Localization of Quantitative Trait Loci for Yield Components in a Cross Oriental × Occidental Barley Cultivar (*Hordeum vulgare* L.)

Mohammad SAMERI and Takao KOMATSUDA*

Plant Genome Research Unit, National Institute of Agrobiological Sciences (NIAS) (Tsukuba, Ibaraki 305–8602, Japan)

Abstract

Ninety-nine recombinant inbred lines (RILs) were grown for three years and yield components were evaluated. Composite interval mapping showed that the *vrs1* and *uzu* genes consistently controlled grain weight per plant. The analyses also identified a new QTL of kernel length on chromosome 3H, a QTL for kernel width on chromosome 6H and a QTL for thousand-kernel weight on chromosome 2H. The QTL generated in this study may be a useful target for marker-assisted selection in breeding programs.

Disciplines: Biotechnology / Plant breeding

Additional key words: composite interval mapping, dense spike (*dsp1*), six-rowed spike (*vrs1*), *uzu* or semibrachytic (*uzu1*)

Introduction

Oriental and occidental barley cultivars are genetically divergent from each other and they harbor genes characteristic of each region. We developed recombinant inbred lines (RILs) from a cross between ‘Azumamugi’ and ‘Kanto Nakate Gold’. Azumamugi was bred from a cross between two Japanese landraces (‘Tochigi Sekitori 1’ and ‘Torano-o Sai 1’). Kanto Nakate Gold was bred by the following pedigree: Golden Melon/Shikoku//Kinki Shu, where Golden Melon is a two-rowed erectum-type cultivar (origin Northern Europe), Shikoku is a Japanese landrace, and Kinki Shu is a selection from Golden Melon. Because Azumamugi and Kanto Nakate Gold belong to oriental and occidental cultivars respectively, the two genotypes are highly polymorphic for morphological, physiological, and molecular markers.

The objective of the present study is to identify QTLs for yield components, especially grain size in barley. We estimated genetic effects of known major genes and considered to detect new genes even with minor effects on these traits.

Materials and methods

Ninety-nine RILs (F12 and F13) were developed from *F2* plants of Azumamugi × Kanto Nakate Gold by single seed descent. Azumamugi is six-rowed (*vrs1.a*), winter type (*sgh1/sgh2*) and *uzu* or semi-brachytic (*uzu1*). Kanto Nakate Gold is two-rowed (*Vrs1.b*), spring type (*sgh1/Sgh2*) and non-uzu (*Uzu1*). Plants were sown in a field of NIAS, Tsukuba, in October 2000, 2001 and 2002, and grown until maturity in summer of 2001, 2002 and 2003 (thereafter referred to as ‘Year 2001’, ‘Year 2002’ and ‘Year 2003’, respectively). Each RIL was represented by a row of ten seeds and spaced 20 cm apart. Rows were spaced 80 cm apart. Four central plants of each RIL were harvested as a bundle of single plants, air-dried for one week at 30°C, and used for measuring quantitative traits. For seed sizes, 10 seeds per RIL were measured using a projector Movias NAC 5200 (Image Technical Center, Yokohama, Japan).

The heritabilities (h^2) were estimated by the formula: h^2 = \delta_g^2 / (\delta_g^2 + \delta_e^2 / r), where \delta_g^2 = (MS_{genotype} - MS_{error}) / r, \delta_e^2 = MS_{error} and r is number of replications. The correlation coefficients were computed using the computer program SPSS (Version 10.01, SPSS Corp., Chicago, IL).

Molecular linkage map and base map of these
RILs were constructed previously. Map distances were estimated by using Kosambi’s map function. Composite interval mapping (CIM) was performed using the computer program QTL Cartographer version 1.14. A LOD score threshold of 3.0 was used to identify QTLs. The position of detected QTLs were compared with the ones of former QTL studies consulting integrated maps.

Results and discussion

RILs showed a wide range of variation for all the traits (Table 1). ANOVA showed that genetic variances for all the traits were significant, and heritability ranged from 0.73 to 0.99 (Table 2). Correlation coefficients between two years ranged from 0.61 to 0.98, which were significant for all the traits (Table 3). Grain number per spike (GNS) and grain number per plant (GNP) were positively correlated with grain weight per plant (GWT), but negatively correlated with thousand-kernel weight (KWT) (Table 3).

