Roberto Papa, Laura Nanni, Delphine Sicard, Domenico Rau, and Giovanna Attene

CHAPTER 6

Evolution of Genetic Diversity in *Phaseolus vulgaris* L.

Among domesticated plant species, the common bean (*Phaseolus vulgaris* L.) is the most important protein source for direct human consumption (Singh, 2001; Broughton et al., 2003). It is a diploid (2n = 2x = 22), annual species and is predominantly self-pollinating, with the occasional occurrence of cross-pollination by pollinators such as the bumblebee, *Bombus* spp. (Free, 1966). Many studies have been aimed at determining the origins, domestication, and evolution of the genetic diversity of *P. vulgaris*. Since seed storage proteins first became important in bean research, the advent of molecular techniques has had a major impact on our understanding of the *P. vulgaris* evolutionary history (Gepts, 1988b). The presence of geographically isolated gene pools in *P. vulgaris* that originated from at least two independent domestication events and the overlapping distribution with other domesticated and wild species that have different mating systems and are at various degrees of reproductive isolation make *P. vulgaris* and the genus *Phaseolus* a unique model for studies of plant evolution. Therefore, in addition to a brief illustration of the major aspects of the evolutionary history of *P. vulgaris* (for further details, see Gepts, 1996, 1988a; Debouck, 1999; Singh, 2001; Broughton et al., 2003; Snoeck et al., 2003), we focus here on recent studies highlighting the roles of the various evolutionary forces in shaping the genetic diversity of *P. vulgaris*. These include the potential

role of introgressive hybridization between *P. vulgaris* and *P. coccineus* in Mesoamerica, the effects of gene flow and selection between wild and domesticated bean populations, the evolution of disease resistance, and the effects of the introduction of the bean into the Old World.

The genus belongs to the tribe *Phaseolae* (subfamily *Papilionoideae,* family *Leguminosae*), which includes two other genera with domesticated species: *Glycine* (soybean) and *Vigna* (cowpea). Verdecourt (1970) redefined *Phaseolus* as a large, diverse genus of at least 50 species, as was later confirmed by further studies (Maréchal et al., 1978; Lackey, 1981, 1983). *Phaseolus* is strictly of the New World, and it grows naturally in the warm tropical and subtropical regions from Mexico (Sousa and Delgado-Salinas, 1993) to Argentina (Delgado-Salinas, 1985; Debouck et al., 1987).

Phaseolus includes five domesticated species: *P. vulgaris* (common bean), *P. lunatus* (lima bean), *P. acutifolius* A. Gray (tepary bean), *P. coccineus* ssp. *coccineus* (runner bean), and *P. coccineus* L. ssp. *polyanthus* Greenman = *P. polyanthus* (= *P. coccineus* ssp. *darwinianus*) (year-long bean). Each of these has a distinct geographic distribution, life history, and reproductive system (Maréchal et al., 1978; Delgado-Salinas, 1985). The phylogenetic relationships between these *Phaseolus* species have been investigated using a number of morphological (Maréchal et al., 1978; Debouck, 1991), biochemical (Sullivan and Freytag, 1986; Jaaska, 1996; Pueyo and Delgado-Salinas, 1997), and molecular (Delgado-Salinas et al., 1993; Schmit et al., 1993; Llaca et al., 1994; Hamann et al., 1995; Vekemans et al., 1998) tools. In particular, a recent phylogenetic analysis of *Phaseolus* and its close relatives combined molecular (internal transcribed spacer [ITS]/5.8S DNA sequences) and nonmolecular data (vegetative, floral, and fruit morphological characters and chromosome numbers) (Delgado-Salinas et al., 1999) and confirmed that *Phaseolus* is monophyletic. This is consistent with several studies of both wild and domesticated species of *Phaseolus* that have used a wide range of tools, including seed proteins, isozymes, and nuclear, chloroplast, and mitochondrial DNA (Debouck, 1999). Delgado-Salinas et al. (1999) also revealed that there may be anywhere from two to nine subclades within *Phaseolus,* with the cultivated species falling into two distinct lineages. In one, the domesticated species *P. vulgaris, P. coccineus, P. polyanthus,* and *P. acutifolius* are found together with two wild species, *P. albescens* and *P. costaricensis.* Another clade contains *P. lunatus* and wild species of both Andean and Mesoamerican distributions (Fofana et al., 1999; Maquet and Baudoin, 1996; Delgado-Salinas et al., 1999).

