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ORIGINAL PAPER

Development of IRAP- and REMAP-derived SCAR markers for marker-assisted selection of the stripe rust resistance gene *Yr15* derived from wild emmer wheat

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Abstract

Key message ***Yr15*** **provides broad resistance to stripe rust, an important wheat disease. REMAP- and IRAP-derived co-dominant SCAR markers were developed and localize *Yr15* to a 1.2 cM interval. They are reliable across many cultivars.**

Abstract Stripe rust [*Pucinia striiformis* f.sp. *tritici* (*Pst*)] is one of the most important fungal diseases of wheat, found on all continents and in over 60 countries. Wild emmer wheat (*Triticum dicoccoides*), which is the tetraploid progenitor of durum wheat, is a valuable source of novel stripe rust resistance genes for wheat breeding. *T. dicoccoides* accession G25 carries *Yr15* on chromosome 1BS. *Yr15* confers resistance to virtually all tested *Pst* isolates; it is effective in durum and bread wheat introgressions and their derivatives. Retrotransposons generate polymorphic insertions, which can be scored as Mendelian markers using techniques such as REMAP and IRAP. Six REMAP- and IRAP-derived SCAR markers were mapped using 1,256 F_2 plants derived from crosses of the susceptible *T. durum* accession D447 (DW1) with its resistant BC_3F_9 and BC_3F_{10} (B9 and B10) near isogenic lines, which carried *Yr15* introgressed from G25. The nearest markers segregated 0.1 cM proximally and 1.1 cM distally to *Yr15*. These markers were also mapped and validated at the same position in another 500 independent F_2 plants derived from crosses of B9 and B10 with the susceptible cultivar Langdon (LDN). *SC2700* and *SC790*, defining *Yr15* on an interval of 1.2 cM, were found to be reliable and robust co-dominant markers in a wide range of wheat lines and cultivars with and without *Yr15*. These markers are useful tags in marker-assisted wheat breeding programs that aim to incorporate *Yr15* into elite wheat lines and cultivars for durable and broad-spectrum resistance to stripe rust.

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Introduction

Wheat exceeds all other food crops worldwide in hectares planted (FAOSTAT 2012) and becomes ever more important as the global population increases. However, modern plant breeding practices have narrowed the genetic diversity in wheat and in the germplasm reservoirs of resistance to biotic and abiotic stresses (Laidò et al. 2013). Stripe

rust, caused by *Puccinia striiformis* f.sp. *tritici* West. (*Pst*), is one of the most devastating diseases of wheat throughout the world (Chen 2005; Chen et al. 2010). It can reduce yield by 70 % or, in severe cases, 100 % and lead to shriveled grains and stunted spikes (Chen 2005; McIntosh et al. 1995). Breeding resistant cultivars is the most economical method for controlling stripe rust.

Wild emmer wheat (*Triticum dicoccoides*) harbors extensive genetic resources for wheat improvement including genes for rust resistance (Fahima et al. 1998; Gerechter-Amitai and Grama 1974; Nevo et al. 2002). An accession highly resistant to stripe rust, *T. dicoccoides* G25, was earlier described (Gerechter-Amitai and Grama 1974; Gerechter-Amitai and Stubbs 1970); its resistance was later shown to be conferred by a single dominant gene, designated *Yr15* (Gerechter-Amitai et al. 1989a, b). *Yr15* was shown to be effective against 24 *Pst* races from 18 countries worldwide (Gerechter-Amitai et al. 1989a) and to 26 international isolates and Chinese races (Niu et al. 2000). More recent work showed that *Yr15* was effective against all *Pst* races identified in the USA (Chen et al. 2010; Murphy et al. 2009). Wide-scale introgression of *Yr15* into cultivated hexaploid bread wheat and tetraploid pasta wheat began in the 1980s following initial demonstration of the efficacy of *Yr15* (Gerechter-Amitai and Grama 1974); it continues today (Yaniv et al. 2014).

Chen (2005) proposed combining all-stage (seedling) and HTAP (high-temperature adult plant) resistance as the most effective strategy for durable resistance to stripe rust. However, it is relatively difficult to combine genes for both forms of resistance into a single cultivar on the basis of phenotypic selection, and almost impossible, if the introgression involves two or more resistance genes to the same disease. Molecular markers that are closely linked to the genes of interest help to minimize introgression of unwanted flanking genes from the wild germplasm and thereby accelerate the process of developing wheat cultivars with stronger and more durable resistance. Fine genetic mapping in a large population is a prerequisite for both the application of marker-assisted selection and map-based gene cloning.