GWT was controlled predominantly by *vrs1* and *uzu* as detected previously. The allele of Azumamugi at the *vrs1* locus increased GWT by 9.15–11.75 g (Table 4). KWT of six-rowed lines was 74% of two-rowed lines, but GNP of six-rowed lines was 2.7 times of two-rowed lines, therefore GWT of six-rowed lines was 1.5 times of two-rowed lines, showing six-rowed lines have a potential of higher yield than two-rowed lines (Fig. 1). The allele of Azumamugi at the *uzu* locus decreased GWT by 9.9–10 g (Table 4). However, the *uzu* locus did not control the GNP. Another QTL for GWT was detected on chromo-

### Table 1. Means, coefficient of variations and standard deviations for agronomic traits observed in azumamugi, Kanto Nakate Gold and their 99 RILs

<table>
<thead>
<tr>
<th>Traits</th>
<th>GWT (g)</th>
<th>STW (g)</th>
<th>HI (%)</th>
<th>KLN (mm)</th>
<th>KWD (mm)</th>
<th>KS</th>
<th>KA (mm²)</th>
<th>GNS</th>
<th>GNP</th>
<th>KWT (g)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Year 2002</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Azumamugi</td>
<td>61.3</td>
<td>52.3</td>
<td>53.9</td>
<td>7.5</td>
<td>3.4</td>
<td>2.3</td>
<td>25.1</td>
<td>92.1</td>
<td>1,953</td>
<td>35.2</td>
</tr>
<tr>
<td>Kanto Nakate Gold</td>
<td>80.0</td>
<td>106.3</td>
<td>43.0</td>
<td>10.0</td>
<td>3.8</td>
<td>2.7</td>
<td>37.3</td>
<td>39.0</td>
<td>1,278</td>
<td>53.0</td>
</tr>
<tr>
<td>RIL – mean</td>
<td>54.9</td>
<td>70.9</td>
<td>43.1</td>
<td>8.6</td>
<td>3.5</td>
<td>2.5</td>
<td>29.9</td>
<td>66.4</td>
<td>1,864</td>
<td>38.5</td>
</tr>
<tr>
<td>RIL – SD</td>
<td>22.5</td>
<td>24.6</td>
<td>9.5</td>
<td>0.7</td>
<td>0.3</td>
<td>0.3</td>
<td>3.5</td>
<td>30.2</td>
<td>905</td>
<td>7.3</td>
</tr>
<tr>
<td>RIL – minimum</td>
<td>12.5</td>
<td>25.0</td>
<td>18.8</td>
<td>7.1</td>
<td>3.0</td>
<td>1.8</td>
<td>22.9</td>
<td>25.8</td>
<td>399</td>
<td>24.4</td>
</tr>
<tr>
<td>RIL – maximum</td>
<td>117.5</td>
<td>145.0</td>
<td>62.5</td>
<td>10.4</td>
<td>4.2</td>
<td>3.5</td>
<td>37.4</td>
<td>108.0</td>
<td>3,933</td>
<td>54.4</td>
</tr>
<tr>
<td>RIL – CV</td>
<td>41.0</td>
<td>34.6</td>
<td>22.1</td>
<td>8.5</td>
<td>7.6</td>
<td>11.1</td>
<td>6.6</td>
<td>45.5</td>
<td>48.6</td>
<td>18.9</td>
</tr>
<tr>
<td><strong>Year 2003</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Azumamugi</td>
<td>50.0</td>
<td>25.0</td>
<td>66.7</td>
<td>7.6</td>
<td>3.3</td>
<td>2.3</td>
<td>25.2</td>
<td>96.0</td>
<td>3,192</td>
<td>34.0</td>
</tr>
<tr>
<td>Kanto Nakate Gold</td>
<td>47.5</td>
<td>46.3</td>
<td>50.7</td>
<td>9.9</td>
<td>3.7</td>
<td>2.7</td>
<td>36.6</td>
<td>36.0</td>
<td>1,593</td>
<td>49.6</td>
</tr>
<tr>
<td>RIL – mean</td>
<td>43.1</td>
<td>57.4</td>
<td>42.3</td>
<td>8.2</td>
<td>3.6</td>
<td>2.3</td>
<td>29.6</td>
<td>62.1</td>
<td>1,759</td>
<td>33.4</td>
</tr>
<tr>
<td>RIL – SD</td>
<td>17.6</td>
<td>17.6</td>
<td>10.8</td>
<td>0.7</td>
<td>0.2</td>
<td>0.2</td>
<td>3.6</td>
<td>27.5</td>
<td>826</td>
<td>6.9</td>
</tr>
<tr>
<td>RIL – minimum</td>
<td>6.3</td>
<td>20.0</td>
<td>10.4</td>
<td>6.8</td>
<td>3.0</td>
<td>1.8</td>
<td>22.4</td>
<td>21.0</td>
<td>578</td>
<td>18.0</td>
</tr>
<tr>
<td>RIL – maximum</td>
<td>87.5</td>
<td>127.5</td>
<td>61.9</td>
<td>9.9</td>
<td>4.2</td>
<td>3.4</td>
<td>37.2</td>
<td>105.0</td>
<td>3,600</td>
<td>47.6</td>
</tr>
<tr>
<td>RIL – CV</td>
<td>40.9</td>
<td>30.6</td>
<td>25.5</td>
<td>8.6</td>
<td>6.7</td>
<td>10.5</td>
<td>12.0</td>
<td>44.3</td>
<td>47.0</td>
<td>20.8</td>
</tr>
</tbody>
</table>