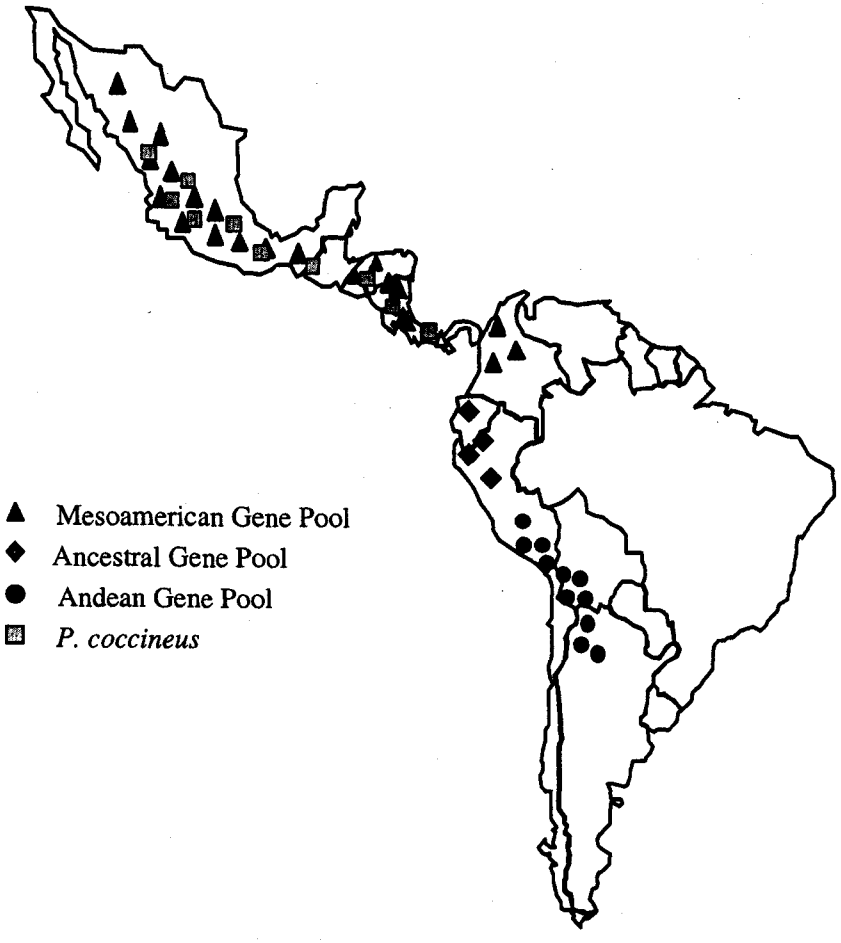

FIGURE 6.1 Distributions of the wild populations of *P. vulgaris* and *P. coccineus.*

The intraspecific organization of genetic variation in *P. vulgaris* has been investigated in detail. The presence of two distinct gene pools was suggested by analyses of seed morphology (Evans, 1973, 1980), of hybrid nonviability in crosses between *P. vulgaris* from Mesoamerica and South America, and of outbreeding depression (see Singh, 2001, for review). The analyses of variations in seed storage proteins (e.g., phaseolin) also supported the presence of distinct Mesoamerican and Andean gene pools, with the presence of parallel geographic patterns in both the domesticated and the wild beans indicating the occurrence of independent domestication in Mesoamerica and South America (Gepts et al., 1986; Gepts and Bliss, 1988; Koenig and Gepts,

1989; Koenig et al., 1990; Singh et al., 1991). A different type of phaseolin (type I) has been observed in wild accessions from north Peru and Ecuador, and sequence analyses of the locus coding for these proteins revealed that type I phaseolin is the ancestral form from which the other phaseolins evolved. This indicated that the populations from north Peru and Ecuador were the closest descendants of the ancestor of the common bean (Kami et al., 1995). Overall, these studies indicated three different wild gene pools (Mesoamerican, Andean, and Ancestral) (figure 6.1), with evidence of domestication events only in the Mesoamerican and Andean gene pools. Both the independent domestication and the origins of wild *P. vulgaris* have been confirmed by various studies based on other molecular markers (Khairallah et al., 1992; Becerra and Gepts, 1994; Caicedo et al., 2000; Papa and Gepts, 2003).