Yr15 was localized to chromosome 1BS using cytogenetic analysis (McIntosh et al. 1996) and mapped using molecular markers (Chague et al. 1999; Peng et al. 2000; Sun et al. 1997) and shown to be flanked by two RAPD and RFLP markers in an interval of 7 cM (Peng et al. 2000). These markers have the disadvantages of being neither closely linked nor easily used for marker-assisted selection (MAS); they are ineffective against introgression of unwanted flanking traits ("linkage drag"). In wheat, 1 cM of genetic distance is approximately equivalent to 4.4 Mb (Delaney et al. 1995), although it has been shown to vary from 0.36 to 20 Mb (Saintenac et al. 2011). Moreover, the large monoploid genomes (5.6 to 6.2 Gbp) of wheats and the correspondingly high proportion of repetitive DNA, predominantly retrotransposons (Breen et al. 2013a; International Wheat Genome Sequencing Consortium 2013; Paux et al. 2006), complicates map-based cloning in wheat (Feuillet et al. 2003; Uauy et al. 2006). Finding markers very closely linked to a target gene by mapping in large segregating populations can ease the isolation of a locus on one or a few BAC clones and narrow the pool of candidate genes therein. We therefore set out to develop new molecular markers for refinement of the mapping interval carrying *Yr15* for MAS and for map-based cloning of this gene.

Retrotransposons not only comprise most of large genomes like that of wheat, but also may be harnessed as molecular markers (Kalendar and Schulman 2006; Schulman et al. 2012). Direct comparisons of retrotransposon marker methods with AFLP indicate that the former are some 25 % more polymorphic (Waugh et al. 1997; Yu and Wise 2000). While regions rich in retrotransposons tend to have lower rates of recombination (He and Dooner 2009), they are also expected to have more polymorphism arising from retrotransposon insertions. Although retrotransposons in grass genomes can be found in large arrays depauperate of genes (Kronmiller and Wise 2007), they also nevertheless frequently flank plant genes (White et al. 1994). A comparison found that retrotransposons comprise only 8 % less (67 vs. 75 %) of the sequence of gene-bearing BACs than of random BACs (International Barley Genome Sequencing Consortium et al. 2012). These properties make retrotransposons well suited for gene mapping (Leigh et al. 2003; Manninen et al. 2000; Queen et al. 2004; Tanhuanpää et al. 2007).

Various molecular marker systems have been used for fine mapping of stripe rust resistance genes (Huang et al. 2003; Kota et al. 2006; Ling et al. 2003; Mago et al. 2005; Stein et al. 2000; Yan et al. 2003). In the current study, six IRAP- (inter-retrotransposon amplified polymorphism) and REMAP (retrotransposon-microsatellite amplified polymorphism) -derived SCAR markers tightly linked to *Yr15* were developed. These are both co-dominant and locus-specific. Two of these markers are highly polymorphic across different genetic backgrounds and can be used reliably to introgress *Yr15* into elite wheat lines and cultivars and through MAS.

Materials and methods

Plant materials

The plant materials used for this study consisted of an F_2 population of 1,256 individuals derived from crosses between the susceptible *T. durum* accession D447

(LD393/2*Langdon ND58-322) and its resistant BC_3F_9 (B9) and BC_3F_{10} (B10) near isogenic line (NIL) derivatives. B9 and B10 carry *Yr15* on a 1BS chromosome segment introgressed from *T. dicoccoides* accession G25, which had been produced by selection for resistance and for morphological similarity to the cultivar in each generation (Yaniv et al. 2014). From this population, 598 individuals were screened by PCR with SSR markers *Xgwm911* and *Xgwm18*, which flank *Yr15*, and 33 F_2 recombinants within the interval were identified. The 33 recombinants were used to develop new SSRs in the interval. The remaining 658 individuals were then screened with the new SSR markers, *Xbarc8* and *Xgwm413*, which flank *Yr15* in an interval of 7.7 cM, and 87 F_2 recombinants were identified. All of the 120 (33 + 87) F_2 recombinants served as the population for fine mapping.

