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Cytogenetic and Molecular Identification of a New Wheat-*Thinopyrum intermedium* Addition Line with Resistance to Powdery Mildew

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Thinopyrum intermedium, which has many useful traits, is valuable for wheat breeding. A new wheat-*Thinopyrum* addition line, SN100109, was developed from the progeny of common wheat cultivar Yannong 15 and *Th. intermedium*. It was resistant to most races of *Blumeria graminis* f. sp *tritici* (*Bgt*), which caused powdery mildew in wheat, and its reactions were different from the reactions of gene *Pm40* and *Pm43*. Genomic *in situ* hybridization (GISH) and molecular marker analysis were used to identify the genomic composition of SN100109. GISH results showed that SN100109 was a wheat-*Th. intermedium* disomic addition line containing one pair of J chromosomes, and the resistance gene was located on the alien additional chromosomes of SN100109. And four molecular markers *BE425942*, *BF482714*, *Xgdm93* and *BV679214* which were assigned to homologous group 2, were specific molecular markers of the additional chromosomes. All the results indicated that SN100109 contained one pair of 2J chromosomes. SN100109 can be used as a novel germplasm source for introducing powdery mildew resistance genes to wheat in breeding programs.

Keywords: *Thinopyrum intermedium*, addition line, powdery mildew, genomic *in situ* hybridization, molecular markers

Introduction

Thinopyrum intermedium is a perennial hexaploid species (2n = 42, $E^eE^eE^bE^bStSt$ or JJ^sJ^sStSt), carrying many potentially favorable traits for wheat improvement including high resistance to wheat leaf rust, stripe rust, stem rust, powdery mildew and eyespot; immunity to smut, leaf blight, root rot, and yellow dwarf, and stripe mosaic viruses; and tolerance to low temperature, salinity, and drought (Friebe et al. 1996; Li et al. 2005; Li and Wang 2009; Li et al. 2012; Zeng et al. 2013). All of these characters make this species a potential source of gene pool for wheat improvement. To date, various addition, substitution, and translocation lines have been developed and used to transfer useful genes into wheat as a bridge parent. Genes for resistance to barley yellow dwarf virus (Zhang et al.

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1999, 2009; Ohm et al. 2005; Lin et al. 2007; Kong et al. 2009; Ayala-Navarrete et al. 2010; Wang et al. 2010), wheat stripe mosaic virus (Friebe et al. 1991; Chen et al. 1999), and wheat leaf rust, stripe rust, and stem rust (Friebe et al. 1996; Tang et al. 2000; Liu et al. 2013) have been successfully transferred to wheat.

Wheat powdery mildew, caused by *Blumeria graminis* f. sp *tritici* (*Bgt*), is one of the most serious wheat diseases. It reduces kernel weight and damages grain quality. With the promotion of semi-dwarf wheat varieties, powdery mildew is increasingly prevalent in the main wheat production area of China and has become a major obstacle to wheat production (Wang et al. 2001). Until now, 67 resistance genes or alleles at 46 loci have been designated (*Pm1 – Pm50, Pm18 = Pm1c*, *Pm22 = Pm1e*, *Pm23 = Pm4c*, and *Pm31 = Pm21*) (McIntosh et al. 2011; Mohler et al. 2013). Of these genes or alleles, 31 genes were derived from common wheat and the remaining were derived from wild relatives. Among them, genes *Pm40* (Luo et al. 2009) and *Pm43* (He et al. 2009) were derived from *Th. intermedium*. However, most genes become ineffective within a short period of use in agriculture because of rapid changes in the virulence of pathogen populations (Hsam and Zeller 2002). For this reason, discovering and deploying new genes for resistance to powdery mildew are of great significance for the genetic improvement in resistance to wheat powdery mildew.

In studies of wheat genetic improvement using *Th. intermedium* as valuable resources, our group have bred 12 octaploid amphiploid lines (Liu et al. 2005a; Wang et al. 2006; Bao 2010) and various addition lines (Zhao et al. 2005; Lin et al. 2005) as well as substitution lines (Liu et al. 2005b). In the present study, an alien disomic addition line, SN100109, was developed from a cross between wheat cultivar Yannong 15 and *Th. intermedium*. It was resistant to most powdery mildew *Bgt* races. Genomic *in situ* hybridization (GISH) and molecular marker analysis were conducted in this study in order to identify the genomic composition of SN100109.

