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Bacterial, Fungal, and Viral Disease Resistance Loci Mapped in a Recombinant Inbred Common Bean Population ('Dorado'/XAN 176)

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Abstract. Understanding the genomic associations among disease resistance loci will facilitate breeding of multiple disease resistant cultivars. We constructed a genetic linkage map in common bean (*Phaseolus vulgaris* L.) containing six genes and nine quantitative trait loci (QTL) comprising resistance to one bacterial, three fungal, and two viral pathogens of bean. The mapping population consisted of 79 $F_{5:7}$ recombinant inbred lines (RILs) derived from a 'Dorado'/XAN 176 hybridization. There were 147 randomly amplified polymorphic DNA (RAPD) markers, two sequence characterized amplified region (SCAR) markers, one intersimple sequence repeat (ISSR) marker, two seedcoat color genes *R* and *V*, the *Asp* gene conditioning seed brilliance, and two rust [*Uromyces appendiculatus* var. *appendiculatus* (Pers.:Pers) Unger] resistance genes: one conditioning resistance to Races 53 and 54 and the other conditioning resistance to Race 108. These markers mapped across eleven linkage groups, one linked triad, and seven linked pairs for an overall map length of 930 cM (Kosambi). Genes conditioning resistance to anthracnose (*Co-2*) [*Colletotrichum lindemuthianum* (Sacc. and Magnus) Lams.-Scrib.], bean rust (*Ur-5*), and bean common mosaic virus (*I* and *bc-3*) (BCMV) did not segregate in this population, but were mapped by inference using linked RAPD and SCAR markers identified in other populations. Nine previously reported quantitative trait loci (QTL) conditioning resistance to a variety of pathogens including common bacterial blight [*Xanthomonas campestris* pv. *phaseoli* (Smith) Dye], ashy stem blight [*Macrophomina phaseolina* (Tassi) Goid.], and bean golden mosaic virus (BGMV), were located across four linkage groups. Linkage among QTL for resistance to ashy stem blight, BGMV, and common bacterial blight on linkage group B7 and ashy stem blight, BGMV, and rust resistance loci on B4 will complicate breeding for combined resistance to all four pathogens in this population.

With recent advances in DNA marker technologies there has been a corresponding influx of studies in common bean (*Phaseolus vulgaris*) concerning genetic linkage maps (Adam-Blondon et al., 1994; Gepts et al., 1993; Jung et al., 1996; Nodari et al., 1993a; Vallejos et al., 1992), identification and characterization of quantitative trait loci (QTL) conditioning disease resistance (Ariyarathne et al., 1999; Jung et al., 1996; Nodari et al., 1993b), and development of diagnostic markers for indirect selection of monogenic resistance genes (Kelly and Miklas, 1998). Collectively, this work is leading to integration of resistance loci into a single core map for common bean (Freyre et al., 1998) with direct implications for breeding resistant cultivars. For instance, development of multiple disease resistant cultivars is often complicated by repulsion (*trans)* linkages among desired resistance genes and QTL used to control different pathogens in a particular production region (Kelly and Miklas, 1998). The development of an integrated map of resistance loci is just beginning, however, as many loci have yet to be mapped and the genomic relationships among several mapped loci are still unknown; thus, a comprehensive map of disease resistance loci in common bean remains a primary goal of breeders.

Bean breeders have concentrated on constructing mapping populations that segregate for many disease resistances, in hopes of understanding genomic relationships among desired resistance factors. Resistance genes for common bacterial blight (*Xanthomonas campestris* pv. *phaseoli*), web blight [*Thanatephorus cucumeris* (Frank) Donk], and rust (*Uromyces appendiculatus* var. *appendiculatus*) diseases limiting bean production in the Dominican Republic were localized on a single map (Jung et al., 1996). This paper describes how a single population was used to detect linkages among mapped loci

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conditioning resistance to six major diseases of common bean, one bacterial (common bacterial blight), three fungal [anthracnose (*Colletotrichum lindemuthianum*), ashy stem blight (*Macrophomina phaseolina*) and rust], and two viral [bean common mosaic virus (BCMV) and bean golden mosiac virus (BGMV)]. Various combinations of these diseases reduce production and seed quality of a fundamental food staple worldwide. The linkage information obtained and its partial integration in the core map will further facilitate development of multiple disease resistant bean cultivars.

Materials and Methods

Plant materials. The mapping population consisted of 79 randomly derived $F_{5:7}$ recombinant inbred lines (RILs) generated by single-seed descent from a 'Dorado'/XAN 176 hybridization. Phenotypic characterization and QTL analysis of this mapping population for response to common bacterial blight, ashy stem blight, and bean golden mosaic virus was reported previously (Miklas et al., 1996b, 1998). This cross is relatively narrow as each parent developed by breeders at the International Center for Tropical Agriculture (CIAT), Cali, Colombia, has a complex pedigree dominated by progenitors from the Mesoamerican race (CIAT, 1995). 'Dorado' (formerly DOR 364) (Honduras 46/Negro 150/5/Honduras 46/Venezuela 54//Desarrural I/Cornell 49-242/4/Pompadour Checa/Turrialba 1/3/Porrillo Sintetico/Honduras 46), a Honduran small red dry edible bean cultivar was bred specifically for resistance to BGMV (Beebe, 1994). XAN 176 (Porrillo Sintetico/Compuesto Chimaltenango-2/4/22-G-4/Gentry 21439//51052/Cornell 49-242/3/Negro/Jules), a black dry edible bean breeding line was bred for resistance to common bacterial blight (CIAT, 1995). 'Dorado' also has resistance to rust, and XAN 176 has moderate resistance to ashy stem blight.