The B9 and B10 NILs were screened by PCR for markers *Xbarc8* (Song et al. 2002) and *Xuhw252* (E. Yaniv, unpublished data), which flank *Yr15*, to select recombinants in the interval. Ten F_3 seeds from each F_2 recombinant were screened with flanking markers to select homozygous recombinants. Eight F_4 seeds of each homozygous recombinant were tested for seedling response to *Pst* race 38E134 as described by Gerechter-Amitai et al. (1989a). In addition, 61 bread and durum wheat lines and cultivars, some containing *Yr15* and some not, were used to validate and confirm two tightly linked markers, which flank *Yr15*, as candidates for MAS.

DNA isolation and SSR analysis

Ten-day-old seedling leaves were collected for use in DNA preparation. DNA was prepared by the CTAB method (Ausubel et al. 1995) with RNase A treatment. Primers for SSR markers *Xgwm413* and *Xbarc8*, assigned to chromosome 1B between *Xgwm911* and *Xgwm18*, were used for PCR amplification. Primer sequences were obtained from the GrainGenes website (http://wheat.pw.usda.gov/cgi-bin/graingenes/browse.cgi?class=marker). Amplifications were performed on 60 ng genomic DNA in 20 μL volumes containing 1× buffer (BioTools B&M Labs, Madrid) containing 2 mM $MgCl_2$, 200 nM of each primer, 200 μM dNTP, and 1 U Taq polymerase (Biotools or Fermentas). Amplification was performed for 34 cycles. After initial denaturation for 4 min at 94 °C, each cycle consisted of 40 s at 94 °C, 40 s at 60 °C, and 2 min at 72 °C. A 5 min final extension at 72 °C followed. Amplification products were separated by electrophoresis on 2 % agarose (RESolute Wide Range, BIOzym) and detected by ethidium bromide staining. Gel pictures were scanned with the FLA-5100 imaging system (Fuji photo film GmbH). The PCR fragments produced by *Xgwm413* were resolved on an automated laser fluorescence (ALF) sequencer 3130XL-ABIprism. To allow this, one primer of each pair was labeled at the 5′ end with Fam fluorescein dye. Fragment sizes were calculated with Peak Scanner software v 1.0 (Applied Biosystems) by comparison to the internal size standards of GS120 L12™ that were added to each lane in the loading buffer.

ISSR, IRAP, REMAP and EST-SSR analysis

A total of 42 ISSR and 100 IRAP single primers as well as 200 REMAP primer combinations (derived from 10 ISSR primers in combination with 20 IRAP primers) were screened on the mapping parents. ISSR primers are anchored at the 3′ or 5′ ends of SSR repeats with a nucleotide at the 3′ end of the primer that does not match the repeat itself. Polymorphic markers potentially linked to *Yr15* were tested on recombinants to determine genetic distances. Conditions for PCR, electrophoresis, staining, and gel scanning were, with minor modifications, as described by Kalendar and Schulman (2006). The IRAP and REMAP primers are specified in Supplementary Table S1. Five wheat EST sequences that contain SSRs and that match genes within the rice genomic region collinear to the wheat region of *Yr15* were used to develop EST-based SSR markers. The sequences, based on ESTs TC8999, TC90000, TC67764, TC78819 and TC64667, were obtained from the International Triticeae Mapping Initiative (ITMI) website (http://wheat.pw.usda.gov/ITMI/EST-SSR/LaRota/Table3_est-ssr%20designed%20primers.xls). The conditions used for PCR, electrophoresis, staining, and gel scanning were the same as for the IRAP and REMAP markers.

Development of IRAP- and REMAP-derived SCAR markers

IRAP and REMAP PCR products were separated by electrophoresis, excised from agarose gels, purified by gel extraction, and cloned. From colonies containing recombinant plasmids, suspended in 200 μL colony storage solution (10 mM NaCl, 5 mM $MgCl_2$, 10 mM Tris–HCl pH 7), 2 μL were tested by PCR amplification using the corresponding IRAP or REMAP primers (Supplementary Table S1) in a reaction volume of 20 μL. Following an initial denaturation for 5 min at 95 °C, the PCRs consisted of 23 cycles of 20 s at 95 °C, 40 s at 60 °C, and 10–120 s at 72 °C (depending on the insert fragment size), with a 5-min final extension at 72 °C. Electrophoresis, staining, and gel scanning were carried out as above. Clones yielding PCR products of correct size were sequenced. Specific primers were designed for each sequence and tested again on the parents and recombinants in order to verify that the correct PCR products were cloned.