Materials and Methods

Plant materials

Materials included the common wheat cultivars Yannong 15, and Huixianhong; *Th. intermedium* (Host) (2n = 42, JJJ^{s}J^{s}StSt, PI547333); the resistant powdery mildew addition line SN100109, an individual from BC_2F_3 progeny of the hybrid between common wheat cultivar Yannong 15 and *Th. intermedium* by cytology method; a SN100109/Huixianhong F_2 population (327 individuals); *Th. elongatum* (Host, Á. Löve) (2n = 14, EE, PI380626); *Th. bessarabicum* (2n = 14, JJ, PI531711); *Pseudoroegneria strigosa* (Á. Löve) (2n = 14, StSt, Z2774); the wheat cultivar Chinese Spring; Chinese Spring-*Th. elongatum* disomic addition lines (DA1E-DA7E); and Chinese Spring-*Th. bessarabicum* disomic addition lines (DA1J–DA7J). Among these lines, *Th. intermedium* was provided by Prof. Zhensheng Li, Institute of Genetics and Developmental Biology, Chinese Academy of Sciences; *Ps. Strigose* was provided by Prof. Lihui Li, the Institute of Crop Sciences, the Chinese Academy of Agricultural Sciences (CAAS); *Th. bessarabicum* was provided by

Prof. Zengjun Qi, State Key Laboratory of Crop Genetics and Germplasm Enhancement, Nanjing Agricultural University, China; Chinese Spring-*Th. elongatum* disomic addition lines (DA1E–DA7E) were kindly provided by Dr. J. Dvorak, University of California, Davis, CA, USA; and Chinese Spring-*Th. bessarabicum* disomic addition lines (DA1J–DA7J) were provided by International Maize and Wheat Improvement Center (CIMMYT). The other lines were obtained from our laboratory.

Evaluation of resistance to powdery mildew

Seedling reactions to 23 *Bgt* races of SN100109 with 42 lines as control were assessed by the Institute of Plant Protection, CAAS. The *Bgt* races used were as follows: E01, E02, E05, E06, E07, E09, E11, E13, E15, E16, E17, E18, E20, E21, E23-1, E23-2, E26, E30, E30-1, E30-2, E31, E32, E49, and E50. And the 42 lines used were as follows: Chancellor(–), Axminster/8cc (*Pm1a*), Ulka/8cc (*Pm2*), Maris Huntsman (*Pm2+6*), Maris Dove (*Pm2+MLD*), Asosan/8cc (*Pm3a*), Chul/8cc (*Pm3b*), Sonora/8cc (*Pm3c*), Kolibri (*Pm3d*), W150 (*Pm3e*), Mich. Amber/8cc (*Pm3f*), Khapli/8cc (*Pm4a*), Armada (*Pm4b*), Hope/8cc (*Pm5a*), Aquila (*Pm5b*), Mission (*Pm4b+5b*), Baimian3 (*Pm4+8*), Coker 983 (*Pm5+6*), Timgalen (*Pm6*), Coker 747 (*Pm6*), CI14189 (*Pm7*), Kavkaz (*Pm8*), Normandie (*Pm1+2+9*), Wembley (*Pm12*), R4A (*Pm13*), Brigand (*Pm16*), Amigo (*Pm17*), MIN (*Pm18=Pm1c*), XX186 (*Pm19*), TAM104/THATCHER (*Pm20*), Yangmai5/Sub.6V (*Pm21*), Virest (*Pm22 = Pm1e*), 817241 (*Pm23 = Pm4c*), Chiyacao (*Pm24*), NCA5 (*Pm25*), 5P27 (*Pm30*), NCD7 (*Pm34*), NCD3 (*Pm35*), NCA4 (*Pmnon-1*), Xiaobaidongmai (*PmXBD*), GRY19 (*Pm40*), CH5025 (*Pm43*).

The reactions to powdery mildew of the F_2 population from the progeny of resistant line SN100109 and susceptible line Huixianhong were evaluated in the greenhouse at Shandong Agricultural University, with Huixianhong as a susceptible control and infection line. Pathogen inoculation of *Bgt* race E09 for F_2 population was carried out by dusting *Bgt* conidia on the infection line when they were on jointing-stage. When Huixianhong was fully infected, the resistance reaction of the parents and the F_2 population to powdery mildew were rated according to a six-level (0, 0;, 1, 2, 3, 4) scale of Baoqin Sheng (Qi et al. 2010), in which types 0, 0;, 1, and 2 were resistant to powdery mildew and types 3 and 4 were susceptible. The physiological *Bgt* E09 was provided by Dr. Hongjie Li, Institute of Crop Sciences, CAAS, Beijing, China.