The Andean and Mesoamerican gene pools have different structures and levels of genetic diversity in both the wild and domesticated populations, where the occurrence of different races has also been described (Singh, 2001). Indeed, there is a higher genetic diversity in the Mesoamerican than the Andean gene pool for both wild and domesticated populations (Koenig and Gepts, 1989; Beebe et al., 2000, 2001; Papa and Gepts, 2003; McClean et al., 2004). Additionally, a higher interpopulation component of genetic variance has been indicated for the Mesoamerican wild populations (using amplified fragment length polymorphism [AFLP]; Papa and Gepts, 2003), in comparison with the Andean wild populations (using random amplified polymorphic DNA [RAPD]; Cattan-Toupance et al., 1998). A much higher level of genetic differentiation has also been observed between the domesticated races from Mesoamerica (using RAPD; Beebe et al., 2000) than between those from South America (using AFLP; Beebe et al., 2001). However, further direct comparisons may be needed because of the use of different types of molecular markers.

Interspecific Hybridization

In contrast to South America, in Mesoamerica *P. vulgaris* often is sympatric with other species that are partially sexually compatible. For this reason, one possible explanation for the differences in the levels of genetic diversity between the gene pools is the occurrence of introgressive hybridization between *P. vulgaris* and the other *Phaseolus* species. Indeed, in Mesoamerica the distribution of *P. vulgaris* overlaps with that of *P. coccineus* and *P. polyanthus*. Molecular studies have shown that *P. polyanthus*, which was formerly included in *P. coccineus*, is intermediate in its morphological

features between these other two species (Hernandez-Xolocotzi et al., 1959), and a hybrid origin has indeed been suggested (Piñero and Eguiarte, 1988; Kloz, 1971; Llaca et al., 1994). At the molecular level, *P. polyanthus* is closer to *P. coccineus* by nuclear DNA comparison (Piñero and Eguiarte, 1988; Delgado-Salinas et al., 1999) but more similar to *P. vulgaris* by chloroplast DNA comparison (Llaca et al., 1994). Thus *P. polyanthus* probably originated from a cross that involved *P. vulgaris* as the maternal parent, with successive backcrosses to *P. coccineus* as the paternal donor (Schmit et al., 1993; Llaca et al., 1994). This interpretation is consistent with studies showing that in artificial crosses between *P. coccineus* and *P. vulgaris*, fertile F_1 progeny can be produced, particularly when *P. vulgaris* is the maternal parent (Singh, 2001; Broughton et al., 2003). This suggests that introgression between *P. coccineus* and *P. vulgaris* occurred in the evolutionary history of both species in Mesoamerica.

Using nuclear and chloroplast microsatellites (simple sequence repeats; SSRs), there is evidence of introgression in sympatric populations of *P. coccineus* and *P. vulgaris* from Morelos, Mexico (Sicard and Papa, unpublished data), which suggests that gene flow might still be important in shaping the structure of the genetic diversity of these two species in Mesoamerica. Through an analysis that used the same SSR loci of wild and domesticated germplasm accessions of these two species and included the Andean gene pool of *P. vulgaris*, the level of introgression was seen to be highly locus specific. Thus loci that displayed higher similarities between *P. vulgaris* and *P. coccineus* from Mesoamerica also showed a stronger differentiation between Andean and Mesoamerican *P. vulgaris*. Because only microsatellites designed from genic regions were used, it was not possible to discriminate between the effects of selection and gene flow in driving this introgression. Nevertheless, these results may have strong implications for our understanding of the structure and level of genetic diversity in the common bean. In particular, they suggest that introgression from *P. coccineus* probably was one of the causes of both the higher genetic diversity present in Mesoamerica (as compared with the Andes) and the partial reproductive isolation between the gene pools. However, other possible explanations, such as homoplasy and convergent evolution, remain to be investigated.