Statistical analysis

Marker order was determined using Mapmaker software (Lander et al. 1987). Because most of the markers analyzed were very closely linked (Ling et al. 2003), recombination frequencies was equal to centiMorgans (cM). Sequence-specific primers were designed using FastPCR software (Kalendar et al. 2014; http://primerdigital.com/fastpcr.html).

Results

Fine genetic mapping at the *Yr15* locus

The SSR markers *Xgwm91-1*, *Xgwm413*, and *Xgwm18* (Röder et al. 1998) flanked *Yr15* over an interval of 20 cM, which was later narrowed to 7.2 cM with markers *Xbarc8* and *Xgwm413* (Peng et al. 2000). That interval was more precisely mapped here. First, we tested the five EST-SSR primer pairs that match genes within the rice genome that are collinear to the wheat region of *Yr15*; no informative polymorphism was linked to *Yr15*. Five new ISSR markers were potentially linked to *Yr15* on 1BS, but were not closer than the SSR markers reported previously (Peng et al. 2000; Röder et al. 1998). A total of 28 IRAP primers and 40 REMAP primer combinations were then tested; they yielded 70 markers that showed linkage to *Yr15*. One IRAP marker, *IR2107*, and five REMAP markers, *RE425–485*, *RE443–495*, *RE443–834*, *RE438–483*, and *RE440–679* (named according to the IRAP or REMAP primers used to develop them), were closely linked to *Yr15* (Fig. 1). These six were taken forward for conversion to locus-specific and co-dominant SCAR markers.

The *IR2107* and *RE443–495* markers produced, in resistant lines, locus-specific and co-dominant markers with fragment sizes of 1,600 and 790 bp, respectively; these were implemented as SCAR markers *SC1600* and *SC790*. REMAP markers *RE425–485* and *RE443–834* yielded co-dominant markers in resistant lines and were developed as SCAR markers *SC2700* and *SC1028*. The REMAP markers *RE438–483* and *RE440–679* were developed as SCAR markers *SC800* and *SC338*. *Yr15* was flanked on the distal side by *SC790* (1.1 cM, Fig. 2a) and on the proximal side by *SC2700* (0.1 cM; Fig. 2b). Markers *SC1600* and *SC1028*, which co-segregated, mapped at a distance of 1.2 cM proximal to *Yr15*. Markers *SC800* and *SC338* were proximal to the gene, respectively, at distances of 2.1 and 2.8 cM (Fig. 3a). The SCAR markers were also mapped and validated on 41 F_3 homozygous recombinants, derived from 500 F_2 individuals from crosses of B9 and B10 with Langdon (LDN). The SCAR marker positions relative to *Yr15* were the same as those in the above population, but the genetic distances were slightly different (Fig. 3b).

IRAP and REMAP-derived SCAR markers are highly efficient for MAS of *Yr15*

To evaluate the effectiveness of the newly identified, IRAP- and REMAP-derived SCAR markers, we assayed the four closest markers (*SC790*, *SC2700*, *SC1600*, and *SC1028*)

