

RESEARCH ARTICLE

Molecular mapping of a stripe rust resistance gene in wheat line C51

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Abstract

Stripe rust, a major disease in areas where cool temperatures prevail, can strongly influence grain yield. To control this disease, breeders have incorporated seedling resistance genes from a variety of sources outside the primary wheat gene pool. The wheat line C51, introduced from the International Center for Agricultural Research in the Dry Areas (ICARDA), Syria, confers resistance to all races of *Puccinia striiformis* f. sp. *tritici* (PST) in China. To map the resistant gene(s) against stripe rust in wheat line C51, 212 F₈ recombinant inbred lines (RILs) derived from the cross X440 × C51 were inoculated with Chinese PST race CYR33 (Chinese yellow rust, CYR) in the greenhouse. The result showed that C51 carried a single dominant gene for resistance (designated *YrC51*) to CYR33. Simple sequence repeat (SSR) and resistance gene-analogue polymorphism (RGAP) markers that were polymorphic between the parents were used for genotyping the 212 F₈ RILs. *YrC51* was closely linked to two SSR loci on chromosome 2BS with genetic distances of 5.1 cM (*Xgwm429*) and 7.2 cM (*Xwmc770*), and to three RGAP markers C51R1 (XLRR For / NLRR For), C51R2 (CLRR Rev / Cre3LR-F) and C51R3 (Pto kin4/ NLRR-INV2) with genetic distances of 5.6, 1.6 and 9.2 cM, respectively. These RGAP-linked markers were then converted into STS markers. Among them, one STS marker, C51STS-4, was located at a genetic distance of 1.4 cM to *YrC51* and was closely associated with resistance when validated in several populations derived from crosses between C51 and Sichuan cultivars. The results indicated that C51STS-4 can be used for marker assisted selection (MAS) and would facilitate the pyramiding of *YrC51* with other genes for stripe rust resistance.

[Zheng J., Yan Z., Zhao L., Li S., Zhang Z., Garry R., Yang W. and Pu Z. 2014 Molecular mapping of a stripe rust resistance gene in wheat line C15. *J. Genet.* 93, 443–450]

Introduction

Stripe rust caused by *Puccinia striiformis* f. sp. *tritici* (PST), is a devastating disease in common wheat (Roelfs *et al.* 1992). This disease causes significant economic losses in terms of reduced production and/or high costs for chemical control (<http://www.globalrust.org>; Wellings 2007). This disease can be devastating under favourable conditions with yield losses of more than 90% on susceptible cultivars being recorded (Chen 2005; Sharma-Poudyal and Chen 2011). China has the largest stripe rust epidemic area in the world with the northwestern and southwestern areas of China being particularly prone, especially in autumn-sown crops (Wan *et al.* 2004). Although growing resistant cultivars is the most

effective, economic and environment friendly way to control stripe rust (Line and Chen 1995), the rapid development of new PST races often circumvents resistance in cultivars (Line and Qayoum 1991; Chen *et al.* 2003; Zhang *et al.* 2011). Finding new germplasm with new resistance genes and pyramiding different resistance genes to breed multiline cultivars may increase the durability of resistance (Ma *et al.* 2001; Wen *et al.* 2008).

Today, marker-assisted selection (MAS) is commonly used in breeding organizations. This technology greatly facilitates the process of resistance gene pyramiding (Chen *et al.* 2003). Identification of molecular markers that cosegregate or are closely linked with the resistance genes is most useful for MAS (Madan *et al.* 1997). During the last decades, molecular mapping has identified chromosome regions carrying important genes in wheat using different markers.

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Keywords. *Puccinia striiformis*; yellow rust; SSR; RGAP; STS; marker-assisted selection; *Triticum aestivum*.

However, these genetic markers are often at some distance from the targeted genes and can be population specific or parent related. The degree of linkage can affect their predictive value, and allelic variations can limit the value in specific populations (Bagge *et al.* 2007; Liu *et al.* 2012). As a result, relatively few linked markers are used in breeding. Gene specific markers (or functional markers) are ideal for MAS in wheat breeding as they are derived directly from the gene conferring the phenotype. To date, more than 30 resistance genes have been cloned in common wheat (Liu *et al.* 2012), but only three stripe rust resistance genes, including *Yr17* (Helguera *et al.* 2003), *Yr18* (Lagudah *et al.* 2009) and *Yr36* (Fu *et al.* 2009), have been cloned. Although this is promising, there are relatively few functional markers for stripe rust, and of those, several are associated with defeated genes that are no longer of any use in breeding. Identification of new resistances along with tightly linked markers is still critical in effective breeding for resistance.

