

Alien introgressions represent a rich source of genes for crop improvement

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Our major crop species represent the relatively few species that were selected by our ancestors from a multitude of extant species, and the resulting narrow germplasm forms the basis of modern monoculture in many areas of the world. Crop monoculture over vast areas inevitably leads to boom and bust cycles alternating between susceptible and resistance genotypes, disease epidemics, and loss of productivity after the appearance of new virulent races of pathogens. Breeders and geneticists have increasingly sought new sources of resistance in diverse germplasm, often involving distant wild relatives. Often, such alien introgressions are accompanied by linkage drag where recombination is suppressed in the target gene region, and standard recombination-based approaches cannot be used in the molecular dissection of the target genes. In PNAS, Cao et al. (1), however, circumvent the problems and report the isolation of a resistance gene from an alien introgression in wheat using a combination of cytology and gene expression analysis.

As a rule, polyploids containing more than two sets of chromosomes have highly buffered genotypes, are more tolerant, and are more likely to benefit from alien introgression than diploids. Common bread wheat (*Triticum aestivum* L.; $2n = 6x = 42$, genomes AABBDD) is hexaploid, consisting of A, B, and D progenitor genomes. Most of the highest yielding wheat cultivars carry portions of alien chromosomal introgression from related weedy species. The process of alien introgression is illustrated in Fig. 1. The alien introgression T1BL-1RS, for example, resulted from the breakage of wheat chromosome 1BL-1BS (the long and short arms of chromosome 1 of the B genome) at the centromere and the replacement of the 1BS arm of wheat by the 1RS arm of rye. The 1RS arm in T1BL-1RS carries a battery of resistance genes specifying resistance to leaf rust (*Lr26*), stem rust (*Sr31*), stripe rust (*Yr9*), powdery mildew (*Pm8*) (2), and genes for adaptation to abiotic stresses, including a robust drought-tolerant root system (3). Because of linkage on the 1RS arm, the genes are inherited as a single supergene linkage block.

The *Pm21* gene, specifying broad-spectrum resistance to all known races of powdery mildew fungus *Blumeria graminis*

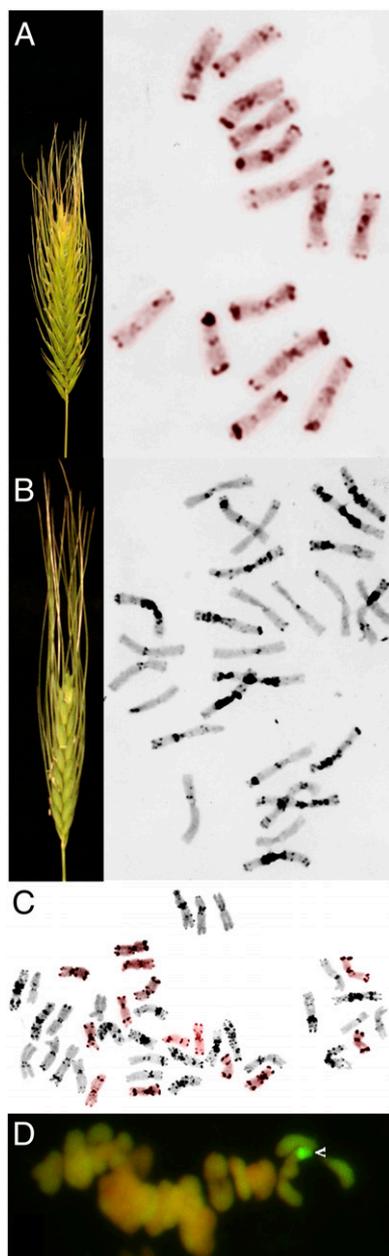


Fig. 1. The scheme of alien introgression used to transfer *Pm21* from *Dasyprum villosum* (syn *Haynaldia villosa*) to wheat. (A) *D. villosum* spike (Left) and C-banded chromosomes (Right; $2n = 2x = 14$, genome VV, size = 4,900 Mb). (B) Tetraploid wheat (*Triticum turgidum*) spike (Left) and C-banded chromosomes (Right; $2n = 4x = 28$, genome AABB, size = 11,200 Mb). (C) The amphiploid of *T. turgidum* × *D. villosum* ($2n = 6x = 42$) contains the complete chromosome complement of *T. turgidum* and *D. villosum* and is fertile. It was