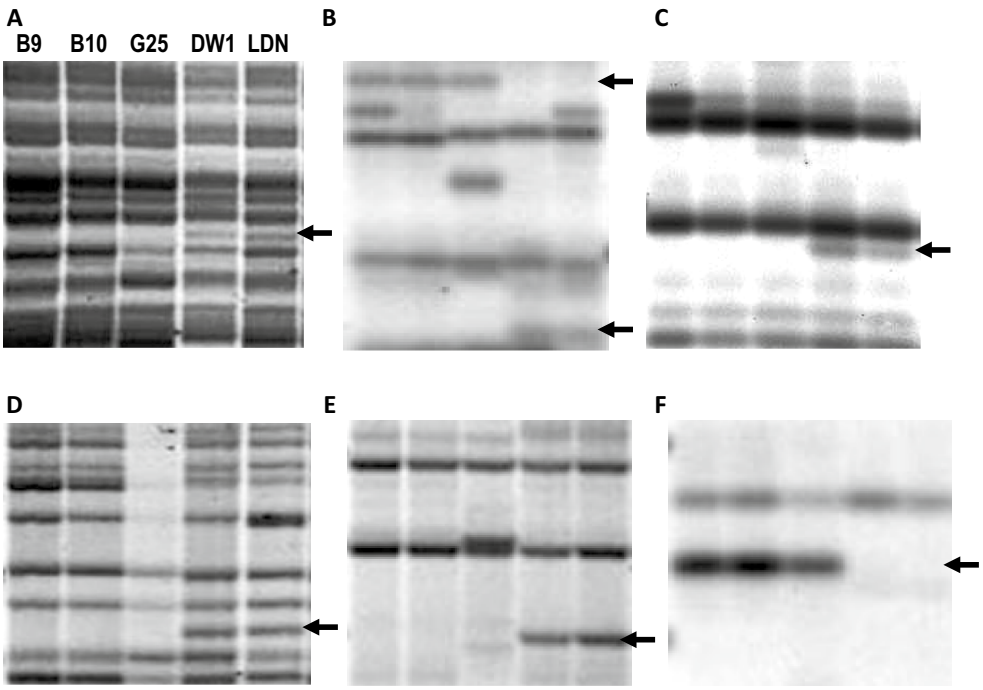

Fig. 1 IRAP and REMAP polymorphisms developed into SCAR markers. The SCAR markers were developed as follows: **a** IRAP, *IR2107*; **b** REMAP, *RE425–485*; **c** REMAP, *RE443–495*; **d** REMAP, *RE443–834*; **e** REMAP, *RE438-483*; **f** REMAP, *RE440-679*. In **b–f**, the sample order is the same as in **a** B9 and B10 are resistant RILs; G25 is the resistant donor of *Yr15*; DW1 and LDN are the susceptible *T. durum* cultivars. *Arrows* point to the polymorphic bands developed into SCAR markers

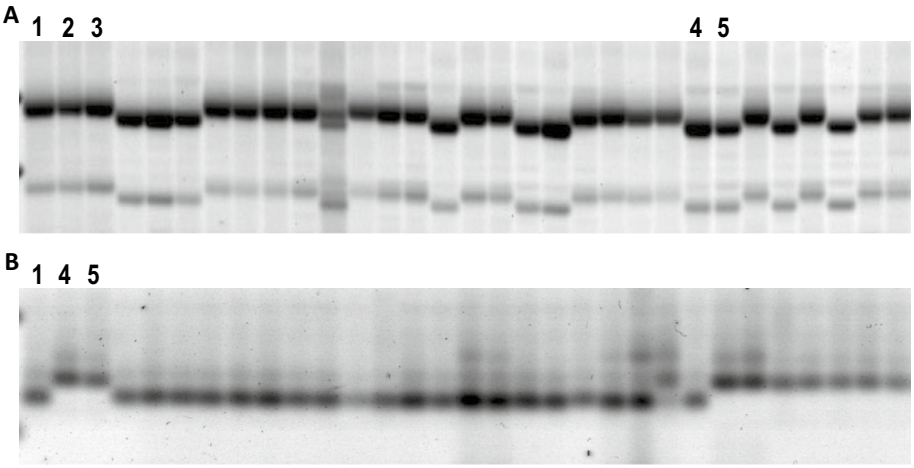

Fig. 2 Polymorphism of retrotransposon-derived SCAR markers on resistant and susceptible F_3 homozygous recombinant plants. **a** Marker *SC790*; lines carrying *Yr15* (B9, B10, G25, respectively, *lanes 1*, *2*, *3*) can be distinguished by the higher MW band of the susceptible cultivars (DW1, LDN, respectively, *lanes 4*, *5*) and F_3 lines thereby genotyped (*unlabeled lanes*). **b** SCAR *SC2700*, where the parental and recombinant lines carrying *Yr15* display a lower MW band (labeled as in **a**)

on 25 wheat lines containing *Yr15* and ten lines that lack it (Fig. 4; Tables 1, 2). All except two of the lines (HSB2408 and HSB2955) were distinguishable by *SC790* and *SC2700*. All of the remaining lines and cultivars showed complete correspondence between these two markers and response to *Pst*. Marker *SC1600* was not polymorphic in any tested line or cultivar. It amplified the resistance-associated allele in all lines, except cv. Boston, which by phenotype contains *Yr15*. However, the resistance-associated allele was present in all lines and cultivars except line N163 that did not possess *Yr15*. Marker *SC1028* was clearly not closely associated with the *Pst* resistance phenotype (Table 1). Thus, of the markers tested and developed, *SC2700* and *SC790* appeared to be closely linked and reliable, either alone or in combination, for monitoring *Yr15* in MAS. These markers are available via a material transfer agreement; please contact the corresponding author.

