmh45-2AL) was located on chromosome 2AL (LRS=37.2, Figure 4) and explained 35% of phenotypic variation. The marker loci wPt-5865 and wPt-7024 in the 2AL QTL (QPPO.dmh45-2AL) were both significantly associated with PPO activity in this season. In 2005, significant QTLs for PPO activity were also identified on chromosomes 4A (QPPO.dmh45-4A) and 7B (QPPO.dmh45-7B). The three QTLs that were identified collectively explained 53% of the observed phenotypic variation in this season (Table 1).

Three QTLs were detected in 2006, together accounting for 70% of variation in PPO activity (Table 1). A highly significant (P<0.001) QTL on chromosome 2AL was also associated with PPO activity in 2006. Marker locus wPt-7024, which was located in the 2A QTL (QPPO.dmh45-2A) had a highly significant association with PPO activity (LRS = 59.9; P>0.001) and accounted for 50% of the observed variation. The QTL on 7BL (QPPO.dmh45-7B) explained 10% of the phenotypic variation (LRS value of 9.5) and corresponded with a QTL identified in 2005. In the 2006 growing season, another QTL (QPPO.dmh45-7A) was identified on chromosome 7A. It was not detected in 2005. Marker locus wPt-0004 within this QTL was closely associated with PPO activity. This marker had an LRS value of 8.9 (P>0.01) and explained 10% of the variation in PPO activity in 2006. The 4A QTL (QPPO.dmh45-7A) that was identified in 2005 was not detected in 2006.

The three QTLs that were identified in 2006 (QPPO.dmh45-2AL; QPPO.dmh45-7A and QPPO.dmh45-7B) were also detected when analysing pooled data across both seasons (Table 1). The three QTLs together accounted for 71% of the variation in PPO activity. The association of PPO activity with markers wPt-5865 and wPt-7024 in the 2AL QTL was highly significant (P<0.001; LRS of 63.7) and contributed 52% of the observed phenotypic variation. A suggestive QTL that was
Table 1 Results of interval mapping for PPO activities (A_{450}) in the DM5637*B8 x H45 DH population.

<table>
<thead>
<tr>
<th>Site-year</th>
<th>Chromosome location</th>
<th>QTLs</th>
<th>Closest marker</th>
<th>LRS value(^1)</th>
<th>PVE (%)(^2)</th>
<th>Parent (+ve PPO)</th>
</tr>
</thead>
<tbody>
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<td>QPPO.dmh45-2AL</td>
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<td>H45</td>
</tr>
<tr>
<td></td>
<td>4A</td>
<td>QPPO.dmh45-4A</td>
<td>wPt-7919</td>
<td>8.9*</td>
<td>10</td>
<td>H45</td>
</tr>
<tr>
<td></td>
<td>7B</td>
<td>QPPO.dmh45-7B</td>
<td>wPt-9925</td>
<td>7.1*</td>
<td>8</td>
<td>H45</td>
</tr>
<tr>
<td>Narrabri-2006</td>
<td>2AL</td>
<td>QPPO.dmh45-2AL</td>
<td>wPt-7024</td>
<td>59.9**</td>
<td>50</td>
<td>H45</td>
</tr>
<tr>
<td></td>
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<td>8.9*</td>
<td>10</td>
<td>H45</td>
</tr>
<tr>
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<td>7B</td>
<td>QPPO.dmh45-7B</td>
<td>wPt-9925</td>
<td>9.5*</td>
<td>10</td>
<td>H45</td>
</tr>
<tr>
<td>Pooled</td>
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<td>wPt-7024</td>
<td>63.7**</td>
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<td>H45</td>
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<td>8</td>
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<td>wPt-9925</td>
<td>10.2*</td>
<td>11</td>
<td>H45</td>
</tr>
</tbody>
</table>

\(^{1}\) Likelihood statistic ratio; * and ** indicate significant and highly significant level at P<0.01 and P<0.001, respectively. Non asterisk LRS values indicate the suggestive level (P>0.05).

\(^{2}\) Phenotypic variation explained.

Figure 4 Likelihood Ratio Statistic (LRS) and additive effect plots of the QILs on chromosome 2AL for PPO activity, stability of YAN brightness and yellowness, respectively. LRS scores and additive effect curves (solid and broken respectively) are shown on the partial maps of chromosome. Vertical lines (green) from left to right indicate suggestive, significant and highly significant levels of LRS values, respectively.

identified on chromosome 7A (QPPO.dmh45-7A) (LRS=7.4) explained 8% of phenotypic variation. Analysis of pooled data revealed that the 7B QTL was significantly associated (P<0.01) with PPO activity (LRS=10.2) and explained 11% of phenotype variation. In these QTLs the closest identified markers were consistently identified across seasons. Interval mapping analysis revealed that H45 contributed alleles of the three QTLs that conferred elevated PPO activity.