Genome in situ *hybridization (GISH)*

Total genomic DNA of *Ps. strigosa* was labeled with ChromaTide® Alexa Fluor® 488-5-dUTP (Invitrogen, USA) by the nick translation method. Total genomic DNA of Yannong 15 was used for blocking. Detailed procedures of the chromosome preparation and hybridization mixture were according to the methods by Bao et al. (2009) with a little modified. 4′,6-Diamidino-2-phenylindole (DAPI) was used to counter-stain the wheat chromosomes to blue. Images were taken using an Olympus BX-61 fluorescence microscope equipped with a CCD (DS-Ri1, Nikon, Japan) camera.

Molecular marker analysis

DNA of young leaves of plant was extracted by the SDS-phenol method (Devos et al. 1993). A total of 557 G-SSR, EST-SSR, and STS molecular markers from E and J genomes were screened by our laboratory. The molecular markers and SSR methods followed the methods described in the study by Cui et al. (2012). The reaction volume used was 10 μℓ. G-SSR, EST-SSR and STS amplification were carried out in a T100™ Thermal Cycler (BIO-RAD, USA). PCR products were separated on 8% non-denaturing polyacrylamide gel (39:1). The gels were stained with 0.2% silver nitrate and colored with 3% NaOH solution.

Results

Characteristics of SN100109 resistance to powdery mildew

Using Chancellor as susceptible control, the reactions to 23 *Bgt* races in SN100109 were different from the reactions in 41 accessions with known powdery mildew genotypes (Table S1*). The results showed that SN100109 was immune to E01, E18, E23-2, E30-1, and E32; and highly resistant to E09, E17, E21, E23-1, E26, E31, E49, and E50; and resistant to E02, E13, and E16. Compared with *Pm40* (Luo et al. 2009) and *Pm43* (He et al. 2009), which also derived from *Th. intermedium*, SN100109 and GRY19 (*Pm40*) had different resistance reactions to *Bgt* races E05, E06, E07, E16, and E21. The resistance reactions to *Bgt* races E05, E06, E07, E11, E15, and E31 between SN100109 and CH5025 (*Pm43*) were also different. Therefore, SN100109 showed different reactions to the powdery mildew races than other accessions with known powdery mildew genotypes.

GISH analysis of SN100109

Analysis of root tip cells proved that SN100109 had the chromosome number with 2n = 44. Investigation of the pollen mother cells showed that most observed cells of SN100109 had 22 bivalents at meiotic metaphase I, which indicated that this line was cytological stable.

GISH analysis using labeled genomic DNA of *Ps. strigosa* as probe and genomic DNA of Yannong 15 as block were conducted to the root tip cells and poll mother cells of SN100109. The results of mitotic metaphase I and meiotic metaphase I cells showed that two chromosomes of SN100109 presented greenish-fluorescence hybridization signals in their terminal regions (Figs 1A and 1B), while the remaining 42 chromosomes showed a uniform blue fluorescence of DAPI. The results of meiotic metaphase I showed the same result, which revealed that SN100109 had 42 chromosomes from common wheat plus one pair of J chromosomes. The cytological study and the GISH results proved that SN100109 was a wheat-*Th. intermedium* disomic addition line.

* Further details about the Electronic Supplementary Material (ESM) can be found at the end of the article.