f. sp. *tritici* (Bgt) of wheat and the second example of a whole-arm alien introgression with a high impact in agriculture, is from the distant wheat relative *Dasyprum villosum* (L.) Candargy (syn. *Haynaldia villosa*). In the alien introgression chromosome T6AL-6VS, the 6AS arm of wheat was replaced by the 6VS arm of *D. villosum*. In further evaluations, the 6VS arm in T6AL-6VS and others of independent origin have been shown to harbor other beneficial genes, including resistance to wheat curl mite, stripe rust, *Fusarium* head scab, and soil-borne mosaic virus (4). Plants heterozygous for *Pm21* and the T6AL-6VS segment do not pair with the normal 6AL-6AS in their short chromosome arms during meiosis (Fig. 1). Thus, all of the genes on the 6VS arm are inherited as a single linkage block.

The lack of pairing eliminates a simple marker-assisted chromosome walking strategy for the isolation of *Pm21*. Among the many possibilities for genetic analysis, the most obvious one would have been to survey *D. villosum* accessions for susceptibility to Bgt and map the gene in the parental source material. However, all collections available to Cao et al. (1) proved to be resistant. A feasible second strategy is to induce homologous recombination between wheat and alien chromosomes, but its frequency is low and often localized to specific regions of the alien introgression (5). Instead, Cao et al. (1) used microarray analysis to identify candidate genes induced on infection with Bgt. The *Pm21* gene was localized to a segment using radiation hybrid mapping.

backcrossed with bread wheat (*T. aestivum*; $2n = 6x = 42$, genome AABBDD), and wheat *D. villosum* translocation chromosome T6AL-6VS with *Pm21* was recovered. (D) In meiosis, the alien segment arrowhead does not pair or recombine with any wheat chromosome arm and thus is intractable to genetic analysis. The C-banding (11) was used to identify individual chromosomes, and *D. villosum* chromosomes are pseudocolored to distinguish them from wheat chromosomes.

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The cDNA clones mapping to the *Pm21* interval were used to pull out transformation-competent artificial chromosome (TAC) clones from the *Pm21* source *D. villosum* strain, and their correspondence to *Pm21* locus on T6AL-6VS was verified by FISH. Sequencing of the TAC clone *Stpk-V* (serine threonine protein kinase gene from *Haynaldia villosa*) identified a putative serine and threonine kinase gene, whose role in powdery mildew resistance was verified by transformation and virus-induced gene silencing (1).

In *Stpk-V*, Cao et al. (1) identify another member of the enigmatic receptor-like cytoplasmic kinases (RLCK), the most known being the R gene *Pto* (*Pseudomonas syringae* pv. tomato) of tomato. The resistance response to *Pto* is known to require a specific effector AvrPto (6). Other members of the RLCK group, including PBS1 of *Arabidopsis*, which is the target of the bacterial type III effector protease

AvrPhB and BIK1, interact with the plant immunity receptor kinase FLS2 (7, 8). RLCKs seem, therefore, to be signal transduction components of the pathogen-associated molecular pattern (PAMP)-triggered immunity (PTI) pathway, whereas some members are involved in effector-triggered immunity, which is the case with PTO and indirectly, PBS1 (9). STPK-V mediates a broad immunity to Bgt. Whether resistance to Bgt involves PTI or effector triggered immunity (ETI) because of virulence effectors present in all races of Bgt is unknown, and further characterization of the role played by STPK-V in fungal defense should provide more insight into the design of broad durable resistance.

Returning to the boom and bust cycles, the question of whether the alien genes are durable still remains. A large number of alien genes have been deployed in agriculture, but durability has not always

been realized (2). The *Sr31* gene of rye origin and widely deployed in global wheat production has recently been overcome by new stem rust races originating in Kenya and is now threatening the world wheat crop (10). *Pm21* has provided effective resistance to a broad spectrum of Bgt races in China and elsewhere since 2002. Rare genetic variation has been observed in a collection of 6V chromosomes of diverse origin for resistance to powdery mildew (4). The discovery of a 6V susceptible introgression stock opens the possibility of recombination-based analysis and further studies on the relationship between *Stpk-V* and *Pm21*. Certainly, the pioneering study of Cao et al. (1) provides impetus for the molecular dissection of many high-impact alien introgressions and the search for the many useful genes hiding in the weeds.

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