PCR markers. The DNA sample, representing a line or parent, was extracted from the leaf tissue of three plants macerated together following the procedures of Miklas et al. (1996b). For randomly amplified polymorphic DNA (RAPD) markers, intersimple sequence repeat (ISSR) markers (González et al., 1998), and sequence characterized amplified region (SCAR) markers, the polymerase chain reaction (PCR) consisted of 25 μL reactions containing two units of Stoffel fragment DNA polymerase (Perkin Elmer Cetus, Norwalk, Conn.), 1× Stoffel buffer, 5 mM $MgCl_2$, 200 μM of each 2'-deoxynucleoside 5'-triphosophate, 0.2 μM (25 ng) primer for RAPD and ISSR markers vs. 12 to 17 ng of each primer pair for SCAR markers, and 25 ng template DNA. The amplification profiles performed with a Perkin Elmer 480 DNA thermal cycler consisted of i) RAPD markers = 3 cycles of 94 °C for 1 min, 35 °C for 1 min, and 72 °C for 2 min followed by either 34 cycles of 94 °C for 1 min, 40 °C for 1 min, and 72 °C for 2 min or 31 cycles of 94 °C for 10 s, 40 °C for 20 s, and 72 °C for 2 min, followed by a 1-s autosegment extension for each cycle and a final cycle extension at 72 °C for 5 min; ii) ISSR markers = same as the shorter duration RAPD amplification profile above; and iii) SCAR markers = profiles reported by Melotto et al. (1996) and Melotto and Kelly (1998) were used. Amplified products were separated on 1.4% agarose gels containing ethidium bromide (0.5 $\mu g \cdot mL^{-1}$) for 5 h at 3 $V \cdot cm^{-1}$ constant voltage.

For RAPD markers, the parental DNA was amplified by 800 decamer primers obtained from Operon Technologies, Alameda, Calif. For ISSR markers, the six primers $(GACA)_3GG$, $(GACA)_3AG$, $(GACAC)_2$, $(GATA)_4$, $(GAGA)_4CG$, and $(GAGA)_4TG$ were used. All RAPD markers and ISSR markers identified between the parents were assayed across the mapping population.

Morphological markers. The dominant *V* gene conditioning black seedcoat color and purple flower color was present in XAN 176. The dominant *Asp* gene conditioning shiny seeds, and the dominant *R* gene conditioning red seedcoat color were present in 'Dorado'. The rk^d locus also conditions red seedcoat color. Testcrosses (Bassett, 1998) indicate that the red seedcoat color of the 'Honduran Red' market class is conditioned by a dominant gene, most likely *R* (Bassett unpublished data); therefore, we assumed that Dorado possessed *R* instead of rk^d. The segregation for *V* and *Asp* was evaluated across 78 of the 79 RILs as one RIL had white seeds due to an outcross or seed mixture. Segregation for *R* (red seedcoat color) and *r* (brown seedcoat color) could only be determined for the 34 RILs with the recessive allele (*v*) at *V*.

Linkage analysis and QTL mapping. The Mapmaker/EXP 3.0 (Lander et al., 1987) computer program was used to obtain a linkage map of the RAPD loci. A pairwise linkage analysis of the RAPD data, imposing a minimum LOD of 3.8 and maximum distance of 30 cM, was used to establish the linkage groups. The markers within linkage groups were ordered with three-point and multipoint LOD values of 3.0 and 2.5, respectively, using the Order and Ripple commands. Centimorgan (cM) distances between linked loci were based upon recombination fractions using the Kosambi (1944) mapping function.

For interval mapping QTL, a significant threshold of LOD 3.0 was applied within Mapmaker EXP 3.0/QTL 1.1 (Lander et al., 1987; Lincoln et al., 1992). Additional QTL were assumed if, while the position of a single QTL was fixed, any additional QTL with a LOD increase of 3.0 above the fixed QTL was observed. The phenotypic means of the RILs used for QTL mapping were from the 1993 greenhouse experiments for common bacterial blight reactions in the leaves (CBB-GH-leaf) and pods (CBB-GH-pod) and a 1992 field trial (CBB-field) as reported by Miklas et al. (1996b). The disease reaction means of the RILs for ashy stem blight (ASB) and BGMV were obtained from 1993 field trials (Miklas et al., 1996b, 1998).

Certain linkage groups of this map were partially integrated with the core map either directly or indirectly through shared loci with other maps.

Resistance gene markers. To locate other disease resistance traits on the map, the parental DNA was screened for 23 RAPD and SCAR markers linked previously to 11 resistance genes as listed by Kelly and Miklas (1998). Markers polymorphic between 'Dorado' and XAN 176 were assayed across the RIL mapping population. If a marker was placed on the map, the parents were challenged by an appropriate set of pathotypes for detection of the corresponding linked resistance gene, except for anthracnose where we lacked proper screening facilities and isolates. For instance, we screened the parents against the NL-3 strain (Haley et al., 1994b) of BCMV to detect the *I* gene (top necrosis reaction) or the *bc-3* gene (immune reaction), and against an array of bean rust races 41, 44, 47, 49, 53, 54, 67, 73, and 108, able to detect most rust resistance genes present across the 19 host differentials of common bean (Stavely et al., 1989). If a resistance gene was polymorphic between the parents, then its segregation across the mapping population was determined.