The efficiency of the above four markers for tagging *Yr15* was further assayed in 13 hexaploid cultivars into which *Yr15* was introgressed by backcrossing. Markers *SC1600* and *SC1028* were again unreliable in distinguishing the cultivars and derivatives with and without the introgressed *Yr15*. Markers *SC2700* and *SC790* discriminated all pairs of lines, except for cv. Stiletto and Stiletto *Yr15* (Table 2), thus indicating that they can be used reliably in MAS programs.

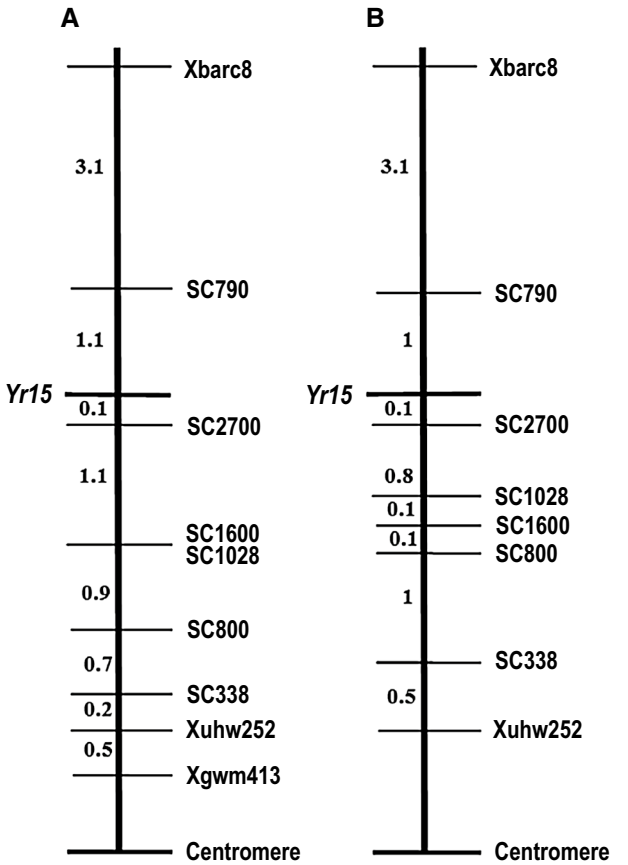

Fig. 3 Fine genetic map of stripe rust resistance gene *Yr15* in chromosome 1BS of wheat. Maps resulting from the analysis of **a** 1,256 F_2 plants of the cross of near isogenic lines B9 and B10 × D447, and **b** 500 F_2 plants of the populations B9 and B10 × LDN

Discussion

MAS and map-based cloning of the stripe rust resistance gene *Yr15* depends on the development of markers closely linked to the gene. *Yr15* was previously mapped using SSR, RFLP, and RAPD markers, but for use in MAS breeding programs these markers suffer from relatively loose linkage to the gene and variable polymorphism among cultivars. In the present study, the availability of co-dominant SSR markers (*Xgwm911*, *Xgwm18*, *Xgwm413*, and *Xbarc8*) assisted in identification of recombinant lines in a high-resolution mapping population. Of available marker systems, the retrotransposon-based IRAP and REMAP methods proved both highly polymorphic and effective.

