

SCAR and CAPS Markers Linked to Quality Traits (Oil, β -glucan, and Protein Content) in Oat

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Introduction

To improve grain quality for different markets, oat breeders are faced with the challenge of selecting varieties with high and low content of oil, β -glucan, protein and other chemical traits. Each of these traits is controlled by multiple genetic loci.

We report on the development of PCR-based Sequence Characterized Amplified Region (SCAR) and Cleaved Amplified Polymorphic Sequence (CAPS) molecular markers in oat for application in genetic studies and marker assisted selection (MAS) breeding (Rines et al. 2006). A set of 8 markers were designed to target oil content (Orr and Molnar 2007) and a set of 15 markers were designed to target β -glucan and protein content (Orr and Molnar 2008). These more robust markers were developed from Random Amplified Polymorphic DNA (RAPD) markers mapped in the Kanota x Ogle (KO) (Wight et al. 2003) or the Terra x Marion (TM) (De Koeyer et al. 2004) recombinant inbred line (RIL) populations and associated with QTLs for grain quality traits.

Materials and methods

The diagnostic band of each RAPD marker (10 bp RAPD primer) was isolated from an agarose gel and sequenced. For development of the derived SCAR or CAPS marker, multiple PCR primers (20-25bp) were designed and optimal PCR conditions determined. Then the best combination was tested for polymorphism and mapped on the KO or TM mapping populations, using post-amplification restriction enzyme digestion if necessary.

Results and discussion

Based on QTL studies reported in the literature, 5 RAPD markers were identified as linked to 4 TM QTLs for oil content and a 6th and a 7th RAPD marker were linked to homologous or homoeologous regions. Likewise 7 RAPD markers are linked to β -glucan content QTLs and 2 others to homologous or homoeologous regions. SCAR or CAPS markers were developed from each and mapped in either KO or TM (Table). While many of the new markers map to the same loci as the original RAPD markers, others map to homologous or homoeologous genomic regions and still others to regions not known to be orthologous to the original RAPD regions.

The naked (*N1*) locus on the TM5 linkage group and several other genomic regions are associated with QTLs for multiple traits (Table), either due to clustering of independent genetic loci or due to pleiotrophic affects. Thus 3 of the new markers are associated with both oil and β -glucan content and 9 with protein content.

Similar results have been obtained for numerous SCARs for other traits (Orr and Molnar, unpublished).

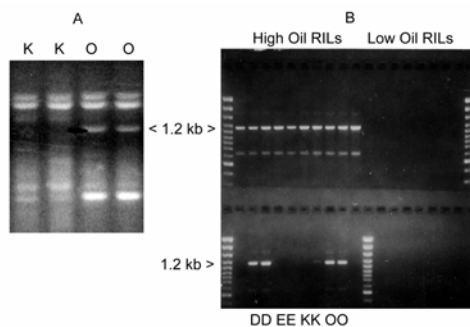


Figure: PCR amplification products separated by electrophoresis in agarose gels give more complex banding patterns for a RAPD marker (A) than for the derived SCAR marker (B). In panel A, RAPD primer *ubc364* produces a 1.2kb product with genomic DNA of oat variety 'Ogle' (lanes 3 and 4) but not with 'Kanota' (1 and 2). This polymorphic band was used to design new SCAR primers. Panel B shows products obtained with SCAR marker *ubc364os* primers and genomic DNAs of 'Dal' (lanes 23 and 24), 'Exeter' (25 and 26), 'Kanota' (27 and 28), and 'Ogle' (29 and 30). All high oil content RILs of the Dal x Exeter population produce the 1.2 kb band (2 to 11) but none of the low oil content RILs (12 to 22) indicating that this SCAR has potential for MAS for oil content. Lanes 1, 20, 21, and 32 are molecular weight standards and lane 31 is a control sample lacking any genomic DNA.

SCAR or CAPS Marker	Map Location (Linkage Group) Linkage to published QTL indicated by underlining
	Original RAPD Derived SCAR
Oil Content QTLs	
<i>ubc121ms</i>	TM11 <u>TM15</u>
<i>ubc167s</i>	<u>TM15</u>
<i>ubc167ts (Hinf I)</i> [#]	TM15
<i>ubc167ms (Hpa II)</i> [#]	<u>TM15</u>
<i>ubc185s</i> [#]	TM5
<i>ubc186s</i>	<u>TM15</u>
<i>ubc189s</i> [#]	<u>TM4_16</u>
<i>ubc198s</i> [#]	<u>TM15</u>
<i>ubc364os</i> [#]	<u>KO15</u> <u>KO11_41+20</u>
β-Glucan Content QTLs	
<i>ubc109as</i>	TM4
<i>ubc109bs</i>	TM2 (<u>KO11 / KM11</u>)
<i>ubc118ms</i>	Unknown
<i>ubc159as</i> [#]	TM5
<i>ubc179os</i>	TM11
<i>ubc185s</i> [#]	TM5
<i>ubc189s</i> [#]	TM4_16
<i>ubc221as</i>	<u>KO17</u>
<i>ubc254s</i>	KO5
<i>ubc263s</i> [#]	TM14
<i>ubc264kas</i> [#]	TM1 (KO33)
<i>ubc267s</i>	TM18
<i>ubc352ks</i>	KO19+27
<i>ubc352os</i>	KO22_44+18, (TM5); TM27
<i>ubc360s</i> [#]	KO33
<i>ubc360s (TagI)</i>	KO2 (TM21), (KO3)
<i>ubc364os</i> [#]	<u>KO11_41+20</u>
<i>ubc372s</i>	<u>KO3</u>
<i>ubc375s</i>	<u>KO20</u>
<i>ubc375s (DdeI)</i>	<u>KO20</u>

Notes: [#] These 3 SCARs are linked to both oil and β -glucan content QTLs.
^{*} These 9 SCAR or CAPS markers are linked to protein content QTLs.

Conclusions

These PCR-based markers have potential to service as anchor markers for comparative mapping, to define homologous and homoeologous relationships in oat, to investigate the complex genetics of grain quality traits, and for marker assisted oat breeding.

Our SCAR and CAPS markers complement marker development that has been done by other research groups and several new initiatives, such as DArT and SSR markers, which are underway to increase the number and utility of PCR-based markers in oat.

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