



Domestication patterns in wild Lima bean (*Phaseolus lunatus* L.) from the Americas

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Introduction

Lima bean (*Phaseolus lunatus* L.) is one of the five cultivated *Phaseolus* bean species, which originated in the Neotropics. Lima beans have two botanical varieties, *var. silvester* and *var. lunatus* that refer to the wild and domesticated ones, respectively. Within the *silvester* variety two gene pools, the *Mesoamerican* and the *Andean*, have long been recognized (1-6). However, recent evidence suggest that the structure of genetic diversity in wild Lima beans may be organized into three gene pools: gene pool AI is restricted to the Andes of Ecuador and northern Peru, gene pool MI is mostly found in Mexico in the area to the north and northwest of the Isthmus of Tehuantepec, and gene pool MII is widespread and found in Mexico (to the south and southeast of the Isthmus of Tehuantepec), Central America, Colombia, southern Peru and Argentina (Serrano et al., unpublished data). Wild Lima bean is one of the few species where multiple domestication events have been documented, which gave rise to three cultigroups: two small-seeded ones called "Sieva" and "Potato" (their place of origin is still controversial) and one large-seeded called "Big Lima" of Andean origin (Ecuador-northern Peru) (1,3,6). It is widely known that the domestication process has important consequences on genetic diversity and phenotypic traits (e.g. the domestication syndrome), therefore the importance to determine where and how domestication events took place in a given species. The purpose of this research was to evaluate domestication patterns in Lima beans with special emphasis on the small-seeded cultigroups, and the effect of domestication on genetic diversity. For this purpose, two intergenic spacers of the chloroplast DNA (cpDNA) (*atpB-rbcL* and *trnL-trnF*) and the internal transcribed spacer of the ribosomal DNA (ITS/5.8S) were sequenced in a sample of wild and domesticated Lima beans. The results confirm a domestication center for the "Big Lima" cultigroup on the western slope of the Andes in Ecuador and northern Peru. The results support a second domestication event in Mexico for the small-seeded cultigroups, in the area to the west and northwest of the Isthmus of Tehuantepec. There was a strong domestication founder effect in both Mesoamerica and the Andes (about 70% for cpDNA and 75% for ITS/5.8S).

Plant material

All plant material used in this study was obtained from the International Center for Tropical Agriculture in Cali, Colombia. A total of 42 wild and 35 domesticated Lima bean accessions were selected on the basis of their geographic distribution and complete passport data. Leaflets were collected from each of 5 seedlings per accession for DNA extraction with a standard CTAB method.

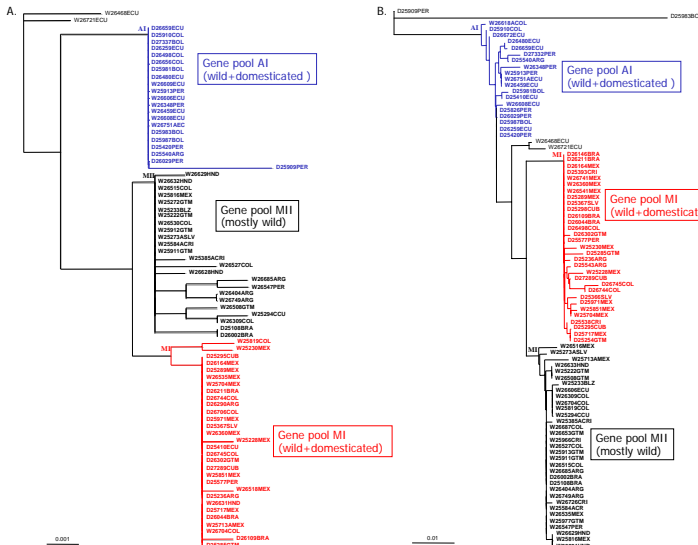


Figure 1. NJ topologies based on cpDNA (A) and ITS/5.8S (B) data for wild (W) and domesticated (D) Lima beans. Gene pools that contain wild and domesticated accessions are colored: AI (blue) and MI (red). Each accession is preceded by a W for wild or a D for domesticated and followed by three capital letters indicating country of origin as follows: MEX= Mexico, GTM= Guatemala, BLZ= Belize, HND= Honduras, SLV= El Salvador, CRI= Costa Rica, COL= Colombia, CUB= Cuba, ECU= Ecuador, PER= Peru, BOL= Bolivia, BRA= Brazil, ARG= Argentina. Gene pool MII contains mostly wild accessions, except for two domesticated accessions from Brazil.