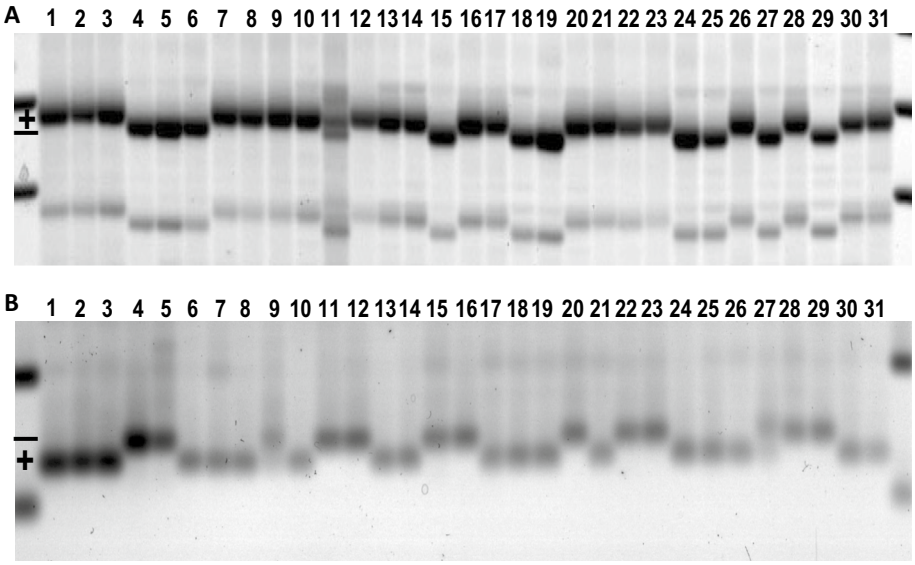

Fig. 4 Polymorphism of retrotransposon-derived SCAR markers on wheat lines and cultivars. **a** Marker SC790. *Lanes from left*: 1, B9; 2, B10; 3, G25; 4, DW1; 5, LDN; 6, Reeves; 7, Sel32; 8, Sel46; 9, Sel20; 10, Legron; 11, Agrestis; 12, Cortez; 13, Boston; 14, Sel7; 15, Maverick; 16, G90; 17, Wed; 18, Baxter; 19, Avocet; 20, Avocet *Yr15*; 21, Sel4; 22, HSB2398; 23, HSB2527; 24, Combat; 25, QT3960; 26, Kern *Yr15*; 27, Kern; 28, UC1107 *Yr15*; 29, UC1107; 30, UC1104 *Yr15*; 31, B1. **b** Marker *SC2700*. *Lanes from left*: 1, B9; 2, B10; 3, G25; 4, DW1; 5, LDN; 6, Legron; 7, Agrestis; 8, Cortez; 9, Kulin; 10, Kulin *Yr15*; 11, Avocet; 12, Kern; 13, Avocet *Yr15*; 14, Kern *Yr15*; 15, UC1107; 16, UC1041; 17, UC1107 *Yr15*; 18, UC1041 *Yr15*; 19, Boston; 20, Reeves; 21, Sel7; 22, M708; 23, Maverick; 24, G90; 25, Wed; 26, Sel32; 27, Combat; 28, Baxter; 29, Ruby; 30, Sel46; 31, Sel20

A total of 70 polymorphic IRAP and REMAP bands, potentially linked to *Yr15*, were generated in our study. To facilitate marker-assisted breeding of wheat against stripe rust, we identified co-dominant PCR-based markers *SC2700* and *SC790* flanking *Yr15* at genetic distances of 0.1 and 1.1 cM, respectively. The reliability of these markers was tested on independent large F_2 populations and also on a range of lines and cultivars with different genetic backgrounds. These tightly linked markers can be easily detected using 2 % agarose gels and readily used to incorporate *Yr15* into elite breeding lines. They may also serve in predicting the presence of *Yr15* in populations and collections. They are therefore expected to be useful for minimizing linkage drag associated with the gene and, for example, for combining *Yr15* and HTAP resistance (Chen 2005) during further breeding.

The high frequency of polymorphic retrotransposon bands suggests that the retrotransposon families chosen for IRAP and REMAP primers have been transpositionally active in durum wheat and its wild ancestor, *T. dicoccoides*. The tight association of *SC2700* and *Yr15* and the high polymorphism of the retrotransposon-based markers linked to *Yr15* may not be coincidental. Sequence analyses of the regions containing disease resistance genes in rice (Song et al. 1998), maize (Ramakrishna et al. 2002), barley (Shirasu et al. 2000; Wei et al. 2002), potato (Ballvora et al. 2007), and soybean (Innes et al. 2008) are consistent with this result. In general, these studies show highly dynamic changes in the retrotransposon content as well as a breakdown in the colinearity of resistance genes, which may be driven by recombination between retrotransposons.

