Gene flow events among bean species of section *Phaseoli* in Colombia and Costa Rica using microsatellites markers

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Introduction

Gene flow events in common bean have been reported in several parts of the Americas where wild and cultivated forms are sympatric, often distant from a few to dozens of meters. After using microsatellite markers to successfully establish gene flow events in weedy forms and to indicate pollen direction from the wild into the cultivated and vice versa (González-Torres et al. 2004), we were interested in testing the hypothesis of participation of alien species into such a flow. Studies (Schmit et al. 1993; Llaca et al. 1994; Delgado-Salinas et al. 1999) using neutral molecular markers have shown that among the dozens of species described (Freytag & Debouck 2002) a small group of species including the common bean – section *Phaseoli* – share the same lineage. These species are sympatric with wild *P. vulgaris* in several mountainous areas of tropical America (Debouck 2000): *P. albescens* in western Mexico (Ramírez-Delgadillo & Delgado-Salinas 1999), *P. costaricensis* in central Costa Rica (Araya-Villalobos et al. 2001), and *P. dumosus* in several parts of Central America and the northern Andes (Schmit & Debouck 1991).

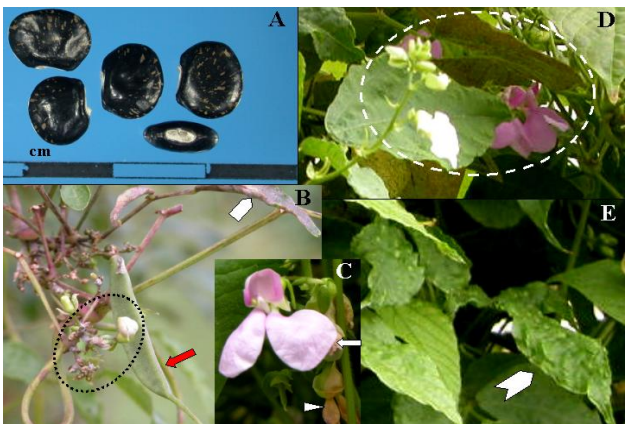

Figure 1. Phenotypic markers of interspecific hybrids.
A. Original seeds of individual FI7031 (atypical seeds can produce plants with hybrid expression
B. Material FI7031 showed floral bud abscission and ovule abortion.
C. G24765 flower with abnormal size, bracteoles , and spatial position of wings (similar to *P. dumosus).*
D. Individual G24666A displays different flower colors (white and purple).
E. The *cripple* phenomenon exhibited by individual G24666A.

Materials and Methods

We tested microsatellites screened at 67 loci (Gaitán-Solís et al. 2002) to evaluate the level of participation of nuclear genes in six rare forms possibly resulting from interspecific hybridizations in natural conditions of Colombia (in contact with *P. dumosus*) and Costa Rica (with *P. costaricensis* and *P. dumosus*). The analysis involved these species as well as *P. coccineus* and *P. albescens* as controls (Table). The atypical materials were selected because of growth abnormalities often seen in artificial interspecific hybrids (shrivelled seeds, ovule abortion, crippled plants) (Hucl & Scoles 1985).

CIAT Identification	Collector Identification	Species	Country	Department	Biological Status
G24765 (Pop9072)	OT-453	P. x *vulgaris*	Colombia	Boyacá	Weedy
G24666A (Pop9077)	OT-229	P. x *vulgaris*	Colombia	Cundinamarca	Weedy
FI7031 (S34124)	DGD-3149	P. x *vulgaris*	Costa Rica	Cartago	Weedy
FI7033 (S34124)	DGD-3149	P. x *vulgaris*	Costa Rica	Cartago	Weedy
FI7034 (S34124)	DGD-3149	P. x *vulgaris*	Costa Rica	Cartago	Weedy
FI7035 (S34124)	DGD-3149	P. x *vulgaris*	Costa Rica	Cartago	Weedy
S29699	DGD-2102	P. *costaricensis*	Costa Rica	San Jose	Wild
G36285 (Coc-1718)	DGD-3087	P. *coccineus*	Guatemala	Quezaltenango	Wild
G36290 (Coc-1440)	OT-811	P. *dumosus*	Colombia	Caldas	Cultivated
PL3592	ROL-141	P. *albescens*	Mexico	Jalisco	Wild
G23418 (FI6846)	DGD-2111	P. *vulgaris*	Costa Rica	Cartago	Wild