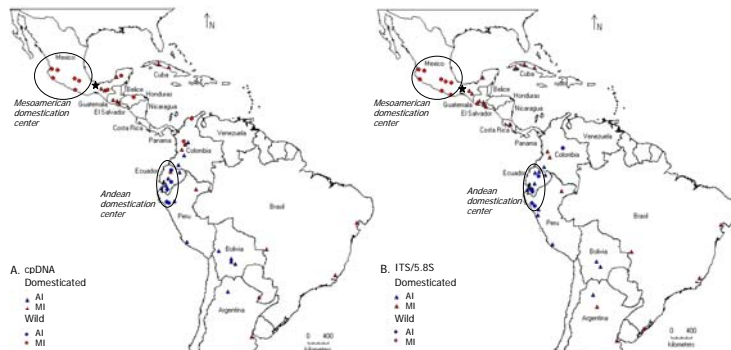


Figure 2. Geographic distribution of wild and domesticated clusters AI (blue) and MI (red) (see Figure 1) that reveal the Mesoamerican and Andean domestication centers in wild Lima beans according to cpDNA (A) and ITS/5.8S (B) evidence. Circles indicate distribution of wild accessions, triangles indicate the distribution of domesticated accessions. Star=Isthmus of Tehuantepec.

Domestication centers in wild Lima bean

Two intergenic spacers of the cpDNA (*atpB-rbcL* and *trnL-trnF*) and the ITS/5.8S region were sequenced in the sample of wild and domesticated Lima beans. In order to evaluate the genetic relationships among wild and domesticated Lima beans, two genetic distance matrices were obtained (for cpDNA and ITS/5.8S data) under the F84 model of molecular evolution. From these distance matrices, two dendrograms were built by using the NJ algorithm (Figure 1). As it can be seen from the cpDNA and ITS/5.8S NJ topologies, most of the domesticated accessions group together with wild Lima beans in only two clusters: cluster AI (blue color) and MI (red color) (Figure 1). The cluster AI contains wild accessions from the wild gene pool AI (as defined by Serrano et al. unpublished data) restricted to the western side of the Andes of Ecuador and northern Peru, and domesticated accessions distributed in Colombia, Ecuador, Peru, Bolivia and Argentina (Figure 2). Domesticated accessions from this cluster are large-seeded (100-seed weight=70g) and have been identified as the "Big Lima" cultigroup. These results suggest a domestication event in the Andes of Ecuador-northern Peru, as it has been reported previously (5,6). The cluster MI contains wild accessions from the wild gene pool MI (as defined by Serrano et al. unpublished data) that occur in Mexico, in the area to the west and northwest of the Isthmus of Tehuantepec, and small-seeded domesticated accessions widely distributed from Mexico to Argentina (Figure 2). These results reveal a second domestication event from small-seeded wild populations, possibly in an area restricted to Mexico (as stated above). Previous studies (6) have already suggested a possible domestication event for the small-seeded domesticated forms in the Mexico-Guatemala area (including the Caribbean and the Isthmus of Panama). In the present study the possible domestication area is narrowed to the north part of the distribution of wild Lima beans. However, in order to locate more precisely domestication sites of small-seeded cultivars, germplasm collections of wild populations should be improved, specially in Mexico, where sampling is still deficient. One of the purposes of the present study was to validate the existence of two different small-seeded cultigroups ("Sieva" and "Potato") (3). The present results (Figure 1) do not support the existence of two small-seeded cultigroups, therefore, it is recommended to group all the small-seeded domesticated forms in only one cultigroup.

Founder effect due to domestication

The founder effect due to domestication was calculated on the basis of nucleotide diversity indexes (π) (7) estimated for wild and domesticated Lima beans (Table 1). For the Andean domestication event, the reduction of genetic diversity was estimated as 48% and 75% according to cpDNA and ITS/5.8S data, respectively. For the Mesoamerican domestication event, the reduction in genetic diversity was even greater and estimated for cpDNA as 91.87% and for ITS/5.8S as 75.75%. The average reduction in genetic diversity for cpDNA (about 70%) and for ITS/5.8S (around 75%) for the two domestication events indicate that in general there was a strong founder effect due to domestication with a magnitude that is about the same or larger to that reported for other crops such as common bean (38-55%) (8) and maize (20-65%) (9). These results suggest that wild Lima beans contain unique genetic diversity not present in domesticated Lima beans and therefore special attention should be taken to preserve these genetic resources.