Yellow Alkaline Noodle Color Components

Interval mapping analysis identified four genomic regions that controlled ΔL* for YAN in 2005 (Table 2). The major QTL was located on chromosome 2AL (LRS=48.5) and explained 64% of phenotypic variation. The QTL (QΔL*.dmh45-1AS) on chromosome 1AS explained 27% of phenotypic variation (LRS=13.1). QTL on chromosome 1B (QΔL*.dmh45-1B) and 7AL (QΔL*.dmh45-7A) were also detected. These QTL explained 14% (LRS=7) and 20% (LRS=10.7), of phenotypic variation, respectively.

In 2006, QΔL*.dmh45-2AL was the only QTL detected (LRS value of 30.2) and it explained 47% of variation. Analysis of pooled data detected all of the four QTLs that were identified in 2005 (Table 2). Each was of a similar significance level to that
Table 2 Results of interval mapping of yellow alkaline noodle (YAN) color stability CIE \( \Delta L* \) and \( \Delta b* \) at 0-24 hours in the DM5637*B8 x H45 DH population.

<table>
<thead>
<tr>
<th>Site-year</th>
<th>Chromosome location</th>
<th>QTLs</th>
<th>Closest marker</th>
<th>LRS value(^1)</th>
<th>PVE (%)(^2)</th>
<th>Contribution parent</th>
</tr>
</thead>
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<tr>
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<td></td>
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</tr>
<tr>
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<td>H45</td>
</tr>
<tr>
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<td>10.7*</td>
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<td>H45</td>
</tr>
<tr>
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<td>H45</td>
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</table>

\(^1\) Likelihood statistic ratio; \* and \** indicate significant and highly significant level at \( P<0.01 \) and \( P<0.001 \), respectively. Non asterisk LRS values indicate the suggestive level (\( P>0.05 \)).

\(^2\) Phenotypic variation explained.

for year 2005. Interval mapping analysis revealed a positive additive effect of the QAL*.dmh45-1B and the major QTL (QAL*.dmh45-2A) on chromosome 2AL which indicated that parent DM5637*B8 contributed a positive allele at each of these QTLs. The additive effect for the other QTLs (QAL*.dmh45-1AS and QAL*.dmh45-7AL) were negative in both growing seasons, indicating that parent H45 contributed the positive allele for the QTLs.

QTL for YAN trait \( \Delta b^* \) were detected on chromosomes 1AS (QA\(b^*\).dmh45-1AS), 2AL (QA\(b^*\).dmh45-2AL) and 7AL (QA\(b^*\).dmh45-7AL) were detected (Table 2). QA\(b^*\).dmh45-1AS was detected only in 2005 and explained 17% of observed variation in that season (LRS=7.6). The QTL on 2AL (Figure 4) and 7AL were detected in both years. The 2AL was highly significant (\( P<0.001 \)) in both years, accounting for 63% and 57% of variation in \( \Delta b^* \) (with LRS values of 47.1 and 39.2 in 2005 and 2006, respectively, Figure 4).

This QTL had a positive additive effect which was significantly associated with brightness stability of yellow alkaline noodles that was contributed by the parent DM5637*B8. Regression analysis identified that marker wPt-7024 was significantly (\( P<0.001 \)) associated with \( \Delta b^* \). The other significant QTL on chromosome arm 7AL was detected across the growing seasons and accounted for 22% and 17% of phenotypic variation (LRS=12.1 and 8.9), in 2005 and 2006, respectively. This QTL explained 23% of variation across seasons. The marker wPt-0004 was closely linked to locus controlling this QTL. The negative additive effect recorded for QTLs located on chromosomes 1AS and 7AL indicate that the high PPO parent (H45) contributed the positive allele for these QTLs. In pooled environments, the three QTLs (QA\(b^*\).dmh45-1AS, QA\(b^*\).dmh45-2AL and QA\(b^*\).dmh45-7AL) were significant (\( P<0.01 \)) explaining 17%, 68% and 23% of phenotypic variation, respectively.
Discussion

Both ΔL* and Δb* were significantly (P<0.01) correlated with PPO activity across growing seasons. The findings of this study are in agreement with the reports of the role of PPO on time dependent darkening (darkening due to aging) on yellow alkaline noodles (Kruger et al., 1994; Morris 1995; Fuerst et al., 2006).