Results and Discussion

Linkage map. Of the 800 decamer primers screened, 712 amplified discernible DNA fragments in the parental lines 'Dorado'

and XAN 176. In total, 124 primers detected 165 RAPD markers that were segregating in the mapping population. A relatively high frequency (31%) of primers detected more than one RAPD marker, and 10 RAPD markers were apparently codominant. The average heterozygosity (4.8%) detected by these codominant markers approached the 6% expected for an $F_{5:7}$ RIL population (Hallauer and Miranda, 1981). One ISSR marker (900 bp) was detected by primer $(GACAC)_2$.

In total, 155 loci were identified. They included 147 RAPD markers (139 dominant and 8 codominant), two SCAR markers, one ISSR marker, two seedcoat color genes *R* and *V*, the *Asp* gene for seed brilliance, and two rust resistance genes: one conditioning resistance to Races 53 and 54 and the other conditioning resistance to Race 108. They were assigned to 11 linkage groups (US1 to US11), one linked triad (LT12), and seven linked pairs (LP13 to LP19) (Fig. 1). The other 18 RAPD markers were unlinked. For *P. vulgaris*,11 linkage groups would be expected (2n = 22).

The overall map length was estimated at 930 cM, with an average 6-cM distance between markers, which approximates the 1056 and 963 cM lengths for the Davis (Gepts et al., 1993) and Florida (Vallejos et al., 1992) maps, respectively. Our map gave nearly twice the genomic coverage as the Nebraska (545 cM; Jung et al., 1996) map, even though both populations were based on narrow crosses and had a similar number of mapped markers, 175 (75 plus 100 additional markers mapped subsequent to their report, which did not significantly increase the length) for the Nebraska map. Perhaps, complex pedigrees contributed to an increased level of sequence diversity between our parental lines, thus giving rise to a larger map. Still there was not enough sequence diversity, as obtained in Middle American x Andean gene pool crosses, to define clearly 11 complete linkage groups or incorporate the numerous pairs and triads of linked markers into linkage groups.

Two large linkage groups, US6 and US7, contained 43% of the mapped loci, which is consistent with the Paris map (Adam-Blondon et al., 1994), where the linkage groups P1 and P8 contained 42% of the mapped markers, and the Nebraska map, where linkage groups 1 and 2 contained 45% of the mapped markers. The Davis and Florida maps exhibit the same general trend of a few linkage groups containing a large proportion of the mapped markers. Interestingly, the marker saturation and length of corresponding linkage groups varied between maps as D1 [39 (no. of markers on linkage group)/204 (no. of markers for the