2): Coefficient of Variation (%).
3): Data in italic were scored in the Year 2001.

### Table 2. Analysis of variance for agronomic traits in the RILs from the cross of Azumamugi × Kanto Nakate Gold

<table>
<thead>
<tr>
<th>Source</th>
<th>df</th>
<th>GWT</th>
<th>STW</th>
<th>HI</th>
<th>KLN</th>
<th>KWD</th>
<th>KS</th>
<th>KA</th>
<th>GNS</th>
<th>GNP</th>
<th>KWT</th>
</tr>
</thead>
<tbody>
<tr>
<td>Genotype</td>
<td>98</td>
<td>693.67***</td>
<td>721.08***</td>
<td>178.19***</td>
<td>0.93***</td>
<td>0.11***</td>
<td>0.12**</td>
<td>21.04**</td>
<td>1,647.30**</td>
<td>1,406,931.00**</td>
<td>95.70**</td>
</tr>
<tr>
<td>Year</td>
<td>1</td>
<td>6,913.64***</td>
<td>9,102.58***</td>
<td>35.33ns</td>
<td>7.18**</td>
<td>0.68**</td>
<td>1.53**</td>
<td>5.33ns</td>
<td>932.75**</td>
<td>540,248.70*</td>
<td>1,311.57**</td>
</tr>
<tr>
<td>Error</td>
<td>98</td>
<td>124.40</td>
<td>191.11</td>
<td>29.00</td>
<td>0.10</td>
<td>0.02</td>
<td>0.01</td>
<td>3.74</td>
<td>20.98</td>
<td>95,717.71</td>
<td>5.57</td>
</tr>
</tbody>
</table>

Heritability | 0.82 | 0.73 | 0.84 | 0.89 | 0.82 | 0.90 | 0.82 | 0.99 | 0.93 | 0.94 |

*, **: significant at the 5% and 1% level respectively.
ns: not significant.
The major factor of GNP, GNS and KWT was the vrs1 locus (Table 4). Six-rowed lines decrease KWT but considerably increase GNP, thus six-rowed spikes increase the yield of grains (Fig. 1). The vrs1 locus contributed 25 to 80% of phenotypic variance for KWT in separate studies and 63–72% in this study. In spite of the major effect by the vrs1 locus on KWT, we detected a new QTL for KWT named qKWT.ak-2H on the short arm of chromosome 2H (Table 4). The allele of Azumamugi at this QTL increased KWT.