In hexaploid wheat, SSRs, also known as microsatellites show a much higher level of polymorphism and are more informative than any other marker system (Plaschke *et al.* 1995; Ma *et al.* 1996; Röder *et al.* 1998). Further, their genetic location is known, they are accurate, repeatable and are PCR based. These advantages mean that SSR loci are more frequently employed than other marker systems (Karakousis *et al.* 2003; Somers *et al.* 2004; Li *et al.* 2006). The resistance gene-analogue polymorphism (RGAP) technique employs primers designed from conserved motifs of resistance genes and has also been widely used to develop molecular markers in wheat (Chen *et al.* 1998a, 1999; Chen and Line 1999; Toojinda *et al.* 2000; Shi *et al.* 2001; Yan *et al.* 2003; Yan and Chen 2006; Wen *et al.* 2008). However, as their genetic location is generally unknown, they must be mapped through linkage disequilibrium (LD) with markers of known location.

C51, a spring wheat line collected from International Center for Agricultural Research in the Dry Areas (ICARDA),

Syria, has been grown in Sichuan province for over 15 years and maintains a high level of resistance of the Chinese PST races. The objective of this study was to identify and map the gene(s) for stripe rust resistance in wheat line C51 using SSR and RGAP markers, and establish effective MAS breeding methods for the gene(s).

Materials and methods

Plant materials

A total of 212 F₈ recombinant inbred lines (RILs) were derived from the cross X440 × C51 by advancing random individual F₂ plants to F₈ generation by single-seed descent. C51 (pedigree: dove's'/buc's'//star's'), originating from ICARDA, is highly resistant to all Chinese PST races in seedling and adult stages. X440 (pedigree: Tai1287/SW1862//Chuanyu12), bred within Sichuan, is highly susceptible to current PST races. Lines containing important stripe resistance genes, Sichuan wheat cultivars, some homozygous resistant F_{3,4} lines derived from C51/susceptible materials used in validation studies, are listed in table 1.

Resistance assessments

The 212 F₈ RILs along with the parents were planted in a growth chamber and inoculated with PST race CYR33 using the method described by Stubbs (1988). Inoculations were performed on the first fully expanded leaf by brushing fresh urediniospores from a fully infected susceptible genotype onto the seedlings. Inoculated seedlings were placed in plastic covered cages and incubated at 9°C and 100% relative humidity (RH) for 24 h and then transferred into a growth chamber under conditions of 14 h of light (22 000 lux) at 17°C, and 10 h of darkness at 12°C, with 70% RH. Infection types (IT) were scored 16–17 days after inoculation when the

Table 1. *Yr* genes, cultivars and advanced lines used to validate STS markers.

Lines or cultivars	Pedigree	Origin	<i>Yr</i> gene	Resistant performance to PST CYR33
99-1	Yr5/6*Avocet S	Australia	<i>Yr5</i>	Resistant
99-2	Yr10/6*Avocet S	Australia	<i>Yr10</i>	Resistant
99-3	Yr15/6*Avocet S	Australia	<i>Yr15</i>	Resistant
99-7	Yr24/3*Avocet S	Australia	<i>Yr24</i>	Susceptible
99-8	Yr26/3*Avocet S	Australia	<i>Yr26</i>	Susceptible
02-08	Yr27/3*Avocet S	Australia	<i>Yr27</i>	Susceptible
Chuanmai42	SynCD768/SW3243//Chuan6415	Sichuan, China	<i>YrCH42 (Yr26)</i>	Susceptible
Chuanmai104	Chuanmai42/Chuannong16	Sichuan, China	<i>Yr26</i> , +, ?	Resistant
13-02-1	Chuanmai28/C51 F _{3,4}	Sichuan, China	<i>YrC51</i>	Resistant
13-03-1	Taichuang29/C51 F _{3,4}	Sichuan, China	<i>YrC51</i>	Resistant
13-06-1	Mingxian169/C51 F _{3,4}	Sichuan, China	<i>YrC51</i>	Resistant
99-15	Chuanmai28	Sichuan, China	–	Susceptible
20-108	Taichung29	Netherlands	–	Susceptible
98-105	Mingxian169	Chinese landrace	–	Susceptible
99-18	Avocet S	Australia	–	Susceptible