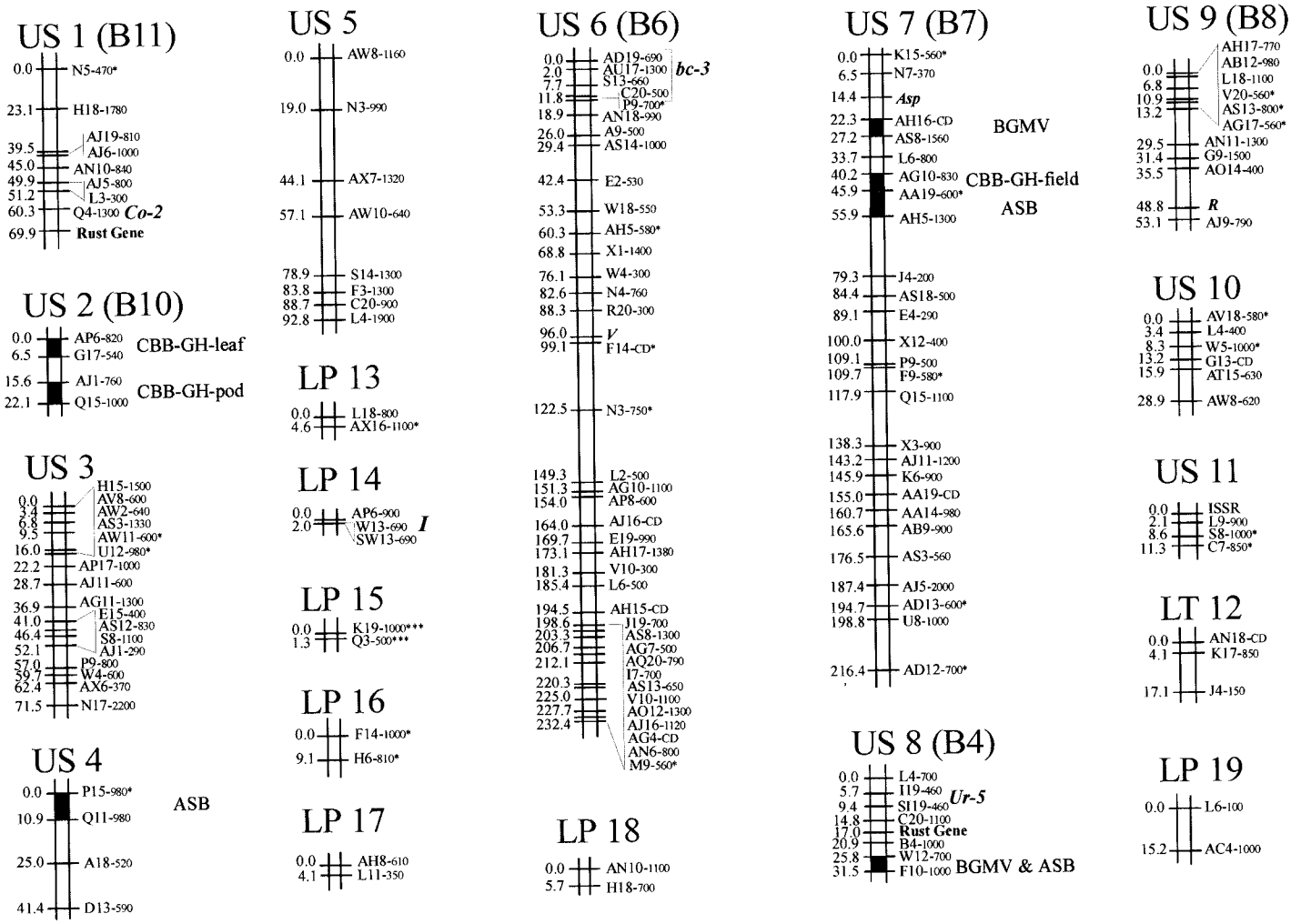

Fig. 1. Genetic linkage map based on the segregation of 147 RAPD marker loci; one ISSR marker; two SCAR markers; three seedcoat traits *R, V,* and *Asp;* and two rust resistance genes across 79 $F_{5:7}$ recombinant inbred lines ('Dorado'/XAN 176). The resistance genes *Co-2, Ur-5, bc-3,* and *I* were positioned based on linkage to previously identified markers. The cumulative map distances are shown in Kosambi centimorgans. Markers followed by * or *** had distorted segregations from the expected (1:1) at $P < 0.05$ or 0.001, respectively. CD signifies a codominant RAPD marker. Marker intervals containing major-effect QTL (LOD > 3.0) are indicated by solid bars within the linkage groups. Corresponding linkage groups of the core map are in parentheses.

Table 1. List of quantitative trait loci having pronounced effects (LOD >3.0) on disease resistance in an $F_{5:7}$ RIL population ('Dorado'/XAN 176).

Trait[z]	QTL interval	LOD score	Linkage group[y]	R^2
CBB-GH-leaf	$AP6_{820}$-$G17_{540}$	20.5	US2 (B10)	73.0
	$AA19_{600}$-$L6_{800}$ (with AP6 fixed)	4.8	US7 (B7)	7.6
CBB-GH-pod	$AJ1_{760}$-$Q15_{1000}$	6.3	US2 (B10)	52.4
CBB-field	$AA19_{600}$-$L6_{800}$	3.9	US7 (B7)	36.5
ASB	$AH5_{1300}$-$AA19_{600}$	4.8	US7 (B7)	29.4
	$P15_{980}$-$Q11_{980}$ (with AH5 fixed)	3.3	US4	13.8
	$W12_{700}$-$F10_{1000}$ (with AH5 fixed)	3.4	US8 (B4)	13.8
BGMV	AS81560-AH16-$_{CD}$	7.7	US7 (B7)	38.6
	$W12_{700}$-$F10_{1000}$	7.8	US8 (B4)	39.4

[z]ASB = ashy stem blight, BGMV = bean golden mosaic virus, CBB = common bacterial blight, and GH = greenhouse.
[y]Corresponding linkage group of the consensus map in parenthesis.

entire map); 210 cM (length of linkage group] and P1 (35/150; 110 cM) had the highest proportion of mapped loci for the respective Davis and Paris maps, whereas US1 contained fewer mapped markers (9/155; 70 cM). Conversely, D6 (15/204 = 106 cM) and P6 (6/150 = 31 cM) had a lower proportion of mapped markers than US6 (38/155 = 232 cM). Discrepancies in saturation and length of corresponding linkage groups attests to the need for integrating maps to obtain complete linkage groups.

Three markers deviated drastically ($P < 0.001$) from the 1:1 ratio expected for dominant markers in an $F_{5:7}$ RIL population. Two of these extremely distorted markers formed linkage pair LP-15, and one was unlinked. Twenty-four other markers with mildly distorted ($0.05 > P > 0.01$) segregation patterns were dispersed across eight linkage groups and two linked pairs (Fig. 1). Of the markers exhibiting distorted segregation, 67% had a higher than expected proportion of the XAN 176 allele. This proportion of distortion among segregating markers (16%) is within the 7% to 23% range reported for other maps (Adam-Blondon et al., 1994; Jung et al., 1996; Nodari et al., 1993a; Vallejos et al., 1992).

Mapped resistance genes and QTL. Of the 23 RAPD and SCAR markers linked previously to 11 resistance genes that we screened (Kelly and Miklas, 1998), eight markers, representing four resistance gene loci (*Co-2*, *Ur-5*, *bc-3*, and *I*) segregated in our population (Fig. 1). The $OQ4_{1440}$ RAPD marker, linked to *Co-2* (Young and Kelly, 1996), mapped to an end of US1. The $F10_{970}$ and $I19_{460}$ RAPD markers (Haley et al., 1993) and $SI19_{460}$ SCAR (Melotto and Kelly, 1998) linked to *Ur-5* were located on US8. The $SI19_{460}$ SCAR (Melotto and Kelly, 1998) mapped 3.7 cM from the originating RAPD $I19_{460}$ (Haley et al., 1994b). The $AD19_{690}$, $S13_{660}$, and $C20_{460}$ markers (Haley et al., 1994a; Johnson et al., 1997), linked to *bc-3*, all mapped within 11.8 cM at one end of US6. The $W13_{690}$ RAPD (Haley et al., 1994b) and $SW13_{690}$ SCAR (Melotto et al., 1996) markers, linked to the *I* gene, mapped to the same location on LP-14.

