

Progress Report

Output 1: Improved small-seeded Middle American bean germplasm with less dependence on inputs

Activity 1.1 Developing germplasm tolerant to abiotic stresses of drought and low soil fertility

Highlights:

- Multiple stress resistance to abiotic constraints such as drought, aluminum toxicity, and low nutrient supply was confirmed in several genotypes, especially in G 21212, which presents efficient transport and utilization of photosynthates and nutrients toward grainfilling.
- Two bred lines (SEA 5 and CAP 4) and two landraces (G 21212 and G 1977) were very well adapted to drought stress because of their ability to mobilize photosynthates for grain production. Drought resistance equal to or better than that of SEA 5 was obtained in bred lines with superior plant type and acceptable grain color.
- Modest levels of drought resistance were recovered from interspecific crosses with *Phaseolus acutifolius*.
- Lines and landraces tolerant to toxic level of aluminum in soil and tolerant to low nutrients supply were identified.
- Two bred lines and one Mexican landrace were tolerant to waterlogging.

1.1.1 Evaluation of a set of selected landraces and bred lines for multiple stress resistance

Rationale: In other crops, materials have been identified with resistance to multiple abiotic stresses. In bean there is a long history of studying abiotic stress tolerance, but as individual stresses (drought, phosphorus, nitrogen, aluminum, or generalized low fertility). In 1998, a Multiple Stress Nursery was established that combined materials that had expressed tolerance to different types of physiological stress. The purpose of this nursery was to seek genotypes that express tolerance to multiple stresses, with the expectation that some mechanisms of physiological tolerance could be effective against more than one type of abiotic stress.

Materials and methods: Forty-nine elite materials that had been superior under conditions of low phosphorus, aluminum toxicity, or drought were identified, including landrace germplasm, bred lines, and commercial checks. The nursery was evaluated in Popayán, Santander de Quilichao, and Darien under different edaphic stresses. In Popayán apparently low P and possibly low micronutrients primarily cause the stress, while liming over years has eliminated aluminum toxicity. In Santander de Quilichao

there is a combined stress of aluminum toxicity and low P, and occasionally manganese toxicity, depending on the field site and climatic conditions. In Darien the stress is exclusively caused by low P. Evaluations were also carried out in Palmira for drought tolerance. All stressed trials were accompanied by a control treatment with high fertilizer inputs, but at times the background soil fertility was so low that inputs resulted in an intermediate level of stress.

Results: Data from 15 trials are reported here. Table 1 presents data of the best 18 of the 49 materials that were selected for further study.

In the high- and mid-fertility and irrigated treatments, materials that stood out for their responsiveness included Brazilian cultivar Carioca (which is well known for its responsiveness), FEB 192, A774, VAX 1 and VAX 2. AM 38, a Flor de Mayo type for the Mexican highlands, also responded well in Popayán where temperature was more moderate.

In the low fertility trials, some of these same materials continued to perform very well. VAX 1 in particular stood out again, and A 774 and FEB 192 performed acceptably. However, in the low fertility treatment the germplasm accession G 21212 came to the forefront, and was often the best or second best among all materials (e.g., in trials 7, 8, 10 and 12, as well as mid-fertility trial (6). This unique material has been highlighted in past reports for its low-P tolerance (see IP-1 1999 report) and for its QTL with major effects on yield under P stress (see SB-2 1999 report). It has been the object of physiological analysis and appears to have a stress tolerance mechanism for efficient transport of photosynthate to grain.

Two drought trials were carried out, one under moderate stress in 1998 (trial 14) and one under severe stress in 1999 (trial 15). The line SEA 5 that was bred for drought tolerance was excellent under both levels of stress, expressing very good grain-filling traits. BAT 477, long recognized for its tolerance, expressed relatively good yield as expected. Again, materials like A 774 and FEB 192 performed quite acceptably, and under moderate stress the line A 785 stood out. However, under severe stress the accession G 21212 again was outstanding, displaying its trait of excellent grain filling that it also expresses under low P. This suggests that its mechanism of low-P tolerance might also function for tolerance to drought stress, in which case its genes are especially valuable. This topic will be the object of further investigation. Under severe drought, the germplasm accession G 1977 also stood out and displayed vigorous growth, suggesting that it was accessing moisture that other genotypes could not. G 1977 was also selected originally for low-P tolerance, and reasonable performance in trials 2, 3, 4, and 6.

Table 1. Yield (kg ha⁻¹) of selected common bean lines evaluated under multiple stresses.

Genotypes	Trials ^a															Mean
	1 P-98B	2 D-99A	3 Q-99A	4 Q-99B	5 P-98C	6 P-99A	7 D-99B	8 Q-99A	9 Q-99B	10 Q-98B	11 Q-98B	12 P-98C	13 Pa-99B	14 Pa-98B	15 Pa-98b	
A 774	3667	3522	1193	1734	1061	879	532	303	328	339	904	127	2190	1306	705	1252
Carioca	3358	2846	1131	1132	924	1387	497	395	369	416	703	173	2202	1217	446	1146
DICTA 17	2985	2754	839	939	1094	1715	770	296	368	264	589	301	1986	1314	637	1123
FEB 192	3279	3442	1682	1773	421	827	675	235	222	336	886	74	2039	847	725	1164
MAM 38	3579	2949	1012	1244	1460	1280	1074	386	262	155	635	352	1824	1111	564	1192
G 3513	1319	2360	1087	1155	170	950	643	471	180	228	578	216	1238	1038	607	816
BAT 477	2189	2517	867	1342	271	595	548	478	266	323	1110	60	2011	1407	718	980
G 18479	2805	2454	915	965	632	1151	934	319	348	260	594	147	1955	1076	259	987
G 92	3645	2596	1196	1555	811	1595	756	269	404	202	749	231	1891	1278	93	1151
G 19227A	1498	2196	536	495	224	1122	903	606	158	176	345	113	1039	787	280	698
VAX 1	3289	2966	1607	1634	1168	1568	783	389	385	279	690	36	1984	1116	649	1236
G 21212	2542	3003	901	935	1095	1988	1198	505	152	452	736	437	1529	1176	822	1164
A 785	3062	2301	1171	936	1190	1417	870	521	276	306	592	510	2133	1356	334	1131
VAX 2	3812	2579	1085	1675	1403	1232	1123	352	303	254	721	92	1896	1060	523	1207
G 1977	2397	3132	1295	1595	994	1984	710	316	226	446	515	176	1675	1068	801	1155
MAR 1	3626	2884	971	1460	414	298	485	436	480	255	659	107	1864	1164	308	1027
V 8025	2438	2845	691	1248	1182	1379	809	306	179	220	357	387	1675	1142	728	1039
SEA 5	1475	1643	800	629	152	418	631	477	216	442	813	71	2058	1478	820	808
Mean	2629	2401	864	972	697	1098	718	290	230	206	569	139	1605	1009	408	
LSD	734	677	382	375	360	494	388	234	145	209	446	252	326	292	180	

- a. 1 and 2 = high fertility, 3-6 = medium fertility, 7-12 = low fertility, 13 = irrigated check treatment for drought trial, 14 = intermediate drought stress, and 15 = severe drought stress. PxxA = Popayán March, PxxB = Popayán June, PxxC = Popayán October; DxxA = Darien April, DxxB = Darien September; QxxA = Santander de Quilichao April, QxxB = Santander de Quilichao September; and PxxB = Palmira June.

Conclusions: The multiple stress nursery has been highly useful for identifying materials with multiple stress tolerance, especially G 21212, VAX 1 and A 774. Materials such as A 774 and VAX 1 have also expressed tolerance to flooding in the spring of 2000. These experiences validate the hypothesis that was the basis for establishing the nursery, that multiple stress tolerance is feasible and that common mechanisms might exist for tolerance to several stresses. Lines such as FEB 192, A 774, and VAX 1 also present excellent yield without stress, thus allaying fears that stress tolerance is necessarily associated with low yield potential. Based on the data available, 18 elite entries were selected from among the original 49, and another 18 were added to conform a nursery of 36 entries. This is presently being evaluated under stressed and unstressed conditions.

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1.1.2 Drought resistance

Identification of traits associated with drought resistance

Rationale: A set of 49 genotypes was assembled to evaluate for tolerance to low soil fertility conditions. Several of the genotypes included in this set of materials were known to be good performers under drought conditions. Last year we reported results from a field trial that evaluated genotypic differences in adaptation to moderate drought stress among 49 genotypes. This year we report results obtained from the same 49 genotypes for their adaptation to greater levels of drought stress.

Materials and methods: A field trial was conducted at Palmira in 1999 (June to September) to determine differences in tolerance to high levels of water stress. The field trial included 49 bean genotypes. Details on planting and management of the trial were similar to those reported in the 1997 Bean Project Annual Report, p 33-34. The incidence of *Macrophomina phaseolina* and *Sclerotium rolfsii* was recorded at physiological maturity. A number of plant attributes were measured at mid-podfilling in order to determine genotypic variation in tolerance to water stress. These plant traits included leaf area index, canopy dry weight per plant, shoot nutrient (N, P, K, Ca, and Mg) uptake, shoot ash content, and shoot total nonstructural carbohydrates (TNC). At the time of harvest, grain yield and yield components (number of pods per plant, number of seeds per pod, 100 seed weight) were determined. Seed N, P, ash content, and TNC were also measured.

Results and discussion: During the crop growing season, air temperatures were maximum 32.1 °C and minimum 16.4 °C, while incident solar radiation ranged from 12 to 25 MJ m⁻² d⁻¹. The total rainfall during the 2 months (July-August) of active crop growth was 38 mm compared to the potential pan evaporation of 318 mm. These results on rainfall and pan evaporation indicated that the crop suffered a high level of water stress during active growth and development.

Under high water stress conditions in the field, the seed yield of 49 genotypes ranged from 15 to 820 kg ha⁻¹ (Figure 1). Among the genotypes tested, two land races (G 21212 and G 1977) and two bred lines (SEA-5 and CAP 4) were outstanding in their adaptation to high levels of water stress conditions. These four genotypes were also less affected by the incidence of soil-borne pathogens (*Macrophomina phaseolina* and *Sclerotium rolfsii*) as determined by the percentage of infected plants (Figure 2). Five out of 49 genotypes including MAM 46, A 36, G 3096, Compuesto Chimaltenango, Negro INIFAP, and G 92 showed greater sensitivity to soil-borne pathogens. Several genotypes that yielded well under rainfed conditions were also found to be less affected by the incidence of soil-borne pathogens (Figure 3).

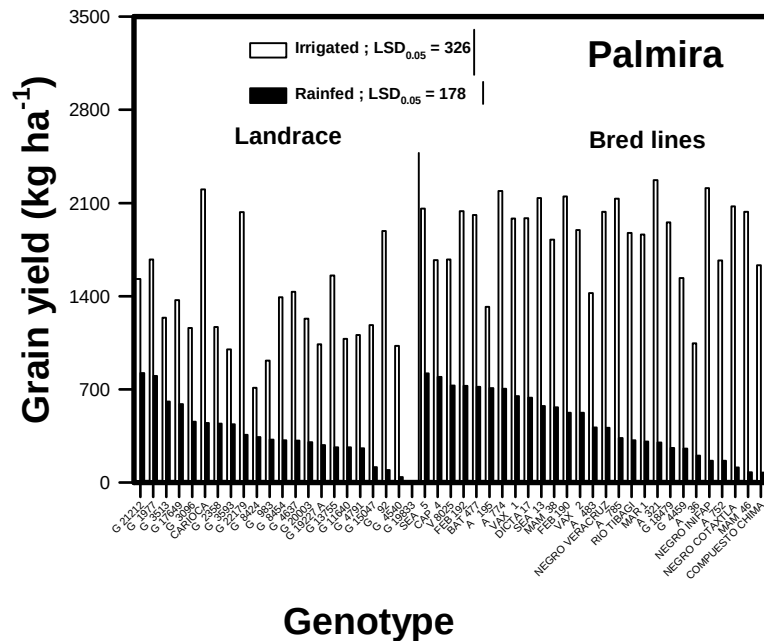


Figure 1. Genotypic variation in adaptation to rainfed and irrigated conditions of 49 common bean genotypes grown in a Mollisol at Palmira, Colombia.

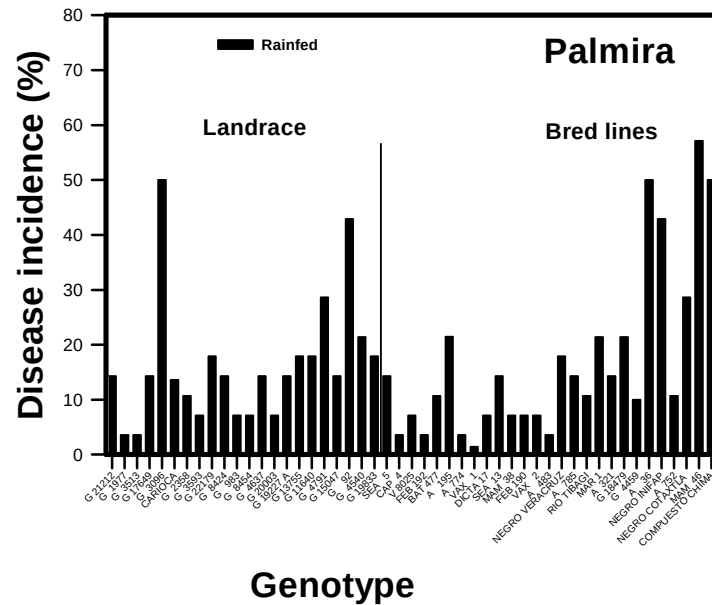


Figure 2. Genotypic variation in incidence of soil-borne pathogens caused by *Macrophomina phaseolina* and *Sclerotium rolfsii* on 49 genotypes of common bean grown in a Mollisol at Palmira.

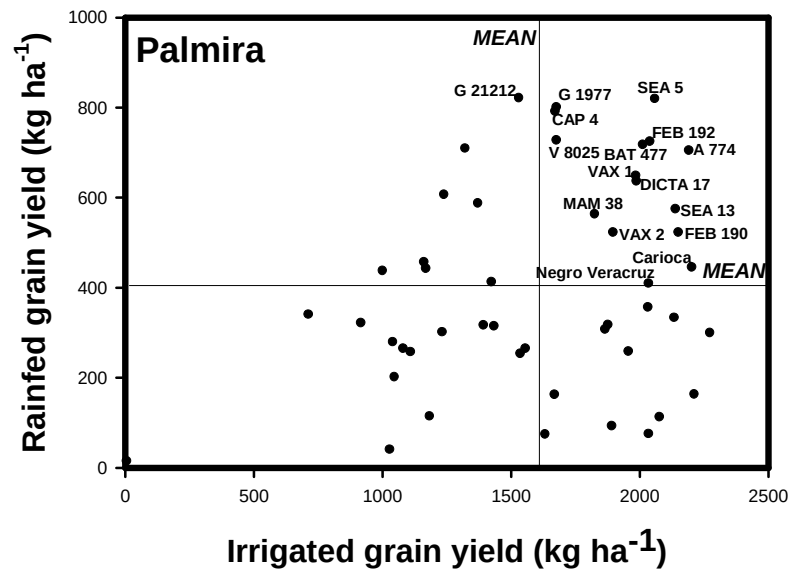


Figure 3. Identification of genotypes that are adapted to rainfed conditions and are less sensitive to soil-borne pathogens in a Mollisol at Palmira. Genotypes that yielded superiorly with drought and were less sensitive to soil-borne pathogens (*Macrophomina phaseolina* and *Sclerotium rolfsii*) were identified in the upper box of the left-hand side.

The relationship between grain yield of rainfed and irrigated treatments indicated that SEA 5 was both adapted to a high level of water stress and responsive to irrigation (Figure 4). SEA-5 was also found to be very well adapted to moderate and severe water stress conditions (1997 and 1998 Bean Project Annual Reports). Among the 49 genotypes tested, G 19833 was the most poorly adapted genotype. This Andean landrace was also the poorest performer at moderate water stress conditions (1998 Bean Project Annual Report). Eighteen of the 49 genotypes were superior in their performance to the most widely planted cultivar, Carioca.

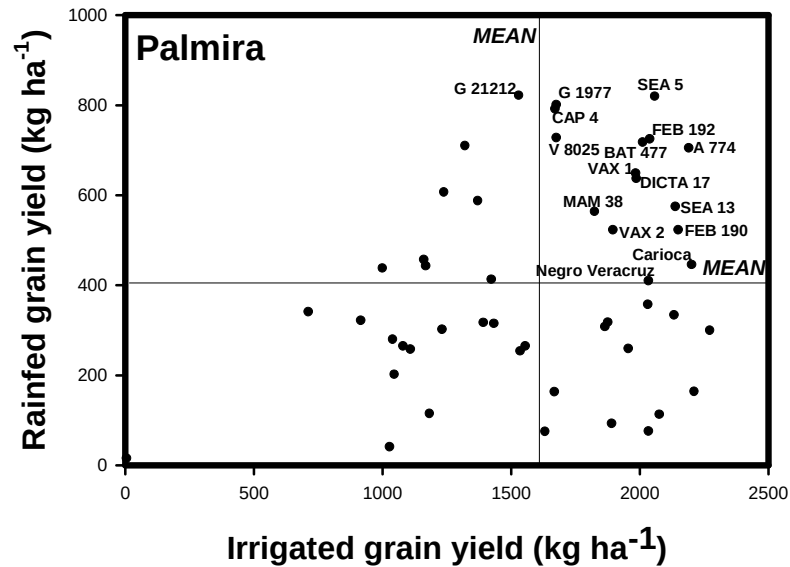


Figure 4. Identification of genotypes that are adapted to rainfed conditions and are responsive to irrigation to a Mollisol at Palmira. Genotypes that yielded superiorly with drought and were also responsive to irrigation were identified in the upper box of the right-hand side.

Measurements of leaf area index indicated that several genotypes had greater leaf area values than the best performers such as SEA 5 and G 21212 under high water stress conditions (Figure 5). This shows that these two genotypes had a better transport system for mobilizing photosynthates to developing grains.

Last year, we reported that the superior performance of SEA 5 in severe drought was related to lower seed P content. We verified this observation with the evaluation of 49 genotypes by measuring seed ash (mineral) content and seed N, P, K, Ca, and Mg content. The relationship between grain yield and seed ash content indicated that the land race G 1977 was outstanding in combining low seed ash content with high grain yield under water stress conditions (Figure 6). The seed ash content of the bred line SEA 5 was intermediate while A 774 was very high. Thus seed ash may not be a very useful indicator of water stress resistance in common bean. Nevertheless, seed P content showed somewhat better negative relationship with grain yield under water stress conditions (Figure 7). Except for the bred lines such as CAP 4 and FEB 192, other superior

performers combined high seed yield with lower seed P content. However, several genotypes also showed lower grain yield and lower seed P content. This indicates that seed P content alone may not be adequate as a selection method for water stress.

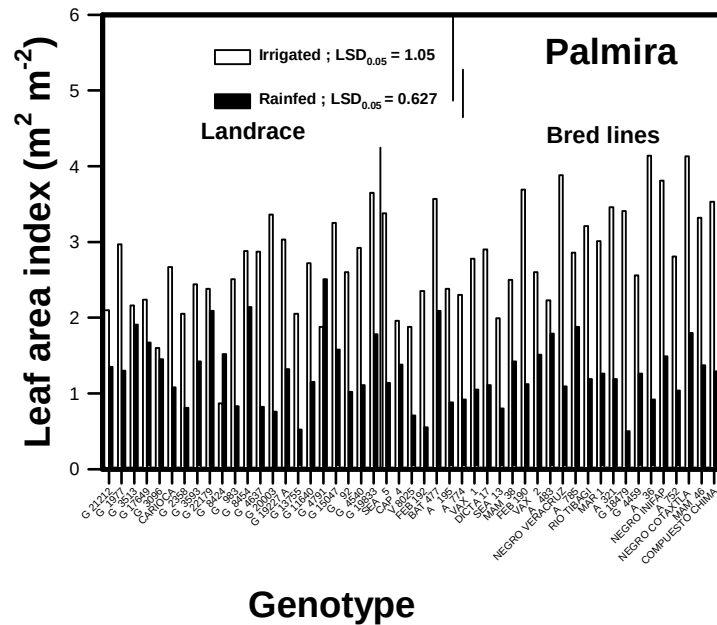


Figure 5. Genotypic variation in leaf area index of 49 genotypes of common bean grown in a Mollisol at Palmira, Colombia.

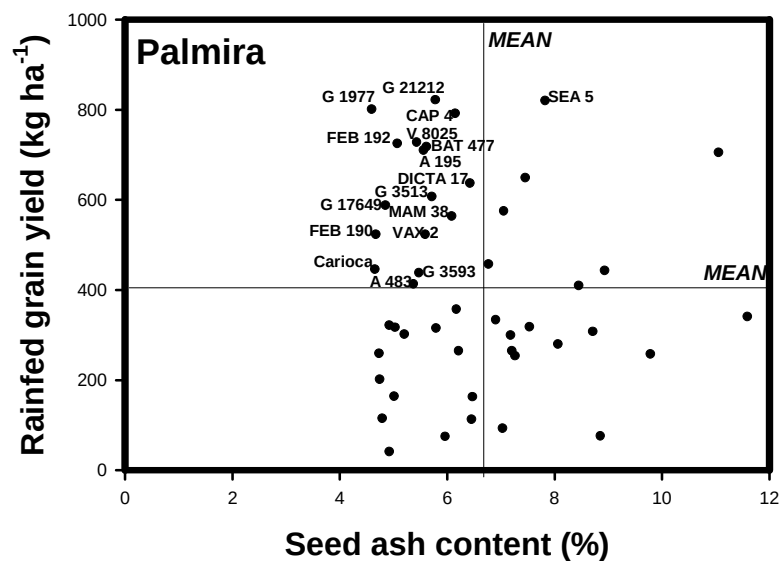


Figure 6. Identification of genotypes that combine superior seed yield with lower ash (mineral) content in seed when grown under rainfed conditions in a Mollisol at Palmira. Genotypes that were superior in grain yield and lower in seed ash were identified in the upper box of the left-hand side.

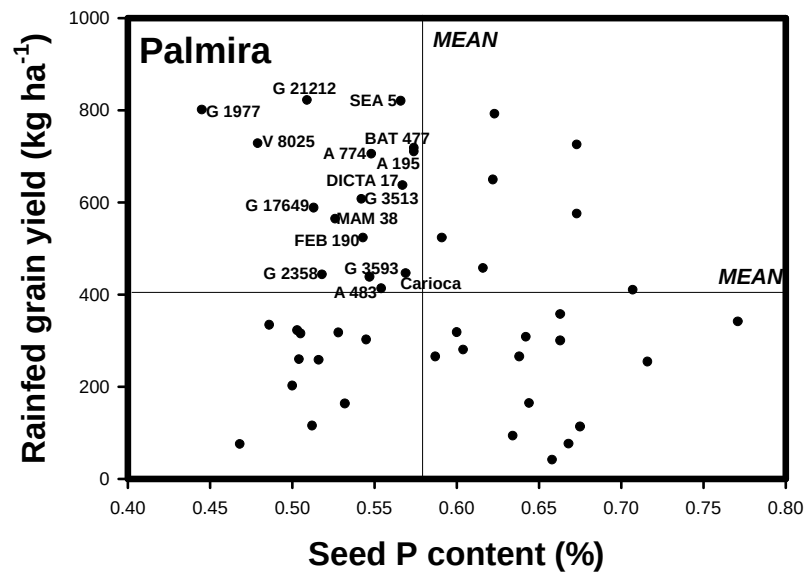


Figure 7. Identification of genotypes that combine superior seed yield with lower P content in seed when grown under rainfed conditions in a Mollisol at Palmira. Genotypes that were superior in grain yield and lower in seed P were identified in the upper box of the left-hand side.