Results and Discussion

The microsatellites were powerful enough to separate the species though they belong to the same evolutionary phylum (Fig. 2). The characterization of the different bean species through microsatellite *loci* evaluation according to Gaitán-Solís et al. (2002) is confirmed and extended to *P. albescens* and *P. costaricensis* for the first time (Fig. 2, 3).

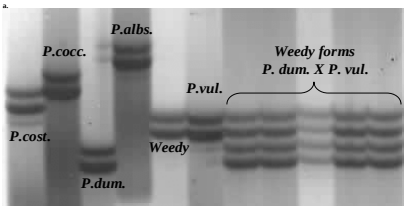

Figure 2. Specific microsatellites characterizing each species and observed in the evaluated interspecific hybrids. *Locus* of microsatellites BM181 shows different allelic forms in all species and the shared alleles in hybrids individuals between *P. vulgaris* and *P. dumosus*.

These molecular markers indicated that the putative natural interspecific hybrids actually were hybrids (Fig. 2, 3). The data indicate that the evaluated hybrids result from gene flow between the common bean and *P. dumosus* (as pollen donor) in the Central Valley of Costa Rica as well as in Boyacá, Colombia (Fig. 4).

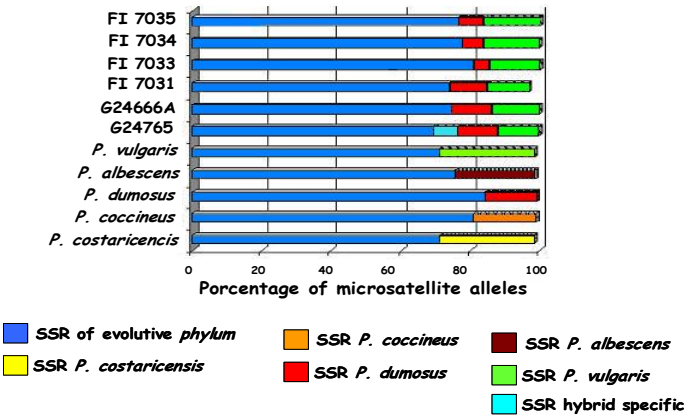

Figure 3. Graphic representation of the alleles found in the individuals.

A different allelic frequency, namely of *P. dumosus*, suggests that the hybrids are of different generations (Fig. 3, 4). The cluster obtained using MCA established the hybrid group structure. This group is spatially near to *P. vulgaris* and *P. dumosus* indicating that the evaluated *loci* have been a recombination among these species by gene flow events. The natural interspecific hybrids are rare, and their reduced fertility might imply that the species of this *phylum* are "good biological species". They can be valued as natural genetic bridges in improvement programs (Singh 2001).

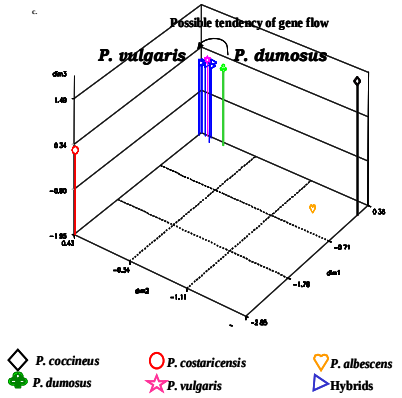

Figure 4. Spatial distribution of individuals using multiple correspondence analyses.

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