Haley et al. (1994a) observed no recombination between $S13_{660}$ and $AD19_{690}$ in an F_2 population, whereas we observed a spacing of 7.7 cM, perhaps because our recombinant inbred mapping population, which had undergone more meioses, experienced a few crossover events between the marker loci (Burr and Burr, 1991). The 2.0-cM distance between $S13_{660}$ and $C20_{460}$ was similar to the 2.9-cM distance reported by Johnson et al. (1997), and the placement of $C20_{460}$ internal to $S13_{660}$ at the end of the linkage group was the same for both maps. The utility that other RAPD markers, like $AU17_{1300}$ and $P9_{700}$, may have as selectable markers for the *bc-3* gene warrants investigation in lieu of the gene-pool specificity of the previously identified markers (Haley et al., 1994a; Johnson et al., 1997; Miklas et al., 1996a).

The positions of *Co-2*, *Ur-5*, *bc-3*, and *I* on our linkage map can only be inferred because they did not segregate, as neither parent expressed *bc-3* or *Ur-5* and both parents expressed the *I* gene, and reaction to anthracnose was not tested. However, the inferred location of these resistance genes is reinforced by i) the placement of *Co-2*, *I* and *bc-3* toward the ends of linkage groups, which coincides with their positions on other maps (Adam-Blondon et al., 1994; Gepts et al., 1993; and Johnson et al., 1997); ii) the mapping of multiple markers linked to an individual gene to the same genomic region within a linkage group, as for *bc-3* and *Ur-5*; and iii) the RAPD and SCAR for the *I* gene mapped to the same position.

It is not uncommon for a RAPD marker tightly linked to a disease resistance gene to be present in susceptible or lacking in resistant germplasm (Miklas et al., 1996a), as observed herein for the *bc-3*, *Ur-5*, and *I* gene RAPD markers. Although the $SW13_{690}$ SCAR is considered diagnostic for the *I* gene (Melotto et al., 1996), and is even used in our breeding program as such, its absence in 'Dorado', which has the *I* gene, should be duly noted.

Nine QTL with major affects (LOD > 3.0) on disease resistance were identified (Table 1, Fig. 1). Four QTL conditioned resistance to common bacterial blight: two on leaves (CBB-GH-leaf), one on pods (CBB-GH-pod), and one on field reaction (CBB-field). Three QTL conditioned field resistance to ashy stem blight. Lastly, two QTL conditioned field resistance to BGMV.

Differential reactions of the parents to the nine rust races were not attributable to any known genes. Two rust resistance genes derived from 'Dorado' were observed to segregate in the RIL population (Table 2, Fig. 1). The rust gene located on US1 conditioned a combined hypersensitive and small pustule (2, 3 on a 1 to 6 scale; Stavely, 1983) resistance reaction to races 53 and 54 and had susceptible large pustule reactions 4, 5, or 6 to seven races. The rust gene on US8 conditioned a combined hypersensitive and small pustule reaction 2, 3 to race 108 and 4, 5, and 6 reactions to eight races. This rust gene appears loosely linked with the *Ur-5* (formerly B-190; Kelly et al., 1996) rust resistance gene block shown to consist of six tightly linked dominant genes in coupling (Stavely, 1984). To determine whether these rust genes represent new loci or alleles of existing rust resistance loci (Kelly et al., 1996) would require allelism tests. The differential reaction of the gene on US1 most resembles that of the *Ur-3* gene, such that this should be the first test of allelism for this particular locus. The rust resistance gene on US8 should first be tested for allelism with the *Ur-4* and *Ur-5* genes, which occur on the same linkage group.

Partial map integration. Sixty-seven percent of our map can be integrated into previous maps, but only partially because in most instances the integration is based on only one or two markers instead

Table 2. Comparison of differential rust reactions of two unknown rust resistance genes segregating in a 'Dorado'/XAN 176 RIL population to known rust resistance genes tested against nine rust races.

	Rust race								
Gene	41	44	47	49	53	54	67	73	108
Unknown (US1)[z]	S[y]	S	S	S	R	R	S	S	S
Unknown (US8)	S	S	S	S	S	S	S	S	R
Ur-3	R	S	S	S	R	R	S	S	R
Ur-4	S	R	S	R	S	S	S	R	R
Ur-5	R	R	R	S	R	R	S	R	S
Ur-6	S	S	R	S	S	S	S	R	S
Ur-11	R	R	R	R	R	R	R	R	S

[z]The unknown resistance genes are on linkage groups US1 and US8, respectively.
[y]Ratings of S and R represent susceptible (grades 4, 5, and 6) and resistant (grades 1, 2, and 3) rust reactions, respectively.

of the recommended two to three shared markers per linkage group. The US1, US6, US7, and US9 linkage groups and linked pair LP-14 containing the genes *Co-2*, *bc-3* and *V*, *Asp*, *R*, and *I*, respectively, correspond with the defined linkage groups B11, B6, B7, B8, and B2 of the core map (Adam-Blondon et al., 1994; Freyre et al., 1998; Nodari et al., 1993a; Vallejos et al., 1992). The US2 and US8 linkage groups correspond with B10 and B4 through resistance QTL shared with other maps. The other five linkage groups, US3, US5, US8, US10, and US11, have not been fully integrated with another map.