Relationship between seed N and grain yield under water stress conditions indicated that BAT 477 was superior in combining greater seed N content with high seed yield (Figure 8). It is important to note that G 21212 and SEA 5 showed lower levels of seed N. Under rainfed conditions, grain yield was greater and shoot TNC content at mid-podfilling was lower for two landraces (G 21212 and G 1977). This observation indicates that these two land races could mobilize photosynthates better than the other genotypes tested (Figure 9). Two bred lines, SEA 5 and CAP 4 were also moderate in their shoot TNC content. The superior adaptation of these four genotypes to drought was found to be because of their efficient utilization of N and P for grain production (Figure 10) in addition to the mobilization of photosynthates.

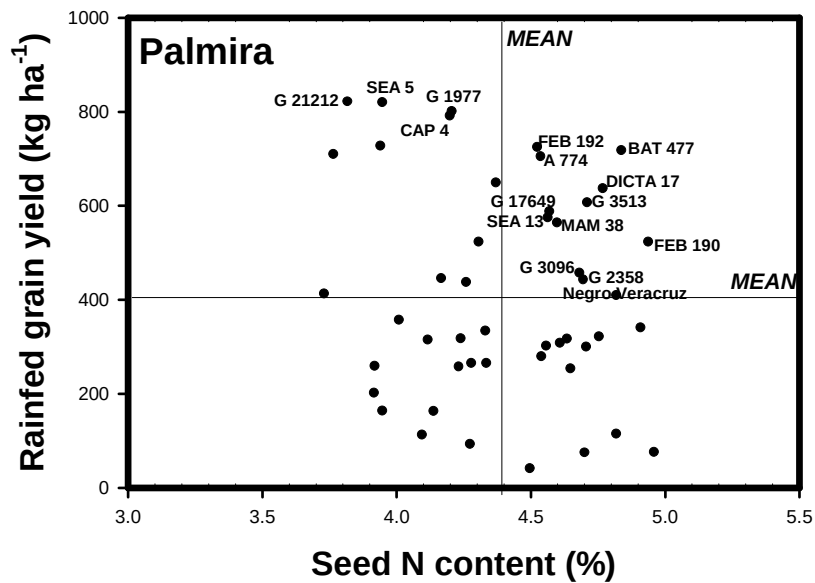


Figure 8. Identification of genotypes that combine superior seed yield with greater N content in seed when grown under rainfed conditions in a Mollisol at Palmira. Genotypes that were superior in grain yield and higher in seed N were identified in the upper box of the right-hand side.

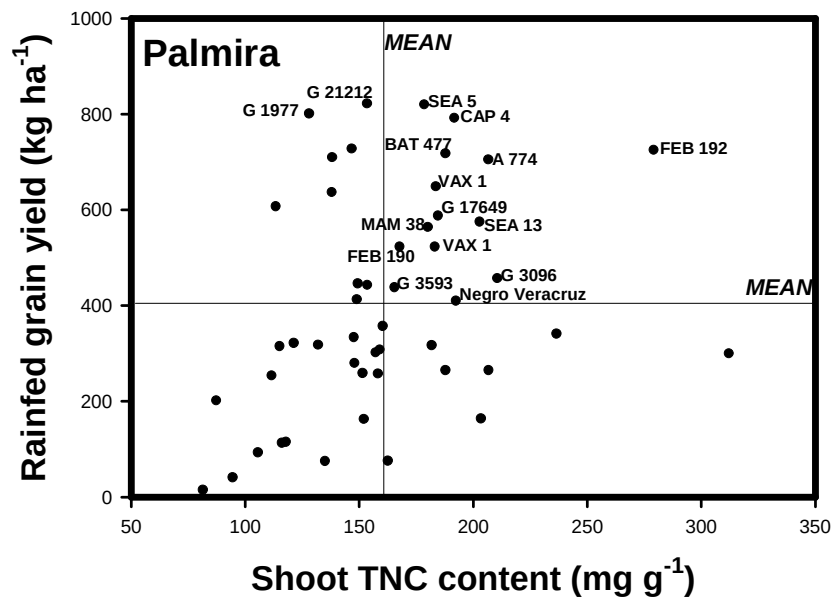


Figure 9. Identification of genotypes that combine superior seed yield with greater amount of total nonstructural carbohydrates (TNC) in the shoot when grown under rainfed conditions in a Mollisol at Palmira. Genotypes that were superior in grain yield and higher in shoot TNC were identified in the upper box of the right-hand side.

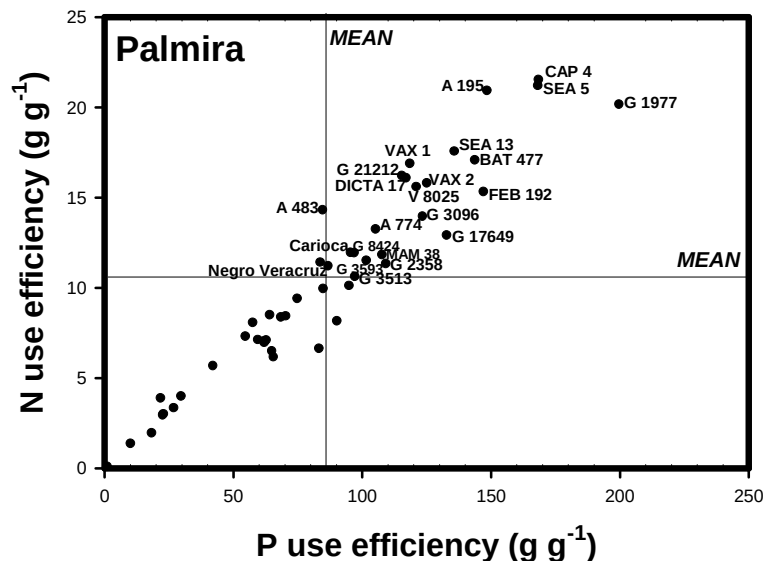


Figure 10. Identification of genotypes that are efficient in the utilization of N and P to produce greater seed yield when grown under rainfed conditions in a Mollisol at Palmira. Genotypes that were superior in both N and P use efficiency were identified in the upper box of the right-hand side.

Correlation coefficients between final grain yield and other plant attributes indicated that leaf area production, shoot biomass production, and shoot nutrient uptake were highly related to seed yield with rainfed conditions (Table 2). Significant positive relationship was also observed between seed yield and shoot TNC content under rainfed conditions. This observation indicates that the superior performers maintained a higher level of photosynthates in the shoot tissue. A negative relationship was observed between seed yield and seed nutrients (N and P) or seed ash content. Although few drought-adapted genotypes showed lower seed P content as observed from another field study that was reported last year; the relationship was not significant. We need to test this relationship using recombinant inbred lines.

Conclusions: This field study indicated that two bred lines (SEA 5 and CAP 4) and two landraces (G 21212 and G 1977) were very well adapted to drought stress. The superior performance of these four genotypes under drought was associated with resistance to soil-borne pathogens combined with their ability to mobilize photosynthates to developing grain and to utilize the acquired N and P more efficiently for grain production. Further work is needed to evaluate the usefulness of seed P as a selection method for identifying bean genotypes adapted to drought.

Table 2. Correlation coefficients (*r*) between final grain yield (kg ha⁻¹) and other plant attributes of 49 genotypes of common bean grown under rainfed conditions in a Mollisol at Palmira, Colombia.

Plant traits	Rainfed ^a	Irrigated ^a
Leaf area index	0.39***	0.18*
Shoot biomass	0.42***	0.44***
Shoot N uptake	0.36***	0.41***
Shoot P uptake	0.38***	0.40***
Shoot K uptake	0.38***	0.39***
Shoot Ca uptake	0.40***	0.29***
Shoot Mg uptake	0.31***	0.29***
Shoot Ca content	0.16	-0.31***
Shoot Mg content	0.02	-0.29***
Shoot TNC ^b	0.23**	0.19*
Shoot ash content	-0.15	-0.14
Seed N content	-0.11	-0.22**
Seed P content	-0.17	-0.07

a. * significant at *P* = 0.05, ** at *P* = 0.01, and *** at *P* = 0.001.

b. TNC = total nonstructural carbohydrate.

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Development and screening of lines for drought resistance

The drought selection project is focussed at present on Middle American types, especially for lowland Central America where the Niño effect has resulted in three consecutive dry years and serious losses in agricultural production. Although drought tolerance presents large genotype x environment (G x E) interaction, results in Palmira tend to correlate better with Central America than with other drought areas, such as highland Mexico. In January, all drought trials were lost because of excessive rainfall that resulted from the Niña effect. In the June planting, drought was much more reliable and the trials received only 60 mm of rainfall until about 62 days after planting, when rainfall resumed. Thus, stress in the June planting was moderately severe. Several trials were evaluated in this period.

Rationale: Several years of drought trials in CIAT had identified promising parental material in both the Mesoamerican and Durango races. Prior breeding had demonstrated that combining these two races could result in superior resistance, as in the case of the check variety, SEA 5.

Materials and methods: A set of 64 lines and checks were planted in a lattice design with three repetitions. These populations had from two to five different drought sources in their pedigrees and thus represented an attempt to recombine genes from several sources and bean races of the Middle American gene pool. In particular, race Durango figured prominently in these crosses. Most lines that were selected for testing have commercial grain color for Central America (red brilliant or black opaque).

Results: Lines that were comparable to the tolerant check (SEA 5) were identified in commercial colors (Table 3). In particular, the line identified as SX 12008-167 presented excellent tolerance and also good plant type, contrary to many other lines derived from race Durango. This represents substantial progress in the development of drought sources for this region, because in the past most sources were of cream or brown color. These new lines have already been incorporated into the crossing program to combine their drought tolerance with the necessary disease resistance, especially resistance to BGMV, which is present in most drought-prone tropical environments.

Table 3. Superior lines for drought tolerance derived from crosses among common bean races.

Line	Code	Color seed	Yield (kg ha ⁻¹)	Harvest days	100-seed weight (g)
13	SX 12008-180	Black	1479	69	23
12	SX 12008-167	Black	1429	72	17
23	SX 12008-261-2	Cream	1381	69	23
9	SX 12008-35-5	Brown	1364	72	24
2	SX 12293-121	Roxo	1349	63	25
41	SX 12010-40-1	Roxo	1291	73	23
22	SX 12008-261-1	Cream	1278	73	22
19	SX 12008-248-2	Black	1265	72	21
7	SX 12008-35-3	Brown	1248	72	25
8	SX 12008-35-4	Brown	1248	75	23
11	SX 12008-154	Cream mottled	1226	72	23
5	SX 12008-35-1	Brown	1224	77	19
	SEA 5 (tolerant check)	Cream	1189	67	23
	Tio Canela 75 (red check)	Red	1051	72	18
	DOR 390 (black check)	Black	886	79	17
	A 750 (susceptible check)	Purple mottled	115	88	26
	Mean		1063	72	22
	LSD (<i>P</i> = 0.05)		264	3	1

Conclusions: In the past, breeding for drought resistance had been managed as an independent project, outside the mainstream breeding program. The lines identified in this trial are much closer to commercial phenotype than lines identified in the past and can be used readily in the mainstream breeding program for Central America and the Caribbean.

Red-seeded lines from the mainstream breeding program

Rationale: Over the past 2 years, a set of red-seeded lines have been developed with resistance to important bean diseases of Central America, especially BGMV and CBB. These had been selected in conditions of medium fertility stress in early generations, but had not been evaluated systematically for drought resistance.

Materials and methods: Eighty-three of these lines were evaluated for drought tolerance in this same planting as the other trials described above, in a lattice design with two repetitions.

Results: A surprising level of tolerance was found in a number of lines, especially in those derived from variety Catrachita (Table 4). These lines were selected under fertility stress and possibly they have acquired a superior root system that is beneficial under drought as well. They have already been shipped to Central America for testing and these data will be shared with colleagues for use in the selection of promising materials.

Conclusions: Several lines presented a surprising level of drought tolerance and could be tested directly in Central America. As in the case of the lines mentioned above, they can be used readily in the mainstream breeding program for Central America.

Table 4. Best bean lines in the Central American red-seeded class under drought stress.

Line	Code	Color seed	Yield (kg ha ⁻¹)	Harvest days	100-seed weight (g)
33	MR 12439-18	Red	1767	76	23
78	MR 12326-53	Red	1709	74	25
30	MR 12439-105	Red	1702	72	27
71	MR 12326-40	Red	1641	73	21
27	MR 12439-105	Red	1615	73	27
79	MR 12326-53	Red	1612	72	23
51	MR 12438-93	Red	1592	76	25
18	MR 12439-6	Red	1535	72	23
15	MR 12746-48	Red	1521	79	19
72	MR 12326-48	Red	1498	74	21
13	MR 12826-31	Red	1494	75	22
80	MR 12326-53	Red	1485	73	23
	SEA 5 (tolerant check)	Cream	1407	71	24
	Catrachita (red check)	Red	1373	74	24
	Tio Canela 75 (red check)	Red	1044	79	18
	Orguloso (red check)	Red	672	72	22
	A 750 (susceptible check)	Purple mottled	228	88	26
	Mean		1213	74	22
	LSD ($P = 0.05$)		437	3	2

Lines derived from interspecific crosses

Rationale: The tepary bean (*Phaseolus acutifolius*) is highly resistant to drought. *P. parvifolius* is a close relative of *acutifolius*. Both *P. acutifolius* and *P. parvifolius* are desert species and can be crossed with *P. vulgaris* using embryo rescue. Interspecific lines were developed in past years to introgress resistance to CBB into common bean, but last year we initiated evaluations for drought tolerance with the same lines.

Materials and methods: A second drought trial contained lines derived from interspecific crosses of *P. vulgaris* with *P. acutifolius*. Several accessions of *P. acutifolius* that had been evaluated as especially resistant were included in the trial. In the present trial, 90 lines and checks were included in a lattice design with three repetitions, representing different amounts of contribution of the three species involved.

Results: Among the accessions of *P. acutifolius*, at least one yielded better than the tolerant *P. vulgaris* check, SEA 5 (Table 5). However, none of the lines presented yields comparable to the *P. vulgaris* check, SEA 5, but two significantly outyielded ICA Pijao, the *P. vulgaris* parent in all crosses and thus a more proper comparison to measure the impact of introgression of drought tolerance genes. Drought tolerance is assumed to be

governed by multiple quantitative genes, and thus the introgression of genes from interspecific crosses is an important milestone. However, the gap between the best lines and the best *P. acutifolius* is still wide, indicating that much remains to be done to take advantage of this tolerance. The lines were developed by the congruity backcross system in which the two species are alternated in each generation of backcrossing. This system is thought to favor the genetic recombination among homologous chromosomes and thus the introgression of foreign genetic material. A study of DNA markers (amplified fragment length polymorphism [AFLP]) has confirmed that introgression has been extensive across several lines (see SB-2 report).

Table 5. Yields of *Phaseolus acutifolius* and interspecific lines under drought stress.

Line	Code	Yield (kg ha ⁻¹)	Harvest days	100-seed weight (g)
41	4V3A1	1014	73	20
16	3V1A1P	921	80	17
	ICA Pijao	675	80	17
	SEA 5 (tolerant check)	1497	67	24
	G 40068 (<i>P. acutifolius</i>)	1742	62	13
	G 40159 (<i>P. acutifolius</i>)	1684	62	12
	A 750 (susceptible check)	237	88	26
	Mean	769	77	16
	LSD ($P = 0.05$)	246	5	2

Conclusions: The recovery of genes from tepary bean for drought resistance is an important advance, but at present these materials are still inferior to lines derived among common bean races. Much more work is necessary to introgress drought resistance from tepary to common bean.

Yield potential of early maturing lines

Rationale: Another solution to the problem of drought in Central America is the use of early maturing varieties that produce within a limited period of rainfall. Traditional Central American landraces typically are early maturing and are favored for this reason. However, early maturing beans normally have poor yield potential. Thus, improving yield capacity is an important aspect of selecting for earliness.

Materials and methods: A set of 36 lines was yield tested with and without supplemental lighting (18-hr daylength), to verify the photoperiod insensitivity of the lines. Lines were planted in the CIAT station at Santander de Quilichao with high fertility rates to alleviate the soil limitations that normally are present there.

Results: The treatment without supplemental lighting averaged 887 kg ha⁻¹. The bred line, BAT 304, was used as an early maturing check, because it is relatively early

compared to other commercial materials (maturing in 70 days in this trial). BAT 304 was in fact the latest material among the lines tested in this trial, and also was the best yielding (1992 kg ha⁻¹). However, two lines in particular (nos. 26 and 19) were noteworthy, not because of their absolute yields, but because their yields almost equaled that of BAT 304 in far less time—59-61 days (Table 6). These lines yielded substantially more than the Central American check varieties, Desarrural and Orguloso, especially under natural light.

Table 6. Best-yielding, early maturing bean lines under two treatments of daylength.

	Artificial light		Natural light	
	Days to harvest	Yield (kg ha ⁻¹)	Days to harvest	Yield (kg ha ⁻¹)
BAT 304	70	1373	70	1992
Line 26 ^a	61	1179	62	1851
Line 19 ^b	58	888	59	1641
Bola 60 Días	62	1104	64	1483
MCD 2004 (Desarrural)	63	1064	63	1092
Orguloso	59	490	57	873
Trial average	63	673	61	887
LSD (<i>P</i> = 0.05)	8.5	596	4.3	410

- a. (Othello x ICA Pijao) x ((A57 x XAN 159) x (BAT 477 x G 17341)).
b. (Chase x PEF 13) x (Early Ray x G 17341).

For reasons that are not understood, the lighted treatment presented lower yields (average = 673 kg ha⁻¹) than the unlighted treatment in general, although the better materials in the unlighted treatment showed the largest yield reductions in this treatment, and the poorest yielders without lights actually presented slightly better yields. Thus, in the lighted treatment, the differences between the best and the worst tended to be narrower. The apparent differential varietal response to lighting suggests that lighting had a real effect on yield, although it had no effect on flowering date for most materials. If this effect were to be confirmed, it would suggest some effects of photoperiod are independent of flowering date, which has been the standard criterion of photoperiod response in bean. Photoperiod response may be affecting photosynthate transport because of feedback inhibition of photosynthesis.

Conclusions: Yield potential of early maturing bean lines was substantially more than check cultivars under natural light and daylength. These materials will be incorporated into the Central American breeding program.

Contributors: IM Rao, S Beebe, H Terán, JM Osorno

1.1.3 Identification of traits associated with aluminum resistance

Rationale: The major soil-fertility related constraints to bean production in the tropics are low availability of P and N, and toxicity of Al and Mn associated with low pH in soil. Toxicity of Al in subsoils is a serious problem and amending subsoils with lime is not only difficult but also prohibitively expensive for resource-poor farmers. Last year, we reported results from a field evaluation of 77 genotypes for resistance to Al-toxic soils and tolerance to low nutrient supply, particularly low P and micronutrients. Based on those results, we selected 49 genotypes for further studies.

Previous research indicated significant genotypic variation in seed yield when grown in Al-toxic soils. These genotypic differences in seed yield could be related to differences in tolerance to Al, acquisition of nutrients, and utilization of nutrients for transport of photoassimilates to developing seeds. Genotypes that are adapted to Al-toxic soils are capable of acquiring essential macro- and micronutrients in a low-pH and high-Al environment.

Field studies were continued at Santander de Quilichao (990 m, Oxisol – Plinthic Kandiodox) to identify Al-resistant genotypes.

Materials and methods: A set of 49 genotypes, including germplasm accessions and bred lines, was evaluated in the field for identification of plant attributes for adaptation to infertile, acid soil conditions. These materials were evaluated in two seasons and the mean values from two trials are reported. Two levels of fertilizer (high and nil) input were applied. Plots with high fertilizer input (HFI) received banded application of P (40 kg ha^{-1}) in the form of triple superphosphate and foliar application (twice) of urea (1 kg ha^{-1}). Plots with no fertilizer input (NFI) received no application of nutrients. Soil characterization data of NFI plots showed toxic levels of exchangeable Al (66% Al saturation) and Mn (8 to 10 ppm) and low availability of Ca ($1.4 \text{ cmol}_c \text{ kg}^{-1}$) and Mg ($0.51 \text{ cmol}_c \text{ kg}^{-1}$) while P availability was more than adequate for plant growth and development. Plots of HFI treatment showed toxic levels of Mn (14 to 21 ppm) with very low levels of exchangeable Al.

A number of plant attributes were measured at mid-podfilling to determine genotypic variation in tolerance to toxic level of Al in soil. These plant traits included leaf area index, and canopy dry weight per plant and shoot nutrient (N, P, K, Ca, and Mg) uptake. At the time of harvest, grain yield and yield components (number of pods per plant, number of seeds per pod, 100-seed weight) were determined. Seed N and P contents were also determined.

Results and discussion: Among the 49 genotypes tested, three landraces (G 19227A, G 21212, and G 3513) and three bred lines (A 785, BAT 477, and SEA 5) were found to be outstanding in their adaptation to Al-toxic soil conditions (Figure 11). With NFI treatment, grain yield ranged from 67 to 606 kg ha^{-1} , while with HFI the range was from 236 to 1682 kg ha^{-1} . Grain yield of A 784 was 33% greater than that of a widely adapted genotype, Carioca, with NFI treatment.

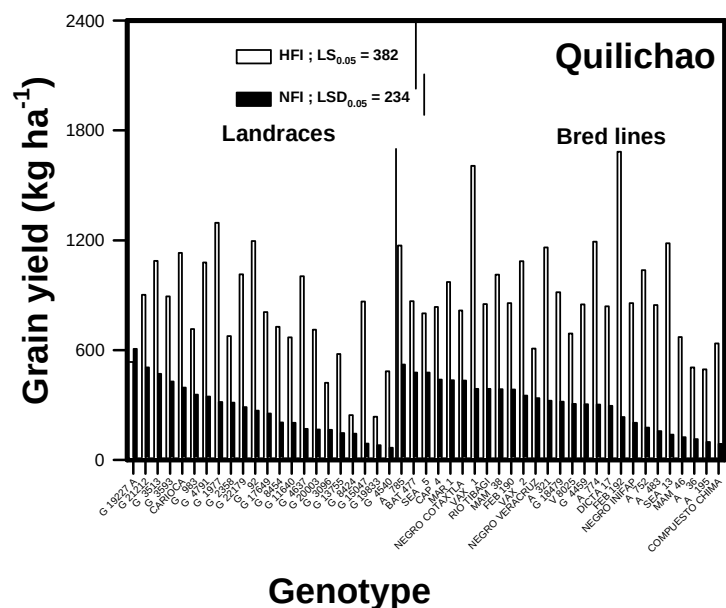


Figure 11. Genotypic variation in adaptation to Al-toxic soil at Santander de Quilichao, Colombia. (NFI = no fertilizer input, HFI = high fertilizer input).