The vrs1, uzu and dsp1 have been reported to control the kernel sizes and our result was in agreement with these reports. Behind the pronounced effects of these major genes, we detected a new QTL for kernel length (KLN) named qKLN.ak-3H on chromosome 3HS and a new QTL for kernel width (KWD) named qKWD.ak-6H on chromosome 6H (Table 4). The QTL of KWD detected on chromosome 7H can be identical with qDHE.ak-7H, which controlled days to heading.

Kernel shape (KS = KLN/KWD) was controlled by vrs1, uzu and dsp1, but kernel area (KA = KLN × KWD) was controlled by vrs1 and uzu.

QTL for straw weight per plant (STW) was detected on chromosome 3HL, at the uzu locus, and the allele of Azumamugi decreased STW by 6–11.8 g (Table 4). Another QTL for STW was detected on chromosome 3HL (may coincide with denso), together with Ppd-H1 (Eam1) and Sgh2 (spring growth habit), which primarily controlled days to heading. The QTL of harvest index (HI = 100 × GWT/(GWT + STW)) detected on chromosome 5HL could correspond to qTPN.ak-5H, which was a QTL for triplet number.

Our study detected new QTLs for controlling seed size. These QTLs are useful for modifying spike architecture moderately rather than the major genes of strong pleiotrophic effects. The QTLs may be introduced to cultivars.
Table 4. QTLs of yield components detected in the RILs of Azumamugi × Kanto Nakate Gold