The partial integration of US1 with B11 is tenuous at best, as it is based solely on the single marker $OQ4_{1440}$ linked with the *Co-2* gene. US6 integrates with B6 via the shared *bc-3*-linked RAPD markers ($S13_{660}$, $AD19_{690}$, and $C20_{460}$) and *V* locus. US7 is partially integrated with B7 through the shared *Asp* locus and a QTL for common bacterial blight resistance, which maps internally to *Asp* (toward the *Phs* locus) as it does in two other populations BAT 93/Jalo EEP 558 (Nodari et al., 1993b) and BAC 6/HT 7719 (Ariyaranthe et al., 1999; Jung et al., 1996). This QTL for common bacterial blight derives from GN No. 1 sel 27 (Miklas et al., 2000). The linked pair LP14 can be partially integrated with B2 of the core map due to occurrence of *I* on linkage group D2 (Davis), P2 (Paris), and D (Florida) of previous maps. The US-9 linkage group can be tenuously integrated with B8 of the core map due to the presence of the *R* gene, which is tightly linked with the previously mapped C locus for seedcoat color (Bassett, 1991).

A SCAR for the $AP6_{820}$ marker (Miklas et al., 2000) on US2 mapped between the $W10_{550}$ and $BC409_{1250}$ SCAR markers (Jung and Coyne, personal communication) on linkage group B10 in another population BAC 6/HT 7719 (Ariyaranthe et al., 1999). All three SCAR markers are associated with common bacterial blight resistance, and the $AP6_{820}$ SCAR has been useful for MAS of common bacterial blight resistance in gamete and backcross selection programs (Miklas et al., 2000). Both BAC 6 and the XAN 176 parent of our population derive resistance to common bacterial blight from GN No. 1 sel 27.

A SCAR (unpublished, available upon request) for the $W12_{700}$ RAPD marker linked with BGMV resistance on US8 was present on linkage group B4 (Beebe, personal communication) of the 'Dorado'/G19833 map (Beebe et al., 1998), where it was also associated with BGMV resistance. The B4 linkage group of 'Dorado'/G19833 has been fully integrated into the core map. The $W12_{700}$ SCAR occurs between anchor RFLP markers Bng 71 and Bng 160 (Beebe, Blair, and Pedraza, personal communication).

Linkages among resistance genes and QTL . Three genomic regions contained linked genes and/or QTL conditioning resistance to different pathogens (Fig. 1). Rust and *Co-2* genes were linked (9.6 cM) on US1 (B11). QTL conditioning resistance to common bacterial blight, ashy stem blight, and BGMV were located within 34 cM on US7 (B7) in the same general region of *Asp* and presumed location of *Phs*, where a QTL conditioning physiological resistance to white mold [*Sclerotinia sclerotiorum* (Lib.) de Bary] was also identified (Miklas et al., 1999). Nodari et al. (1993b) observed resistance to common bacterial blight and *Rhizobium* Frank nodulation, and Geffroy (1997) observed the *Co-9* gene for resistance to anthracnose in this same general region.

The unknown and *Ur-5* rust-resistance genes and QTL conditioning resistance to ashy stem blight and BGMV were located within 25 cM on US8 (B4). The *Ur-4* rust resistance gene also resides on B4 (Gepts et al., 1993), although it segregates independently from *Ur-5* (Kelly et al., 1996). The QTL for resistance to ashy stem blight and BGMV actually mapped to the same marker interval, a result indicative of either a single locus having a pleiotropic effect on resistance to multiple diseases or perhaps a pair of closely linked genes. The latter explanation is most likely because resistance to ashy stem blight derived from XAN 176 was negatively associated ($r = -0.40$, $P < 0.01$) with resistance to BGMV derived from 'Dorado', which is supportive of two genes closely linked in repulsion.

Could these regions on US7 and US8 represent initial visualization of a cluster of genes conferring resistance to a diverse set of pathogens as observed in other crops (reviewed by Michelmore and Meyers, 1998) including soybean [*Glycine max* (L.) Merr.] (Tamulonis et al., 1997) and lettuce (*Lactuca sativa* L) (Witsenboer et al., 1995)? Generally, clusters of resistance genes are considered to be formed from duplication events followed by mutation, divergence, and genetic specificity for different pathogens (Staskawicz et al., 1995). A slightly different mechanism, described by Meyers et al. (1998) and Michelmore and Meyers (1998), emphasizing divergence probably gave rise to the cluster of tightly linked resistance genes that form the *Ur-5* (Stavely, 1984) and *Co-2* (Geffroy et al., 1998) loci in common bean. Caution should be exercised when interpreting the linkages among resistance loci in this study, however, as the small size of the 'Dorado'/XAN 176 population leads to a reduction in the precision of the location of a QTL or gene (Melchinger et al., 1998).