Conversely, many genetic and molecular studies show that genes for disease resistance are frequently clustered or closely associated (Islam et al. 1989; Joshi and Nayak 2013; Leister 2004; Michelmore and Meyers 1998). Among the named genes conferring resistance to stripe rust, *Yr10*, *Yr15*, *Yr24*, and *YrH52* are reportedly located on chromosome 1BS (McIntosh et al. 1998). Therefore, 1BS is an important carrier of stripe rust resistance genes. Sequencing, annotation, and functional verification will ultimately establish if the 1B resistance genes are indeed clustered. *Yr15* was earlier reported to be in the 1S0.8 region of 1BS, which is both very rich in genes and highly recombinogenic, having only 365 kb per cM (Gill et al. 1996; Sandhu et al. 2001). These features suggest that *SC2700* and *SC790* may be closely linked to *Yr15* physically, particularly on the distal side, and therefore should be reliable markers with which to screen a BAC library for the positional cloning of *Yr15* and possibly other nearby resistance genes.

Table 1 Polymorphism of newly identified IRAP- and REMAP-derived SCAR markers between different hexaploid lines and cultivars

Cultivar/line	Phenotype	Genotype			
	Yr15	*SC2700*	*SC790*	*SC1600*	*SC1028*
Sel32	+	+	+	+	+
Sel46	+	+	+	+	+
Sel20	+	+	+	+	+
Sel7	+	+	+	+	+
Sel07-97 Merav/ N163/G25	+	+	+	+	+
Sel4	+	+	+	+	+
HSB 2398	+	+	+	+	+
HSB 2408	+	−	−	+	+
HSB 2527	+	+	+	+	+
HSB 2944	+	+	+	+	−
HSB 2949	+	+	+	+	+
HSB 2955	+	−	−	+	+
Legron	+	+	+	+	−
Agrestis	+	+	+	+	−
Cortez	+	+	+	+	+
Boston	+	+	+	−	−
B1	+	+	+	+	−
B2	+	+	+	+	+
79W793	+	+	+	+	−
G90	+	+	+	+	−
Wed	+	+	+	+	−
BM383B195	+	+	+	+	+
B70	+	+	+	+	−
B174C93.8	+	+	+	+	−
B176c193.10	+	+	+	+	−
Reeves	−	−	−	+	+
M708	−	−	−	+	+
Merav	−	−	−	+	+
N 163	−	−	−	−	−
Ruby	−	−	−	+	+
Sapphire	−	−	−	+	+
Combat	−	−	−	+	+
QT3960	−	−	−	+	+
Maverick	−	−	−	+	+
Baxter	−	−	−	+	+

+ resistance-associated allele, − susceptibility-associated allele

Table 2 Polymorphism of newly identified IRAP- and REMAP-derived SCAR markers between genotypes with and without *Yr15*

Cultivar	Phenotype	Genotype			
	Yr15	*SC2700*	*SC790*	*SC1600*	*SC1028*
UC1107 *Yr15*	+	+	+	+	+
UC1107	−	−	−	+	+
UC1358 *Yr15*	+	+	+	+	+
UC1358	−	−	−	+	+
UC1128 *Yr15*	+	+	+	+	+
UC1128	−	−	−	+	+
Kern *Yr15*	+	+	+	+	−
Kern	−	−	−	+	+
UC1037 *Yr15*	+	+	+	+	−
UC1037	−	−	−	+	+
UC1110 *Yr15*	+	+	+	+	+
UC1110	−	−	−	+	+
UC1041 *Yr15*	+	+	+	+	−
UC1041	−	−	−	+	+
Avocet *Yr15*	+	+	+	+	+
Avocet	−	−	−	+	+
Corrigin *Yr15*	+	+	+	+	−
Corrigin	−	−	−	+	+
Excalibur *Yr15*	+	+	+	+	+
Excalibur	−	−	−	+	+
Kulin *Yr15*	+	+	+	+	−
Kulin	−	−	−	+	+
Stiletto *Yr15*	+	+	+	+	−
Stiletto	+	+	+	+	−
Suncea *Yr15*	+	+	+	+	−
Suncea	−	−	−	+	+

+ resistance-associated allele, − susceptibility-associated allele

Author contributions A. H. S., E. Y., and R. K. defined the research theme and designed the methods and experiments. E. Y. developed the mapping population and marker *Xuhw252*, the genetic materials and the framework genetic map. B. A. M. developed and mapped the retrotransposon markers, screened the F_2 populations, and developed F_3 lines. D. R. carried out all stripe rust resistance testing. H. B. developed and contributed genetic materials. All authors contributed to the writing.

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Conflict of interest The authors declare that they have no conflict of interest.

Ethical standards The experiments comply with the current laws of the countries in which they were performed.

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