Gene Flow and Selection Between Wild and Domesticated *P. vulgaris*

For beans, as for many other species (Harlan and de Wet, 1971), the wild and domesticated forms belong to the same biological species and are

completely cross-fertile (Koinange et al., 1996). The domestication process has led to a reduction in genetic diversity within each of the bean gene pools (Sonnante et al., 1994), as has been seen for other species (e.g., *Zea mays:* Doebley et al., 1990; Ladizinsky, 1998). This effect, called a domestication bottleneck, is a function of the small samples of individuals that founded the domesticated populations. In addition to this founder effect, which has generally affected the whole genome diversity, selection for specific traits probably has also contributed to reductions in genetic diversity at target loci and in the surrounding genomic regions. This results from the combined

FIGURE 6.2 Close-range sympatry between wild and domesticated common bean (*P. vulgaris* L.) in Teopisca, Chiapas, Mexico. Wild and the domesticated common beans have a similar climbing growth habit and phenology. Pods of wild and domesticated beans. (Photo courtesy of Papa and Gepts.)

effects of selection and recombination (e.g., hitchhiking; Maynard Smith and Haigh, 1974; Kaplan et al., 1989). Thus the effects of domestication at neutral loci that are linked to those selected during domestication are likely to be strictly related to the breeding system of a given species (allogamous versus autogamous), along with other factors affecting the amount of recombination (e.g., population size). For instance, in the allogamous plant species *Zea mays,* the role of hitchhiking appears to have affected restricted genomic regions around selected sites (Wang et al., 1999, 2001; Tenaillon et al., 2001; Clark et al., 2003). A higher level of linkage disequilibrium probably would be expected in autogamous species, such as the common bean. The traits that distinguish the domesticated from the wild form are collectively called the domestication syndrome (Hammer, 1984), and they are shared by most domesticated crop species. These key traits include the lack of seed dispersal and dormancy, a compact plant architecture, a higher yield, a synchronicity, and an early flowering. The majority of these domestication traits have simple Mendelian determinism with, in most cases, complete or semidominance of the wild allele. Indeed, with few exceptions, domesticated alleles are associated with a lack of gene function (Gepts, 2002; Gepts and Papa, 2002).

Wild and domesticated forms often are found in sympatry throughout the distribution of the common bean (figure 6.2), from North Mexico to Argentina. Several examples of introgression have been documented, along with the occurrence of weedy populations that colonize highly disturbed areas, such as abandoned fields (Freyre et al., 1996; Beebe et al., 1997). Even if the autogamous breeding system is a limiting factor, the observed level of outcrossing (2–3%) (Ibarra-Pérez et al., 1997; Ferreira et al., 2000) suggests that, as found in other highly selfing species (Ellstrand et al., 1999), gene flow is likely to limit the independent evolution of wild and domesticated populations. A significant level of gene flow between wild and domesticated *P. vulgaris* has recently been observed in Puebla, Mexico, using inter–simple sequence repeats (ISSRs) (González et al., 2005), and in Michoacán and Guanajuato, Mexico, using phenotypic markers and ISSRs (Payró de la Cruz et al., in press).

The introgression between the wild and the domesticated common bean (*P. vulgaris* L.) in Mesoamerica has also been studied using genetically mapped AFLP markers (Papa and Gepts, 2003; Papa et al., in press). AFLPs have been positioned on a molecular linkage map (Freyre et al., 1998) where several genes and quantitative trait loci have been located, including those responsible for the genetic control of the domestication

syndrome (Koinage et al., 1996). Diversity for the same markers was thus analyzed in two samples of wild and domesticated populations from Mexico. Gene flow occurred principally in close-range sympatry, that is, when two populations grew in close proximity (figure 6.2). Through both phenetic and admixture population analyses, introgression was found to be about three to four times higher from domesticated to wild populations than in the reverse direction (Papa and Gepts, 2003). Mapping of AFLP markers has also shown that differentiation between wild and domesticated populations is highest near the genes for domestication and is lower farther from these genes. Concurrently, the genetic bottleneck induced by domestication was strongest around these genes. Therefore selection may be a major evolutionary factor in the maintenance of the identities of wild and domesticated populations in sympatric situations. Furthermore, domesticated alleles appear to have displaced wild alleles in sympatric wild populations, thus leading to a reduction in genetic diversity in such populations (Papa et al., in press).

Evolution of Disease Resistance

The common bean is one of the few plant species for which population genetics and molecular genetics have both been used to study the evolution of resistance and the defense against parasites at both the ecological and molecular levels (de Meaux and Mitchell-Olds, 2003; Seo et al., 2004).