From a breeder's perspective, linkages among resistance loci are beneficial when they derive from an individual parental line (*cis*-configuration). When crossed with susceptible parents, these resistances will be inherited as a single unit and likely remain intact in subsequent progeny. When linked resistance genes are donated by different parents (*trans*-configuration), recombination between the loci is necessary to obtain both resistances into a single line. In the 'Dorado'/XAN 176 population, recombination between disease resistance loci on US7 and US8 would be

required to obtain lines with multiple disease resistance against rust, common bacterial blight, ashy stem blight, and BGMV. Thus, breeding multiple disease resistant cultivars is complicated initially by these *trans*-linkages, and subsequently simplified once the linked genes are recombined in *cis*-orientation. As additional resistance genes and QTL are incorporated in the common bean linkage map, it will become ever more useful as a blueprint for breeding multiple disease resistant cultivars.

Literature Cited

Adam-Blondon, A.-F., M. Sevignac, M. Dron, and H. Bannerot. 1994. A genetic map of common bean to localize specific resistance genes against anthracnose. Genome 37:915–924.

Ariyarathne, H.M., D.P. Coyne, G. Jung, P.W. Skroch, A.K. Vidaver, J.R. Steadman, P.N. Miklas, and M.J. Bassett. 1999. Molecular mapping of disease resistance genes for halo blight, common bacterial blight, and bean common mosaic virus in a segregating population of common bean. J. Amer. Soc. Hort. Sci. 124:654–662.

Bassett, M.J. 1998. A test cross protocol for determining the genotype of dark red seedcoat colors in common bean. J. Amer. Soc. Hort. Sci. 123:1048–1052.

Beebe, S. 1994. Breeding for resistance to bean golden mosaic virus: History and perspectives, p. 148–150. In: F. Morales (ed.). Bean golden mosaic: Research advances. Proc. Workshop PROFRIJOL and CIAT, Guatemala City 9–13 Nov. 1992. International Center of Tropical Agriculture, Cali, Colombia.

Beebe, S., F. Pedraza, M. Rojas, J. Gutierrez, and J. Tohme. 1998. A genetic map of common bean combining RFLP, RAPD, SCAR and AFLP markers. Annu. Rpt. Bean Improvement Coop. 41:95–96.

Burr, B. and F.A. Burr. 1991. Recombinant inbreds for molecular mapping in maize: Theoretical and practical considerations. Trends Genet. 7:55–60.

Freyre, R., P.W. Skroch, V. Geffroy, A.F. Adam-Blondon, A Shirmohamadali, W.C. Johnson, V. Llaca, R.O. Nodari, P.A. Pereira, S.M. Tsai, J. Thome, M. Dron, J. Nienhuis, C.E. Vallejos, and P. Gepts. 1998. Towards an integrated linkage map of common bean. IV. Development of a core linkage map and alignment of RFLP maps. Theor. Appl. Genet. 97: 847–856.

Geffroy, V. 1997. Dissection génétique de la résistance à *Colletotrichum lindemuthianum*, agent de l'anthracnose, chez deux génotypes représentatifs des pools géniques de *Phaseolus vulgaris*. Thèse de Doctorat, Institut National Agronomique, Paris-Grignon.

Geffroy, V., F. Creusot, J. Falquet, M. Sévignac, A.-F. Adam-Blondon, P. Gepts, and M.A. Dron. 1998. A family of LRR sequences at the *Co-2* locus for anthracnose resistance in *Phaseolus vulgaris* and its potential use in marker-assisted selection. Theor. Appl. Genet. 96:494–502.

Gepts, P., R. Nodari, S.M. Tsai, E.M.K. Koinange, V. Llaca, R. Gilbertson, and P. Guzmán. 1993. Linkage mapping in common bean. Annu. Rpt. Bean Improvement Coop. 36:xxiv–xxxv.

González, A., J. León, A. Salinas, and P. Gepts. 1998. Determination of genetic diversity among wild and domesticated beans using inter simple sequence repeats (ISSRs). Annu. Rpt. Bean Improvement Coop. 41:99–100.

Haley, S.D., L.K. Afanador, and J.D. Kelly. 1994a. Selection for monogenic resistance traits with coupling- and repulsion-phase RAPD markers. Crop Sci. 34:1061–1066.

Haley, S.D., L.K. Afanador, and J.D. Kelly. 1994b. Identification and application of a random amplified polymorphic DNA marker for the I gene (Potyvirus resistance) in common bean. Phytopathology 84:157–160.

Haley, S.D., P.N. Miklas, J.R. Stavely, J. Byrum, and J.D. Kelly. 1993. Identification of RAPD markers linked to a major rust resistance gene block in common bean. Theor. Appl. Genet. 86:505–512.

Hallauer, A.R. and J.B. Miranda. 1981. Quantitative genetics in maize breeding. Iowa State Univ. Press, Ames.

International Center of Tropical Agriculture. 1995. Catalog of advanced bean lines from CIAT. 2nd ed. Compiled by M.A. Rodríguez, H.F. Ramírez, M.C. Valencia, O.V. Voysest, and J.W. White. CIAT, Cali, Colombia.

Johnson, W.C., P. Guzmán, D. Mandala, A.B.C. Mkandawire, S. Temple, R.L. Gilbertson, and P. Gepts. 1997. Molecular tagging of the *bc-3* gene for introgression into Andean common bean. Crop Sci. 37:248–253.

Jung, G., D.P. Coyne, P. Skroch, J. Nienhuis, E. Arnaud-Santana, J. Bokosi, H.M. Ariyarathne, J.R. Steadman, J.S Beaver, and S.M. Kaeppler. 1996. Molecular markers associated with plant architecture and resistance to common blight, web blight and rust in common bean (*Phaseolus vulgaris* L.). J. Amer. Soc. Hort. Sci. 121:794–803.