susceptible check Mingxian 169 showed abundant sporulation. IT scoring was based on a 0–4 scale system, where 0, no visible symptoms or no signs of infection; 0, necrotic flecks; 1, necrotic and chlorotic areas with restricted sporulation; 2, moderate sporulation with necrosis and chlorosis; 3, sporulation with chlorosis; 4, abundant sporulation without chlorosis, (Bariana and McIntosh 1993). Plants with ITs 0–1 were considered resistant, and those with ITs 2–4 susceptible.

DNA extraction

Genomic DNA from the parents and 212 F₈ RILs of X440 × C51 and the validation lines was extracted from leaf tissue samples following the procedure described by Li *et al.* (2006). Each RIL genomic DNA was extracted from mixed adequate amount leaf tissue samples of three plants.

SSR and RGAP marker analysis

Resistant and susceptible bulks comprising equal amounts of DNA from 10 resistant and 10 susceptible lines from the 212 F₈ RILs were used for bulked segregant analysis (Michelmore *et al.* 1991). Wheat microsatellite primers were synthesized by Sangon Biotech (Shanghai, China) according to the sequences published in the GrainGenes database (<http://wheat.pw.usda.gov>). The RGAP primers were designed from the conserved domains of cloned resistance gene (Leister *et al.* 1996; Yu *et al.* 1996; Chen *et al.* 1998a, b; Shi *et al.* 2001).

The SSR and RGAP markers were used to identify linkage to the resistance locus (Röder *et al.* 1998; Chen *et al.* 1998a; Shi *et al.* 2001). PCRs were performed in a Biorad (Hercules, USA) S1000™ Thermal Cycler. The 15 µL reaction mixture contained 30 ng of template DNA, 1.5 mM Mg²⁺, 10× PCR buffer, 0.75 U of Taq DNA polymerase, 3 mM each of dNTPs (Transgen, Beijing, China) and 30 ng of each primer. PCR conditions were 95°C for 5 min denaturation, followed by 34 cycles of 30 s at 94°C, 1 min at 45–60°C (depending on the individual primer pairs), and 1 min at 72°C and a final extension at 72°C for 10 min.

The PCR product was mixed with 3 µL formamide loading buffer (98% formamide, 10 mM EDTA, 0.05% bromophenol blue, 0.05% xyleneol, pH 8.0) and denatured at 95°C for 5 min. After preelectrophoresis (50 W, 30 min), 2 µL of each sample was loaded on a 6% denaturing polyacrylamide gel and run at 50 W for 1.2 h. The gels were silver stained according to the method of Bassam *et al.* (1991).

Cloning and sequencing of target PCR fragments amplified from RGAP markers

Cloning of PCR fragments followed the method described by Xu *et al.* (2001). Briefly, target PCR fragments amplified from RGAP markers were excised from a dry gel and transferred to a PCR tube. The gel containing the target DNA bands was eluted twice with 200 µL of 1× TE buffer

(pH 8.0) for 30 min, and once with 200 µL of double distilled water for 30 min. The gels were then soaked in 50 µL double distilled water, and kept in a PCR thermocycler at 95°C for 10 min to release the DNA. The sample was centrifuged at 4000 rpm for 10 min to spin down gel debris and the supernatant was used as template DNA for PCR amplification following the same RGAP primer and PCR conditions.

PCR products were separated on 1.5% agarose gels and purified by an agarose gel extraction kit (Tiangen, Beijing, China). PCR products were ligated into pGEM-T vector (Tiangen) according to manufacturer's recommendations. DNA lengths of 20 individual white colonies from each transformation were reamplified with corresponding RGAP primers and examined in a 4% denaturing polyacrylamide gel. The correct clones were determined as those that contained fragments of the same size as previously identified by corresponding RGAP markers. Three positive clones with expected insert size were sequenced.