At the phenotypic level, genetic variation for resistance against parasites has been reported between and within *Phaseolus vulgaris* gene pools. The two cultivated common bean gene pools are differentiated by their resistance to the fungi responsible for anthracnose, *Colletotrichum lindemuthianum* (Sicard et al., 1997a, 1997b); for rust, *Uromyces appendiculatus* (Steadman et al., 1995); and for angular leaf spot, *Phaeoisariopsis griseola* (Guzman et al., 1995). In each of these interactions, the plants of one cultivated gene pool were more resistant to the fungus coming from the other gene pool than to the fungus isolated from the same gene pool. Similar results were obtained in natural populations where different sets of resistance genes against *C. lindemuthianum* were found in the three gene pools (Geffroy et al., 1999). Natural populations of the three gene pools maintained resistance genes that were overcome by local fungi but remained useful against possible invaders (Geffroy et al., 1999). Within centers of diversity, natural populations of *P. vulgaris* were differentiated for resistance to the fungus

C. lindemuthianum in both Mexico and Argentina (Cattan-Toupance et al., 1998; Sicard, unpublished data). In Mexico, natural populations of *P. vulgaris* were maladapted to the fungus *C. lindemuthianum* and had a greater resistance to allopatric strains than to local strains (Sicard, unpublished data).

The effects of parasite selection pressure on the molecular diversity of *P. vulgaris* have been studied by comparing the diversity between phenotypic resistance, neutral markers, and molecular markers located on both resistance candidate and defense-related genes. For resistance genes, restriction fragment length polymorphism (RFLP) markers located in a nucleotide-binding site (NBS) and AFLPs located on a leucine-rich repeat (LRR) domain of two families of resistance genes have been developed (Neema et al., 2001; de Meaux and Neema, 2003). For defense-related genes, three microsatellites located in genes encoding pathogenesis-related protein and located in different linkage groups have been used (Yu et al., 2000; Sicard and Papa, unpublished data). Population structures (i.e., the population differentiation) at the gene pool level and on the regional scale were conserved for all three: the phenotypic resistance markers, the resistance or defense gene-tagged markers, and the neutral markers. This suggests that the history of the common bean and its lifecycle (autogamous, low seed migration) influences molecular polymorphism at both neutral and defense or resistance loci (Neema et al., 2001; de Meaux et al., 2003; de Meaux and Neema, 2003). The levels of population differentiation and the levels of within-population diversity differed between the neutral and resistance gene-tagged markers. Plants of the Mesoamerican and Andean centers of diversity were shown to be more differentiated for RAPD markers than for NBS-tagged RFLP markers, which suggests a homogenizing effect of selection on the NBS region of two resistance gene candidate families, as was also found from DNA sequence data (Neema et al., 2001; Ferrier-Cana et al., 2003). In Mexico, a comparison of neutral markers and markers tagged on the LRR domain of one resistance gene family revealed that the average level of diversity within populations was higher for resistance gene candidate–tagged markers than for RAPD markers, suggesting diversifying selection or higher mutation rates in the LRR region of these resistance loci. This is consistent with the hypothesis that the LRR domains of resistance proteins form a versatile binding domain that is involved in parasite recognition (de Meaux and Neema, 2003).

Altogether, these data show that population history, population dynamics, and parasite selection pressure are all shaping the phenotypic and molecular polymorphism at resistance genes.

Introduction into the Old World

After Columbus's voyage in 1492, intense biological exchanges occurred between the Old World and the New World. Several crops were introduced, mainly into the Iberian Peninsula, from which they spread into the rest of Europe and around the world (Simmonds, 1976). The common bean probably arrived in Spain and Portugal from Central America in 1506 (Ortwin-Sauer, 1966). In 1528, Pizarro explored Peru, and the introduction of accessions from the Andes probably started after 1532 (Brucher and Brucher, 1976). The first description of the common bean in a European herbal was by Fuchs (1543) in Germany, around which time it also started its expansion into the Mediterranean area. Birri and Coco (2000) report on the contents of a manuscript published by Pierio Valeriano Bolsanio in 1550 (Biblioteca Vaticana Codice Latino 5215 C 8–9) that described his travels in 1532, from Rome to Belluno (northeast Italy); a bag of beans was received from the pope, Giuliano de Medici (Pope Clemente VII, 1523–1534), with the specific objective of its introduction as a crop plant. As Gepts (2002) notes, the bronze portals of the cathedral of Pisa, which have been dated to 1595, include realistic representations of the common bean. This all suggests that *P. vulgaris* was well known in Italy by the end of the 16th century. *P. vulgaris* probably arrived in Turkey and Iran at the beginning of the 1600s. In the 17th and 18th centuries, the Arabs introduced the common bean into East Africa, and in 1669 it was being cultivated on a large scale in the Netherlands (Van der Groen, 1669). Overall, this demonstrates that the pathways of dissemination of beans into Europe were very complex, with several introductions from the New World combined with direct exchanges between European and other Mediterranean countries.