Kelly, J.D. and P.N. Miklas. 1998. The role of RAPD markers in breeding for disease resistance in common bean. Molecular Breeding 7:1–11.

Kelly, J.D., J.R. Stavely, and P.N. Miklas. 1996. Proposed symbols for rust resistance genes. Annu. Rpt. Bean Improvement Coop. 39:25–31.

Kosambi, D.D. 1944. The estimation of map distances from recombination values. Ann. Eugenics 12:172–175.

Lander, E.S., P. Green, J. Abrahamson, A. Barlow, M.J. Daley, S.E. Lincoln, and L. Newburg. 1987. MAPMAKER: An interactive computer package for constructing primary genetic linkage maps of experimental and natural populations. Genomics 1:174–181.

Lincoln, S.E., M.J. Daly, and E.S. Lander. 1992. Mapping genes controlling quantitative traits with Mapmaker/QTL 1.1. Whitehead Institute Technical Report. 2nd ed. Whitehead Institute for Biomedical Research, Cambridge.

Melchinger, A., H. Utz, and C. Schön. 1998. Quantitative trait loci (QTL) mapping using different testers and independent population samples in maize reveals low power of QTL detection and large bias in estimates of QTL effects. Genetics 149:383–403.

Melotto, M., L. Afanador, and J.D. Kelly. 1996. Development of a SCAR marker linked to the *I* gene in common bean. Genome 39:1216–1219.

Melotto, M. and J.D. Kelly. 1998. SCAR markers linked to major disease resistance genes in common bean. Annu. Rpt. Bean Improvement. Coop. 41:64–65

Meyers, B.C., K.A. Shen, P. Rohani, B.S. Gaut, and R.W. Michelmore. 1998. Receptor-like genes in the major resistance locus of lettuce are subject to divergent selection. Plant Cell 11:1833–1846.

Michelmore, R.W. and B.C. Meyers. 1998. Clusters of resistance genes in plants evolve by divergent selection and a birth-and-death process. Genome Res. 8:1113–1130.

Miklas, P.N., L. Afanador, and J.D. Kelly. 1996a. Recombination-facilitated RAPD marker-assisted selection for disease resistance in common bean. Crop Sci. 36:86–90.

Miklas, P.N., R. Delorme, W.C. Johnson, and P. Gepts. 1999. Dry bean G122 contributes a major QTL for white mold resistance in the straw test. Annu. Rpt. Bean Improvement Coop. 42:43–44.

Miklas, P.N., E. Johnson, V. Stone, J.S. Beaver, C. Montoya, and M. Zapata. 1996b. Selective mapping of QTL conditioning disease resistance in common bean. Crop Sci. 36:1344–1351.

Miklas, P.N., J.R. Smith, R. Riley, K.F. Grafton, S.P Singh, G. Jung, and D.P. Coyne. 2000. Marker-assisted breeding for pyramided resistance to common bacterial blight in common bean. Annu. Rpt. Bean Improvement Coop. 43:(In press).

Miklas, P.N., V. Stone, C.A. Urrea, E. Johnson, and J.S. Beaver. 1998. Inheritance and QTL analysis of field resistance to ashy stem blight in common bean. Crop Sci. 38:916–921.

Nodari, R.O., S.M. Tsai, R.L. Gilbertson, and P. Gepts. 1993a. Towards an integrated linkage map of common bean. II. Development of an RFLP-based linkage map. Theor. Appl. Genet. 85:513–520.

Nodari, R.O., S.M. Tsai, P. Guzmán, R.L. Gilbertson, and P. Gepts. 1993b. Towards an integrated linkage map of common bean. III. Mapping genetic factors controlling host–bacteria interactions. Genetics 134:341–350.

Staskawicz, B.J., F.M. Ausubel, B.J. Baker, J.G. Ellis, and J.D.G. Jones. 1995. Molecular genetics of plant disease resistance. Science 268:661–669.

Stavely, J.R. 1983. A rapid technique for inoculation of *Phaseolus vulgaris* with multiple pathotypes of *Uromyces phaseoli*. Phytopathology 73:676–679.

Stavely, J.R. 1984. Genetics of resistance to *Uromyces phaseoli* in a *Phaseolus vulgaris* line resistant to most races of the pathogen. Phytopathology 74:339–344.

Stavely, J.R., J.R. Steadman, and R.T. McMillan, Jr. 1989. New pathogenic variability in *Uromyces appendiculatus* in North America. Plant Dis. 73:428–432.

Tamulonis, J.P., B.M. Lussi, R.S. Hussey, W.A. Parrott, and R. Boerma. 1997. DNA markers associated with resistance to Javanese root-knot nematode in soybean. Crop Sci. 37:783–788.

Vallejos, C.E., N.S. Sakiyama, and C.D. Chase. 1992. A molecular marker-based linkage map of *Phaseolus vulgaris* L. Genetics 131:733–740.

Witsenboer, H., R.V. Kesseli, M.G. Fortin, M. Stanghellini, and R.W. Michelmore. 1995. Sources and genetic structure of a cluster of genes for resistance to three pathogens in lettuce. Theor. Appl. Genet. 91:178–188.

Young, R.A. and J.D. Kelly. 1996. RAPD markers flanking the *Are* gene for anthracnose resistance in common bean. J. Amer. Soc. Hort. Sci. 121:37–41.