Additional evidence suggests a new map for the distribution of wild-weed-crop complexes of common bean in Colombia

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Introduction

Extensive wild-weedy-cultivated complexes of common bean were observed during collection expeditions in regions of Colombia where wild and cultivated beans are sympatric. Interbreeding complexes may be important mechanisms for the generation of genetic variability in landraces. Such complexes have also been observed in *Phaseolus vulgaris* L, in both Middle American and Andean gene pools (Beebe et al. 1997; González et al. 2003; and Toro and Ocampo, 2004). The ocurrence of introgression between the domesticated form and its wild ancestor or between the Middle American and Andean gene pools seem very probable and has been proposed for this crop, as a consequence of intraspecific hybridisation (Paredes and Gepts, 1995; and Chacón et al. 2002). Here, we report the finding of new complexes (wild-weedy-cultivated) of common bean in Colombian regions where these have not been reported previously. Additionally, we analyzed these complexes from a biochemical (phaseolin and isozyme markers) and morphological viewpoint to estimate the variability as a contribution to their conservation and use.

Materials and Methods

Plant material. Ten wild-weedy-crop complexes were selected after a geographic sampling in Colombia. In addition three accessions were chosen as controls: two cultivated *P. vulgaris* from the Andes and Mesoamerica (G4494 and G5773, respectively) and a Colombian wild (G24408). For the morphological and biochemical analysis, we only took the multiplied and conserved seed in the Phaseolus germplasm bank held in CIAT (Table 1).

Morphological analysis. For the analysis of morphological traits of seed phenotypes, we focused on seed size, seed shape, 100-seed weight, color and color pattern.

Biochemical analysis. The seed storage proteins were analyzed as selfed materials of phaseolin type found for each analyzed seed. This variation was first analyzed in 1D-SDS-PAGE (Brown et al. 1981) and confirmed later in 2D-IEF-SDS-PAGE (O'Farrel, 1975). For the isozyme analysis only a complex was selected (G50849), being used for it thirty selfed materials of phaseolin type. We used only two polymorphic enzymatic complexes: peroxidase (PRX; 1.11.1.7) and diaphorase (DIA; 1.6.4.1). The methodology for isozyme analysis was the one reported by Ramirez et al. (1987).

Results

Seed morphological variation of the complexes. The original seed of these populations was collected and classified as cultivated materials. However, during the initial seed increase, we observed segregation for seed size and colors indicating possible wild-weedy-crop complexes. The materials (1,182 in total) were classified as cultivated [642 (54%)], intermediate [432 (37 %)] and wild [108 (9 %)] (Table 1).

Table 1. Description of the wild-weed-crop complexes from domesticated Colombian populations of common bean.

CIAT No.	*Department*	Generación Go (seed original)		Generation advanced (increased seed)	
		S. W.[1]	Gene pool	B. S.[2]	Phaseolin types (frecuency in parenthesis)
G50711	Antioquia	64.2 g.	Andean	Cultivated	S (1), B (2), C (4), Car (2)
				Weedy	S (6), B (2), C (5), H_1 (1)
				Wild	S (5), C (3)
G50849	Antioquia	31.0 g.	Mesoamerican	Cultivated	S (37), C (41), H_1 (6), H_2 (3), T (4)
				Weedy	S (15), C (6), H_1 (2), H_2 (1)
				Wild	S (6), C (3)
G50632	Antioquia	50.5 g.	Andean	Cultivated	S (36) , CH (5), C (41), T (55), L (1)
				Weedy	S (3), B (17), C (3), T (1)
				Wild	B (6), T (1)
G50646	Antioquia	64.8 g.	Andean	Cultivated	S (14), B (2), CH (1), T (37), C (24), H_1 (1), H_2 (1)
				Weedy	S (13), T (9), C (5)
				Wild	T (1), C (6)
G50785	Antioquia	60.6 g.	Andean	Cultivated	S (16), B (3), CH (1), C (41), T (67), H_1 (8)
				Weedy	S (19), B (12), CH (10), T (30), C (45), H_1 (2)
				Wild	S (4), B (4), CH (3), T (5), C (13)
G50879	Caldas	62.5 g.	Andean	Cultivated	B (13), C (49), T (2), H_1 (22), H_2 (1)
				Weedy	B (16), C (4), H_1 (4)
				Wild	B (2), C (1), H_1 (1)
G50983	Cundinamarca	21.0 g.	Mesoamerican	Cultivated	S (6), C (2), Mu (1)
				Weedy	S (24), B (48), CH (13), C (9), H_2 (1), Mu (34)
				Wild	S (3), B (2), Mu (1)
G50988	Boyaca	35.4 g.	Mesoamerican	Cultivated	S (3), T (2), C (10), H_1 (5)
				Weedy	S (10), C (4), H_1 (2)
				Wild	S (5), C (6), H_1 (1)
G50797	Tolima	61.0 g.	Andean	Cultivated	S (1)
				Weedy	S (6), C (3), H_1 (4)
				Wild	S (9), C (2), H_1 (4)
G50859	Cauca	33.0 g.	Mesoamerican	Cultivated	S(5),B (24),T(10),C(18),Ca_1(4),H_1(2),H_2(1),Car (7)
				Weedy	B (36), C (6), H_1 (1)
				Wild	B (11)