The relationship between grain yield with NFI and that with HFI indicated that A 785 and G 3513 were better adapted to both low and high input of fertilizer (Figure 12).

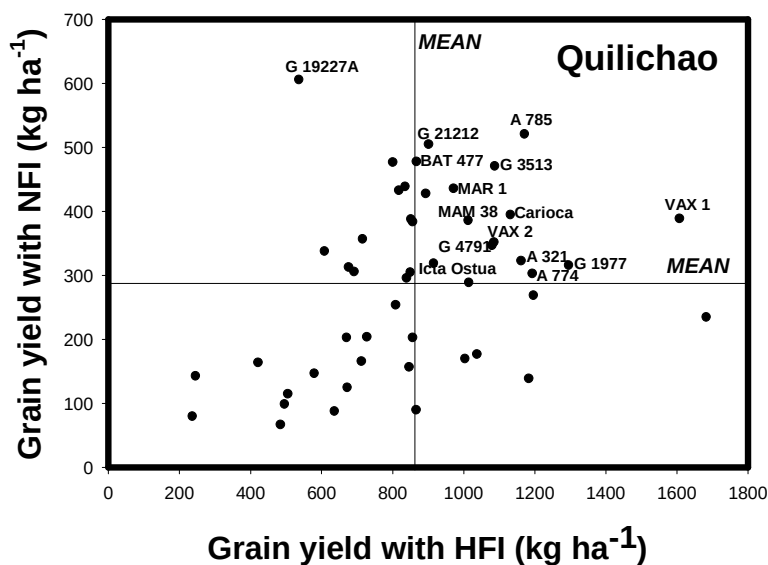


Figure 12. Identification of genotypes that are adapted to Al-toxic soil and are responsive to application of lime and P inputs to an Oxisol at Santander de Quilichao. Genotypes that gave superior yield with no fertilizer inputs (NFI) and were also responsive to application of high fertilizer inputs (HFI) were identified in the upper box of the right-hand side.

With NFI treatment, BAT 477 exhibited greater value of leaf area index at mid-podfilling (Figure 13). This was mainly because of the ability of this genotype to acquire greater amounts of P, Ca, and Mg from soil in the presence of toxic levels of Al (Figures 14 to 17). Shoot Ca content of G 19227A was markedly greater than that of the most of the genotypes tested (Figure 15).

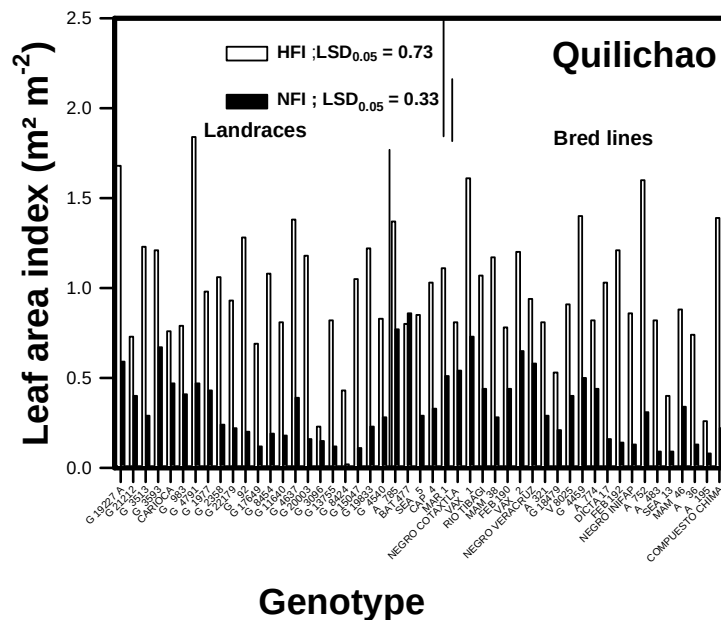


Figure 13. Genotypic variation in leaf area index of 20 genotypes of bean grown in an Oxisol at Santander de Quilichao. NFI= no fertilizer input, HFI= high fertilizer input.

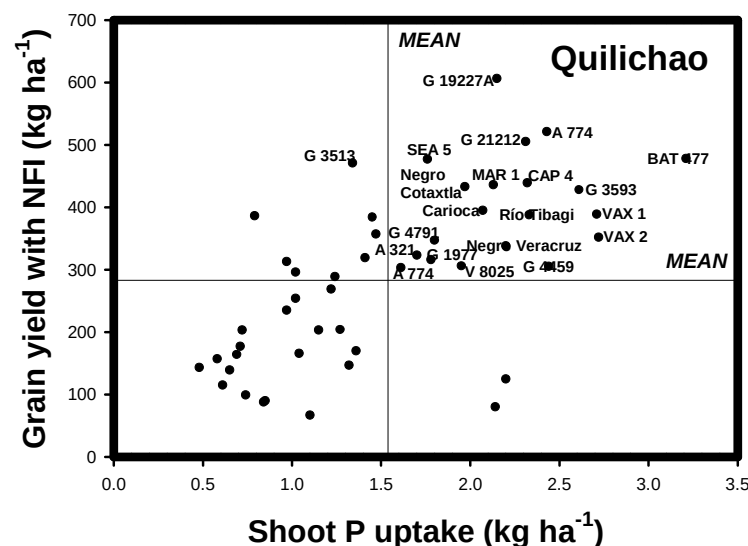


Figure 14. Identification of genotypes that combine superior seed yield with greater P uptake when grown with no fertilizer input (NFI) to an Oxisol at Santander de Quilichao. Genotypes that were superior in grain yield and P uptake were identified in the upper box of the right-hand side.

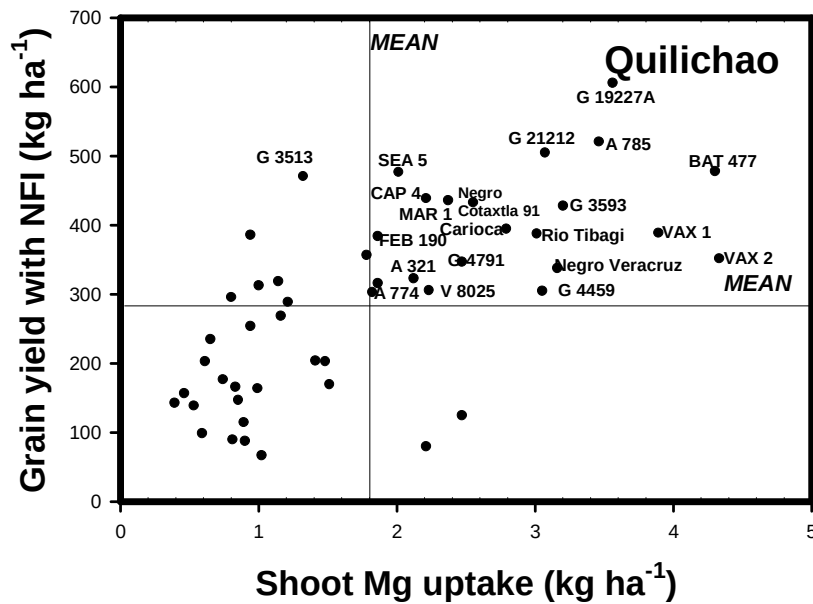


Figure 17. Identification of genotypes that combine superior seed yield with greater Mg uptake when grown with no fertilizer input (NFI) to an Oxisol at Santander de Quilichao. Genotypes that were superior in grain yield and P uptake were identified in the upper box of the right-hand side.

Genotypes that were adapted to Al-toxic soil conditions were also superior in their ability to acquire Ca and Mg from NFI treatment. This observation indicates the importance of Ca and Mg acquisition to seed yield when grown in Al-toxic soil.

Among the three best performers with NFI treatment, BAT 477 and SEA 5 were outstanding in combining greater seed yield with high content of N in seeds (Figure 18). It appears that these two bred lines not only are tolerant to toxic levels of Al in soil, but also are capable of mobilizing a greater proportion of shoot N to developing seeds. Among the land races and bred lines, FEB 190 was outstanding in its high N content in seed with moderate seed yield with NFI treatment. The bred line A 785 and the landrace G 21212 were outstanding in seed yield with NFI treatment, but were low in seed N content.

Correlation coefficients between final grain yield and other plant attributes indicated that leaf area production was highly related to seed yield with NFI treatment (Table 7). Seed yield was also significantly related to shoot nutrient uptake with NFI treatment. Significant negative relationship was observed between seed yield and seed P content indicating that greater P-use efficiency has contributed to superior adaptation to Al-toxic soil conditions.

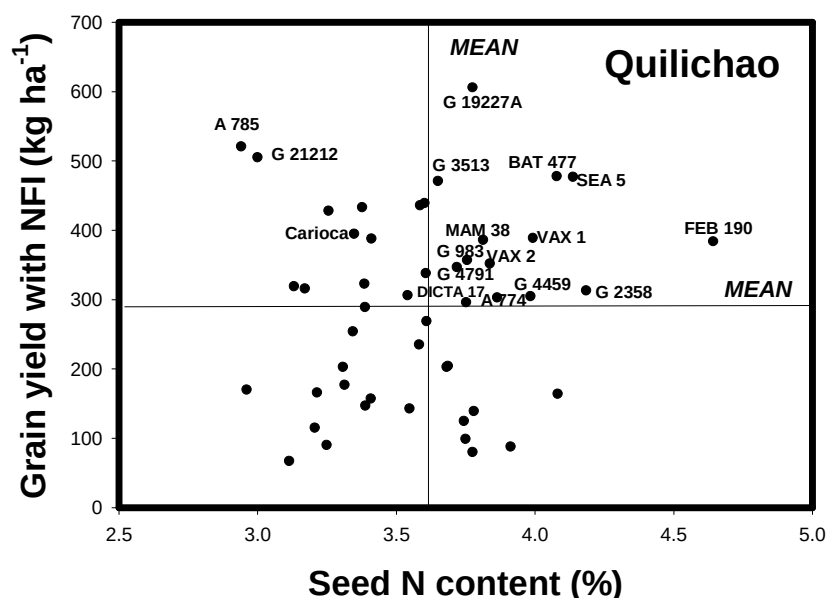


Figure 18. Identification of genotypes that combine superior seed yield with greater N content in seed when grown with no fertilizer input (NFI) to an Oxisol at Santander de Quilichao. Genotypes that were superior in grain yield and seed N were identified in the upper box of the right-hand side.

Table 7. Correlation coefficients (r) between final grain yield (kg ha^{-1}) and other plant attributes at mid-podfilling of 49 genotypes of common bean grown with no fertilizer input (NFI) or high fertilizer input (HFI) to an Oxisol at Santander de Quilichao.

Plant traits	NFI ^a	HFI ^a
Leaf area index	0.71***	0.41***
Shoot biomass	0.73***	0.52***
Shoot N uptake	0.73***	0.46***
Shoot P uptake	0.73***	0.45***
Shoot K uptake	0.73***	0.37***
Shoot Ca uptake	0.69***	0.54***
Shoot Mg uptake	0.73***	0.52***
Shoot Ca content	0.54***	0.31***
Shoot Mg content	0.48***	0.17*
Seed N content	-0.08	-0.16*
Seed P content	-0.32***	-0.35***

a. * = significant at $P = 0.05$, ** at $P = 0.01$, and *** at $P = 0.001$.

Conclusions: Results from this field study in Santander de Quilichao indicate that three bred lines (A 785, BAT 477, and SEA 5), and three germplasm accessions (G 19227A, G 21212, and G 3513) are superior in their resistance to Al. This study also showed that Al resistance can be combined with high seed N content.

Contributors: IM Rao, S Beebe, J Ricaurte, JM Osorno, H Terán, R García

1.1.4 Identification of traits associated with nutrient efficiency

Rationale: Last year, we reported results from a field evaluation of 77 genotypes for tolerance to low nutrient supply, particularly to low phosphorus and micronutrients. Based on those results, we selected 49 genotypes for further studies. Previous research indicated significant genotypic variation in seed yield with no fertilizer input (NFI) to a P-fixing Inceptisol. These genotypic differences in seed yield were associated with differences in acquisition of nutrients and utilization of nutrients for transport of photoassimilates to developing seeds. We also found that some genotypes were capable of combining high seed yield with higher level of N in the seed with NFI treatment.

Field studies were conducted at Popayán (1750 m, Inceptisol – Typic Dystropept) to identify nutrient efficient genotypes.

Materials and methods: The trial at Santander de Quilichao was duplicated at Popayán with the same genotypes and fertilizer inputs. Soil characterization data indicated low P availability (5 ppm) in NFI plots and deficiency of some microelements (Mn, Cu, Zn, and B) in both NFI and HFI treatments. These deficiencies of microelements resulted from heavy applications of lime over several years. The HFI treatment received 60 kg ha⁻¹ of P application. Micronutrients were applied by foliar application of Kelatex B, Zn, and Cu at the rate of 2 g L⁻¹ of water.

A number of plant attributes were measured at mid-podfilling to determine genotypic variation in tolerance to low nutrient supply in soil. These plant traits included leaf area index, canopy dry weight per plant, shoot nutrient (N, P, K, Ca, and Mg) uptake, and shoot TNC. At the time of harvest, grain yield and yield components (number of pods per plant, number of seeds per pod, 100 seed weight) were determined. Seed N and P contents and seed TNC were also determined.

Results and discussion: Among the 49 genotypes tested, four bred lines (A 785, V 8025, MAM 38, and DICTA 17) and one landrace (G 21212) were tolerant to low nutrient supply, particularly phosphorus and micronutrients (Figure 19). With HFI treatment, grain yield ranged from 152 to 1460 kg ha⁻¹, while with NFI the range was from 22 to 510 kg ha⁻¹. With NFI, grain yield of G 21212 was 2.5-fold greater than that of a widely adapted genotype, Carioca. Relationship between seed yield of NFI and HFI treatments indicated that the bred line A 785 was outstanding with NFI and at the same time highly responsive to application of fertilizer inputs (Figure 20). Acquisition of P by A 785 and G 21212 was only moderate, but seed yield was greater than that of the other genotypes

with NFI treatment (Figure 21). Two bred lines, MAM 38 and V 8025, were outstanding in their ability to acquire P from low-P soil.

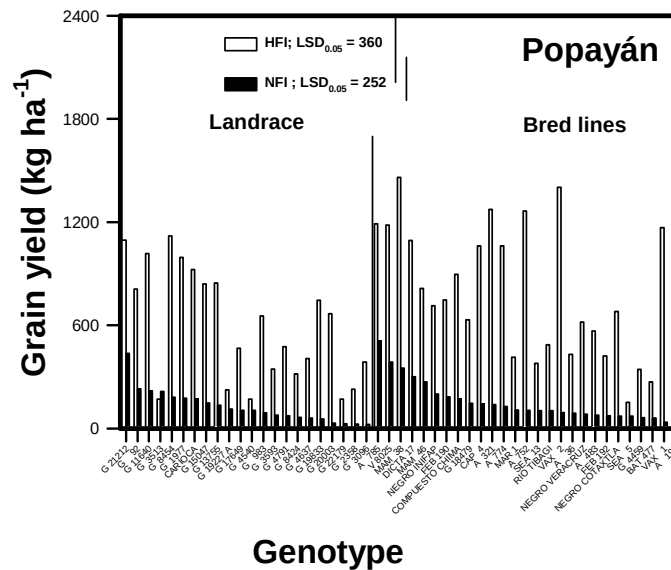


Figure 19. Genotypic variation in adaptation to low nutrient supply in an Inceptisol at Popayán. NFI = no fertilizer input, HFI = high fertilizer input.

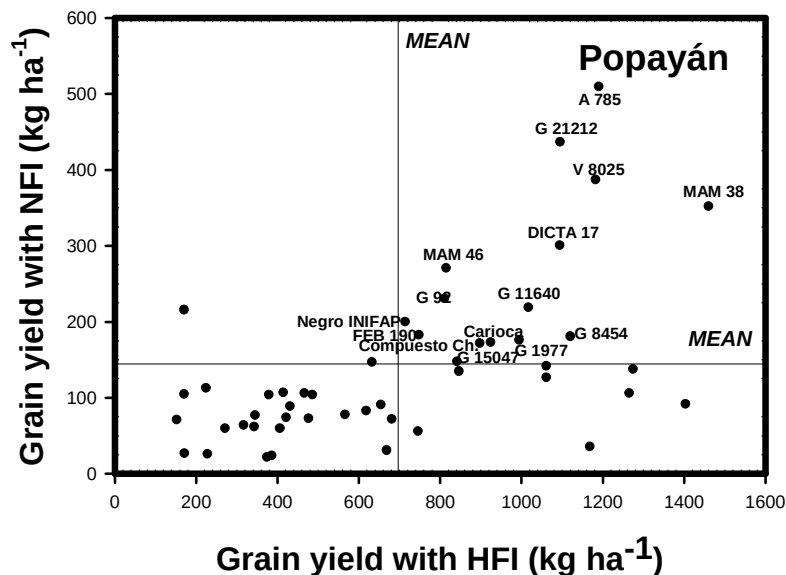


Figure 20. Identification of genotypes that are adapted to low supply of nutrients in soil and are responsive to application of lime and P inputs to an Inceptisol at Popayán. Genotypes that gave superior yield with no inputs and were also responsive to application of inputs were identified in the upper box of the right-hand side.

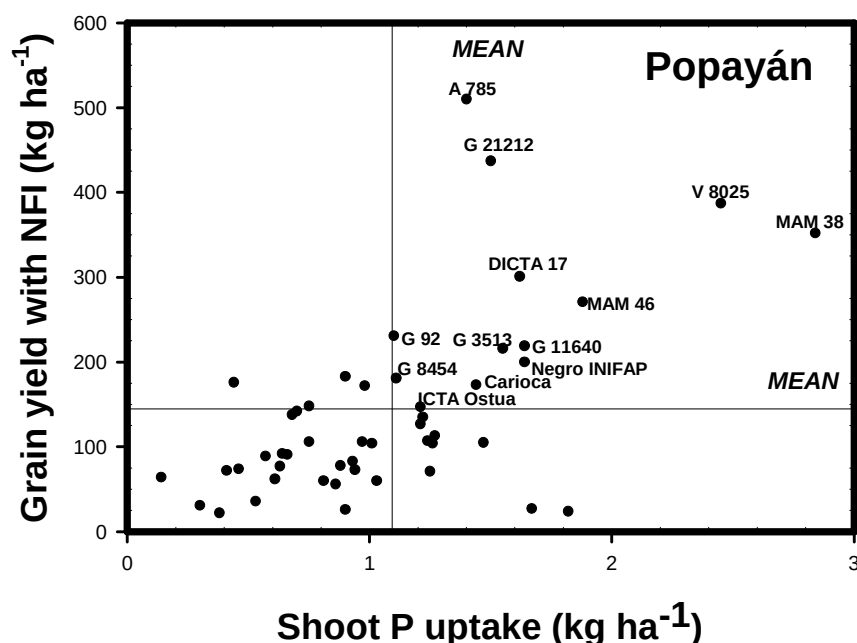


Figure 21. Identification of P-acquiring genotypes from soil with low P supply to an Inceptisol at Popayán. Genotypes that gave superior yield with no fertilizer input (NFI) and were also superior in acquiring P from low P supply in soil were identified in the upper box of the right-hand side.

The relationship between seed yield with NFI and seed P content indicated that the land race G 21212 yielded high with lower seed P content, while the bred line A 785 had moderate seed P-content and greatest seed yield (Figure 22). Moderate level of seed P is a desirable trait for seedling vigor when planted in low-P soils. High seed P-content could sometimes be considered less desirable in terms of nutritional value. Two bred lines, A 785 and BAT 477, combined greater seed yield with high seed N content with NFI treatment (Figure 23).

Genotypes that are efficient in mobilization of photosynthates or TNC can perform better with low P supply in soil (Figure 24). Relationship of grain yield and shoot TNC content with NFI treatment indicated that one bred line (A 785) and one land race (G 21212) were particularly efficient in mobilizing photosynthates to developing seeds. Of these two genotypes, G 21212 showed excellent grain filling under field conditions.

Three genotypes (G 1977, Negro Cotaxtla 91, and A 785) were found to be outstanding in their ability to combine greater N and P use efficiency (Figure 25). Among these three genotypes, G 1977 was particularly outstanding in efficient use of both N and P, both at this site and at Palmira under water stress conditions. One bred line (A 785) and one land race (G 1977) were identified as less P-demanding genotypes per unit seed yield production (Figure 26).

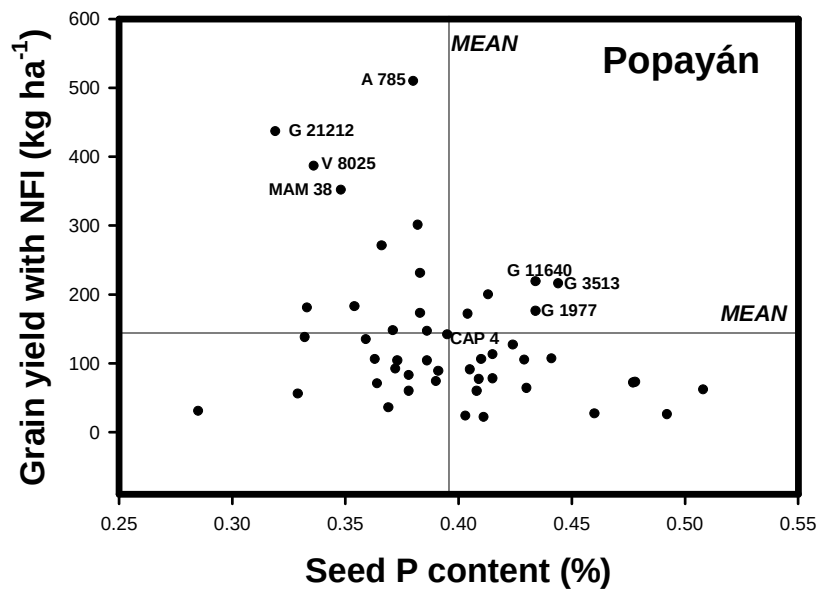


Figure 22. Identification of genotypes that combine superior seed yield with greater P content in seed when grown with no fertilizer input (NFI) in an Inceptisol at Popayán. Genotypes that were high in grain yield and low in seed P were identified in the upper box of the left-hand side.