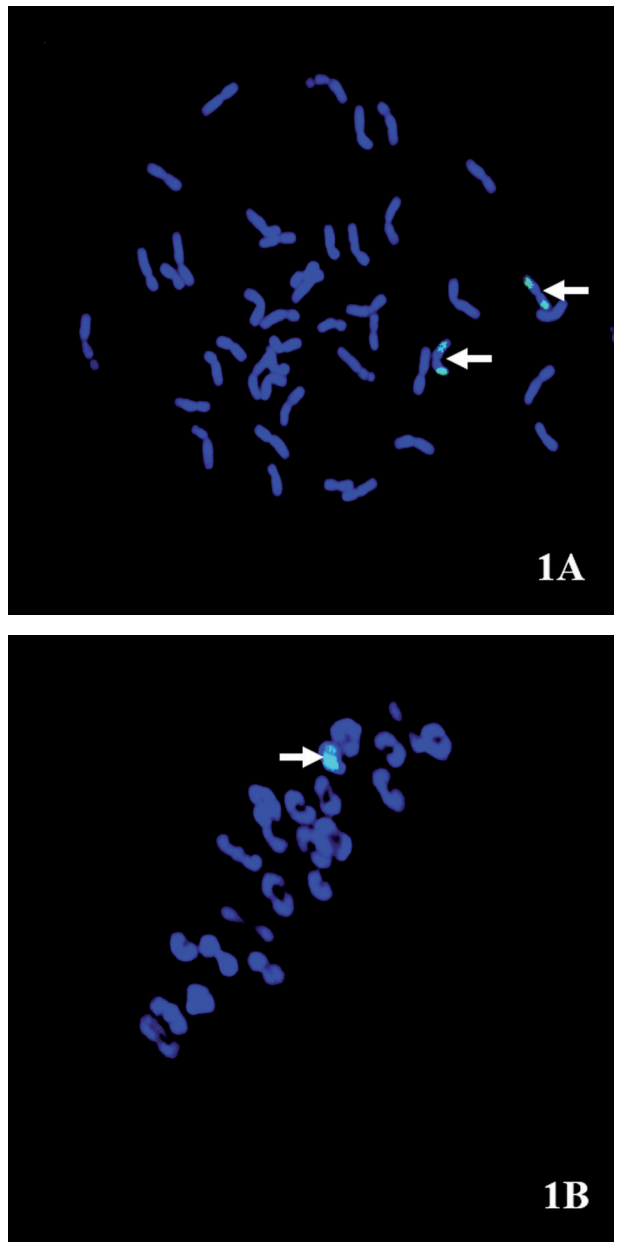

Figure 1. GISH patterns of root tip cell (A) and pollen mother cell (B) of SN100109 (2n = 22II), using labeled genomic DNA of *Ps. strigosa* as probe and genomic DNA of Yannong 15 as block. Arrows indicate that one pair of J chromosomes from *Th. intermedium* were added

Molecular markers specific to the additional chromosomes

A set of 557 molecular markers were applied to reveal amplified polymorphisms in *Th. intermedium*, SN100109, Yannong15 and Chinese Spring. Four markers (*BE425942*, *BF482714*, *BV679214*, and *Xgdm93*) amplified unique bands in *Th. intermedium* and SN100109, these unique bands were 380, 400, 640, and 100 bp in length. Figure 2 presents the amplification products of *BE425942*.

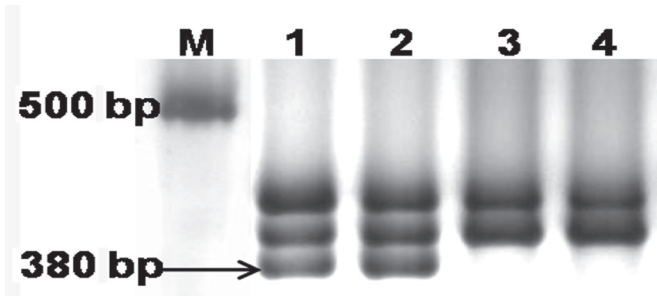

Figure 2. Amplification products of *BE425942* in *Th. intermedium*, SN100109, Yannong 15, Chinese Spring
M: DL2000 marker; 1: *Th. intermedium*; 2: SN100109; 3: Yannong 15; 4: Chinese Spring

In order to attribute these four markers to specific homologous groups, Chinese Spring-*Th. elongatum* disomic addition lines (DA1E–DA7E) and Chinese Spring-*Th. bessarabicum* disomic addition lines (DA1J–DA7J) were used to assign these markers. The results indicated that the unique bands of four markers ($BE425942_{380}$, $BF482714_{400}$, $BV679214_{640}$ and $Xgdm93_{100}$) were present in *Th. intermedium*, *Th. elongatum*, and DA2E (Fig. 3). Thus, the four markers were assigned to homologous group 2, and the additional chromosomes of SN100109 were coming from 2J chromosomes.

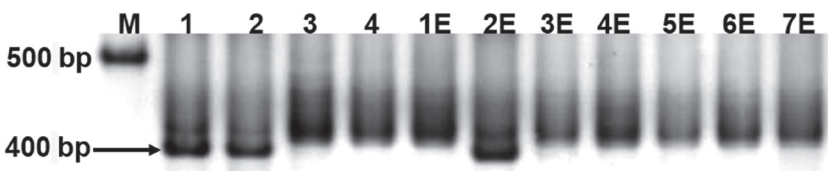

Figure 3. PCR amplification pattern of marker *BF482714* on *Th. intermedium*, *Th. elongatum*, *Th. bessarabicum*, Chinese Spring and wheat-*Th. elongatum* addition lines
M: DL2000 marker; 1: *Th. intermedium*; 2: *Th. elongatum*; 3: *Th. bessarabicum*; 4: Chinese Spring; 1E: DA1E; 2E: DA2E; 3E: DA3E; 4E: DA4E; 5E: DA5E; 6E: DA6E; 7E: DA7E