[1]S. W. : Is the seed weight derived from 100 seeds [2]B. S. : Biological Status;

These segregating populations were considered to be complexes, since they involve wild and weedy stabilized forms. These complexes showed a great diversity in seed size (from small to large) and color (Figure 1).

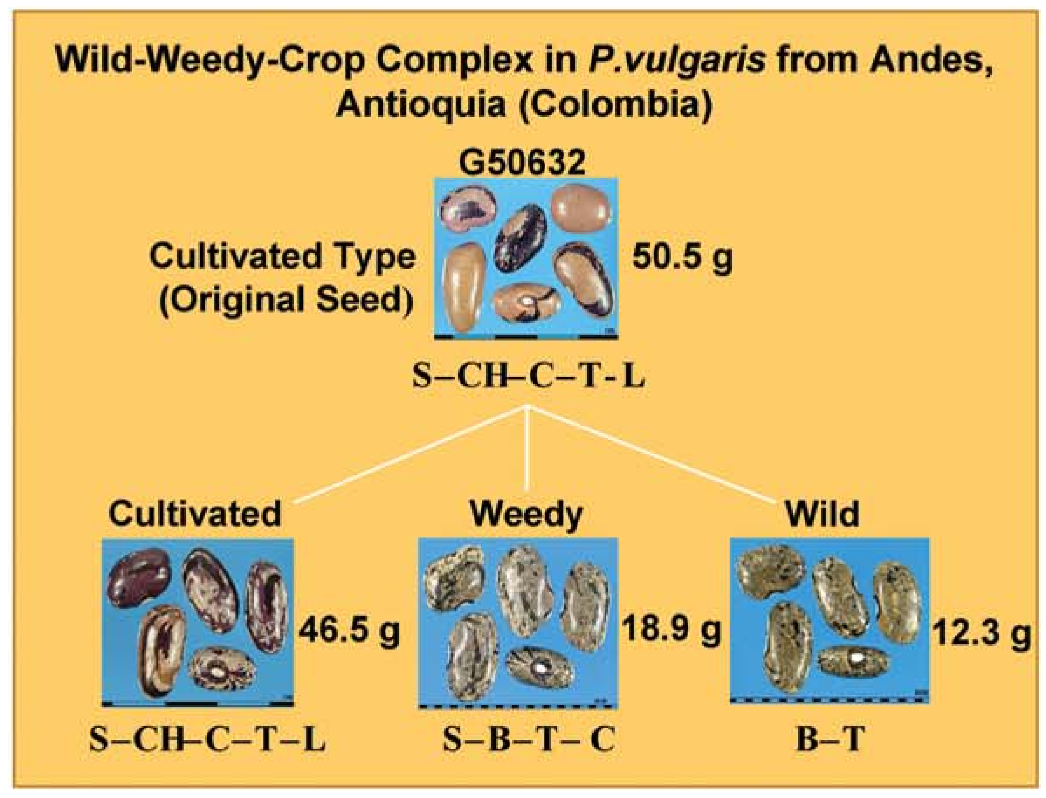

Figure 1. Example of Seed morphological variation and biochemical for wild-weedy-crop complexe G50632. In the right side of imagenes shows 100 seed weight and down side is the phaseolin types.

Seed protein variation of the complexes. a great diversity for phaseolin types was found within these complexes. The patterns found so far were: five Andean (C, T, H1, H2 and Ca), two Mesoamerican-Colombian (B, CH), a Mesoamerican (S) and three Colombian (L, Car and Mu), with a frequency of 55%, 20%, 21% and 4% respectively. In these complexes, the 'S', 'B', 'C', 'T', and 'Mu' phaseolins form a continuum across the full range of biological status (Table 1). The 'C' phaseolin was present at the highest frecuency (30%). Followed by the 'S' type with 21%, 'T' type with 20% and 'B' type with 17%. The higher ocurrence of Andean phaseolin types (55%) compared to that of Middle American 'S' phaseolin type (21%) may have resulted from a selection for larger seed size in the sampled Colombian regions for this study. The phaseolin types such as 'L' (Beebe et al. 1997), 'Mu' and 'Car' (Toro et al. 2007) have been found so far only in Colombian materials (Figure 2).