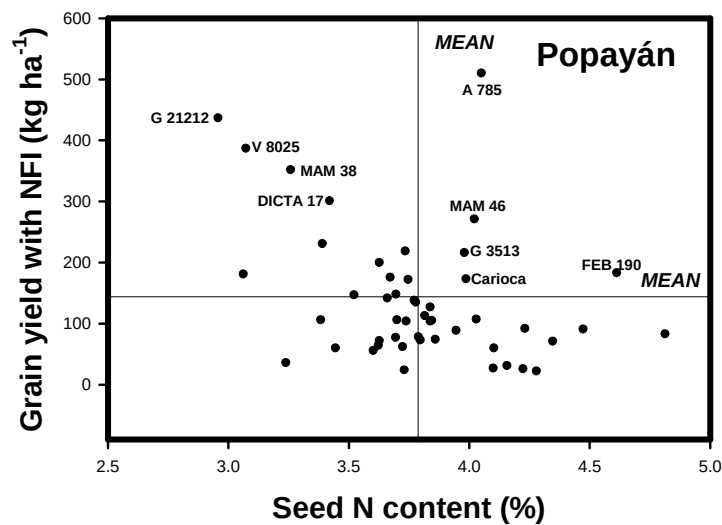


Figure 23. Identification of genotypes that combine superior seed yield with greater N content in seed when grown with no fertilizer input (NFI) in an Inceptisol at Popayán. Genotypes that were superior in grain yield and seed N were identified in the upper box of the right-hand side.

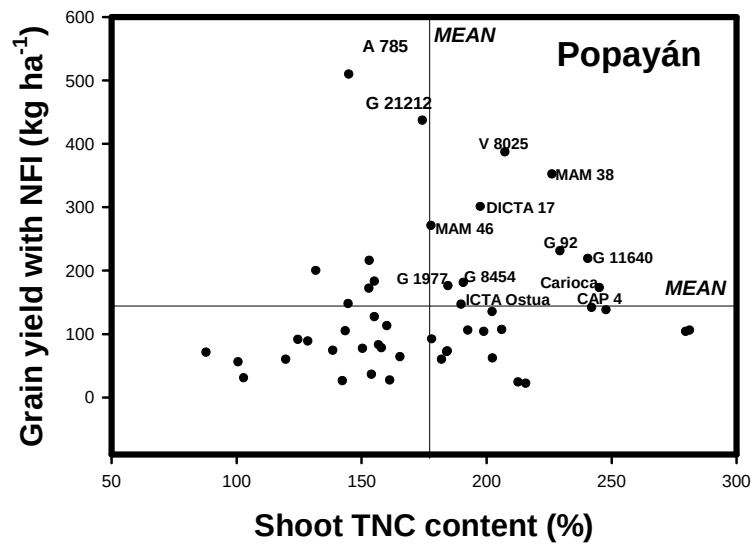


Figure 24. Identification of genotypes that combine superior seed yield with lower amount of total nonstructural carbohydrates (TNC) in the shoot when grown with no fertilizer input (NFI) in an Inceptisol at Popayán. Genotypes that were superior in grain yield and lower in shoot TNC were identified in the upper box of the left-hand side.

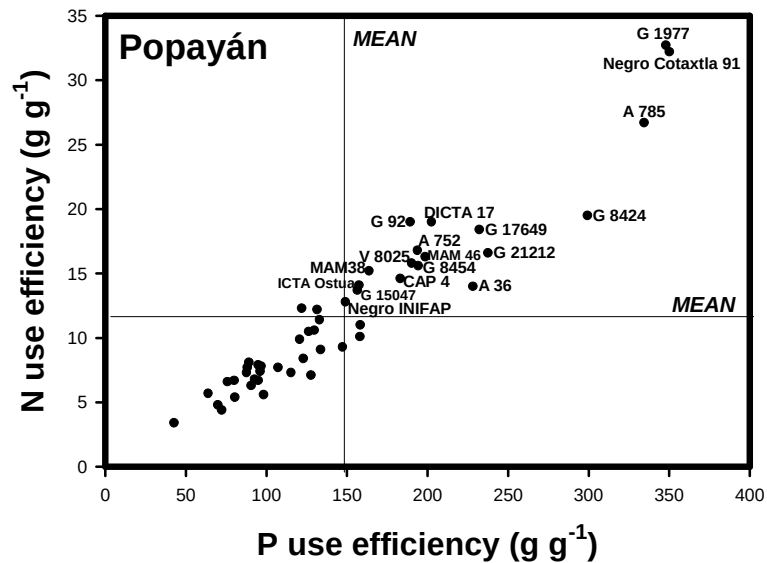


Figure 25. Identification of genotypes that are efficient in the utilization of N and P to produce greater seed yield when grown in low-P soil at Popayán. Genotypes that were efficient in N and P utilization were identified in the upper box of the right-hand side.

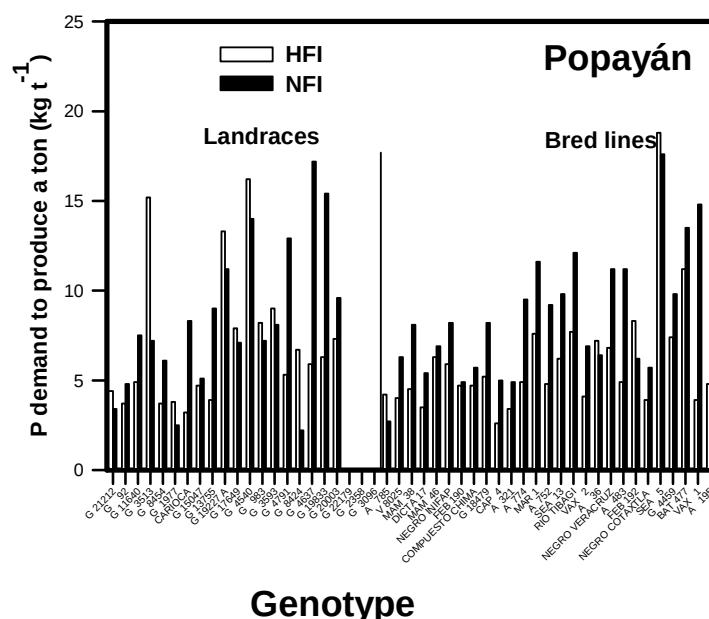


Figure 26. Genotypic differences in P demand to produce a ton of grain yield among 49 genotypes of common bean grown under nil (NFI) and high (HFI) fertilizer inputs to an Inceptisol at Popayán. P demand was defined as kg of shoot P uptake needed to produce one ton of seed yield.

Correlation coefficients between final grain yield and other plant attributes indicated that shoot biomass production, shoot N uptake, and shoot P uptake were highly related to seed yield with NFI treatment (Table 8). Significant negative relationship was observed between seed yield and shoot N and P content, indicating that greater N and P use efficiency has contributed to superior adaptation to low-P soil conditions.

Table 8. Correlation coefficients (r) between final grain yield (kg ha^{-1}) and other plant attributes of 49 genotypes of common bean grown under nil (NFI) or high (HFI) fertilizer input to an Inceptisol at Popayán, Colombia.

Plant traits	NFI ^a	HFI ^a
Shoot biomass	0.78***	0.76***
Shoot N uptake	0.72***	0.63***
Shoot P uptake	0.74***	0.62***
Seed N content	- 0.30***	- 0.29***
Seed P content	0.05	- 0.50***
N content in shoot biomass	- 0.47***	- 0.17*
P content in shoot biomass	- 0.19*	- 0.48***
TNC in shoot biomass	0.09	0.23**
Seed TNC content	0.14	- 0.03

a. * = significant at $P = 0.05$, ** at $P = 0.01$, and *** at $P = 0.001$.

Conclusions: This field study indicated that four bred lines (A 785, V 8025, MAM 38, and DICTA 17) and one landrace (G 21212) were tolerant to low nutrient supply, particularly phosphorus and micronutrients. One of the bred lines (MAM 38) was highly responsive to high fertilizer input in terms of seed yield.

Contributors: IM Rao, S Beebe, J Ricaurte, JM Osorno, H Terán, R García

1.1.5 Identification of genotypes with tolerance to waterlogging

Rationale: Climatic fluctuations have become more accentuated in Central America over the last few years. While climatologists still do not agree whether this is a long-term trend, farmers are in fact facing alternating periods of drought and excess water that seem to be more extreme than in the past. Hurricane Mitch was a graphic example of this.

Materials and methods: No trials were consciously planned to study flooding tolerance. However, the Niña effect in the April planting season produced record rainfall in the Santander de Quilichao station and plots were flooded once or twice a week. This led to an unusual level of stress on the plots and marked genotypic differences were observed.

Results: Some populations were lost entirely, but great differences in tolerance to these conditions were observed consistently across studies and fields. The line A 774, a mulatinho type developed for Brazil, presented the best tolerance, followed by the CBB-resistant line, VAX 1. Both lines are used as checks across trials and fields, and thus their superiority was evident and confirmed. Garbancillo Zarco, a Mexican climbing bean, also performed very well. Red-seeded populations presented broad variability, while the commonly used black-seeded parents (DOR 390, A 785, etc) were largely sensitive to flooding.

Conclusions: Flooding tolerance could well be relevant for regions such as Central America if climatic fluctuations become more radical. However, the precise stress in these trials is not clear. Although flooding and poor root aeration was undoubtedly a primary stress, it might have been confounded with nitrogen deficiency and with soil toxicities, especially manganese.

Contributors: S Beebe, H Terán, JM Osorno

1.1.6 Evaluation of symbiotic nitrogen fixation (SNF) in recombinant inbred lines

Rationale: Nitrogen is the most limiting nutrient in bean production after phosphorus. The bred line, BAT 477, was recognized long ago as superior in SNF capacity, including under drought and phosphorus stress. It had been used to develop recombinant inbred lines (RILs) that were evaluated in past years for SNF capacity in the greenhouse, and for yield under P stress and under drought, with the corresponding unstressed treatments. However, an evaluation of SNF capacity in the field had not been carried out.

Materials and methods: In the past year, another trial with the same RILs was financed by Agency for International Development (AID) funds with the University of Minnesota. The RILs were planted in small plots in four repetitions at a field site with sandy soil and severe N-deficiency. Data were taken on total N accumulated and the yield on small plots. Nitrogen accumulation in the Minnesota trial was compared to yield in phosphorus and drought stress trials in Colombia.

Results: When data were compiled across yield trials and the SNF trial, lines were identified with multiple stress tolerance (Table 9). One of these (line 98) was particularly outstanding across stress treatments and was incorporated into the breeding program. This once again highlights the potential for obtaining multiple stress tolerance. A broad subset of 30 lines was selected for physiological analysis for a Cuban MSc student. A comprehensive QTL analysis is pending to evaluate QTL for several traits: SNF capacity in the greenhouse with low-P stress, SNF in the field, yield under low-P stress, yield under drought, and seed nitrogen content.

Table 9. Total nitrogen as measured in a sandy soil in Minnesota, USA, and yield under drought and phosphorus stresses, of recombinant inbred lines developed from the cross BAT 477 x DOR 364.

Line	Total N (mg N per 4 plants) 4 reps	Yield (kg ha ⁻¹)			Color
		High P 3 reps	Low P 3 reps	Drought 2 reps	
DOR 364	1543	3331	683	297	
BAT 477	2282	3721	998	383	
SEA 5	-	-	-	648	
G3513	-	3188	990	-	
Carioca	-	3626	901	-	
Line 4	2580	3261	553	1028	Cream brilliant
Line 14	2392	3661	671	703	Brown brilliant
Line 16	2456	3712	549	875	Pink semi-br
Line 25	2544	3527	958	669	Black brilliant
Line 49	2509	3415	851	648	Cream-brown br
Line 61	2712	3899	794	685	Brown brilliant
Line 98	2668	3769	996	724	Black opaque
Mean	2388	3448	723	516	
LSD (<i>P</i> = 0.05)		714	318	229	

Conclusion: BAT 477 continues to be an excellent source of multiple stress tolerance. Progenies of BAT 477 demonstrate resistance to multiple stresses including low P, and drought and nitrogen stress, and can make an important contribution to yield stability.

Contributors: S Beebe, H Terán (IP-1); P Graham (University of Minnesota)

Progress towards achieving output milestones:

➤ Parents/populations/lines tolerant to drought/low soil fertility available

We have identified a number of genotypes that are better adapted to drought and low soil fertility conditions. Among these, G 21212, a landrace from Colombia, was outstanding in its adaptation to abiotic stress factors such as drought, low P supply in soil and Al toxicity. In collaboration with partners from national agricultural research systems (NARS) and nongovernmental organizations (NGOs), we are evaluating a set of 49 genotypes including landraces and bred lines. This will allow the breeders from national programs to genetically recombine abiotic with biotic stress adaptation and with commercial grain quality.

Activity 1.2 Developing germplasm with multiple resistance to diseases

Highlights:

- The selection of the *bgm-1* gene was applied to about 9000 individual plants and the backcross program to improve commercial varieties with intermediate resistance to BGYMV is nearing completion.
- Sources of resistance to BGMV, BCMV, BSMV, and CCMV were identified in parental materials, and in intermediate and advanced common bean breeding lines.
- Resistance to ALS was identified in a number of *P. vulgaris*, *P. polyanthus*, and *P. coccineus* genotypes.
- Potential sources of ALS resistance were identified in interspecific crosses between *P. vulgaris* and *P. polyanthus* or *P. coccineus*.
- 25 bean genotypes with high levels of resistance to *Phaeoisariopsis griseola* races from Africa, Central America, and South America were identified.
- 10 bean genotypes with high levels of resistance to *Macrophomina phaseolina* were identified.
- Breeding for resistance to *Ascochyta* blight has been successful in creating families with superior resistance that will serve to improve both major gene pools and snap bean types.

1.2.1 Mainstream breeding F₁, F₂, F₃, including *bgm-1* selection

Rationale: Central America is a priority region for poverty alleviation and for bean production. Therefore, the mainstream breeding program continued to focus on small red and small black beans, largely for Central America, with a smaller effort in Carioca type. Bean golden yellow mosaic virus continues to be an indispensable breeding priority for this region.

Materials and methods: Selection was practiced at four principal sites: Palmira (drought, molecular markers, and seed increase), Santander de Quilichao (poor soil fertility, CBB, and ALS), Popayán (anthracnose), and Darien (adaptation to a mid-altitude site and low P). Selection of the *bgm-1* gene for resistance to BGMV continued, as reported last year, as part of the gamete selection scheme with the purpose of identifying F₁ plants that carry the gene, and on advanced lines. As a complement to the mainstream selection program, certain elite commercial varieties with an intermediate level of resistance to BGMV, but lacking the *bgm-1* gene were selected for introducing this gene and thereby improving their resistance. A source of *bgm-1*, SAM 1, also is highly resistant to CBB.

Results: In 2000, about 60 red-seeded lines were selected across these sites and were shipped to the Escuela Agrícola Panamericana (EAP-Zamorano) for inclusion in regional nurseries in Central America through the PROFRIJOL network. These were the same lines that were evaluated under drought stress in Palmira (Table 4). Compared to previous lines, these represent advances in resistance to CBB and in lesser degree to ALS, in combination with resistance to BGYMV and modest resistance to poor soil and drought. In general, resistance to ALS still needs to be fortified with new sources of resistance. Meanwhile, early generation populations incorporating an increasing contribution of abiotic stress tolerance were selected in F₁ to F₄ generations. About 1200 lines of red and black seed types are being evaluated in Darien (the environment that is most similar to Central American production environments) to develop a set of 150-200 lines for distribution in 2001.

Selection of the *bgm-1* gene for resistance to BGMV was applied to another 9000 F₁ plants. The plan to introduce the *bgm-1* gene by backcrossing into commercial varieties advanced to the F₂BC₂ generation. In the F₁BC₁, phenotypic selection was also practiced for resistance to CBB, prior to making the BC₂. At present, the most advanced lines are in BC₂F₃ and they are being purified for *bgm-1* and for CBB reaction. Because SAM 1 was not phenotypically distant from the recurrent parents, two cycles of backcrossing are deemed sufficient to recover the phenotype of the recurrent parent.

Conclusions: Marker-assisted selection continues to be an indispensable part of the breeding effort and has served to create lines with multiple disease resistance. It should be extended to other critical disease resistance genes as these are identified and tagged with reliable PCR markers. Resistance to ALS, to regionally important viruses (BSMV and CCMV), and to the abiotic stresses still needs to be strengthened.

Contributors: S Beebe, G Mahuku, H Terán, C Jara, JM Osorno, C Cajiao (IP-1);
C Quintero, J Tohme (SB-2)

1.2.2 Bean common mosaic virus (BCMV)

Resistance to viral diseases of common bean in the tropics remains an important breeding activity in the Bean Project because these diseases can severely affect bean production in the main target regions. Bean virology activities seek to identify sources of resistance to emerging bean diseases, such as “amachamiento” (bean sterility), caused by CCMV. Also, effective disease screening methodologies are implemented to select breeding lines possessing resistance to the endemic (BCMV and BGMV) and sporadic (BSMV) viral diseases that limit bean production in the tropics.

Rationale: The BCMV is the most widely distributed pathogen of common bean in the world. Thus genetic resistance continues to be incorporated in all breeding lines developed by CIAT in collaboration with national agricultural research institutes (NARIs), as the main prerequisite for germplasm development.

Materials and methods: The screening methodology implemented at CIAT for BCMV targets segregating populations in order to select homozygous BCMV-resistant lines in the F₃ generation (dominant monogenic resistance) or in subsequent generations (dominant/recessive genes). Hence, 1200 entries from segregating populations were screened this year for their reaction to BCMV. Also, 960 lines from multiple crosses were evaluated this year for their resistance to this virus. Incorporating BCMV resistance in red-seeded Central American cultivars has been a difficult task because of genetic linkage problems between genes determining BCMV-susceptibility and the red seed-coat color. This year, we evaluated 83 Central American, red-seeded materials for their reaction to BCMV.

Results: These evaluations showed that 91% of the materials possessed monogenic dominant resistance to BCMV, 5% were susceptible to the virus, and 4% were segregating for this trait (R/S). This proportion of BCMV-resistant lines would be a major achievement for other bean-producing regions of Latin America. However, this type of resistance is not adequate for those Central American countries, such as El Salvador, where the commercial red-seeded varieties are preferred (varieties with monogenic dominant resistance to BCMV have a darker red seed-coat color, which reduces their commercial value). Also, a different viral disease (BSMV) selectively attacks these genotypes.

Conclusions: The strategy proposed to counteract this problem was the gradual replacement of bean genotypes possessing monogenic dominant resistance by genotypes protected against BCMV by the recessive *bc 3* gene. This gene is not selectively attacked by BSMV and does not have a linkage problem related to seed color and BCMV susceptibility. However, 44 potential *bc 3* donor parents evaluated under field conditions in El Salvador last year succumbed to BGMV. This experience clearly points out the need to breed for multiple disease resistance, in this case to BGMV, BCMV, and BSMV simultaneously. This approach was followed this year with 96 advanced materials possessing multiple disease resistance to different pathogens, including bacterial blight and viral diseases. Of the 96 lines screened, 28% showed multiple resistance to the biotic factors mentioned.

Contributors: FJ Morales, M Castaño, CJ Alvarez

1.2.3 Bean severe mosaic virus (BSMV)

Rationale: Bean severe mosaic is a generic name given to a complex disease of common beans caused by different strains of cowpea severe mosaic virus (CPSMV) in Latin America. The problem is more “severe” in bean cultivars possessing monogenic dominant resistance to BCMV because of hypersensitive reactions between the pathogen and the dominant *I* gene. Also, the disease is more prevalent in Central America, where the chrysomelid vectors of the causal viruses are common in bean plantings. In bean cultivars possessing recessive resistance to BCMV, the disease is not as severe, and some genotypes only show mild mosaic symptoms.

Materials and methods: We have finished the evaluation of a Bean Core Collection of 1218 accessions for their reaction to CPSMV. These evaluations were conducted under controlled conditions by standard mechanical inoculation methods.

Results: About 88% of the accessions evaluated reacted with CPSMV, thus indicating that those genotypes were devoid of the dominant *I* gene; whereas 10% of the accessions inoculated reacted with mosaic and necrosis (bean severe syndrome), demonstrating the presence of the dominant *I* gene in these genotypes. About 2% of the accessions evaluated reacted with top necrosis, a reaction that is conditioned by yet another dominant gene previously characterized at CIAT (Morales and Singh 1997¹) as *Anv*, apparently epistatic to the dominant *I* gene.

Conclusions: No immune genotypes were detected in these evaluations, but many of the accessions that reacted with mild mosaic could be used as potential sources of resistance or tolerance to CPSMV. Nevertheless, it is important to replace the dominant *I* gene with the recessive *bc 3* gene for resistance to BCMV/BCMNV in the breeding project for the improvement of Central American common bean cultivars.

Contributors: FJ Morales, M Castaño, CJ Alvarez

1.2.4 Bean “sterility” (amachamiento)

Rationale: “Amachamiento” is a syndrome associated with a marked yield reduction of bean plantings in Costa Rica and Nicaragua. The etiology of this syndrome was elucidated in 1998, as a viral disease caused by the chrysomelid-transmitted CCMV, often found associated with BCMV in local bean cultivars. The CCMV was known to attack beans in the lowlands of Central America and the Caribbean, where it caused the disease previously known as “bean yellow stipple”. At higher altitudes, where this virus is now located, the characteristic yellowing disappears, to be replaced by foliar distortion. However, the main effect of the virus continues to be the significant yield reduction it causes both in the lowland and mid-altitude bean production areas.

Materials and methods: A yield loss experiment was conducted under controlled conditions over a 2-year period, with eight selected bean cultivars manually inoculated in six replications. The line DOR 364 is a BGYMV-resistant cultivar widely adopted in Central America. We also evaluated three genotypes of the Durango race of common beans, previously shown to have field resistance to a variety of different bean viruses.

Results: The red-seeded landrace “Sacapobres” suffered an average 52% yield loss, whereas the improved “Talamanca” cultivar showed an average yield loss of 34% under the experiment glasshouse conditions of the study. The DOR 364 is a BGYMV-resistant line (cultivar) widely adopted in Central America. We tested this line for its reaction to CCMV, and observed an average yield reduction of 45% in five replications. We also

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¹ Morales FJ, Singh SP. 1997. Inheritance of the mosaic and necroses reactions induced by bean severe mosaic common viruses in *Phaseolus vulgaris* L. *Euphytica* 93:223-226.

evaluated three genotypes of the Durango race of common beans, previously shown to have field resistance to a variety of different bean viruses. Under the test conditions selected, Pinto 114 suffered average yield reduction of 6.6%, Great Northern 123 of 37%, and Red Mexican 35 of 52.4%. Another source of resistance to bean viruses, belonging to the Nueva Granada (Andean) race, cultivar “Red Kloud”, showed the highest level of resistance in these tests, with an average yield loss of 0% and a range between 0% and 25%.