In recent years, molecular markers have contributed to our understanding of the origins and dissemination pathways of *P. vulgaris* from its areas of domestication into Europe. The phaseolins have been used to characterize a European collection of *P. vulgaris* that was mainly from Portugal, Spain, France, and the Netherlands. This revealed that the European common bean arose from the introduction of domesticated beans from both of the American gene pools, with a higher frequency of Andean phaseolin types (76%; T, C, and H types) than of the Mesoamerican types (24%; S and

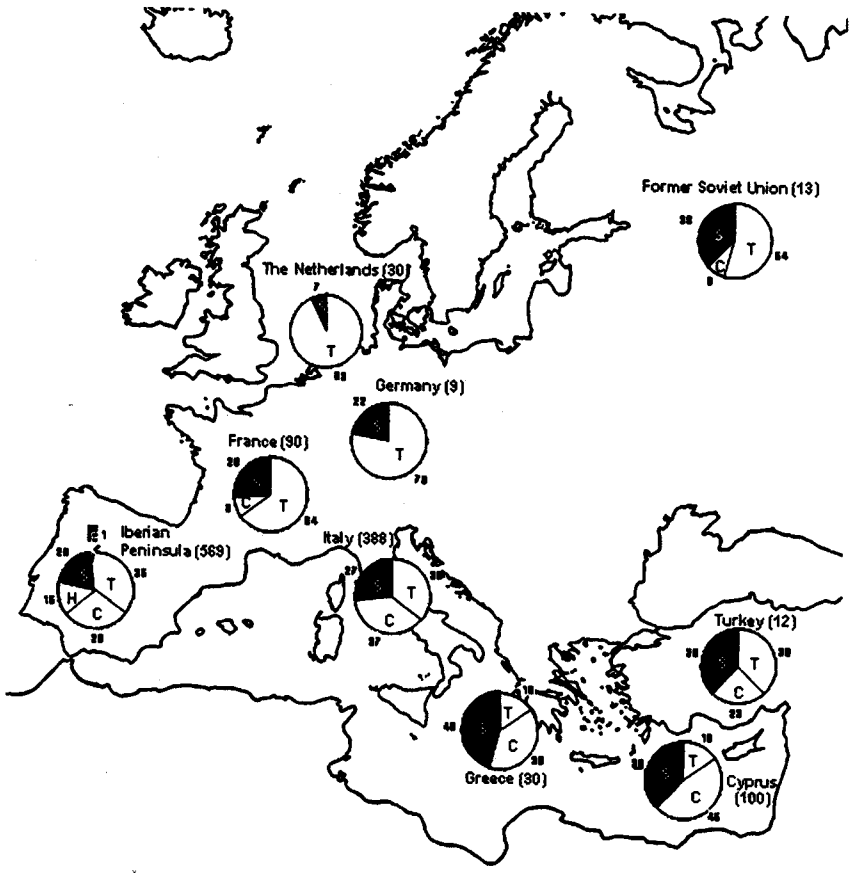

FIGURE 6.3 Distribution of phaseolin types across Europe (%). *White background:* Andean phaseolin types (T, C, and H). *Black background:* Mesoamerican phaseolin types (S and B). The sample sizes are given in parentheses after the country names. For the Iberian Peninsula, the data were obtained as weighted means of the results of the experiments of Gepts and Bliss (1988), Lioi (1989), Ocampo et al. (2002), and Rodiño et al. (2003). The data for France and the Netherlands are from Gepts and Bliss (1988). The data for Germany, Italy, Greece, Cyprus, Turkey, and the former Soviet Union are from Lioi (1989). When pooled samples were used, the calculations did not take into account the possible redundancy between different collections.