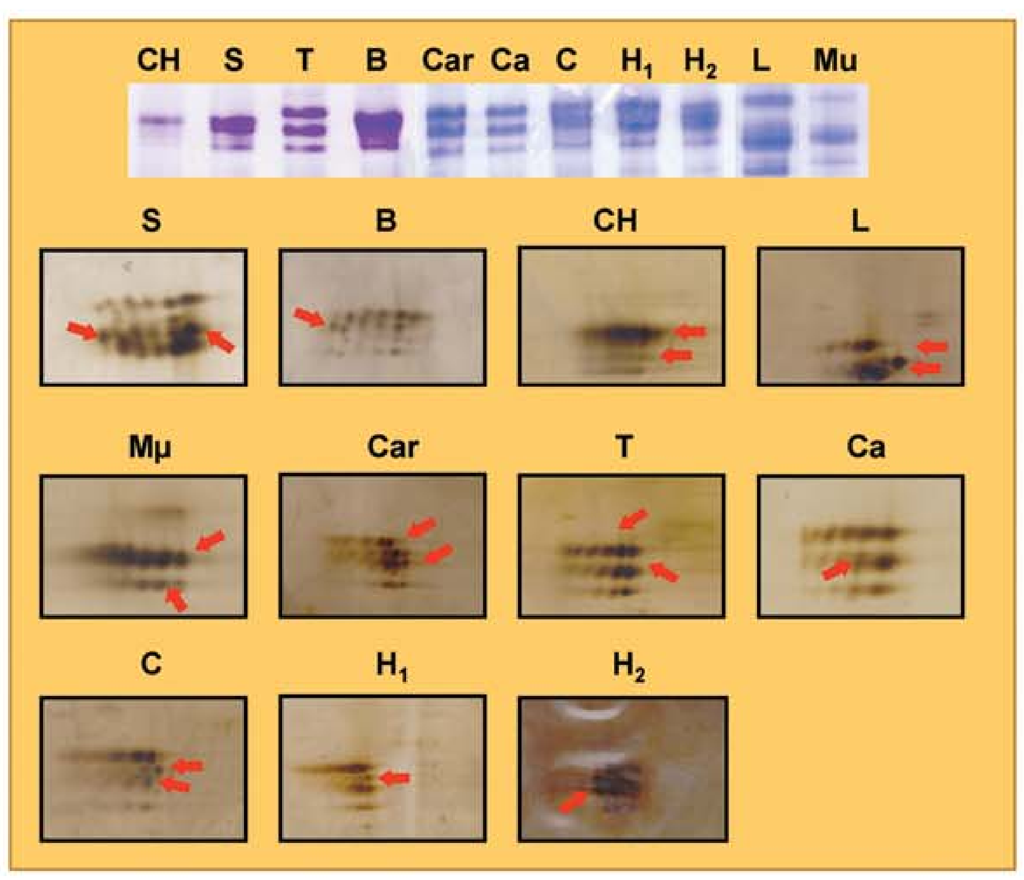

Figure 2. One-dimensional SDS-PAGE (upper line) and 2D-IEF-SDS-PAGE (second line downwards) gels of wild-weed-crop complexes showing phaseolin types found in them . For the 2D gels, arrows point to key peptides.

Isozyme variation of the complexes. For the isozyme analysis, both allozymes (Mesoamerican and Andean) are found in the analyzed complex (G50849) (Table 2). The selected isozyme loci carry alleles from both Mesoamerican and Andean gene pools:

Table 2. Allozyme constitution and seed size of the wild-weedy-crop complex G50849.

Biological material	Analyzed "selfed materials"	100 seed weight (g)	Isozyme loci	
			Prx	Dia-1
G50849 Cultivated	23	23.4-47.8	100 (5) 98 (14) 100/98 (4)	100 (17) 95 (6)
G50849 Weedy	4	10.0-24.0	100 (0) 98 (3) 100/98 (1)	100 (4) 95 (0)
G50849 Wild	3	5.3-7.2	100 (1) 98 (0) 100/98 (2)	100 (3) 95 (0)

The Dia-1 (95), PRX98 alleles are considered to be Mesoamerican and the Dia-1 (100), PRX100 alleles are of Andean origin (Koenig and Gepts, 1989; Debouck et al. 1993). Nevertheless, only two allozymes were found in all phases of the complex: a heterozygote allozyme (PRX 100/98) and an Andean allozyme (Dia-1 [100/100]).