Conclusions: Possibly the significant yield loss of “Sacapobres” could be further aggravated by its susceptibility to BCMV under field conditions. We will test this possibility next year. In field visits made to the affected Brunca region of southern Costa Rica, it was evident that “Talamanca” had excellent vegetative growth, but very few, if any, pods per plant. The “amachamiento” symptoms on this cultivar are expressed mainly as a mild mottle, so possibly the cultivar is also showing symptoms of poor adaptation to that region. Talamanca is a black-seeded genotype developed in Colombia. However, it was tested for adaptation in the region of Perez Zeledón, not far from the affected areas visited in Costa Rica, which were located at a higher altitude. Whereas it was apparent that yield under the artificial test conditions of this study was significantly influenced by the varying environmental conditions found during the year, the averages given here reflect the expected level of resistance to CCMV in each of the common bean genotypes tested. Thus, and in anticipation of further germplasm screening work, we recommend the genotypes Porrillo Sintético and Red Kloud for crop improvement purposes in the presence of CCMV.

Contributors: FJ Morales, M Castaño, CJ Alvarez

1.2.5 Bean golden yellow mosaic virus (BGYMV)

Rationale: This virus is one of the most devastating problems of common bean in the lowlands and mid-altitude valleys of Central America.

Materials and methods: This year, 83 selected red-seeded advanced lines bred for Central America were evaluated for their reaction to BGYMV under controlled conditions, following a standard mechanical inoculation methodology.

Results: Of the 83 lines, 21 (about 25%) showed a high level of virus resistance, 37 (44%) were moderately resistant, and the remaining lines (31%) were susceptible to BGYMV.

Conclusions: The high proportion of virus-resistant lines (>50%) reflects the use of BGYMV-resistant parents in their crosses, namely DOR 482, ICA Pijao, DOR 364, MD-30-75, Red MEX 35, and Tio Canela (DOR 364, 367, 391, 483).

Contributors: FJ Morales, M Castaño, CJ Alvarez

Summary: Table 10 shows the reaction of the selected 83 red-seeded lines to four different viral diseases prevalent in Central America. As can be concluded from these results, most of the red-seeded genotypes possess monogenic dominant resistance to BCMV, which renders these materials susceptible to BSMV, and creates genetic linkage problems that hinder the selection of commercial red-seeded grain types. An effort must be made to rapidly change the breeding strategy to replace the dominant *I* gene with the recessive *bc 3* gene.

Table 10. Reaction^a of 83 selected red-seeded lines to four different viral diseases^b prevalent in Central America.

Entry	Code	Identification	BCMV	BGMV	BSMV	CCMV
1	MN12917-26	ICA Pijao x (XAN 252 x MAR 1) x (DOR 482 x J117)	R(I)	S	S	I
2	MN12917-22	ICA Pijao x (XAN 252 x MAR 1) x (DOR 482 x J117)	R(I)	S	S	I
3	MN12917-22	ICA Pijao x (XAN 252 x MAR 1) x (DOR 482 x J117)	R(I)	R	S	I
4	MN12917-22	ICA Pijao x (XAN 252 x MAR 1) x (DOR 482 x J117)	R(I)	R	S	I
5	MN12917-22	ICA Pijao x (XAN 252 x MAR 1) x (DOR 482 x J117)	R(I)	S	S	I
6	MN12917-4	ICA Pijao x (XAN 252 x MAR 1) x (DOR 482 x J117)	R(I)	S	S	I
7	MR12826-8	VAX3 x MD30-75 x DOR 364 x MAR 1 x BELD x XAN 309	R(I)	S	S	I
8	MR12826-8	VAX3 x MD30-75 x DOR 364 x MAR 1 x BELD x XAN 309	R(I)	S	S	I
9	MR12826-8	VAX3 x MD30-75 x DOR 364 x MAR 1 x BELD x XAN 309	R(I)	S	S	I
10	MR12826-8	VAX3 x MD30-75 x DOR 364 x MAR 1 x BELD x XAN 309	R(I)	S	S	I
11	MR12826-8	VAX3 x MD30-75 x DOR 364 x MAR 1 x BELD x XAN 309	R(I)	R	S	I
12	MR12826-8	VAX3 x MD30-75 x DOR 364 x MAR 1 x BELD x XAN 309	R(I)	S	S	I
13	MR12826-31	VAX3 x MD30-75 x DOR 364 x MAR 1 x BELD x XAN 309	R(I)	R	S	I
14	MR12826-32	VAX3 x MD30-75 x DOR 364 x MAR 1 x BELD x XAN 309	R(I)	S	S	I
15	MR12746-48	MD30-75 x PVPA 9576 x XAN 310 x CATRACH x MAR 1	R(I)	R	S	I
16	MR12747-10	MD30-75 x PVPA 9576 x XAN 310 x CATRACH x MAR 1	R(I)	S	S	S
17	MR12910-15	XAN 252 x MAR 1 x DOR 482 x J 117	R(I)	S	S	I
18	MR12439-6	Catrachita x XAN 309 x Orgullo x Tio Canela	R(I)	S	S	I
19	MR12439-31	Catrachita x XAN 309 x Orgullo x Tio Canela	R(I)	R	S	I
20	MR12439-31	Catrachita x XAN 309 x Orgullo x Tio Canela	R(I)	R	S	I
21	MR12439-31	Catrachita x XAN 309 x Orgullo x Tio Canela	R(I)	R	S	I
22	MR12439-31	Catrachita x XAN 309 x Orgullo x Tio Canela	R(I)	R	S	I
23	MR12439-31	Catrachita x XAN 309 x Orgullo x Tio Canela	R(I)	R	S	I
24	MR12439-31	Catrachita x XAN 309 x Orgullo x Tio Canela	R(I)	R	S	I
25	MR12439-31	Catrachita x XAN 309 x Orgullo x Tio Canela	R(I)	R	S	I
26	MR12439-105	Catrachita x XAN 309 x Orgullo x Tio Canela	R(I)	R	S	I
27	MR12439-105	Catrachita x XAN 309 x Orgullo x Tio Canela	R(I)	R	S	I
28	MR12439-105	Catrachita x XAN 309 x Orgullo x Tio Canela	R(I)	R	S	I
29	MR12439-105	Catrachita x XAN 309 x Orgullo x Tio Canela	R(I)	R	S	I
30	MR12439-105	Catrachita x XAN 309 x Orgullo x Tio Canela	R(I)	R	S	I
31	MR12439-18	Catrachita x XAN 309 x Orgullo x Tio Canela	R(I)	R	S	I
32	MR12439-18	Catrachita x XAN 309 x Orgullo x Tio Canela	R(I)	R	S	I
33	MR12439-18	Catrachita x XAN 309 x Orgullo x Tio Canela	R(I)	R	S	I
34	MR12439-18	Catrachita x XAN 309 x Orgullo x Tio Canela	R(I)	R	S	I
35	MR12437-3	MD30-75 x PVPA 9576 x XAN 310	R(I)	R	S	I
36	MR12437-3	MD30-75 x PVPA 9576 x XAN 310	R(I)	R	S	I
37	MR12437-3	MD30-75 x PVPA 9576 x XAN 310	R(I)	R	S	I
38	MR12437-2	MD30-75 x PVPA 9576 x XAN 310	R(I)	R	S	I
39	MR12437-2	Orgullo x Tio Canela x XAN 309 x G 17341 x DCelaya	R(I)	R	S	S
40	MR12437-2	Orgullo x Tio Canela x XAN 309 x G 17341 x DCelaya	R(I)	R	S	S
41	MR12437-135	Orgullo x Tio Canela x XAN 309 x G 17341 x DCelaya	R(I)	S	S	S
42	MR12437-135	Orgullo x Tio Canela x XAN 309 x G 17341 x DCelaya	R(I)	S	S	I
43	MR12437-135	Orgullo x Tio Canela x XAN 309 x G 17341 x DCelaya	R(I)	R	S	I
44	MR12437-135	Orgullo x Tio Canela x XAN 309 x G 17341 x DCelaya	S	S	M	I
45	MR12437-168	Orgullo x Tio Canela x XAN 309 x G 17341 x DCelaya	R(I)	S	S	I
46	MR12438-76	RMEX 35 x Rojo Seda x Tio Canela x XAN 309 x Orgullo	R(I)	S	S	I
47	MR12438-81	RMEX 35 x Rojo Seda x Tio Canela x XAN 309 x Orgullo	R(I)	R	V	I
48	MR12438-93	RMEX 35 x Rojo Seda x Tio Canela x XAN 309 x Orgullo	R(I)	R	S	I

Continued.

Table 10. Continued.

Entry	Code	Identification	BCMV	BGMV	BSMV	CCMV
49	MR12438-93	RMEX 35 x Rojo Seda x Tio Canela x XAN 309 x Orgullo	R(I)	R	S	I
50	MR12438-93	RMEX 35 x Rojo Seda x Tio Canela x XAN 309 x Orgullo	R(I)	R	S	I
51	MR12438-93	RMEX 35 x Rojo Seda x Tio Canela x XAN 309 x Orgullo	R(I)	R	S	I
52	MR12438-93	RMEX 35 x Rojo Seda x Tio Canela x XAN 309 x Orgullo	R(I)	S	S	I
53	MR12438-97	RMEX 35 x Rojo Seda x Tio Canela x XAN 309 x Orgullo	R(I)	S	S	I
54	MR12438-97	RMEX 35 x Rojo Seda x Tio Canela x XAN 309 x Orgullo	V	S	S	I
55	MR12438-97	RMEX 35 x Rojo Seda x Tio Canela x XAN 309 x Orgullo	R(I)	S	S	I
56	MR12438-97	RMEX 35 x Rojo Seda x Tio Canela x XAN 309 x Orgullo	R(I)	S	S	S
57	MR12438-27	RMEX 35 x Rojo Seda x Tio Canela x XAN 309 x Orgullo	R(I)	S	S	I
58	MR12438-78	RMEX 35 x Rojo Seda x Tio Canela x XAN 309 x Orgullo	R(I)	S	S	I
59	MR12438-78	RMEX 35 x Rojo Seda x Tio Canela x XAN 309 x Orgullo	R(I)	R	S	I
60	MR12438-46	RMEX 35 x Rojo Seda x Tio Canela x XAN 309 x Orgullo	S	R	M	I
61	MR12438-46	RMEX 35 x Rojo Seda x Tio Canela x XAN 309 x Orgullo	S	R	M	I
62	MR12438-46	RMEX 35 x Rojo Seda x Tio Canela x XAN 309 x Orgullo	V	R	S	I
63	MR12438-96	RMEX 35 x Rojo Seda x Tio Canela x XAN 309 x Orgullo	R(I)	S	V	I
64	MR12438-96	RMEX 35 x Rojo Seda x Tio Canela x XAN 309 x Orgullo	R(I)	S	V	I
65	MR12440-22	Rojo Seda x Tio Canela x XAN 309 x G 17341 x G 1345	R(I)	R	S	I
66	MR12440-22	Rojo Seda x Tio Canela x XAN 309 x G 17341 x G 1345	R(I)	R	S	I
67	MR12440-22	Rojo Seda x Tio Canela x XAN 309 x G 17341 x G 1345	R(I)	R	S	I
68	MR12440-22	Rojo Seda x Tio Canela x XAN 309 x G 17341 x G 1345	R(I)	S	S	I
69	MR12326-40	XAN 309 x Orgullo x Tio Canela x XAN 309	R(I)	S	S	I
70	MR12326-40	XAN 309 x Orgullo x Tio Canela x XAN 309	R(I)	R	S	I
71	MR12326-40	XAN 309 x Orgullo x Tio Canela x XAN 309	R(I)	R	S	I
72	MR12326-48	XAN 309 x Orgullo x Tio Canela x XAN 309	R(I)	S	S	I
73	MR12326-1	XAN 309 x Orgullo x Tio Canela x XAN 309	R(I)	R	S	I
74	MR12326-1	XAN 309 x Orgullo x Tio Canela x XAN 309	R(I)	S	S	I
75	MR12326-7	XAN 309 x Orgullo x Tio Canela x XAN 309	S	S	M	I
76	MR12326-53	XAN 309 x Orgullo x Tio Canela x XAN 309	R(I)	S	S	R
77	MR12326-53	XAN 309 x Orgullo x Tio Canela x XAN 309	R(I)	S	V	I
78	MR12326-53	XAN 309 x Orgullo x Tio Canela x XAN 309	R(I)	S	S	I
79	MR12326-53	XAN 309 x Orgullo x Tio Canela x XAN 309	R(I)	S	S	I
80	MR12326-53	XAN 309 x Orgullo x Tio Canela x XAN 309	R(I)	S	S	I
81	MR12326-53	XAN 309 x Orgullo x Tio Canela x XAN 309	R(I)	S	S	R
82	MR12326-13	XAN 309 x Orgullo x Tio Canela x XAN 309	R(I)	S	S	I
83	MR12326-14	XAN 309 x Orgullo x Tio Canela x XAN 309	R(I)	S	S	I

- a. R = resistant, M = medium susceptibility, I = *I* gene, V = segregating, and S = susceptible.
b. Bean common mosaic virus (BCMV), bean golden yellow mosaic virus (BGMV), bean severe mosaic virus (BSMV), and cowpea chlorotic mottle virus (CCMV).

1.2.6 Identifying genotypes with resistance to angular leaf spot

Rationale: Because of the high variability that is exhibited by most pathogens, identification of sources of resistance to major diseases is a continuous activity. Understanding the mechanisms underlying disease resistance is essential for developing appropriate breeding strategies. This year, we screened materials from the germplasm bank to look for sources of ALS resistance in the small-red type of bean that are grown widely in Central America. In addition, we screened the primary and secondary gene pools for possible sources of ALS resistance and interspecific crosses derived from crossing *P. vulgaris* with *P. polyanthus* and/or *P. coccineus*.

Materials and methods: 117 bean germplasm representing small-red materials that were identified as having resistance to local *P. griseola* isolates in Santander de Quilichao in 1999 were screened again in Darien, using other races found in this area. In a separate experiment, 233 small reds that were not evaluated in 1999 were evaluated this year in

Santander de Quilichao. In both locations, these materials were inoculated with a mixture of the widest genetic diversity of the pathogen, locally isolated from the same area (Tables 11 and 12). Evaluations for disease severity were assessed four times, starting 2 weeks after inoculation, using a CIAT 1 – 9 scale, where 1 represents no visible symptoms and 9 represents severe symptoms and disease expression (van Schoonhoven and Pastor-Corrales 1987²). Plants that had a rating of 3 or less were considered resistant, 4-6 were intermediate, and a rating greater than 6 were considered susceptible.

Table 11. Virulence phenotype of *Phaeoisariopsis griseola* isolates used for inoculation of potential sources of resistance in Darien, Colombia.

Isolate	Race	Differential cultivars ^a											
		A	B	C	D	E	F	G	H	I	J	K	L
PG 81 COL	5-47	a		c				g	h	i	j		l
PG 261 COL	31-47	a	b	c	d	e		g	h	i	j		l
PG 270 COL	63-0	a	b	c	d	e	f						
PG 289-1 COL	47-0	a	b	c	d		f						

a. Andean differential cultivars: A = Timoteo, B = G 11796, C = Bolón Bayo, D = Montcalm, E = Amendoin, and F = G 5686. Mesoamerican differential cultivars: G = PAN 72, H = G 2858, I = Flor de Mayo, J = MEX 54, K = Bat 332, and L = Cornell 49242.

Table 12. Virulence phenotype of *Phaeoisariopsis griseola* isolates used for inoculation of potential sources of resistance in Santander de Quilichao, Colombia.

Isolate	Race	Differential cultivars ^a											
		A	B	C	D	E	F	G	H	I	J	K	L
PG 3 COL	63-0	a	b	c	d	e	f						
PG 61 COL	31-63	a		c	d			g	h	i	j	k	l
PG 65 COL	1-55	a						g	h	i		k	l
PG 32 COL	31-55	a	b	c	d	e		g	h	i		k	l
PG 1 COL	7-55	a	b	c				g	h	i		k	l

a. Andean differential cultivars: A = Timoteo, B = G 11796, C = Bolón Bayo, D = Montcalm, E = Amendoin, and F = G 5686. Mesoamerican differential cultivars: G = PAN 72, H = G 2858, I = Flor de Mayo, J = MEX 54, K = BAT 332, and L = Cornell 49242.

² van Schoonhoven A, Pastor-Corrales MA. 1987. Standard system for the evaluation of bean germplasm. CIAT, Cali, Colombia. 53 p.

Ninety-six sources of ALS resistance that include bred and germplasm materials were evaluated under field conditions with a mixture of races (Tables 13 and 14) that are encountered in Darien and Santander de Quilichao. These materials had been identified as having good levels of ALS resistance in previous screenings.

Table 13. Bean genotypes resistant to *Phaeoisariopsis griseola* in Darien and/or Santander de Quilichao, Colombia (R = resistant, I = intermediate, and S = susceptible).

Genotype identification	Disease rating	
	Darien	Santander de Quilichao
A 223	R	R
A 384	R	R
AFR 645	R	R
AFR 702	R	R
AFR 703	R	R
AND 1056	R	R
AND 277	R	R
APN 47	R	R
BRU 13	R	R
CAL 143	R	R
CAL 173	R	R
CNF 5558	R	R
G 10474	R	R
G 10736	R	R
G 10909	R	R
G 14301	R	R
G 18970	R	R
G 22257	R	R
G 04333	R	R
G 05698	R	R
G 15396	R	R
G 20523	R	R
G 20743	R	R
G 22255	R	R
G 22267	R	R
A 247	S	R
G 5207	S	I
G 5377	S	I
G 10865	S	R
G 22447	S	R
G 00811	S	R
G 01805	R	S
G 01845	S	I
G 01916	S	R
G 02647	S	R
G 09462	I	S
G 18451	S	R
G 20748	S	R
ICTA Texel	S	I
Jacinto	S	S
NIC 147	S	I
NIC 159	S	I
RWR 222	S	R

Table 14. Response of 96 bean genotypes to inoculation with an angular leaf spot race, 63-47, from Malawi under greenhouse conditions.

Entry	Disease response ^a	Entry	Disease response ^a	Entry	Disease response ^a
A 233	R	G 18970	I	ZAA 91	I
G 18451	R	G 18842	I	G 14301	I
G 11405	R	AFR 188	I	G 8152	I
G 04333	R	AND 277	I	G 14508	I
G 01845	R	NAB 69	I	G 916	I
G 10909	R	NIC 147	I	A 240	I
MAR 3	R	RAB 354	I	CNF 5558	I
RWR 222	R	G 09462	I	G 9282	I
G 10613	R	MAR 1	I	G 22267	S
G 19833	R	G 02647	I	G 6727	S
CAL 173	R	G 4691	I	AND 1056	S
G 22447	R	G 18224	I	CAL 143	S
G 19115	R	A 82	I	G 15396	S
G 01805	R	G 20818	I	CAL 123	S
G 05653	R	G 20523	I	A 384	S
G 05698	R	G 20743	I	Jacinto	S
AFR 735	R	A 339	I	A 247	S
AFR 702	R	AFR 703	I	G 09603	S
G 5207	R	A 791	I	G 20748	S
G 10474	R	A 216	I	BRB 191	S
G 10736	R	G 03991	I	A 785	S
BAT 496	R	G 20939	I	ICA Tex	S
G 01916	R	DE CELA	I	G 11104	S
A 222	R	G 00811	I	G 5377	S
BAT 1458	I	G 04032	I	G 22301	S
NIC 159	I	BRB 190	I	EMP 365	S
G 22255	I	APN 47	I	G 20525	S
G 2923	I	BRU 13	I	Gordo	S
AND 829	I	RIZ 97	I	EMP 364	S
				G 4724	S

a. R = resistant, I = intermediate, and S = susceptible.

The same 96 sources of resistance were also evaluated under greenhouse conditions using race 63-47 from Africa. The objective was to identify ALS-resistant materials with a broad activity that could also be used to manage this disease in Africa.

Sixteen wild and weedy *P. vulgaris* materials that were identified as resistant under field conditions in Darien and Santander de Quilichao during the 1999 season were screened under greenhouse conditions using race 63-63, the most virulent and aggressive identified

to date. In addition, 37 lines from the Costa Rican national breeding program were screened using race 63-63 under greenhouse conditions.

Results and discussion: Of the 177 small-red genotypes selected in Sander de Quilichao during 1999, only one (G 5608) was resistant in Darien, while 17 that had a resistant or intermediate phenotype in Santander de Quilichao (1999 data) were susceptible in Darien. Of the genotypes that were resistant or intermediate in both locations none had a resistant or intermediate reaction to race 63-63 under greenhouse conditions (Table 15, Figure 27). These results show that the small-red type of beans has very little resistance to *P. griseola*. Therefore, resistance has to be introgressed from other genotypes outside this type of bean.

Table 15. Potential sources of angular leaf spot in small-red beans to inoculation with *Phaeoisariopsis griseola* in Darien and Santander de Quilichao, Colombia

Genotype	Disease rating ^a	
	Darien	Quilichao
G 7004	2	4
G 2094	3	6
G 7005	3	5
G 7874	3	7
G 5608	2	3
G 15804	4	2
G 5622	7	2
G 4075	8	2
G 7185	8	2
G 14512	6	3
G 1925	7	3
G 2059	7	3
G 2746	7	3
G 2757	7	3
G 3697	7	3
G 6430	7	3
G 7185	7	3
G 2168	8	3

a. Disease rating on a score of 1-9, where 1 = no visible symptoms and 9 = severe symptoms and disease expression.

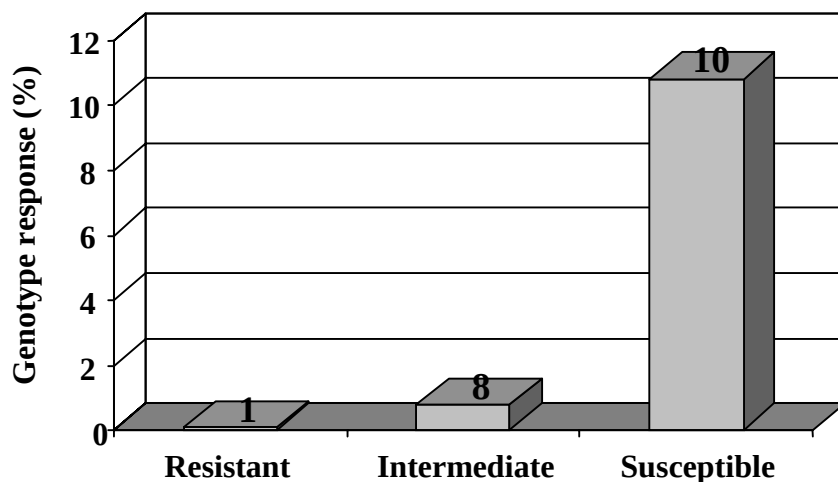


Figure 27. Response of 117 small-red type of common bean to inoculation with *Phaeoisariopsis griseola* under field conditions at Darien and Santander de Quilichao, Colombia.