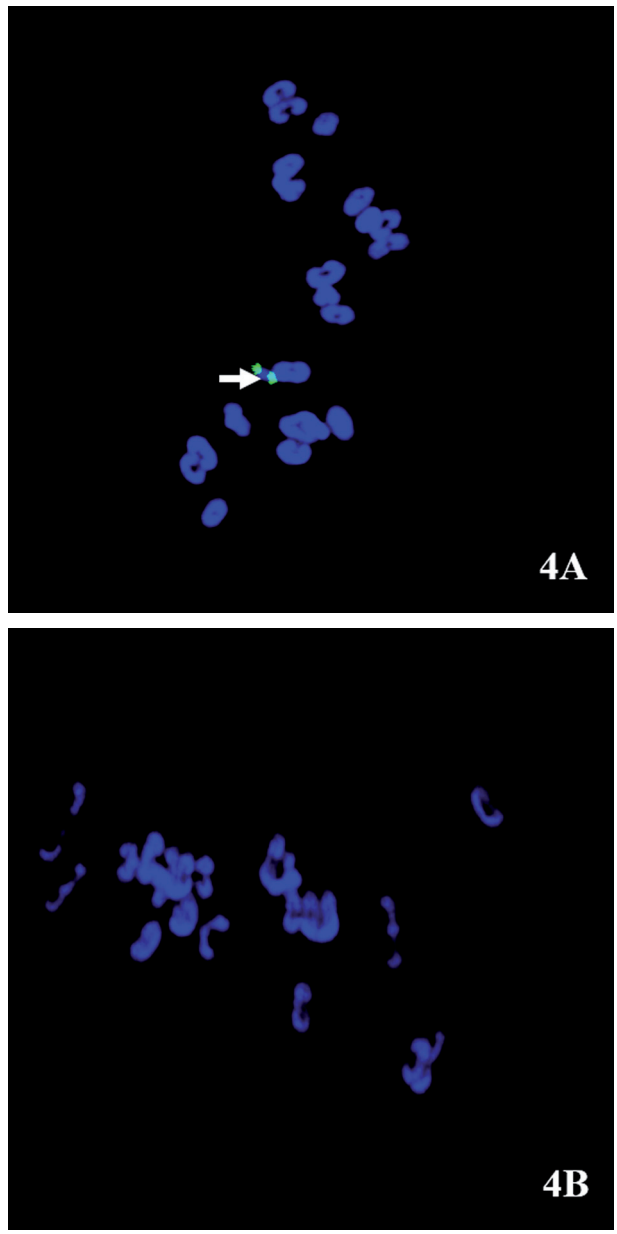

Figure 4. GISH patterns of pollen mother cells of typical resistant and susceptible plants from the F_2 population of SN100109 and Huixianhong. A: GISH pattern of a pollen mother cell of the typical resistant plant, arrow indicate that one J chromosome from *Th. intermedium* was added, B: GISH pattern of a pollen mother cell of the typical susceptible individual, no *Th. intermedium* chromosome was observed

Location of the resistance gene on the chromosomes of SN100109

Under greenhouse condition, SN100109 was almost immune to *Bgt* race E09 and Yannong 15 was highly susceptible while Huixianhong was used as the susceptible control. Among the SN100109/Huixianhong F_2 segregation population (327 individuals), 161 were resistant plants and 166 were susceptible plants.

Fifty plants from F_2 population were selected for the powdery mildew evaluation and GISH analysis. Among them, 21 plants showed typical resistant while the other 29 plants showed typical susceptible. Meiotic analysis showed that among the 21 resistant plants, 8 plants had a chromosome configuration of 2n = 22II = 44 while other 13 plants had 2n = 21II + I = 43. Moreover, the analysis revealed that all 29 susceptible plants had a chromosome configuration of 2n = 21II = 42 (Table S2).

GISH analysis using *Ps. strigosa* genomic DNA as probe were conducted to pollen mother cells from 21 typical resistant plants and 29 susceptible plants in order to identify their chromosome composition. The GISH results showed that all the resistant plants carried 1 or 2 chromosomes from *Th. intermedium* (Fig. 4A), while all the susceptible plants carried no *Th. intermedium* chromosomes (Fig. 4B). The phenotypes for powdery mildew resistance in typical plants matched the results of GISH, suggesting that the resistance gene in SN100109 lies on the additional chromosomes.