Conclusions

The variability at the phaseolin and isozyme levels suggests an important genetic interchange in the study area in Colombia between Mesoamerican and Andean materials. The presence of these complexes that are apparently hybrids between the two gene pools, can be explained by the existence of true wild forms and landraces in Colombia with 'S' phaseolin and 'CH' (Mesoamerican) and 'T' (Andean). These results are concordant with those obtained by Debouck et al. (1993); Paredes and Gepts (1995) and Beebe et al. (1997), using morphological and biochemical markers, and those obtained by Tohme et al. (1996), Chacón et al. (2002), and Ocampo et al. (2005), using molecular markers. However, we are reporting a extensive distribution of these introgressed complexes in Colombia, much more than those reported by Beebe et al. (1997). This distribution includes some departments where wild and cultivated beans are sympatric (Cundinamarca and Boyaca), or in departments where the common bean is an important crop (Antioquia, Caldas, Tolima and Cauca). These results suggest a new map in Colombia for the distribution of these biological complexes of common bean and confirm that a considerable amount of natural hybridization occurs in the areas where these populations were collected (Figure 3).

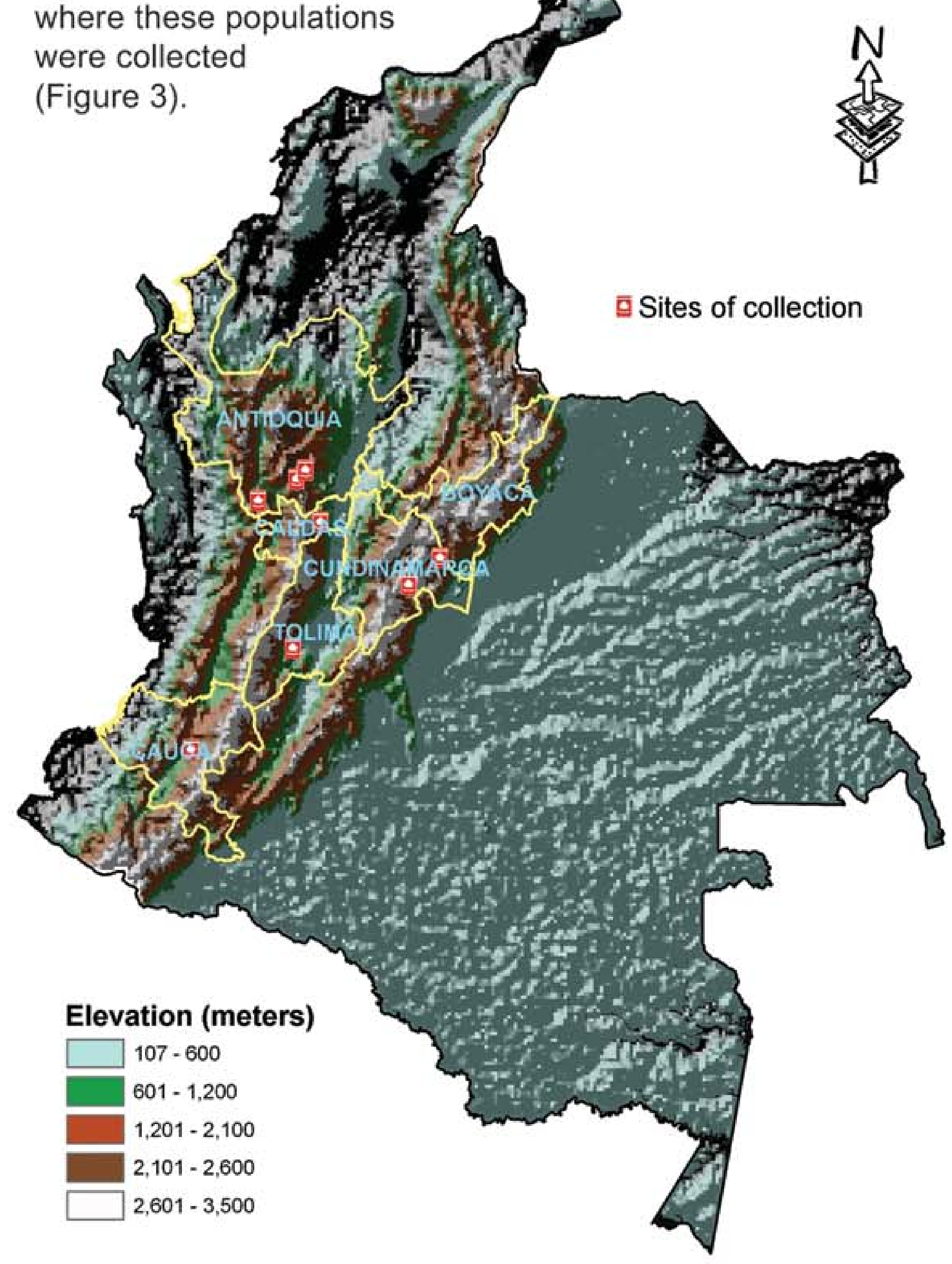

Figure 3. New map for the distribution of the wild-weedy-crop biological complexes of common bean in Colombia.

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