Development of STS markers

Primers of STS markers were designed based on the sequences amplified with RGAP markers that were linked to *YrC51*, using the software Premier Primer 5 (<http://www.premierbiosoft.com>). The developed STS markers were subjected to two testing procedures. Firstly, the STS markers were tested for polymorphisms between the resistant and susceptible bulks and their parents. Secondly, the polymorphic bands amplified from STS markers were cloned and sequenced, and aligned with corresponding sequences amplified from RGAP markers using the software DNAMAN (Wen *et al.* 2008, <http://www.lynnon.com>).

Data analysis

The goodness of fit of observed 212 F₈ RILs data to theoretically expected segregation ratios was tested using chi-square tests. All 212 F₈ RILs were genotyped using linked SSR markers, RGAP markers and corresponding STS markers. The linkage map was constructed with the software Mapmaker 3.0 (Lincoln *et al.* 1993) and the Kosambi mapping function (Kosambi 1944) was employed to convert recombination value to genetic distances.

Results

Inheritance of stripe rust resistance in C51

Seedlings of C51 were highly resistant to CYR33 (IT 0), whereas seedlings of X440 were highly susceptible (IT 4). Of the 212 F₈ RILs plants from cross X440×C51 tested with PST race CYR33 in the greenhouse, 115 were resistant and 97 were susceptible, fitting a 1:1 ratio ($\chi^2 = 1.528$, $P = 0.216$). Segregation ratio showed that C51 has a single dominant gene for resistance to the PST race CYR33. The gene was temporarily designated as *YrC51*.

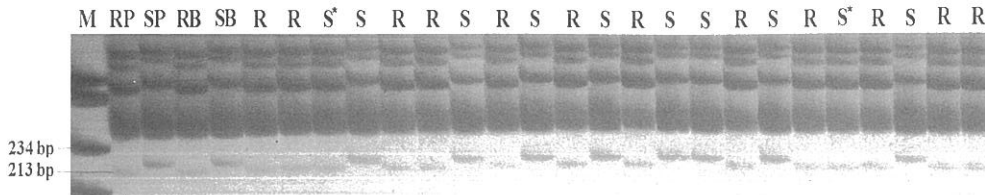


Figure 1. Polymorphism detected by SSR marker *Xgwm429* for *YrC51* in RILs population by silver staining of denaturing polyacrylamide gels. M, marker; RP, resistant parent C51; SP, susceptible parent X440; RB, resistant bulk; SB, susceptible bulk; R, resistance plant; S, susceptible plant; *recombinant

Table 2. Molecular markers linked to *YrC51*, fragment size in C51 and X440.

Marker ^a	Fragment		<i>P</i> _{for1:1}
	C51	X440	
<i>Xgwm429</i>	213	222	0.32
<i>Xwmc770</i>	185	154	0.11
C51R1	548	512	0.21
C51R2	597	—	0.45
C51R3	474	—	0.10
C51STS-4	563	—	0.53

^aFirst two markers are SSRs; next three markers are RGAP markers; and C51STS-4 is a STS marker developed from C51R2-597 bp in this study.

Linkage map for *YrC51*

A total of 420 SSR markers located on all 21 chromosomes of hexaploid wheat were used to test for polymorphism among the parents and two bulks. Two markers (*Xgwm429* and *Xwmc770*) were found to have polymorphisms in both parents and bulks. The phenotypic and genotypic data for the 212 F₈ RIL populations indicated that resistance gene *YrC51* was linked to the two SSR loci. Two codominant alleles *Xgwm429*/213 bp (figure 1), *Xwmc770*/185 bp are present in C51 (table 2). These two SSR loci are located on chromosome 2BS (Somers *et al.* 2004), suggesting that *YrC51* must also be located on this chromosome.

A total of 1711 RGAP primer combinations, developed from 58 single primers, were used to screen the two parents and the corresponding bulks. Among them, only three polymorphic and dominant RGAP markers (C51R1, C51R2 and C51R3) were further used to analyse the 212 RIL populations (table 3). Sizes of the three RGAP markers C51R1, C51R2 and C51R3 were 541, 597 (figure 2) and 242 bp, respectively (table 2).