Discussion

Analysis of genomic composition of SN100109

Using GISH with St genomic DNA as probe, the chromosome composition and structure of *Th. intermedium* in the wheat background were clearly distinguished (Chen et al. 2003, 2005; Bao et al. 2009). About the genomic composition of *Th. intermedium*, Chen et al. (1998) had used GISH with St genomic DNA as probe to redesignate it as JJJ^sJ^sSS. They pointed out that when *Th. intermedium* chromosomes were hybridized with probed St genomic DNA from *Ps. strigosa* in the S-genome chromosomes were labeled uniformly along their entire chromosome; the J-genome chromosomes showed fluorescence signals only in the terminal regions, whereas J^s-genome chromosomes were intensively labeled in the centromere regions, occasionally, in the terminal regions. They also pointed out that the S genome was homologous to the St genome of *Ps. strigosa*; the J-genome chromosomes were related to the E genome of *Th. elongatum* and the J genome of *Th. bessarabicum*; whereas J^s-genome chromosomes were referred to modified E or J type chromosomes. In this study, GISH results showed that the alien additional chromosomes in SN100109 were J-genome chromosomes of *Th. intermedium*. Then four molecular markers *BE425942*, *BF482714*, *Xgdm93* and *BV679214* which were specific molecular markers of the additional chromosomes of SN100109, were assigned to homologous group 2 by the two sets of disomic addition lines (DA1E–DA7E and DA1J–DA7J). All the result indicated that the additional chromosomes in SN100109 were one pair of 2J chromosomes.

Resistance to powdery mildew in SN100109

The line SN100109 was developed from the BC_2F_3 progeny of Yannong 15 and *Th. intermedium*. Greenhouse and field test results showed that SN100109 was almost immune to powdery mildew and its resistance to powdery mildew was stable. A SN100109/Huixianhong F_2 population (327 individuals) in this study was used to locate the resistance gene to specific chromosome. Although we selected a handful of plants from F_2 population, the phenotypes for resistance to powdery mildew in typical individuals matched the results of GISH. The results infer that the resistance gene in SN100109 lies on the additional 2J chromosomes. Gene *Pm40* (Luo et al. 2009) and *Pm43* (He et al. 2009) also derived from *Th. intermedium*. However, *Pm40* had been assigned to 7BS chromosome of wheat (Luo et al. 2009) and originated from one J^s chromosome of *Th. intermedium* (Chang 1999); *Pm43* had been assigned to 2DL chromosome of wheat and also originated from one J^s chromosome of *Th. intermedium* (He et al. 2009). Furthermore, the reactions to 23 *Bgt* races in SN100109, GRY19 (*Pm40*), and CH5025 (*Pm43*) had different resistance reactions. According to the above results, we infer that the powdery mildew resistance gene in SN100109 is a novel resistance gene from *Th. intermedium*.

In this study, SN100109 was identified to one wheat-*Th. intermedium* disomic addition line containing one pair of 2J chromosomes, and the resistance gene was located on the alien additional chromosomes. Four special molecular markers lying on the additional chromosomes of SN100109 can be effectively used to track and select for the chromosomes with the resistance gene to powdery mildew. Chromosome substitution and translocation lines are currently being developed to transfer powdery mildew resistance gene in SN100109, and whether or not the powdery mildew resistance gene in SN100109 is a single dominant gene for resistance also needs further confirmation.

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References

Ayala-Navarrete, L., Thompson, N., Ohm, H. 2010. Molecular markers show a complex mosaic pattern of wheat-*Thinopyrum intermedium* translocations carrying resistance to YDV. Theor. Appl. Genet. **121**:961–970.

Bao, Y.G., Li, X.F., Liu, S.B., Cui, F., Wang, H.G. 2009. Molecular cytogenetic characterization of a new wheat-*Thinopyrum intermedium* partial amphiploid resistant to powdery mildew and stripe rust. Cytogenet. Genome Res. **126**:390–395.

Bao, Y.G. 2010. Molecular cytogenetic identification and hereditary analysis of wheat-*Thinopyrum intermedium* alien chromosome lines. Dissertation. Shandong Agricultural University. Shandong, China. (in Chinese with English summary)

Chang, Z.J. 1999. Production and Molecular Cytogenetic Characterization of Several *Thinopyrum intermedium* Derived Wheat Germplasm Lines. PhD Dissertation. Sichuan Agricultural University. Chengdu, China. pp. 13–88. (in Chinese with English abstract)

Chen, Q., Conner, R.L., Laroche, A., Thomas, J.B. 1998. Genome analysis of *Thinopyrum intermedium* and *Thinopyrum ponticum* using genomic *in situ* hybridization. Genome **41**:580–586.

Chen, Q., Conner, R.L., Laroche, A., Fedak, G., Thomas, J.B. 1999. Genomic origins of *Thinopyrum* chromosomes specifying resistance to wheat streak mosaic virus and its vector, *Aceria tosichella*. Genome **42**:289–295.