Similarly, of the 233 small-red genotypes evaluated in Santander de Quilichao, only six were resistant, while 277 were either susceptible or had an intermediate reaction (Figure 28). These materials (six) will be evaluated in the greenhouse using the most virulent race (63-63) from Central America. However, previous results have shown that very little resistance to ALS exists within the small-red genotypes. Small reds are very important for Central America; therefore, efforts should be made to introgress ALS resistance from other materials that have been identified as having high levels of ALS resistance.

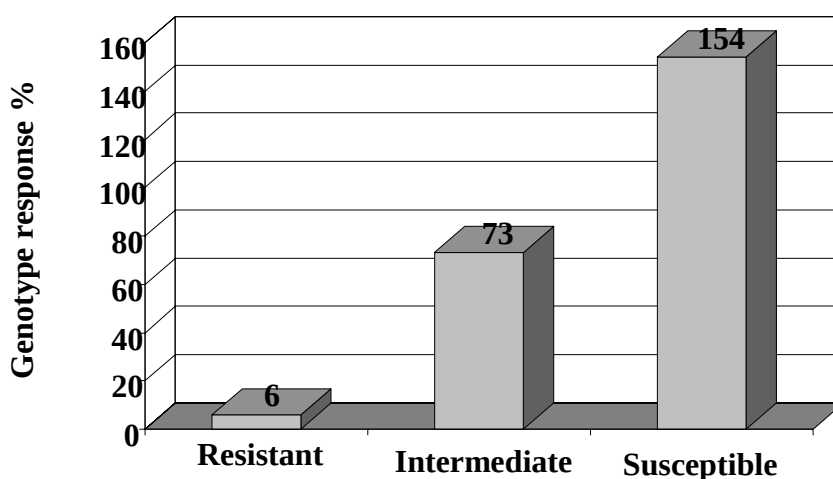


Figure 28. Response of 233 accessions of small-red type of beans to inoculation with *Phaeoisariopsis griseola* under field conditions at Santander de Quilichao, Colombia.

Of the 96 materials previously identified as having some ALS resistance, 25 (12 bred lines and 13 germplasm material) were resistant to the diversity found in both Santander de Quilichao and Darien (Figure 29, Table 13). However, most of the materials that were resistant in Santander de Quilichao were susceptible in Darien, showing the differences in the race structure and composition of *P. griseola*. These results show that simultaneous evaluation of potential sources of resistance in these two areas results in better selection of ALS resistance. Because of the differences in the environmental conditions in Santander de Quilichao and Darien, the race composition is different and using both sites ensures that potential sources of ALS resistance are exposed to the greatest diversity that exists in the pathogen.

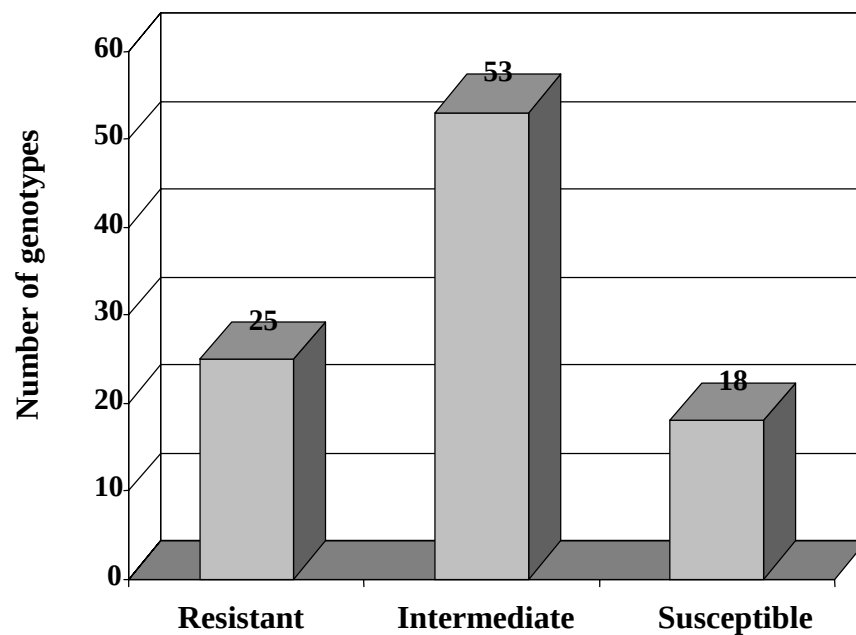


Figure 29. Response of 96 bean materials to inoculation with *Phaeoisariopsis griseola* under field conditions at Darien and Santander de Quilichao, Colombia.

When the same 96 materials were evaluated under greenhouse conditions with race 63-47 from Africa, 25 genotypes were identified as resistant (Table 14). These materials constitute an import source of ALS resistance for African bean researchers.

Of the 16 wild and weedy materials that had been identified as having resistance following field evaluations, only four (G 23435C, G 23477, G 23478, and G 23479) showed high levels of resistance to race 63-63 (Table 16). Two of these materials are from Guatemala and the other is from Mexico. This is very interesting because most of the high levels of resistance found in *P. vulgaris* are materials that originated from the

highlands of Mexico and Guatemala. Future evaluations for ALS resistance should focus on germplasm from these areas.

Table 16. Response of wild and weedy *Phaseolus vulgaris* to inoculation with race 63-63 of *Phaeoisariopsis griseola* under greenhouse conditions.

Accession	Origin	Disease rating (race 63-63) ^a
G 10001	Mexico	S
G 10001A	Mexico	S
G 10002A	Mexico	I
G 10005	Mexico	I
G 11031	Mexico	I
G 12866A	Mexico	S
G 12882A	Mexico	S
G122866	Mexico	I
G 12028	Mexico	S
G 23416	Costa Rica	S
G 23434C	Guatemala	R
G 23435A	Guatemala	I
G 23465	Mexico	I
G 23477	Guatemala	R
G 23478	Guatemala	R
G 23479	Mexico	R

- a. Disease rating on a score of 1-9, where 1 = no visible symptoms and 9 = severe symptoms and disease expression.

Of the 37 lines from the Costa Rican national program, only two materials, UCR-59 and CFR-31, were resistant to race ALS 63-63 (Table 17). Most of these materials were highly susceptible.

Conclusion: Sources of ALS resistance have been identified in *P. vulgaris* and wild and weedy *P. vulgaris*. This group constitutes a very interesting source of ALS resistance and is a first step towards durable ALS resistance. However, the activity spectrum of these genes needs to be established. They must be sufficiently characterized to identify the ones with the broadest activity as well as gene combinations to pyramid. In addition, this information will allow for proper deployment of these resistance genes in ways that will prolong their durability. Inheritance studies for some of these materials have been initiated as well as transferring the identified genes into well-adapted, market-class type beans.

Table 17. Reaction of bean lines from the Costa Rican national program evaluated for reaction to race 63-63 of *Phaeoisariopsis griseola* under greenhouse conditions.

Entry	Disease rating ^a	Entry	Disease rating ^a
UCR-56	S	CFR-25	S
UCR-58	S	CFR-26	S
UCR-59	R	CFR-27	S
UCR-60	S	CFR-28	S
UCR-61	S	CFR-29	I
CFR-11	S	CFR-30	S
CFR-12	S	CFR-31	R
CFR-13	S	CFR-32	S
CFR-14	S	CFR-33	I
CFR-15	S	CFR-34	S
CFR-16	S	CFR-35	S
CFR-17	S	CFR-36	S
CFR-18	S	CFR-37	S
CFR-19	I	CFR-38	S
CFR-20	I	CFR-39	S
CFR-21	S	CFR-40	S
CFR-22	S	CFR-41	S
CFR-23	S	CFR-42	S
CFR-24	S		

a. Disease rating: R = resistant, I = intermediate, and S = susceptible.

Contributors: G Mahuku, C Jara

1.2.7 Screening of the secondary gene pool and interspecific crosses for sources of angular leaf spot resistance

Rationale: Because of the highly variable nature of bean fungal pathogens, beans that are resistant in one location are more likely to be susceptible in another location in the same or different year. This resistance is not durable and the exercise of looking for sources of resistance is therefore a continuous activity. Because some traits are expressed at inadequate levels within the primary gene pool, last year we evaluated 100 genotypes of *P. polyanthus* and *P. coccineus* and found high levels of resistance to ALS in the secondary *Phaseolus* gene pool. With the hypothesis that long-lasting and durable ALS resistance will be found in the secondary and/or tertiary *Phaseolus* gene pools, we screened the *P. polyanthus* and *P. coccineus* Core Collection for resistance to ALS and anthracnose under field conditions. In addition, we screened 1012 interspecific crosses between *P. vulgaris* and *P. polyanthus* and *P. coccineus*, in order to identify lines carrying ALS resistance.

Materials and methods: Materials comprising the Core Collection of *P. polyanthus* and *P. coccineus* (162) were inoculated separately with Andean and Mesoamerican races of *P. griseola* and *Colletotrichum lindemuthianum*. In a separate experiment, 481 interspecific crosses were screened in Darien using local races of *P. griseola*. Crosses were between *P. vulgaris* and *P. coccineus* and *P. polyanthus* (ICA Pijao x G 335172; ICA Pijao x G 35171; ICA Pijao x G 35877; ICA Pijao x G 33720; ICA Pijao x DGD 2119). During the second semester, 534 interspecific crosses that involved wider *P. vulgaris* / *P. polyanthus* or *P. coccineus* crosses were evaluated in Darien. All materials were inoculated, with a mixture of *P. griseola* races found in Darien, four times starting 25 days after planting and every 10 days thereafter. Evaluations for disease severity were assessed four times, starting 2 weeks after inoculation, using a CIAT 1 – 9 scale, where 1 represents no visible symptoms and 9 = severe symptoms and disease expression. Plants that had a rating of 3 or less were considered resistant, 4-6 were intermediate, and a rating greater than 6 were considered susceptible.

Results and discussion: High levels of resistance to ALS were observed among the *P. polyanthus* and *P. coccineus* Core Collections. Andean races of *P. griseola* and *C. lindemuthianum* did not infect any of these materials (Figures 30 and 31). However, Mesoamerican races of *C. lindemuthianum* infected only one genotype, while 11 had an intermediate reaction with a Mesoamerican race of *P. griseola* (Figures 32 and 33). When these materials were evaluated in the greenhouse with race 3841, only two had intermediate reaction, while the rest of the materials were resistant (Figure 34). These results show that the secondary *Phaseolus* gene pool has high levels of resistance to both anthracnose and ALS of common bean and these are potential sources of durable resistance.

When we screened 481 interspecific crosses that had been generated as part of the BGMV improvement program for resistance to ALS, we found low levels of resistance. Only 3.3% of the materials (16) had resistance to mixtures of *P. griseola* races found in Darien (Figure 35). These low levels of resistance were possibly because of the narrow base of the parental materials used in the crosses. In addition, these populations had gone through several cycles of screening for common bean phenotype and possibly the resistance to ALS that was in the original parents might have been lost.

We therefore pulled out 531 interspecific crosses from the germplasm bank that included populations from F₂ to F₉. Only 93 of these materials were resistant to *P. griseola* in Darien (Figure 36). However, most of these materials were segregating and individual selections were made for resistance to ALS.

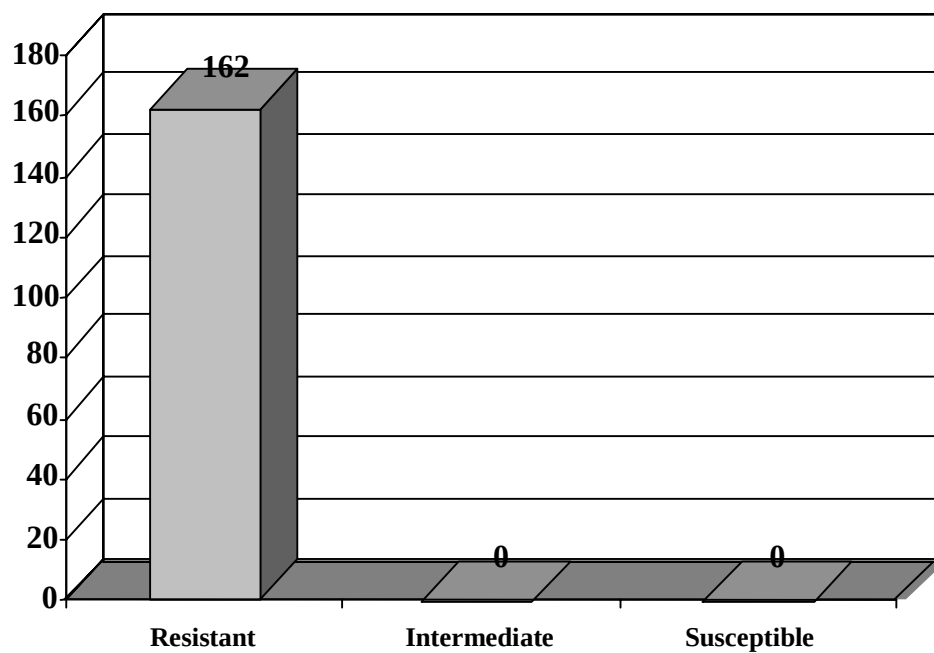


Figure 30. Field evaluation of *Phaseolus coccineus* and *P. polyanthus* using Andean races of *Phaeoisariopsis griseola*.

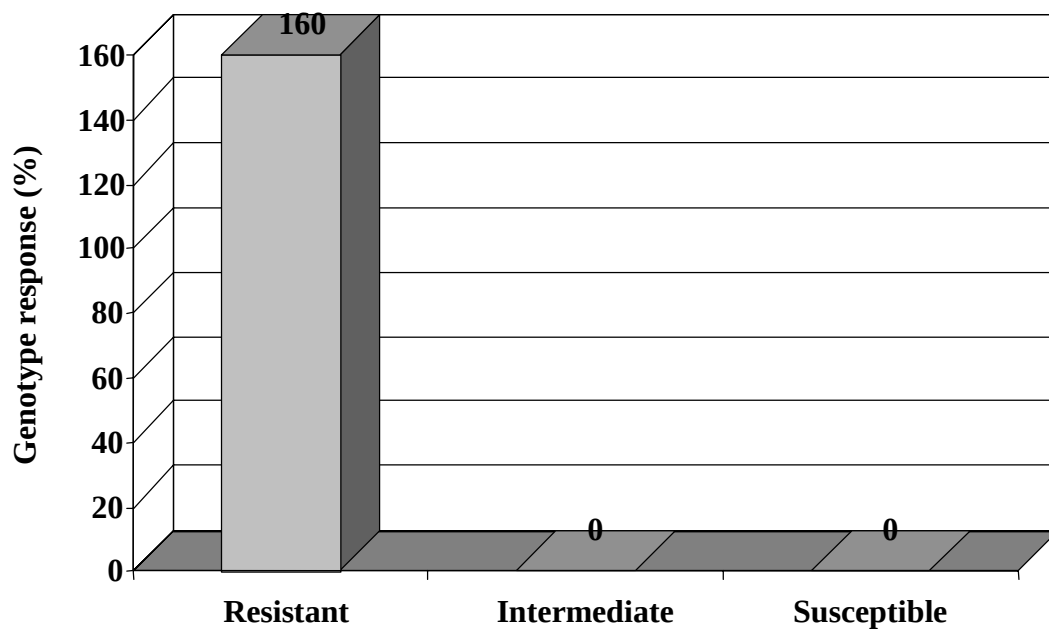


Figure 31. Field evaluation of *Phaseolus coccineus* and *P. polyanthus* using Andean races of *Colletotrichum lindemuthianum*.

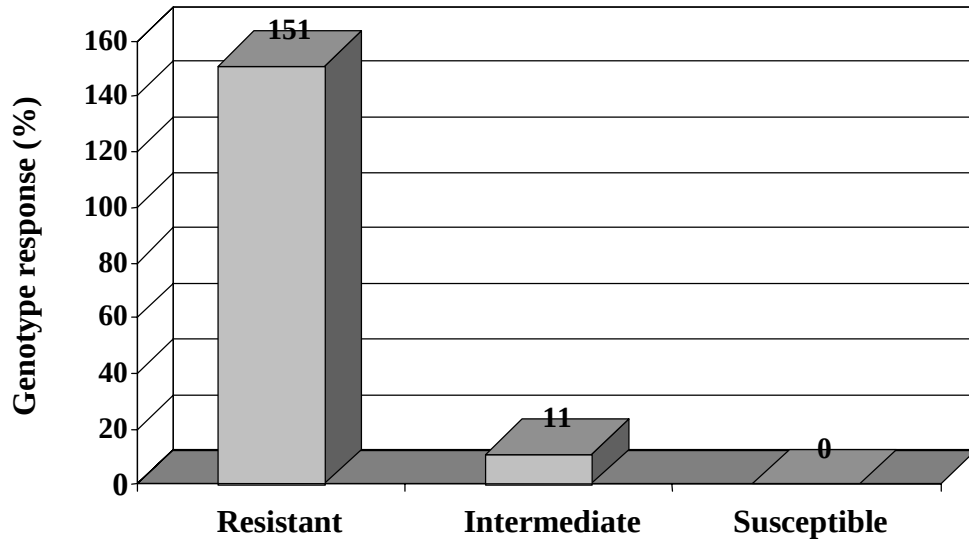


Figure 32. Field evaluation of *Phaseolus coccineus* and *P. polyanthus* using Mesoamerican races of *Colletotrichum lindemuthianum*.

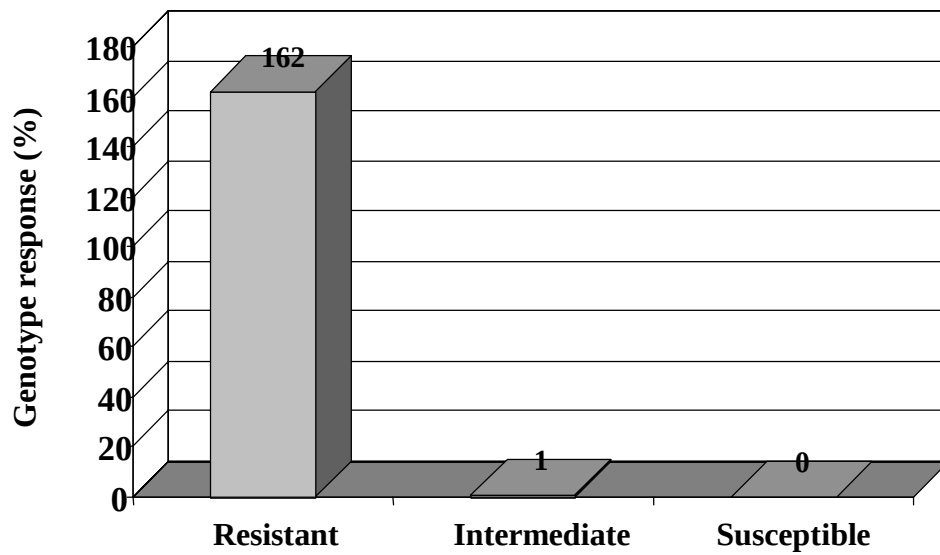


Figure 33. Field evaluation of *Phaseolus coccineus* and *P. polyanthus* using Mesoamerican races of *Phaeoisariopsis griseola*.

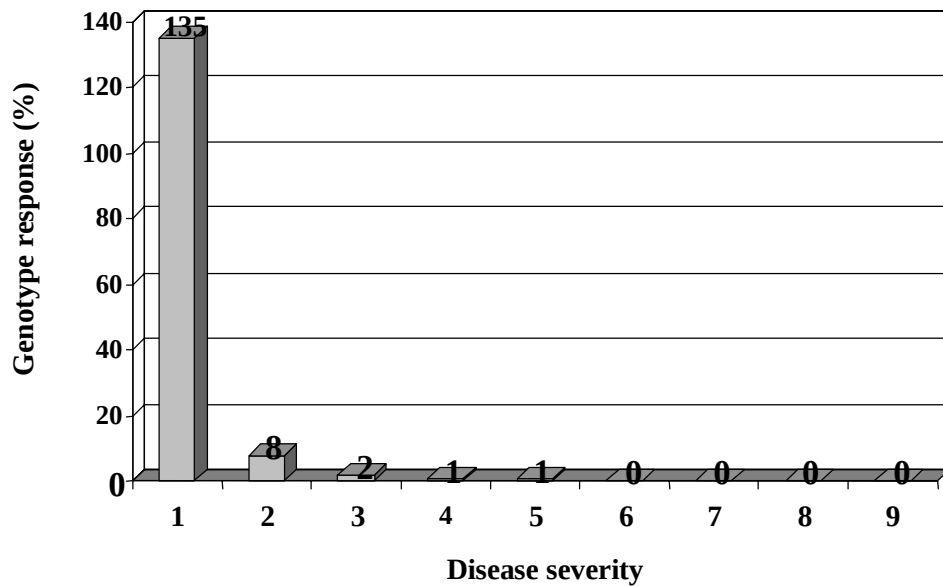


Figure 34. Response of *Phaseolus coccineus* and *P. polyanthus* to inoculation with race 3841 of *Colletotrichum lindemuthianum* under greenhouse conditions using a CIAT 1 – 9 scale, where 1 represents no visible symptoms and 9 = severe symptoms and disease expression.

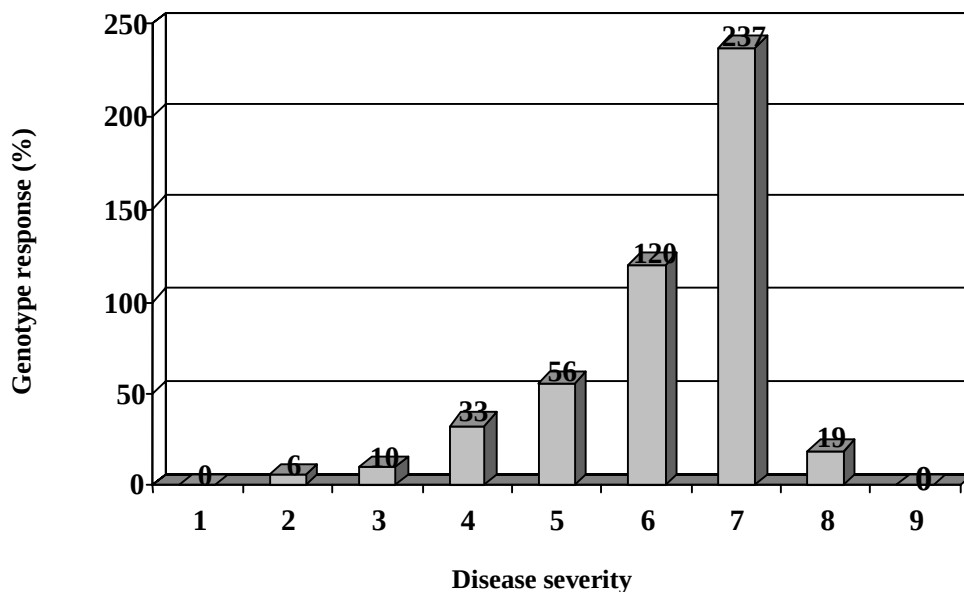


Figure 35. Response of 481 lines derived from interspecific crosses of *Phaseolus vulgaris* and *P. polyanthus* or *P. coccineus*, following field inoculations with mixtures of *Phaeoisariopsis griseola* races using a CIAT 1 – 9 scale, where 1 represents no visible symptoms and 9 = severe symptoms and disease expression.