Development of STS markers

Nine STS primers were designed according to the sequences of the PCR fragments amplified with the three RGAP markers. Only three STS primer pairs generated polymorphic bands with the expected sizes in the parents and bulks (table 4). All three STS markers were dominant, and their polymorphisms were detected in agarose and polyacrylamide gels. Sequence alignments showed that DNA sequences of the PCR products generated by STS markers were consistent with those produced from corresponding RGAP markers. When the three STS markers were used to genotype the RIL population, the results were identical with those obtained using the corresponding RGAP markers. The STS marker C51STS-4 (figure 3) was most closely linked to *YrC51*, being 1.4 cM from the resistance gene (figure 4).

Linkage analysis

Linkage analyses of the 212 F₈ RIL marker data and stripe rust response genotypes showed that the two SSR marker alleles (*Xgwm429*/213 bp, *Xwmc770*/185 bp) were linked in coupling with *YrC51* with genetic distances 5.1 and 7.2 cM, respectively. All the three RGAP markers (C51R1, C51R2 and C51R3) were dominant markers in coupling with *YrC51*, with C51R1 being distal to the gene (5.6 cM), while C51R2 and C51R3 were proximal, with distances to *YrC51* being 1.6 and 9.2 cM (figure 4).

Validation of STS markers

Three STS markers were tested on lines containing several different stripe rust resistant genes and some cultivars as listed in table 1. The 563-bp PCR fragment amplified by C51STS-4 was only present in C51 and its derivatives

Table 3. Three RGAP markers linked to *YrC51* and their primer sequences.

RGAP marker	RGA primer	Sequence (5'–3')	References
C51R1	XLRR For	CCGTTGGACAGGAAGGAG	Chen <i>et al.</i> (1998a)
	NLRR For	TAGGGCCTCTTGCATCGT	Chen <i>et al.</i> (1998a)
C51R2	CLRR Rev	TAACGTCTATCGACTTCT	Chen <i>et al.</i> (1998a)
	Cre3LR-F	CACACACTCGTCAGTCTGCC	Yan <i>et al.</i> (2003)
C51R3	Pto kin4	AGTGTCTTGTAGGGTATC	Chen <i>et al.</i> (1998a)
	NLRR-INV2	TCAGGCCGTGAAAAATAT	Lin and Chen (2007)



Figure 2. Silver stained denaturing polyacrylamide gels showing segregation patterns of RGAP markers C51R2 in RILs population. M, marker; RP, resistant parent C51; SP, susceptible parent X440; RB, resistant bulk; SB, susceptible bulk; R, resistance plant; S, susceptible plant; *recombinant.

Table 4. Sequences of three STS markers and the expected sizes.

STS marker	Sequence (5'-3')	Size (bp)	Annealing temperature (°C)	Corresponding RGAP marker
C51STS-4	F: GGTTCGCTTGGTTACACGGACATC R: CTGCCAGACACCAAGCAAGGGAGG	563	65	C51R2
C51STS-6	F: AGTCGGGAAACCTGTCTGT R: TTTGCTGCCTTTGCTCAC	489	53	C51R2
C51STS-7	F: CGGAAGCATAAAGTGTAAG R: GAGTCAGTGAGCGAGGAA	472	55	C51R3

1	CACACACTCG	TCAGTCTGCC	ACACTAACAT	CGTATTCGTA	CGTACTACGT	TACGTAACAT
61	CCATCTCTCG	CAGGTTGCGT	TGGTTACACG	GACATCCACT	TCTCTTCTTC	GCTGGAACTT
121	GGGAATTGGT	TTGCTATCAA	CAGTGAACGG	TAATGAGTTC	ATGGCAGACA	GACACGTACA
181	CCCCAAACCG	CTGGGACGGG	GGTCGGTGCC	ACCTCACGCT	TGAGACTTGA	GAGAAAGAAA
241	CTTGAGTGCT	GCTATACACA	CGACGTTGTT	CAGCCGGTTT	GTGCACGATC	GCACTCATCC
301	AACCGTGCA	GCCTTGGACA	AAGTGGAAATC	CAGCGGCTAG	ATCAAACATC	GTCCAGTGTT
361	CGGCGGCTGC	TGTGTATCCT	CTGCTTAGTA	CTTCCCAAGA	AACCTTGCCT	CCGGTCGATC
421	GTCCTCACGT	TTATTTGCGA	TTAATTAAAG	GTCGGGTACA	CACGACACGA	GCACCACACC
481	ACAAACAGTGT	TAAGTAAACT	GCTATACTAC	CGAGAACGTT	AATGTTGAG	GTGGACACAC
541	GTCTGCCAGA	CACCAAGCAA	GGGAGGACTG	ACGAGTGTGT	G	