B types) (Gepts and Bliss, 1988). This was confirmed by Lioi (1989) in an analysis of a large collection of accessions that were mainly from Italy, Greece, and Cyprus (66% Andean types) and by Masi and Spagnoletti (unpublished data), who analyzed 544 accessions collected throughout Europe (76% Andean types). Despite a large variance in sample sizes and sampling strategies within and between these studies, at the single-country

level along the Mediterranean Arch (from the Iberian Peninsula to Turkey, throughout France, Italy, Greece, and Cyprus) a prevalence of the Andean phaseolin type has always been observed, with a minimum of 54% for Greece (Gepts and Bliss, 1988; Lioi, 1989; Rodiño et al., 2001, 2003; Ocampo et al., 2002) (figure 6.3). The lack of information for the countries of Central Europe should be noted. When regions within a country are considered, this prevalence of the Andean gene pool is also confirmed for studies in Galicia, Spain (Escribano et al., 1998), Abruzzo in central Italy (Piergiovanni et al., 2000a), Basilicata in southern Italy (Limongelli et al., 1996; Piergiovanni et al., 2000b), and the Marche region in central Italy (using ISSRs and nuclear and chloroplast SSRs; Sicard et al., in press). Thus, the overall frequencies of the Mesoamerican and Andean gene pools appear to be very similar on the continental, country, and regional scales, suggesting large seed exchanges between the European countries.

Differences in the frequencies of each Andean phaseolin type have also been discussed. Gepts and Bliss (1988) showed that in the Iberian Peninsula, phaseolin C was the most common. The prevalence of the C type within Portuguese and Spanish landraces was also observed by Rodiño et al. (2001) and Ocampo et al. (2002). In contrast, Escribano et al. (1998) analyzed landraces from Galicia, Spain, and observed that type T was the most common. This was also seen with a collection of 388 accessions from the Iberian Peninsula (Rodiño et al., 2003). Overall, five phaseolins have been observed in the Iberian Peninsula, including type H (15%) and type B (1%). This may suggest a higher diversity for phaseolin types in this area than in the rest of Europe, although this greater phaseolin variability in the Iberian Peninsula may just be related to the greater number of samples analyzed or differences in the sampling strategies between the studies (figure 6.3).

On a smaller geographic scale, a study conducted in the Abruzzo region of central Italy showed a prevalence of type C (Piergiovanni et al., 2000a), as has also been seen in the Basilicata region in southern Italy (Limongelli et al., 1996; Piergiovanni et al., 2000a). Interestingly, the Hellenic Peninsula has the highest frequency of phaseolin S (46%), a strictly Mesoamerican type; the frequency of phaseolin S, when compared with that of the rest of Europe, is also high (38%) in Cyprus and Turkey (figure 6.3; Lioi, 1989). Therefore, the overall data indicate that in the eastern Mediterranean area there is a high frequency of type S. Finally it should be noted that in France and the Netherlands, type T appears at a very high frequency (Gepts and Bliss, 1988), as in Germany and in the former Soviet Union (Lioi, 1989). It has also been suggested that as well