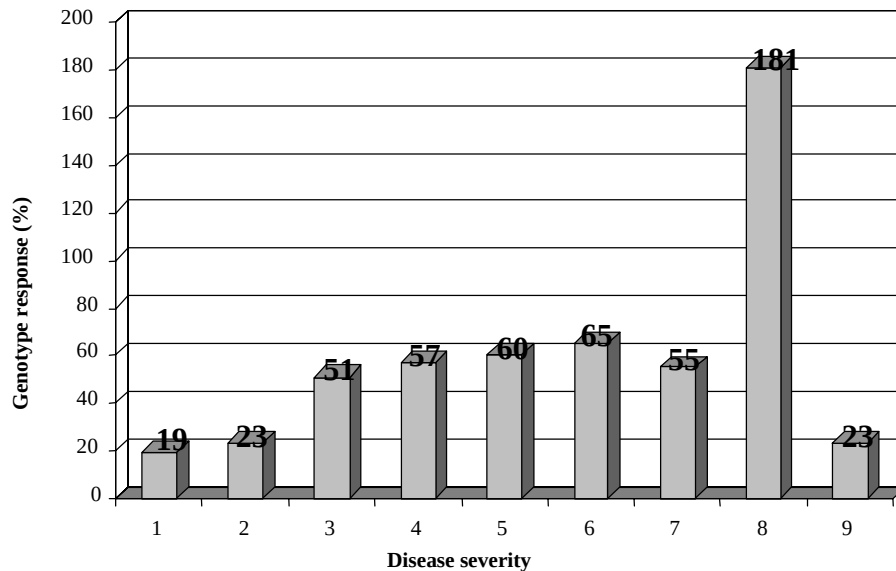


Figure 36. Response of 531 lines derived from interspecific crosses of *Phaseolus vulgaris* and *P. polyanthus* or *P. coccineus*, following field inoculations with mixtures of *Phaeoisariopsis griseola* races using a CIAT 1 – 9 scale, where 1 represents no visible symptoms and 9 = severe symptoms and disease expression.

Conclusion: High levels of resistance to anthracnose and ALS were observed in the secondary *Phaseolus* gene pool. Durable resistance to these diseases potentially might be found within these materials. The interspecific materials identified with ALS resistance form an interesting group of potential sources of ALS resistance. These materials will be screened under greenhouse conditions using isolates of a diverse origin to identify the most promising lines with wide resistance gene activity.

Contributors: G Mahuku, C Jara, C Cajiao, S Beebe

1.2.8 Developing sources of angular leaf spot resistance through breeding

Rationale: Angular leaf spot is considered the most important foliar pathogen of bean in Africa and Brazil, and has become increasingly important in Central America. The development of agronomically acceptable lines that can serve as sources of resistance is an urgent need of breeding programs for these regions.

Materials and methods: Populations were created with several sources of resistance. Several sources are derived from the Andean gene pool. These populations were inoculated under field conditions and have been selected for resistance in Santander de Quilichao for several generations.

In addition to previously identified sources, with the pathologist, germplasm accessions were identified that maintain resistance to the highly virulent race 63-63 that has been identified in Central America. Many of these are climbing beans from Central America of

race Guatemala. These were crossed with other sources (e.g., with G 4691, a Colombian accession with CH phaseolin) and the F₁ in turn was crossed with adapted commercial varieties.

Results: From three superior populations, 79 lines were selected with high resistance under field conditions and with good plant type and adaptation to medium fertility levels. These are largely of Carioca type, but include some black-seeded lines, and are pending an evaluation with races of the ALS pathogen that correspond to other production areas.

Many of the newer populations with climbing bean sources were lost because of flooding in Santander de Quilichao in the first season. However, it appears that these new sources will not produce agronomically useful progeny in the short run and will require two to several cycles of crossing and selection to recover their resistance.

Conclusions: Breeding for ALS resistance was successful in the Santander de Quilichao environment, but the selected lines must now be confronted with races from other regions. It is known that some races, such as the 63-63 race from Honduras, are able to overcome many of the sources that were used in the past and that were apparently resistant in Santander de Quilichao.

Contributors: S Beebe, G Mahuku, H Terán, C Jara, JM Osorno, C Cajiao (IP-1);
C Quintero, J Tohme (SB-2)

1.2.9 Identifying genotypes with resistance to *Macrophomina phaseolina*

Rationale: Field studies showed that bean genotypes that are tolerant or resistant to *Macrophomina phaseolina* yield and perform well under drought conditions. Greenhouse evaluations followed by field experiments showed that, indeed, the genotypes that are resistant to *Macrophomina* have high levels of tolerance to drought and vice versa. This study was carried out to take advantage of, and screen, parents used to develop recombinant inbred lines (RILs) for response to *M. phaseolina*. It was hoped that parents would be contrasting for *M. phaseolina* response and the RILs derived from these parents would be used to tag genes for resistance to *Macrophomina* for subsequent use in developing adapted genotypes. In addition, 49 genotypes that were selected from iron and tannin studies were screened under greenhouse conditions for their response to *M. phaseolina*.

Materials and methods: Sixteen genotypes that were used to develop RILs were screened under greenhouse conditions for their reaction to *Macrophomina* resistance. The following is a list of the parents in their respective combinations: (G 19833 x DOR 364; MAR 1 x VAX 6; BAT 477 x DOR 364; G 21212 x BAT 881; G 3513 x DOR 364; G 19227A x DOR 364; and DOR 476 x SEL 1309).

In a separate experiment, 49 genotypes previously selected for evaluations of tannin content were screened for resistance to *M. phaseolina* under greenhouse conditions. These materials were inoculated with *M. phaseolina sclerotia* at a concentration of

1.5 g kg⁻¹ of soil. The inoculum was put in the top 4 cm of the pot after planting. Incidence and severity of *M. phaseolina* was evaluated starting 12 days after planting until the plants were 20 days old.

Results and discussion: All the parental materials of RILs were either susceptible or resistant to *M. phaseolina* (Table 18). Therefore, we could not take advantage of the existing RILs to characterize and tag genes for resistance to *M. phaseolina*.

Table 18. Response of 16 genotypes used to create recombinant inbred lines for inoculation with *Macrophomina phaseolina* (Mp) under greenhouse conditions.

Genotypes	Mp	Total plants	% incidence	Disease response ^a
G 21212	8	12	67	S
BAT 881	12	12	100	S
G 3513	3	12	25	R
DOR 364	1	12	8	R
G 19227 A	1	12	8	R
G 19833	3	12	25	R
BAT 477	1	12	8	R
MAR 1	9	12	75	S
VAX 6	11	12	92	S
DOR 476	0	12	0	R
SEQ 7	4	12	33	R
AFR 475	3	12	25	R
CARIOCA	7	12	58	S
A 70	12	12	100	S
SEA 5	2	12	17	R
San Cristobal 83	10	12	83	S

a. R = resistant and S = susceptible.

Of the 49 genotypes screened in this study, 10 had high levels of resistance to *M. phaseolina* (Table 19). The most resistant genotypes were G 5841, G 6891, G 7945, G 13778, and G 4790. When we increased the inoculum load to 3g kg⁻¹ of soil, these genotypes had no symptoms, showing that they are highly resistant and probably dominant genes control the resistance. Because of seed shortage, only G 5481, G 13778, and G 4790 were evaluated under drought conditions. These materials had very low incidences of *M. phaseolina* under field drought conditions. These materials will be screened for response to drought conditions during the coming season.

Table 19. Response of bean germplasm to inoculation with *Macrophomina phaseolina* under greenhouse conditions.

Entry	Disease severity rating ^a	Incidence	Entry	Disease severity rating	Incidence
G 4756	S	88	G 6981	R	0
G 16664	R	13	G 18264	I	63
G 17913	R	13	G 4258	S	88
G 5481	R	0	G 17166	S	100
G 3815	I	50	G 2494	R	38
G 7945	R	0	G 23804	S	75
G 16157	S	88	G 20592	I	38
G 13778	R	0	G 12403	S	88
G 6639	R	25	G 12037	S	100
G 22365	I	50	G 2906	S	88
G 18244	I	75	G 22805	I	75
G 4495	I	63	G 2276	S	100
G 4790	R	0	G 12171	S	100
G 58	S	75	G 14778	S	75
G 5285	S	100	G 2769	I	63
G 5758	I	63	G 19515	S	100
G 9384	S	75	G 22291	S	75
G 3821	S	100	G 995	S	75
G 16072	S	100	G 23773	S	100
G 13177	S	88	G 12169	S	88
G 19497	S	100	G 1083	S	100
G 11957	S	100	SEA 5	R	13
G 1400	S	75	BAT 332	I	50
G 11640	S	88	A 70	S	100
G 23283	S	100	BAT 477	S	100

a. R = resistant, I = intermediate, and S = susceptible.

Conclusions: These results are preliminary, but promising. The materials identified will be part of the drought trial next season and their response to drought will be assessed then. A rapid greenhouse technique for screening large volumes of bean genotypes for response to *M. phaseolina* has been developed. More genotypes should be passed through this test, to identify genotypes with resistance to *M. phaseolina* and drought tolerance. We need to start making crosses using contrasting parents with tolerance or resistance to multiple factors for subsequent developing of molecular markers.

Contributors: G Mahuku, C Jara, G Castellanos, IM Rao, S Beebe

1.2.10 Developing breeding lines resistant to *Ascochyta* blight

Rationale: *Ascochyta* is a highly destructive disease of bean in the rainy, higher altitude regions of the Andean zone and Africa. In these areas, *Ascochyta* severely limits the potential use of bean, especially of bush types that are exposed to splashing of inoculum from soil. These are regions that do not produce large volumes of bean, but they are especially poverty stricken regions, and resistance to *Ascochyta* would afford an important crop option to farmers there. Further, in the Andean zone, snap beans are especially susceptible to *Ascochyta*, and an *Ascochyta* attack can trigger a vicious cycle of pesticide abuse.

Materials and methods: In previous years we reported on the progress in implementation of resistance from interspecific crosses. Additionally, a bred variety from Guatemala, ICTA-Hunapú, has proved to be an excellent source. Crosses were created among sources and between sources of resistance and commercial types in several grain classes: Mesoamerican small black-seeded, some Andean grains such as Calima type and Cargamanto, and snap beans. Populations and families were planted in the Popayán station and inoculated three times with isolates obtained locally.

Results: Families derived from crosses among sources produced progenies that were superior to the resistant common bean check, ICTA Hunapú (Table 20). In crosses to susceptible cultivars, observations of F₃ progeny rows suggest that selection was successful in recovering resistance, implying that heritability is acceptably high to permit progress in selection. Families are being purified for subsequent testing and confirmation of resistance. Populations were also sent to Guatemala and promising families will be shared with the national program of Rwanda for selection of locally adapted lines.

Conclusion: For the first time, a level of resistance to *Ascochyta* is available that can permit genetic gain against this disease. Heritability of resistance also appears to be acceptably high. This resistance can make an important contribution in certain high altitude regions of the Andes and Africa.

This research was carried out with the support of the Belgian government.

Table 20. Populations and selected families derived from simple crosses that include the best interspecific selections for resistance to *Ascochyta*, evaluated under inoculated field conditions, 1999B – 2000A, Popayán, Colombia.

Identification	F ₂ families 1998B	F ₂ plants evaluated 1999A	F ₃ families selected 1999B	F ₄ bulks selected 2000A	<i>Ascochyta</i> mean 2000 A
ASC 73 x ASC 77	11	234	0	0	-
ASC 73 x ICTA Hunapú	5	235	12	5	5.0
ASC 74 x ASC 77	10	250	11	2	4.5
ASC 74 x ICTA Hunapú	19	420	15	8	4.7
ASC 75 x ASC 77	4	160	17	2	5.5
ASC 75 x ICTA Hunapú	19	475	48	13	5.3
Total	68	1774	103	30	-
G 35182					2.7
ICTA Hunapú					5.8
G 17723					9.0

Contributors: S Beebe, G Mahuku, C Cajiao

Progress towards achieving output milestones:

- Sources of resistance to the major viral diseases of common bean in Middle America were identified in all cases, and are currently being combined in advanced breeding lines.
- Bean genotypes (25) with high levels of resistance to Andean and Mesoamerican isolates of *Phaeoisariopsis griseola* from Africa, Central America, and South America were identified. These can be used as parents in developing germplasm with multiple resistance to diseases.
- High levels of resistance to ALS and anthracnose were identified in the wild and weedy *Phaseolus vulgaris* materials as well as secondary *Phaseolus* gene pool.
- Of 77 red-seeded lines distributed to Central America, all were resistant to BCMV, 49 were resistant to BGMV, 32 were resistant to CBB, and 20 were resistant to all three diseases.

Activity 1.3 Developing germplasm with resistance to pests: *Zabrotes*, *Acanthoscelides*, *Empoasca*, *Apion*, *Thrips palmi*, and bruchids

Highlights:

- New sources of resistance to *Thrips palmi*, *Empoasca kraemeri*, *Zabrotes subfasciatus*, and *Acanthoscelides obtectus* were identified.
- The methodology to screen for resistance to thrips was refined.
- New lines possessing disease and insect resistance were selected.
- Incorporation of resistance to leafhopper in Andean bean types was reinitiated.

1.3.1 Screening for sources of resistance to major insect pests

Rationale: Identification of sources of resistance to major insect pests is a continuous activity. Understanding of the mechanisms responsible for resistance to insects is essential for the development of appropriate breeding strategies. Apart from bruchids and leafhopper, major emphasis in 2000 was given to the study of resistance to *Thrips palmi*.

Materials and methods: Genotypes are screened under high insect pressure. For bruchids, nurseries are tested in the laboratory using 3-5 replications of 50 seeds per genotype infested with seven pairs of *Z. subfasciatus* or two eggs per seed in the case of *A. obtectus*. Genotypes are classified for resistance according to values for percentage adult emergence and days to adult emergence. Leafhopper and thrips nurseries are planted in the field with 3-4 replications per genotype under high natural infestation levels. Resistance is based on visual damage scores, reproductive adaptation scores, and in some cases, insect counts.

Results and discussion: *Thrips palmi*. This insect is a new pest of beans and many other crops in Latin America. Beans and snap beans are two of its most important host plants. Studies on *T. palmi* included refinement of methodologies to screen for resistance, development of sampling methods, measurement of yield losses and insecticide resistance levels, and preliminary work on management strategies (see Activity 3.3). In 2000, more than 500 genotypes were tested for resistance in replicated nurseries at CIAT and Pradera, a nearby site with high natural infestation (up to 40 adults per leaflet). Materials were rated for visual damage scores using a 1-9 scale (described in the 1999 Annual Report) and reproductive adaptation scores (the ability to set and fill pods under insect pressure). A few genotypes selected in 1999 were reconfirmed for resistance in 2000. These are Brunca, DOR 714, EMP 486, BAT 477, BH 160, FEB 115, among others. As shown in Figure 37, several received damage scores of 6 or less, coupled with reproductive adaptation scores of less than 5. There was a good correlation ($r = 0.71$; $P < 0.001$) between damage scores and reproductive adaptation scores. Five resistant genotypes and a susceptible check were chosen to initiate studies on mechanisms of resistance in collaboration with the Eidgenössische Technische Hochschule-Zentrum (ETHZ) Institute in Switzerland (Andrea Frei's PhD Thesis).

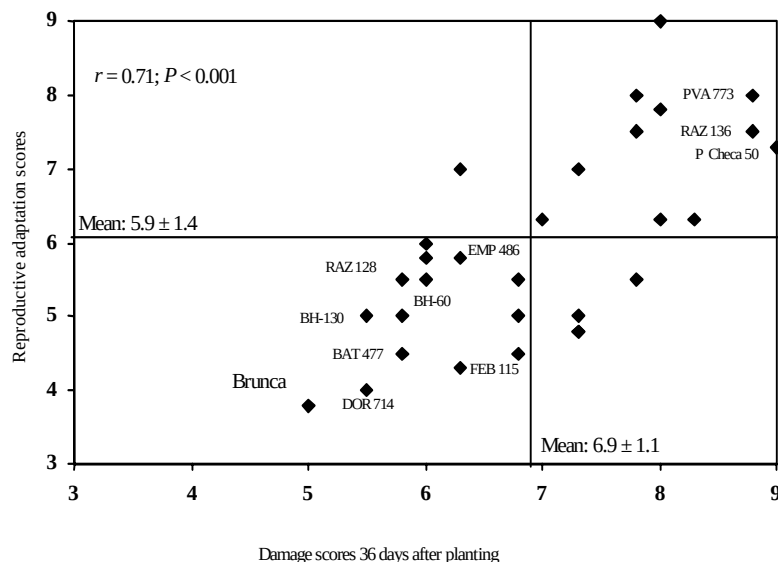


Figure 37. Relationship between damage and reproductive adaptation scores in bean genotypes screened for resistance to the melon thrips, *Thrips palmi*.

Other materials tested for resistance to *T. palmi* included 93 genotypes from the “Vipadogen” and “Potential Parents” nurseries of the Bean Project. Infestation levels in this nursery were high (7.3 adults per leaflet). Damage scores averaged 7.1 ± 1.4 (range: 2.0-8.9); reproductive adaptation scores averaged 7.3 ± 1.3 (range: 4-9). There was a good correlation ($r = 0.72$; $P < 0.05$) between damage and reproductive adaptation scores. As a result of these evaluations, 43 materials were selected for reconfirmation of resistance levels in 2000B. Table 21 shows best genotypes.

Table 21. Response of selected bean genotypes to *Thrips palmi* attack under high natural infestation levels. Means of three replications per genotype.

Genotype	Source (nursery)	Visual damage scores ^a	Reproductive adaptation scores ^b
A 729	Potential parents	2	4
A 730	Potential parents	3	5
Xamego	Potential parents	4	5
A 734	Potential parents	4	6
A 788	Potential parents	5	6
A 725	Potential parents	5	5
A 724	Potential parents	5	6
A 727	Potential parents	5	6
XAN 285	Potential parents	5	6
V 8025	Potential parents	6	6
VAX 3	Vipadogen	6	6
A 722	Potential parents	6	6
A 728	Potential parents	6	6
VAX 5	Vipadogen	6	7
BAT 41	Susceptible check	9	9

- On a 1-9 scale, where 1 = no damage, 3 = initial damage to leaf ribs, 5 = obvious damage to leaf ribs, leaf deformation, 7 = heavy damage along leaf ribs, heavy leaf deformation, silvery appearance of the foliage, stunting of the plant, and 9 = severe damage, all leaves deformed, buds are killed, defoliation.
- On a 1-9 scale, where 1 = good pod formation, 9 = no pods are formed.

As reported in 1999, further studies on resistance to *T. palmi* were conducted using RILS derived from a cross between BAT 881 and G 21212 (a source of low-P tolerance). In a replicated reconfirmation nursery planted at Pradera, under a high level of infestation (7.2 adults per leaflet) 139 RILs and parents showed an array of responses. These fit a distribution skewed towards susceptibility for both visual damage scores and reproductive adaptation scores (Figure 38), with 19% of the lines being rated resistant for damage.

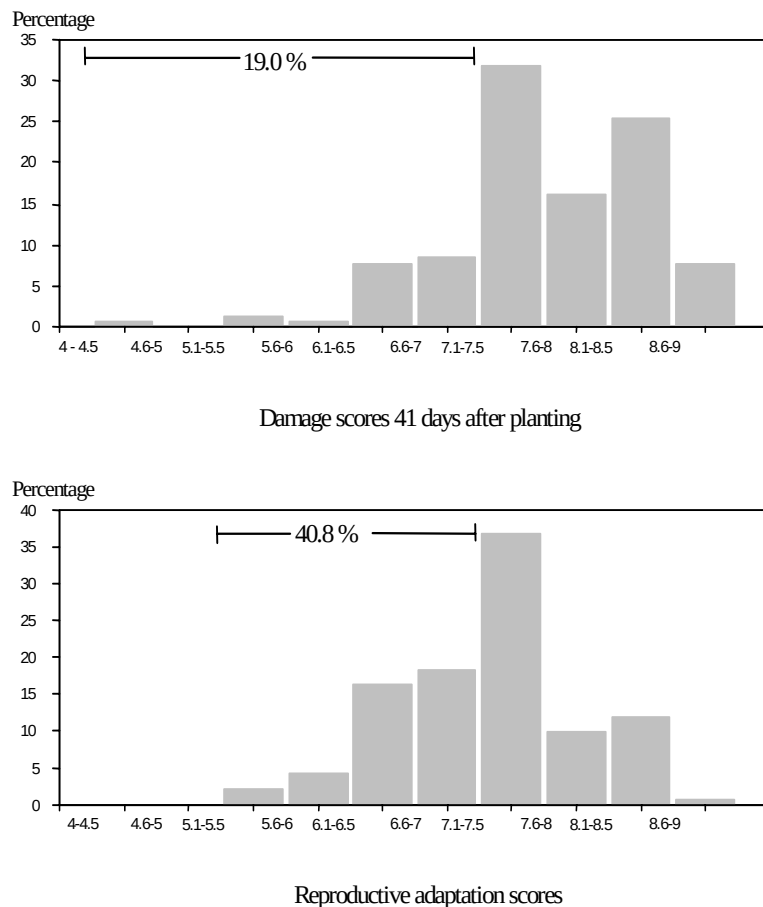


Figure 38. Population distribution for damage and reproductive adaptation scores among 139 recombinant inbred lines derived from the BAT 881 x G 21212 cross-tested for resistance to the melon thrips, *Thrips palmi*.

As in other nurseries, the correlation between damage and reproductive adaptation scores ($r = 0.81$; $P < 0.001$) was high. The rank correlation coefficient between this and the preliminary evaluation conducted in 1999 was high and significant ($r = 0.763$; $P < 0.001$) suggesting that genetic differences tend to show consistency across seasons and infestation levels. Table 22 shows some of the selected genotypes.

The QTL mapping and heritability study of resistance to *Thrips palmi* in common bean is reported in Activity 3.2, section 3.2.7.

Table 22. Levels of resistance to *Thrips palmi* in selected recombinant inbred lines derived from the cross BAT 881 x G 21212. Means of three replications.

Genotype	Damage scores in a 1 (best) to 9 (worst) scale	Reproductive adaptation scores in a 1 (best) to 9 (worst) scale
BH 21134-60-1-1-M-M-M	4.3	5.3
BH 21134-103-1-1-M-M-M	5.3	5.7
BH 21134-144-1-1-M-M-M	5.3	6.0
BH 21134-104-1-1-M-M-M	5.7	6.0
BH 21134-128-1-1-M-M-M	6.0	5.7
BH 21134-46-1-1-M-M-M	6.0	6.0
BH 21134-94-1-1-M-M-M	6.0	6.3
BAT 881	7.6	7.3
G 21212	8.2	7.6
PVA 773 (commercial check)	8.8	8.4

Contributors: C Cardona, JM Bueno, A Frei, MW Blair

Results and discussion: Leafhopper. In 2000, 1087 new germplasm bank accessions were screened for resistance to the leafhopper. Ninety-three accessions were selected for further reconfirmation. To reinitiate the incorporation of resistance to leafhopper in Andean bean types, all EMP lines of this type developed in previous years were evaluated again in 2000. Under high infestation levels (7.1 nymphs per leaf), 16 EMP lines were selected as potential parents for breeding purposes (Figure 39).