Figure 3. Sequence of PCR fragment amplified with the RGAP marker C51R2 and the primer positions of STS marker C51STS-4.

(figure 5). The other two markers could not accurately distinguish the tested lines that have the *YrC51* from other resistant material. This result implied that the STS marker C51STS-4 could be diagnostic to the genotypes with *YrC51*.

Polymorphism of molecular markers closely linked to *YrC51* in various wheat genotypes

A total of 18 wheat lines, representing a part of resistant breeding genotypes and current grown cultivars in the south-western region in China, were tested with race PST CYR33 and the *YrC51*-flanking markers. When tested with PST CYR33, the genotypes *Yr5*, *Yr10*, *Yr15* and *YrC51* were resistant (table 5). When tested with the markers, only one genotype (*Yr24/3**Avocet S) had a similar sized fragment of the *Xgwm429* resistance amplification product, one (Chuanmai42/Chuannong16, *Yr26*, +) had a similar sized fragment

as C51R2/597 resistance associated allele and none had the fragment associated with resistance from C51STS-4/563. None of the genotypes had two or three of the markers together. Thus, when any two of the markers are used in combination, they could detect polymorphism in 100% of the tested genotypes.

Discussion

C51, introduced from ICARDA in 1997, is an exotic resistant germplasm that has always displayed high levels of resistance (IT 0) at all growth stages over the last 15 years in China. Genetic analysis showed C51 carried a single dominant gene for resistance (designed *YrC51*). The *YrC51* gene was located between *Xwm429* and *Xwmc770* on chromosome 2BS (figure 5).

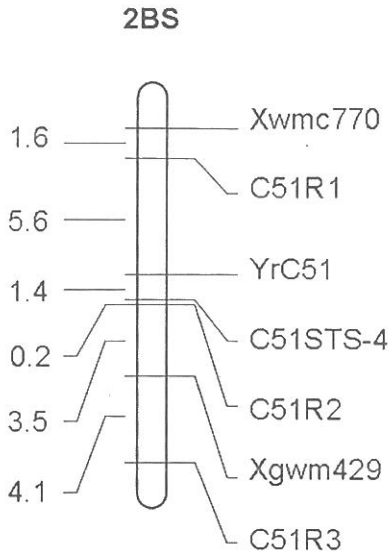


Figure 4. Linkage map of resistance gene *YrC51* on 2BS, constructed with two SSR markers (*Xgwm429*, *Xwmc770*) and three RGAP markers (*C51R1*, *C51R2*, *C51R3*) and a STS marker (*C51STS-4*).

Genes reported on chromosome 2BS include *Yr27*, *Yr31*, *YrSte*, *YrSp*, *YrTp1*, *YrCN19* (*Yr41*) and *YrP81* (<http://wheat.pw.usda.gov>, McIntosh et al. 2010). *Yr27* is present in

common wheat cv. 'Selkirk' (McDonald et al. 2004) and also occurs in a range of lines distributed by CIMMYT and ICARDA. *Yr31* is present in common wheat 'Pastor' (McIntosh et al. 2008). *YrSte* was characterized in the variety 'Stephen' (Luo et al. 2005). *YrSp* in 'Spalding Prolific' was associated with the microsatellite marker *Xwmc441* at a distance of 12.1 cM (Guan et al. 2005). *YrTp1* in wheat line 'A-3' was closely linked with *Xwmc477*/0.4 cM (Yin et al. 2006). *YrCN19* is derived from the Chinese landrace 'Qian 440' and cosegregates with the SSR marker *Xgwm410/391* (Luo et al. 2005). *YrP81* comes from 'Xu29' (ICARDA, Syria, pedigree: Maya S/Sap S) and has been mapped on 2BS with the marker order being *Xgwm429*-1.8 cM-*YrP81*-4.1 cM-*Xwmc770* (Pu et al. 2010). *Yr27*, *Yr31* and *YrSte* are not effective against race CYR32 (Luo et al. 2005; Pu et al. 2010). Based on our observations in the field, *YrCN19* and *YrP81* have been overcome by CYR33 and a new pathotype V26 (data not shown). The SSR markers *Xwmc441* and *Xwmc477*, linked with *YrSp* and *YrTp1*, respectively, were not polymorphic among the parents (C51 and X440) and bulks. From the origin, resistant reaction to races tested, and SSR loci on 2BS, *YrC51* is most likely a new stripe rust resistance gene.