The results of the current study reconfirm that PPO activity in wheat is mainly controlled by a locus on the long arm of chromosome 2A with strong genetic association with DArT marker wPt-7024. Other studies have identified that PPO activity is strongly associated with the SSR marker loci Xgwm312, Xgwm294 and WMC170 which are located on chromosome 2A (Sun et al., 2005; Raman et al., 2005; Watanabe et al., 2006). The chromosome 2A location is consistent with previous findings (Sun et al., 2005; Raman et al., 2007) however it is not certain if the identified QTLs are in precisely the same location as identified in previous studies. The association of DArT marker wPt-9925 on chromosome 7BL with PPO activity in wheat has not yet been reported in the literature. This QTL (QPPO.dmh45-2AL) explained 8-11% phenotypic variation across environments in consecutive years. QTLs on chromosomes 4A and 7A were detected in one season (Narrabri 2005 and 2006, respectively). The QTL on 7A, was also detected when data was pooled across environments. A QTL on chromosome 7A was also detected in other studies involving hexaploid wheat that were grown at the same site (Sadeque 2008). Therefore, it is possible that the minor QTL that is located on chromosome 7A is sensitive to environmental influence. The QTL QPPO.dmh45-4A explained 10% of variation in PPO activity in one season (Narrabri 2005). There are no published reports of the presence of QTL on chromosome 4A, thus the existence of QTL on chromosome 4A needs to be confirmed. The analyses of allelic effects of the QTLs revealed that selection using markers within QPPO.dmh45-2AL may be an efficient means to select low PPO activity genotypes in this DH population.

In the present study, QTL mapping of stability of YAN color components were also performed using DArT markers. A large amount of phenotypic variation was explained by the QTLs for changes in brightness, QΔL*.dmh45-2AL, and yellowness, QΔb*.dmh45-2AL, (64% and 68% respectively) on chromosome 2A. Low PPO wheat line DM5637*B8 exhibited color stability in both years. Not many studies concerned with mapping of color stability of YAN in Australian wheat have been conducted and little information is available. Mares and Campbell (2001) found a QTL on chromosome 2D that was clearly associated with these traits and another on 2A which had some association. These findings are partly in agreement with findings of the current study. The marker locus wPt-7024 was identified as an important DArT marker for all of the color stability traits of YAN, and PPO activity.

Correlations between PPO activity and change in brightness were observed in this population. QTL mapping detected that DArT marker locus wPt-7024 on chromosome 2AL had the closest genetic association with PPO activity and color stability of both ΔL* and Δb* (52% and 68% PVE, respectively). QTL mapping results identified common locations of QTLs for both characters. Also the parental contribution was consistent in each case. The DArT marker locus wPt-7024 had the highest contribution to YAN color components ΔL* and Δb*. Therefore, it can be concluded that PPO activity is the major factor that influences YAN color stability. DArT marker wPt-7024 was associated with both YAN brightness and yellowness along with PPO activity. In addition, two potential QTLs for YAN ΔL* and Δb* have been identified on chromosomes 1AS and 7AL. The QTL on 7AL was detected across environments. A QTL on chromosome 7A was also identified for PPO activity strengthening the hypothesis that it is an important determinant of the change in YAN brightness. It is interesting that a locus on 7B for PPO activity was identified in both seasons but a corresponding QTL for change in brightness was not detected in this analysis.

The identification and mapping of PPO genes in wheat is an important prerequisite for developing low PPO wheat cultivars that exhibit high levels of color stability in wheat end products, especially for yellow alkaline and white salted noodles. This study confirms that PPO activity in wheat is under polygenic control. The DArT marker wPt-7024 that
is located within a QTL on chromosome 2AL and has a high level of significance with PPO activity (P<0.001). In this study a new QTL location on chromosome 7B was identified. A possible QTL location on chromosome 7A was also revealed in this study. The identification of these QTLs and markers associated with PPO activity in this study will enable application of marker assisted selection in plant breeding programs of Eastern Australia which is an efficient approach to pyramid genes for low PPO activity. The results of the current study confirmed earlier findings about the involvement of some genomic regions in controlling the traits investigated. In addition, new genomic regions and associated DArT markers were identified that were linked to loci that control wheat PPO and YAN color stability components.

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