Chen, Q., Conner, R.L., Li, H.J., Sun, S.C., Ahmad, F., Laroche, A., Graf, R.J. 2003. Molecular cytogenetic discrimination and reaction to wheat streak mosaic virus and the wheat curl mite in Zhong series of wheat – *Thinopyrum intermedium* partial amphiploids. Genome **46**:135–145.

Chen, Q. 2005. Detection of alien chromatin introgression from *Thinopyrum* into wheat using S genomic DNA as a probe–A landmark approach for *Thinopyrum* genome research. Cytogenet. Genome Res. **109**:350–359.

Cui, F., Ding, A.M., Li, J., Zhao, C.H., Wang, L., Wang, X.Q., Qi, X.L., Li, X.F., Li, G.Y., Gao, J.R., Wang, H.G. 2012. QTL detection of seven spike-related traits and their genetic correlations in wheat using two related RIL populations. Euphytica **186**:177–192.

Devos, K.M., Millan, T., Gale, M.D. 1993. Comparative RFLP maps of the homeologous group-2 chromosomes of wheat, rye and barley. Theor. Appl. Genet. **85**:784–792.

Friebe, B., Mukai, Y., Dhaliwal, H.S., Martin, T.J., Gill B.S. 1991. Identification of alien chromatin specifying resistance to wheat streak mosaic and greenbug in wheat germplasm by C-banding and *in situ* hybridization. Theor. Appl. Genet. **81**:381–389.

Friebe, B., Jiang, J., Raupp, W.J., McIntosh, R.A., Gill, B.S. 1996. Characterization of wheat-alien translocations conferring resistance to diseases and pests: current status. Euphytica **91**:59–87.

He, R.L., Chang, Z.J., Yang, Z.J., Yuan, Z.Y., Zhan, H.X., Zhang, X.J., Liu, J.X. 2009. Inheritance and mapping of powdery mildew resistance gene *Pm43* introgressed from *Thinopyrum intermedium* into wheat. Theor. Appl. Genet. **118**:1173–1180.

Hsam, S.L.K., Zeller, F.J. 2002. Breeding for powdery mildew resistance in common wheat (*Triticum aestivum* L.). In: Belanger, R.R., Bushnell, W.R., Dik, A.J., Carver, T.L.W. (eds), The Powdery Mildews, a Comprehensive Treatise. APS Press. St. Paul, MN, USA. pp. 219–238.

Kong, L., Anderson, J.M., Ohm, H.W. 2009. Segregation distortion in common wheat of a segment of *Thinopyrum intermedium* chromosome 7E carrying *Bdv3* and development of a *Bdv3* marker. Plant Breeding **128**:591–597.

Li, H., Wang, X. 2009. *Thinopyrum ponticum* and *Th. intermedium*: the promising source of resistance to fungal and viral diseases of wheat. J. Genet. Genomics **36**:557–565.

Li, H.J., Arterburn, M., Jones, S.S., Murray, T.D. 2005. Resistance to eyespot of wheat, caused by *Tapesia yallundae*, derived from *Thinopyrum intermedium* homoeologous group 4 chromosome. Theor. Appl. Genet. **111**:932–940.

Li, H.J, Cui, L., Li, H.L., Wang, X.M., Murray, T.D., Conner, R.L., Wang, L.J., Gao, X., Sun, Y., Sun, S.C., Tang, W.H. 2012. Effective resources of wheat and wheat-*Thinopyrum* derivatives for resistance to *Heterodera filipjevi* in China. Crop Sci. **52**:1209–1217.

Lin, X.H., Wang, L.M., Li, X.F., Zhao, F.T., Gao, J.R., Li, W.C., Lu, W.H., Wang, H.G. 2005. Identification of wheat-*Elytrigia intermedium* alien disomic addition line with powdery mildew resistance. Acta Phytopathologica Sinica **35**:60–65. (in Chinese with English summary)

Lin, Z.S., Cui, Z.F., Zeng, X.Y., Ma, Y.Z., Zhang, Z.Y., Nakamura, T., Ishikawa, G., Nakamura, K., Yoshida, H., Xin, Z.Y. 2007. Analysis of wheat-*Thinopyrum intermedium* derivatives with BYDV resistance. Euphytica **158**:109–118.

Liu, J., Chang, Z.J., Zhang, X.J., Yang, Z.J., Li, X., Jia, J.Q., Zhan, H.X., Guo, H.J., Wang, J.M. 2013. Putative *Thinopyrum intermedium*-derived stripe rust resistance gene *Yr50* maps on wheat chromosome arm 4BL. Theor. Appl. Genet. **126**:265–274.