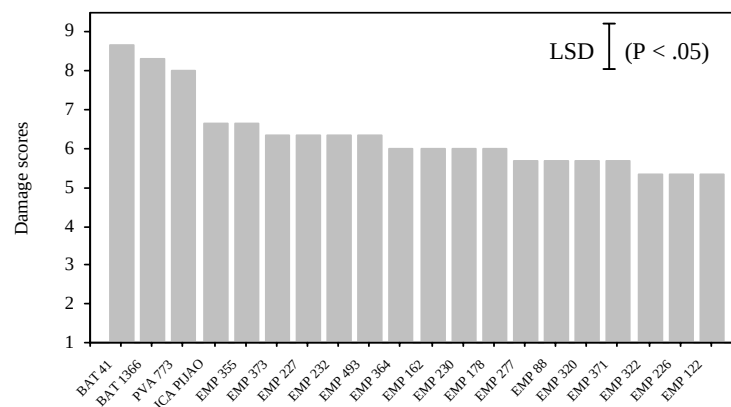


Figure 39. Levels of resistance to the leafhopper (*Empoasca kraemer*) in selected, Andean-type EMP lines.

Contributors: C Cardona, JM Bueno

Results and discussion: Bruchids. The process of identification of sources of resistance to bruchids continued in 2000 with the evaluation of 61 wild *Phaseolus vulgaris* accessions. None of these was resistant to either *Z. subfasciatus* or *A. obtectus*. The *P. acutifolius* accessions G 40202 and G 40244 were resistant to *Z. subfasciatus* while G 40188 was intermediate. No sources of resistance to *A. obtectus* were found among 35 *P. acutifolius* evaluated. In contrast, high levels of resistance to *A. obtectus* were detected in 25 of 143 *P. lunatus* accessions studied.

In preparation for the second phase of the collaborative project with the University of Gent, a comprehensive study on comparative resistance to bruchids in three *Phaseolus* species was undertaken. As shown in Figure 40, high levels of resistance to *A. obtectus* are common in *P. lunatus* while the occurrence of resistance in *P. vulgaris* and *P. acutifolius* is limited to a handful of genotypes.

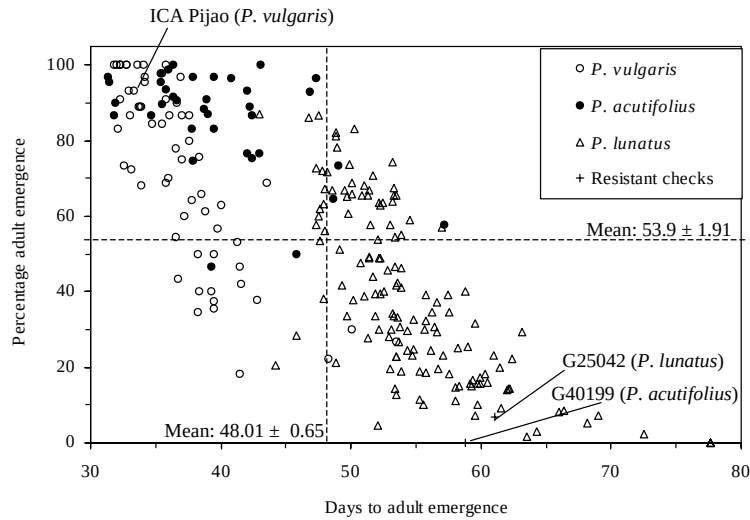


Figure 40. The relationship between days to adult emergence and percentage of adult emergence in accessions of three *Phaseolus* species tested for resistance to the bean weevil, *Acanthoscelides obtectus*. “ICA Pijao” is a commercial susceptible variety. G25042 and G 40199 are resistant checks.

In the case of *Z. subfasciatus*, resistance occurs in a few wild *P. vulgaris* accessions and in a handful of *P. acutifolius* accessions (Figure 41). No resistance to *Z. subfasciatus* has ever been found in *P. lunatus*. Understanding of the mechanisms underlying differential resistance to bruchid species will be attempted within Phase 2 of the Gent Project.

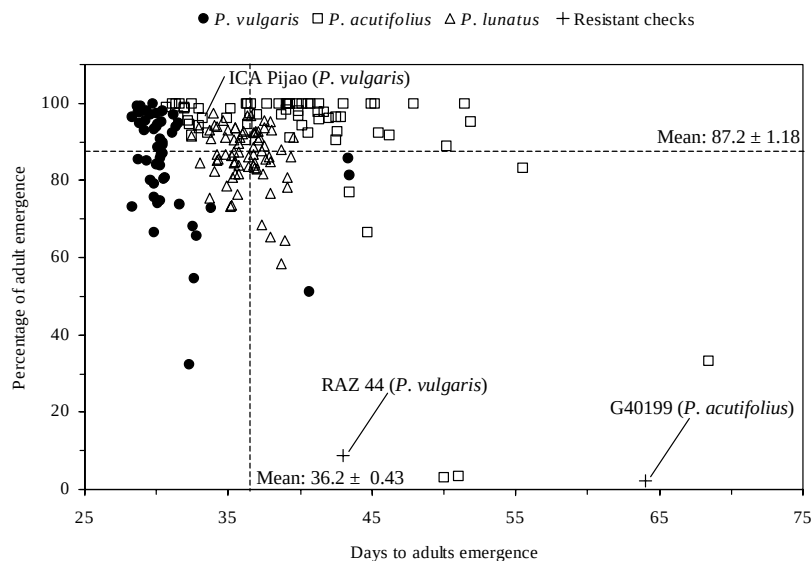


Figure 41. The relationship between days to adult emergence and percentage of adult emergence in accessions of three *Phaseolus* species tested for resistance to the Mexican bean weevil, *Zabrotes subfasciatus*. “ICA Pijao” is a commercial susceptible variety. RAZ 44 and G 40199 are resistant checks.

Contributors: C Cardona, JF Valor

1.3.2 Developing germplasm resistant to insects

Leafhopper: To support breeding efforts within the Project, we evaluated several nurseries developed for adaptation to Andean zone and Mesoamerican conditions. A nursery of 100 F₇ lines combining resistances to BGMV and leafhopper was evaluated under average insect pressure of 6.5 nymphs per leaf. Selections were made for leafhopper resistance, seed types, and agronomic performance. Twenty-four lines were selected for further reconfirmation.

Simultaneously, five F₂ populations were tested for resistance to leafhoppers. Populations EMP 496 x PVA 773 and EMP 250 x PVA 773, were selected. The F₃ populations are being yield-tested at present.

Red lines for Central America and Carioca-type lines for Brazil that combine resistance to leafhopper, BGMV, and CBB were yield-tested in 2000. The best, some of which are shown in Table 23, were coded EMP.

Table 23. Damage scores and yield performance of best Carioca and red lines combining resistances to leafhopper, bean golden mosaic virus, and cassava bacterial blight that were selected for resistance to leafhoppers in 2000. Means of three replications.

Line	Cross ^a	Seed type or color	Damage scores ^b	Yield (kg ha ⁻¹)		Susceptibility index
				Protected	Unprotected	
EMP 584	RM 11493	Carioca	6.3	2531	1076	0.94
EMP 587	RM 11493	Carioca	6.0	2088	1088	0.88
EMP 588	RM 11492	Red	4.1	1662	1037	0.86
EMP 589	RM 11492	Red	5.3	1612	984	0.84
EMP 598	RM 11492	Red	5.8	1700	830	0.97
ICA Pijao	Tolerant check	Black	7.1	2067	831	1.03
Bat 41	Susceptible check	Red	9.0	981	141	1.71

- a. RM 11492 = EMP 473 x ((FEB 200 x XAN 252) F₁ x ((OAC 88-1 x G 5686)F₁ x (A 429 x G 2333)F₁)F₁/- and RM 11493 = EMP 443 x (FEB 188 x XAN 252)F₁ x ((OAC 88-1 x ABA 36)F₁ x (A429 x K2)F₁)F₁/-.
- b. Damage scores on a 1 (best) to 9 (worst) scale.

White-, black-, and cream-seeded new EMP lines (Table 24) were developed in collaboration with the University of Guelph in Canada. These materials were selected for combined resistance to *Empoasca kraemeri* and *E. fabae*.

Table 24. Damage scores and yield performance of best lines combining resistances to *Empoasca kraemeri* and *E. fabae* selected in 2000. Means of three replications.

Line	Cross	Seed color	Damage scores ^a	Yield (kg ha ⁻¹)		Susceptibility index
				Protected	Unprotected	
EMP 547	93-1 x EMP 423-1	White	5.9	1460	862	0.94
EMP 548	EMP 423 x EMP 419-18	White	6.4	1552	759	0.93
EMP 549	EMP 423 x EMP 419-28	White	6.6	1533	876	0.90
EMP 550	Laser x EMP 423-6	White	7.2	1209	599	0.90
EMP 558	Laser x EMP 419-18	White	6.0	1665	705	0.90
EMP 573	2B5 x EMP 423-19	Black	6.3	1626	790	0.85
EMP 580	2B5 x EMP 423-16	Black	6.3	1349	877	0.94
EMP 571	EMP 423 x EMP 419-6	Cream	5.2	1602	665	0.94
ICA Pijao	Tolerant check	Black	7.3	1544	783	0.99
Bat 41	Susceptible check	Red	9.0	1060	197	1.74

- a. Damage scores on a 1 (best) to 9 (worst) scale.

Contributors: C Cardona, JM Bueno

***Z. subfasciatus*:** Backcrossed lines to obtain *Zabrotes*-resistant ICA Pijao lines were tested for resistance to the insect and for yield performance. Some of the lines (Table 25) showed a very high level of resistance and yielded as well as the susceptible recurrent parent. These lines are now available as parents for the incorporation of multiple resistances into Mesoamerican black-seeded cultivars.

Table 25. Levels of resistance to *Zabrotes subfasciatus* and yield performance of five best BC3F₉ lines derived from backcrosses to “ICA Pijao”.

Line	Days to adult emergence	Percentage of adult emergence	Rating	Days to flower	Yield (kg ha ⁻¹)
RAZ 154	44.7	5.4	Resistant	41	1507
RAZ 159	47.4	6.3	Resistant	42	1568
RAZ 162	48.3	1.9	Resistant	42	1573
RAZ 163	49.5	2.3	Resistant	43	1781
RAZ 165	46.3	10.4	Resistant	42	1624
G 12882 ^a	47.0	7.0	Resistant	51	164
ICA Pijao ^b	30.5	94.7	Susceptible	42	1548

a. Donor (Arcelin-1) parent.

b. Recurrent parent.

Contributors: C Cardona, JM Bueno, JF Valor

As reported in 1999, a highlight of the work with *Z. subfasciatus* is the incorporation of arcelin selection criteria into the mainstream-breeding program. Eighteen multiple crosses were developed combining sources of resistance to *Z. subfasciatus*, BGMV, and fungal diseases with Mesoamerican parents. In 2000, we monitored the presence of arcelin in the F₁ seed of these progenies (Table 26) using a nondestructive serological test. Plants positive for arcelin will be planted in the field and screened for other traits within the marker-assisted selection (MAS) approach adopted by the Bean Project.

Table 26. Selection for arcelin in F₁ seeds from crosses for molecular-assisted selection of beans with various useful traits.

Cross	Seeds evaluated (no.)	Seeds selected (no.)
ICTA Ostua x (A 785 x ((RAZ 44 x SEL 1360)F ₁ x (A 429 x XAN 309)F ₁)F ₁)F ₁	100	46
((DOR 390 x (DOR 390 x SAM 3)F ₁)F ₁ x (A 785 x ((RAZ 44 x SEL 1360)F ₁ x (A 429 x XAN 309)F ₁)F ₁)F ₁	100	52
DOR 390 x (Raven x ((RAZ 44 x SEL 1360)F ₁ x (A 429 x XAN 309)F ₁)F ₁)F ₁	99	40
A 801 x (Compuesto Chimaltenango 2 x ((A 429 x XAN 309) F ₁ x (RAZ 44 x Royal red) F ₁) F ₁	26	10
Tio Canela 75 x (DICTA 17 x ((A 429 x XAN 309) F ₁ x (RAZ 44 x Royal red) F ₁) F ₁	135	60
Chingo x (DICTA 17 x ((A 429 x XAN 309) F ₁ x (RAZ 44 x Royal red) F ₁) F ₁	131	63
Rojo de seda x (DICTA 17 x ((A 429 x XAN 309) F ₁ x (RAZ 44 x Royal red) F ₁) F ₁	59	33
A 801 x (Apore x ((RAZ 44 x SEL 1360) F ₁ x (A 429 x XAN 309) F ₁)F ₁)F ₁	81	41
(IPA 7 x (Tio Canela 75 x SAM 3) F ₁) F ₁ x (DOR 500 x ((RAZ 44 x Royal red) F ₁ x (Catrachita x Wilkinson 2) F ₁)F ₁)F ₁	212	106
Tio Canela 75 x (Tio Canela 75 x ((RAZ 44 x SEL 1360) F ₁ x (A 429 x XAN 309) F ₁)F ₁)F ₁	34	20
DOR 364 x (Tio Canela 75 x ((RAZ 44 x SEL 1360) F ₁ x (A 429 x XAN 309) F ₁) F ₁) F ₁	51	19
9825-49-3/F ₆ x (Tio Canela 75 x ((RAZ 44 x SEL 1360) F ₁ x (A 429 x XAN 309) F ₁)F ₁) F ₁	62	31
9825-46-1/F ₆ x (Tio Canela 75 x ((RAZ 44 x SEL 1360) F ₁ x (A 429 x XAN 309) F ₁)F ₁) F ₁	49	18
9824-56-2/F ₇ x (Tio Canela 75 x ((RAZ 44 x SEL 1360) F ₁ x (A 429 x XAN 309) F ₁)F ₁) F ₁	46	26
9824-47-1/F ₇ x (Tio Canela 75 x ((RAZ 44 x SEL 1360) F ₁ x (A 429 x XAN 309) F ₁)F ₁) F ₁	60	32
Chingo x (Tio Canela 75 x ((RAZ 44 x SEL 1360) F ₁ x (A 429 x XAN 309) F ₁)F ₁)F ₁	41	26
Rojo de seda x (Tio Canela 75 x ((RAZ 44 x SEL 1360) F ₁ x (A 429 x XAN 309) F ₁)F ₁) F ₁	32	21
A 801 x (FEB 212 x ((RAZ 44 x A 429 F ₁ x (Catrachita x Wilkinson 2) F ₁)F ₁)F ₁	150	79
Total	1468	723

Contributors: C Cardona, S Beebe, JF Valor, H Terán

1.3.3 Pyramiding *Empoasca* resistance in red-mottled beans

Rationale: Leafhoppers are one of the most serious pests of Andean beans grown in low to mid-elevation areas. Andean beans have traditionally had less *Empoasca* resistance than Mesoamerican beans. The purpose of this research was to transfer the *Empoasca* resistance found in some Mesoamerican genotypes into red-mottled Andean beans.

Materials and methods: Multiple, triple, double, and simple crosses were made between *Empoasca*-resistant lines and susceptible commercial types. The resistant lines included the red-mottled genotypes (EMP 122, EMP 277, EMP 320, EMP 322, and EMP 364) and the Carioca genotypes (EMP 250 and EMP 496). The susceptible lines included large-seeded commercial types for South America (Colombia, Ecuador, and Bolivia), such as PVA 773 and CAL 143. Susceptible lines also included small-seeded types for the Caribbean (Haiti and the Dominican Republic), such as BGMV-resistant lines from the University of Puerto Rico, UPR 9945-138 and 226, and the standard commercial variety, PC 50.

Results and discussion: The crosses with EMP 250 and EMP 496 produced the progeny with the best levels of resistance. The simple crosses with only one Andean parent produced smaller seeded progeny than the triple crosses with two Andean parents. The red-mottled and Carioca seed types combined well to produce progeny with good red-mottled seed types. Some of the progeny of the triple crosses had better seed quality than the current red-mottled EMP lines. Other triple crosses were used to combine Arcelin-based bruchid resistance with the *Empoasca* resistance present in the current red-mottled lines. Many of the double cross progenies that had only one EMP-line parent were eliminated in initial *Empoasca* trials. Similarly, most of the multiple crosses that used a final parent that was *Empoasca*-susceptible produced progeny that were also susceptible.

Conclusions and future plans: It appears necessary to pyramid *Empoasca*-resistant parents to obtain progeny that is resistant. In future multiple crosses we will use final parents that are *Empoasca* resistant in the hopes of obtaining better progeny. The inheritance of quantitative trait loci for leafhopper resistance will be analyzed using the RILs developed by single seed descent from the simple crosses EMP 496 x PVA 773 and EMP 250 x CAL 143. In the future, we hope to develop molecular markers for the selection of *Empoasca* resistance in common beans.

Contributors: MW Blair, C Cardona, JM Bueno

1.3.4 Resistance to *Zabrotes*

Rationale: *Zabrotes* (or the Mexican bean weevil) is one of the most important storage pests of bean. Unusual seed storage proteins called arcelin are derived from wild bean, and confer resistance. In previous reports we have described the selection of arcelin in complex crosses with sources of agronomic traits.

Materials and methods: Complex crosses segregating for arcelin and the *bgm-1* gene were created 2 years ago. The F₁ plants of these crosses were selected first for arcelin with electrophoretic gels and then for the *bgm-1* gene with the SCAR marker. Thus, two important resistance genes were selected by indirect methods to create populations with both genes.

Results and conclusions: Progeny families were selected that are now in the F₃ generation and soon will be selected for anthracnose resistance in Popayán. At that stage they will be selected as individual plant selections for progeny testing. Conclusions are pending.

1.3.5 Resistance to *Apion godmani*

Rationale: *Apion godmani*, the bean pod weevil, is normally controlled in Central America with the use of extremely dangerous pesticides, although resistant varieties DICTA 113 and 122 are reportedly reducing the use of pesticides in Honduras. However,

at present there is no longer an active breeding program for resistance to this pest. Because the pest does not exist in Colombia, selection cannot be practiced here either. An alternative strategy is to improve the DICTA varieties by backcrossing other traits into them, thereby maintaining their level of *Apion* resistance.

Materials and methods: The two DICTA varieties are derived from BGMV-resistant cultivar DOR 364 and present an intermediate reaction to BGMV. To fortify the value of these varieties, they were included in the *bgm-1* backcross program. The introduction of *bgm-1* will make these varieties more viable and attractive for farmers who face BGMV as well as *Apion*.

Results and conclusions: The backcross program with the DICTA lines is in the F₁BC₂ phase. Conclusions are pending.

Contributors: S Beebe, C Cardona, H Terán, JM Osorno, (IP-1);
C Quintero, J Tohme (SB-2)

Progress towards achieving output milestones:

- Studies on insect resistance and development of parental lines with multiple disease and insect resistance contribute to the mainstream breeding efforts of the project.
- Insect resistance to thrips (never studied before on dry beans) can be a key component in the formulation of management strategies for this important pest of the crop.

Activity 1.4 Incorporating wider genetic diversity into beans

1.4.1 Wild quantitative trait loci pursued in population of DOR 390 and G 19892

Rationale: Wild relatives of several crops were investigated as sources of QTL for complex traits, including yield. Wild *P. vulgaris* has been demonstrated to have much broader genetic variability than cultivated bean and feasibly could serve as a source of useful variability for broadening the base of the cultivated bean.

Materials and methods: Several populations involving wild bean were explored using the advanced backcross or backcross selfing method, whereby a large number of lines are generated by backcrossing a wild x cultivated hybrid twice to the cultivated parent. Marker and QTL analyses were initiated in the advanced backcross population of DOR 390 and G 19892. DOR 390 is a widely used Mesoamerican, small, black-seeded cultivar resistant to BGYMV, and G19892 is a wild bean from Argentina that presents Andean DNA patterns. Thus this cross represents a combination of wide genetic diversity within *P. vulgaris*, essentially bridging the two major gene pools. One hundred and thirty eight markers were applied to the population, including 25 SSRs, 111 RAPDs, and two SCARs.

Field trials were established in Santander de Quilichao under moderate fertility stress in the fall of 1999. Subsequently a second trial was planted in Palmira in January 2000 with normal (high) fertility levels. In the first trial, a population of 99 lines was planted in a 10 x 10 lattice, but it was observed that the advanced backcross method had resulted in very few lines with introgression for any given wild bean DNA segment, thus prejudicing the statistical test. Therefore, an additional 35 lines were included in a satellite yield test in the Palmira station and in the DNA analysis.

Results: Among the markers utilized, 17 SSRs, 27 RAPDs, and one SCAR could be mapped to the CIAT mapping population based on DOR 364 x G 19833. These mapped markers were calculated to cover about 692 cM or about 32% of the total genome length. The remaining 93 markers undoubtedly have covered another sizable portion of the genome, but it not yet possible to say how much, or if significant segments of the genome remain unsampled.

In the field, no line presented a statistically significant yield advantage over DOR 390. In the QTL analysis, many significant effects of introgressed DNA were detected by a Student's *t*-test on yield, seed size, and flowering date. However, most of the effects of introgressed DNA were negative, resulting in lower yield or smaller seed size. Indeed, results of negative QTL for yield were relatively consistent across the two sites, although more negative effects were detected in the Santander de Quilichao trial. It appears that the alleles from the wild bean were especially deleterious at the lower fertility level; wild beans have been noted to be sensitive to fertility stress. Only one positive yield QTL from the wild bean was detected in the yield trial that was carried out in Palmira, but it was not confirmed in Santander de Quilichao. This QTL was marked by the RAPD AI1405S, and was mapped to the linkage group B07. In Palmira, this marker was associated with a yield advantage of 175.7 kg ha⁻¹ in lines that carried the marker. It must be stressed that this QTL has still to be confirmed. Lines with the marker are being planted together with DOR 390 to verify if they continue to express any yield advantage. The negative QTL of the wild bean are also being mapped to determine other regions of the genome with important yield effects. The coverage of the map must also be saturated as soon as more microsatellites are available.

Conclusions: As expected, more negative than positive effects proceeded from the wild bean. Unfortunately, no positive yield QTL have yet been confirmed.

Contributors: S Beebe, H Terán, JM Osorno, HF Buendía (IP-1);
MW Blair, F Pedraza, J Tohme (SB-2)

Progress towards achieving output milestones:

- This activity would have contributed toward the milestone of developing improved lines with resistance to diseases, specifically to BGMV, because the improved lines with better yield QTL would carry this resistance derived from the recurrent parent. Given that reliable QTL remain to be identified, little substantial progress can be reported as of yet.