The RGAP technique is an effective method for identifying molecular markers for disease resistance genes (Wen et al. 2008). However, these need to be converted to STS



Figure 5. Validation of the STS marker *C51STS-4* with six wheat lines carried *Yr* genes, three cultivars in Sichuan, lines derived from crosses with C51 and susceptible lines. M, marker; 1, C51; 2, X440; 3, 99-1 (*Yr5*); 4, 99-2 (*Yr10*); 5, 99-3 (*Yr15*); 6, (*Yr24*); 7, 99-8 (*Yr26*); 8, 02-08 (*Yr27*); 9, Chuanmai42 (*Yr26*); 10, Chuanmai104 (*Yr26*, +); 11, 3-02-1 (*YrC51*); 12, 13-03-1 (*YrC51*); 13, 13-06-1 (*YrC51*); 14, Chuanmai28; 15, 20-108 (Taichuang29); 16, 98-105 (Mingxian169); 17, 99-18 (Avocet S).

Table 5. Infection type produced by PST CYR 33 on wheat genotypes and polymorphism of molecular markers flanking the *YrC51* locus in these genotypes.

Wheat genotype	IT	Presence (+) and absence (-) of markers		
		<i>Xgwm429</i>	<i>C51R2</i>	<i>C51STS-4</i>
C51 (<i>YrC51</i>)	0	+	+	+
X440	4	-	-	-
Yr5/6*Avocet S (<i>Yr5</i>)	0;	-	-	-
Yr10/6*Avocet S (<i>Yr10</i>)	2	-	-	-
Yr15/6*Avocet S (<i>Yr15</i>)	1	-	-	-
Yr24/3*Avocet S (<i>Yr24</i>)	3	+/-	-	-
Yr26/3*Avocet S (<i>Yr26</i>)	3	-	-	-
Yr27/3*Avocet S (<i>Yr27</i>)	4	-	-	-
SynCD768/SW3243//Chuan6415 (<i>Yr26</i>)	3	-	-	-
Chuanmai42/ Chuanong16 (<i>Yr26</i> ,+)	0	+	+	-
Chuanmai28/C51 F _{3:4} (<i>YrC51</i>)	0	+	-	+
Taichuang29/C51 F _{3:4} (<i>YrC51</i>)	0	+	+	+
Mingxian169/C51 F _{3:4} (<i>YrC51</i>)	0;	+	+	+
Chuanmai28	4	-	-	-
Taichung29	4	-	-	-
Mingxian169	4	-	+	-
Avocet S	4	-	-	-

markers for use in a breeding context. In the present study, three RGAP markers closely linked to *YrC51* were identified and successfully converted into STS markers. Among them, STS marker C51STS-4 effectively identified the *YrC51* in different genetic backgrounds (figure 5).

In conclusion, a new stripe rust resistance gene on 2BS in wheat line C51 was identified and a closely linked STS marker C51STS-4 was obtained. The closely linked markers C51STS-4, *Xgwm429* and C51R2 to *YrC51* will greatly facilitate the pyramiding of *YrC51* with other resistance genes.

Acknowledgements

This work was supported by the National Natural Science Foundation of China (31071411), the National Key Technologies R&D Programme (2011BAD35B03), the Youth Foundation of Sichuan (2012JQ0066), the National 863 Plan (2011AA10A106) and the Excellent Paper Foundation of Sichuan Academy of Agricultural Science (2011-3).

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Received 29 September 2013, accepted 12 March 2014

Published online: 18 August 2014