Liu, S.B., Wang, H.G., Zhang, X.Y., Li, X.F., Li, D.Y., Duan, X.Y., Zhou, Y.L. 2005a. Molecular cytogenetic identification of a wheat-*Thinopyrum intermedium* (Host) Barkworth & DR Dewey partial amphiploid resistant to powdery mildew. J. Integr. Plant Biol. **47**:726–733.

Liu, S.B., Wang, H.G. 2005b. Characterization of a wheat-*Thinopyrum intermedium* substitution line with resistance to powdery mildew. Euphytica **143**:229–233.

Luo, P.G., Luo, H.Y., Chang, Z.J., Zhang, H.Y., Zhang, M., Ren, Z.L. 2009. Characterization and chromosomal location of *Pm40* in common wheat: a new gene for resistance to powdery mildew derived from *Elytrigia intermedium*. Theor. Appl. Genet. **118**:1059–1064.

McIntosh, R.A., Dubcovsky, J., Rogers, W.J., Morris, C., Appels, R., Xia, X.C. 2011. Catalogue of gene symbols for wheat. http://www.wheat.pw.usda.gov

Mohler, V., Bauer, C., Schweizer, G., Kempf, H., Hartl, L. 2013. *Pm50*: a new powdery mildew resistance gene in common wheat derived from cultivated emmer. J. Appl. Genetics **54**:259–263.

Ohm, H.W., Anderson, J.M., Sharma, H.C., Ayala, N.L., Thompson, N., Uphaus, J.J. 2005. Registration of yellow dwarf virus resistant wheat germplasm line P961341. Crop Sci. **45**:805–806.

Qi, X.L., Cui, F., Yu, L., Ding, A.M., Li, J., Chen, G.L., Wang, H.G. 2010. Molecular tagging wheat powdery mildew resistance gene *Pm21* by EST-SSR. Mol. Plant Breeding 2010 Vol. 1. No. 4. DOI:10.5376/mpb.2010.01.0004. http://mpb.sophiapublisher.com

Tang, S., Li, Z., Jia, X., Larkin, P.J. 2000. Genomic *in situ* hybridization (GISH) analyses of *Thinopyrum intermedium*, its partial amphiploid Zhong 5, and disease resistant derivatives in wheat. Theor. Appl. Genet. **100**:344–352.

Wang, H.G., Liu, S.B., Li, X.F., Gao, J.R., Feng, D.C., Chen, D.H. 2006. Breeding and identification of six octoploid *Trititrigia*. J. Triticeae crops **26**:6–10. (in Chinese with English summary)

Wang, M.J., Zhang, Y., Lin, Z.S., Ye, X.G., Yuan, Y.P., Ma, W., Xin, Z.Y. 2010. Development of EST-PCR markers for *Thinopyrum intermedium* chromosome 2Ai#2 and their application in characterization of novel wheat-grass recombinants. Theor. Appl. Genet. **121**:1369–1380.

Wang, X.Y., Chen, P.D., Zhang, S.Z. 2001. Pyramiding and marker-assisted selection for powdery mildew resistance genes in common wheat. Acta Genetica Sinica **28**:640–646. (in Chinese with English summary)

Zeng, J., Cao, W., Fedak, G., Sun, S., Mccallum, B., Fetch, T., Xue, A., Zhou, Y. 2013. Molecular cytological characterization of two novel durum-*Thinopyrum intermedium* partial amphiploids with resistance to leaf rust, stem rust and *Fusarium* head blight. Hereditas **150**:10–16.

Zhang, Z., Xin, Z., Ma, Y., Chen, X., Xu, Q., Lin, Z. 1999. Mapping of a BYDV resistance gene from *Thinopyrum intermedium* in wheat background by molecular markers. Sci. China Ser. C. **42**:663–668. (in Chinese with English summary)

Zhang, Z., Lin, Z., Xin, Z. 2009. Research progress in BYDV resistance genes derived from wheat and its wild relatives. J. Genet. Genomics **36**:567–573.

Zhao, F.T., Wang, L.M., Li, W.C., Lin, X.H., Li, X.F., Gao, J.R., Wang, H.G. 2005. Cultivation and identification of wheat-*Elytrigia intermedium* alien disomic addition lines. Acta Biologiae Experimentalis Sinica **38**:133–140. (in Chinese with English summary)

Electronic Supplementary Material (ESM)

Electronic Supplementary Material (ESM) associated with this article can be found at the website of CRC at http://www.akademiai.com/content/120427/

Electronic Supplementary *Table S1*. Evaluation of resistance of SN100109 to powdery mildew

Electronic Supplementary *Table S2*. Chromosome numbers and resistance of typical plants in F_2 population