<table>
<thead>
<tr>
<th>Trait</th>
<th>Chr.</th>
<th>Flanking markers</th>
<th>2002</th>
<th>2003</th>
<th>Locus</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Pos. cM</td>
<td>LOD</td>
<td>AE</td>
</tr>
<tr>
<td>GWT (g)</td>
<td>2HL</td>
<td>MWG801 - vrs1</td>
<td>77.56</td>
<td>9.80</td>
<td>11.75</td>
</tr>
<tr>
<td></td>
<td>3HL</td>
<td>uzu - e06m30.10.1</td>
<td>65.53</td>
<td>7.10</td>
<td>-10.00</td>
</tr>
<tr>
<td></td>
<td>3HL</td>
<td>MWG973 - e06m30.8.3</td>
<td>111.39</td>
<td>3.88</td>
<td>7.33</td>
</tr>
<tr>
<td></td>
<td>4HL</td>
<td>vrs1 - MWG503</td>
<td>79.56</td>
<td>36.14</td>
<td>789.63</td>
</tr>
<tr>
<td></td>
<td>5HL</td>
<td>e07m25.3 - e12m19.9.2</td>
<td>116.56</td>
<td>3.10</td>
<td>150.54</td>
</tr>
<tr>
<td>GNP</td>
<td>2HL</td>
<td>vrs1 - MWG503</td>
<td>79.56</td>
<td>70.47</td>
<td>29.99</td>
</tr>
<tr>
<td></td>
<td>2HL</td>
<td>e13m31.7 - e15m19.8.1</td>
<td>57.49</td>
<td>4.09</td>
<td>1.59</td>
</tr>
<tr>
<td></td>
<td>2HS</td>
<td>vrs1 - MWG503</td>
<td>79.56</td>
<td>32.88</td>
<td>-6.57</td>
</tr>
<tr>
<td>KWT (g)</td>
<td>2HL</td>
<td>vrs1 - MWG503</td>
<td>79.56</td>
<td>70.47</td>
<td>29.99</td>
</tr>
<tr>
<td></td>
<td>5HL</td>
<td>e07m25.3 - e12m19.9.1</td>
<td>112.13</td>
<td>4.42</td>
<td>2.02</td>
</tr>
<tr>
<td>KLN (mm)</td>
<td>2HL</td>
<td>ND</td>
<td>65.53</td>
<td>17.96</td>
<td>-0.52</td>
</tr>
<tr>
<td></td>
<td>3HL</td>
<td>uzu - e06m30.10.1</td>
<td>65.53</td>
<td>17.96</td>
<td>-0.52</td>
</tr>
<tr>
<td></td>
<td>7HL</td>
<td>cMWG704 - e11m17.10.2</td>
<td>86.02</td>
<td>7.96</td>
<td>-0.30</td>
</tr>
<tr>
<td>KWD (mm)</td>
<td>2HL</td>
<td>vrs1 - MWG503</td>
<td>79.56</td>
<td>20.08</td>
<td>-0.20</td>
</tr>
<tr>
<td></td>
<td>6H</td>
<td>e11m18.12 - e15m19.9.2</td>
<td>35.78</td>
<td>4.52</td>
<td>0.08</td>
</tr>
<tr>
<td></td>
<td>7HS</td>
<td>ND</td>
<td>65.53</td>
<td>17.96</td>
<td>-0.52</td>
</tr>
<tr>
<td></td>
<td>7H</td>
<td>e11m17.10.2 - e12m19.10.3</td>
<td>97.54</td>
<td>3.90</td>
<td>0.08</td>
</tr>
<tr>
<td>KS</td>
<td>2HL</td>
<td>vrs1 - MWG503</td>
<td>79.56</td>
<td>7.56</td>
<td>0.11</td>
</tr>
<tr>
<td></td>
<td>3HS</td>
<td>ND</td>
<td>65.53</td>
<td>17.96</td>
<td>-0.52</td>
</tr>
<tr>
<td></td>
<td>3HL</td>
<td>uzu - e06m30.10.1</td>
<td>65.53</td>
<td>17.96</td>
<td>-0.52</td>
</tr>
<tr>
<td></td>
<td>6H</td>
<td>ABG458.1 - e11m18.12</td>
<td>34.70</td>
<td>3.86</td>
<td>-0.08</td>
</tr>
<tr>
<td></td>
<td>7HS</td>
<td>cMWG704 - e11m17.10.2</td>
<td>86.02</td>
<td>12.08</td>
<td>-0.15</td>
</tr>
<tr>
<td>KA (mm²)</td>
<td>2HL</td>
<td>vrs1 - MWG503</td>
<td>79.56</td>
<td>13.23</td>
<td>-2.13</td>
</tr>
<tr>
<td></td>
<td>3HL</td>
<td>uzu - e06m30.10.1</td>
<td>67.53</td>
<td>7.13</td>
<td>-1.53</td>
</tr>
<tr>
<td></td>
<td>3HL</td>
<td>ABG602 - e1m32.3</td>
<td>47.29</td>
<td>5.14</td>
<td>-9.20</td>
</tr>
<tr>
<td></td>
<td>5HL</td>
<td>e1m26.9 - e07m25.3</td>
<td>115.39</td>
<td>5.02</td>
<td>9.43</td>
</tr>
<tr>
<td>STW (g)</td>
<td>2HS</td>
<td>e1m32.3 - e1m31.7.1</td>
<td>49.82</td>
<td>6.03</td>
<td>3.04</td>
</tr>
<tr>
<td></td>
<td>2HL</td>
<td>MWG8011 - vrs1</td>
<td>79.56</td>
<td>18.00</td>
<td>6.43</td>
</tr>
<tr>
<td></td>
<td>3HL</td>
<td>ND</td>
<td>65.53</td>
<td>17.96</td>
<td>-0.52</td>
</tr>
<tr>
<td></td>
<td>5HL</td>
<td>e14m27.44 - MWG2230</td>
<td>86.37</td>
<td>3.19</td>
<td>2.14</td>
</tr>
</tbody>
</table>

1): Additive effect of “Azumamugi” allele.
2): Proportion of phenotypic variance explained by the detected QTLs.
3): New QTLs identified in this study are in bold face.
4): Not detected.

M. Sameri & T. Komatsuda
GWT (g)  41 (3)  2007
tivars by means of marker-assisted selection in breeding programs.

Acknowledgments

We thank Kayoko Muromachi for her help in the field evaluations. Research grants from the Ministry of Agriculture, Forestry and Fisheries of Japan (MAFF) and Japan Science and Technology Corporation (JST) are gratefully appreciated.

References