Table 1. Founder effect for the two domestication events in Lima beans (Andean, AI and Mesoamerican, MI) according to cpDNA and ITS/5.8S polymorphisms. N: number of accessions analyzed, π : nucleotide diversity, SD: standard deviation, founder effect was calculated for each domestication event as $(\pi_{wild} - \pi_{domesticated})/\pi_{wild}$.

Domestication event	Population	N	$\pi \pm SD$	Founder effect (%)
cpDNA				
Andean (AI)	Wild	7	0.00050±0.00011	48.00
	Domesticated	14	0.00026±0.00022	
Mesoamerican (MI)	Wild	11	0.00123±0.00037	91.87
	Domesticated	19	0.00010±0.00008	
Average founder effect for cpDNA				
ITS/5.8S				
Andean (AI)	Wild	6	0.00424±0.00119	75.00
	Domesticated	13	0.00106±0.00034	
Mesoamerican (MI)	Wild	7	0.00334±0.00098	75.75
	Domesticated	24	0.00081±0.00031	
Average founder effect for ITS/5.8S				
75.34				

Concluding remarks

In summary, the results support two domestication events for wild Lima beans, one would have occurred on the western side of the Andes in Ecuador and northern Peru from the wild gene pool AI, and the second one would have occurred in Mexico from the wild gene pool MI, in the area to the west and northwest of the Isthmus of Tehuantepec. According to diversity indexes calculated for wild and domesticated Lima beans, in both domestication events there was a strong reduction of genetic diversity due to founder effects which suggest that wild Lima beans may be considered as reservoirs of genetic diversity not present in domesticated Lima beans that may be useful for future breeding programs.

References

- GUTIÉRREZ SALGADO, A., GEPTS, P., DEBOUCK, D.G. (1995). Evidence for two gene pools of the Lima bean, *Phaseolus lunatus* L., in the Americas. *Genetic Resources and Crop Evolution* 42: 15-28.
- DEBOUCK, D.G. (1996). Colombian Common and Lima beans: views on their origin and evolutionary significance. *Revista Colombiana de Biología* 1: 7-15.
- MAQUET, A., VEKEMANS, X., BAUDOIN, J.P. (1999). Phylogenetic study on wild allies of Lima bean, *Phaseolus lunatus* (Fabaceae), and implications on its origin. *Plant Systematics and Evolution* 218: 43-54.
- CAICEDO, A.L., GAITÁN, E., DUQUE, M.C., TORO, O., DEBOUCK, D.G., TOHME, J. (1999). AFLP fingerprinting of *Phaseolus lunatus* L. and related wild species from South America. *Crop Science* 39: 1497-1507.
- FOFANA, B., BAUDOIN, J.P., VEKEMANS, X., DEBOUCK, D.G., du JARDIN, P. (1999). Molecular evidence for an Andean origin and a secondary gene pool for the Lima bean (*Phaseolus lunatus* L.) using chloroplast DNA. *Theoretical and Applied Genetics* 98: 202-212.
- FOFANA, B., du JARDIN, P., BAUDOIN, J.P. (2001). Genetic diversity in the Lima bean (*Phaseolus lunatus* L.) as revealed by chloroplast DNA (cpDNA) variations. *Genetic Resources and Crop Evolution* 48: 437-445.
- NEI, M., and MILLER, J.C. (1990). A simple method for estimating average number of nucleotide substitutions within and between populations from restriction data. *Genetics* 125: 873-879.
- CHACÓN S. M.I., PICKERSGILL, B., DEBOUCK, D.G. (2005). Domestication patterns in common bean (*Phaseolus vulgaris* L.) and the origin of the Mesoamerican and Andean cultivated races. *Theoretical and Applied Genetics* 110: 432-444.
- TENAILLON, M.I., UREN, J., TENAILLON, O., GAUT, B.S. (2004). Selection versus demography: a multilocus investigation of the domestication process in maize. *Molecular Biology and Evolution* 21: 1214-1225.

Acknowledgments

We are grateful to the Universidad Industrial de Santander, Colombia, and the Banco de la República, Colombia, for providing funding to carry out this research. We are also grateful to the Genetic Resources Unit of CIAT for providing the plant material used in this research.