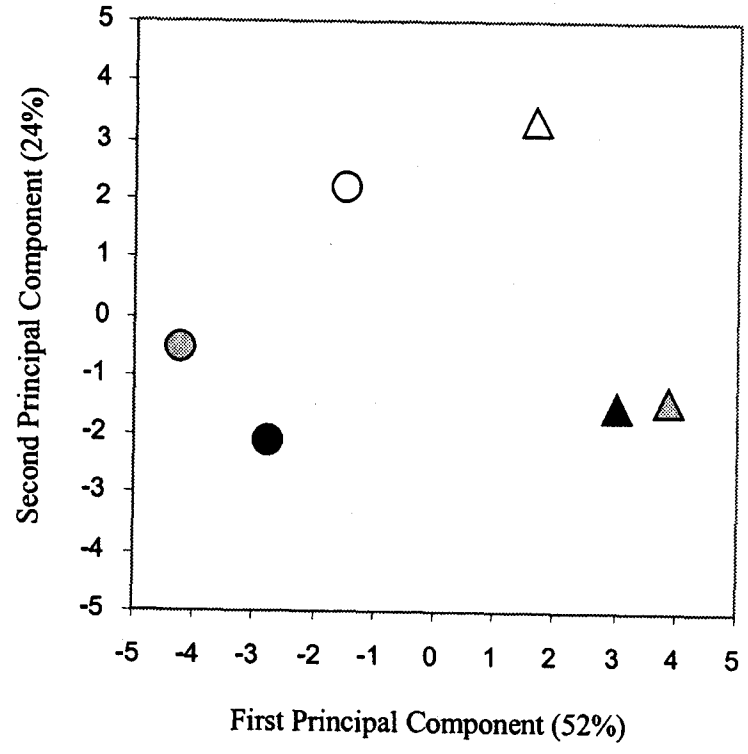

FIGURE 6.4 Relationships between the wild American (*black*), domesticated American (*gray*), and domesticated Iberian (*white*) germplasm of the Mesoamerican (*triangles*) and Andean (*circles*) gene pools. The graph summarizes the differences in isozyme allele frequencies at the eight loci that are common among the studies of Koenig and Gepts (1989), Singh et al. (1991), and Santalla et al. (2002) (Diap-1, Diap-2, Me, Mdh-1, Mdh-2, Prx, Rbcs, and Skdh) and was obtained using JMP 3.1.5 software (SAS Institute, Inc., 1995). For the wild Mesoamerican gene pool, the weighted averages of the Mexican and Central American frequencies (Koenig and Gepts, 1989, table 3) were calculated, but for the wild Andean, only the frequencies for Argentina were considered.

as migration and selection, the phaseolin geographic distribution may be affected by the differential distribution of phaseolin patterns among consumption categories (e.g., dry beans vs. green pod cultivars) (Brown et al., 1982; Gepts and Bliss, 1988). Several studies have shown the occurrence in Europe of markers pertaining to both Andean and Mesoamerican gene pools within the same bean landrace (Piergiovanni et al., 2000a, 2000b;

Rodiño et al., 2001), and molecular evidence of hybridization between gene pools has been obtained by analyzing germplasm from the Marche region in central Italy (Sicard et al., in press). Recently, using isozymes, introgression between the Mesoamerican and the Andean gene pools was observed in the Iberian Peninsula, and two groups of intermediate or putative recombinants (25% of the accessions) between the two gene pools were found (Santalla et al., 2002).

It has been suggested that crop expansion from America to Europe resulted in a reduction in the diversity of the European common bean because of strong founder effects, adaptation to a new environment, and consumer preferences (Gepts, 1999). Isozyme loci have been used to characterize domesticated common beans both from the Americas (Singh et al., 1991) and from the Iberian Peninsula (Santalla et al., 2002). Recalculation of the diversity values using the eight isozyme loci in common between these two studies reveals that the Iberian Peninsula diversity (H_T = 0.25) is about 30% lower than that of the Americas (H_T = 0.37). The difference in diversity (H) of the two gene pools was larger in the Americas (Mesoamerican = 0.23; Andean = 0.16) than in the Iberian Peninsula (Mesoamerican origin = 0.20; Andean origin = 0.21), which results in a much stronger genetic difference between the two gene pools in the Americas (G_{ST} = 0.47) than in the Iberian Peninsula (G_{ST} = 0.18). This has also been shown using principal component analysis (PCA) of the allelic frequencies (figure 6.4), where wild germplasm was also used as the reference (Koenig and Gepts, 1989). Of note, within gene pools, domesticated American germplasm is closer to the wild germplasm than to the domesticated germplasm from the Iberian Peninsula (figure 6.4). This lower differentiation in Europe can be explained by the combined actions of greater gene flow between different gene pools caused by the lack of geographic barriers and convergent evolution.

Overall, the data suggest that the structure of genetic diversity of common bean in Europe has been highly influenced by hybridization between the two gene pools together with homogeneous selection for adaptation to the European environments. For example, this is likely to have been the case for photoperiod insensitivity. In addition, the bottleneck effect of the introduction of the common bean into Europe might not have been as strong as was previously suspected (Gepts, 1999), and it appears that hybridization between the two gene pools of *P. vulgaris* has had a significant impact on the maintenance of the overall level of genotypic diversity. Second, heterogeneous selection for different uses and local adaptation to a wide range of environments and agronomic practices in Europe might also have counteracted the effects of drift and homogeneous selection for adaptation to European environmental conditions. Third, the founding populations might have been highly representative of the diversity present in the American gene pools. This could be because there were several different introductions from the Americas or because the attractiveness of various types of seed color and shape probably has favored the capture of different alleles and genotypes. Extensive studies on the genetic diversity of the European bean populations are still needed to test these hypotheses.

These data suggest that the expansion of *P. vulgaris* into Europe and introgression between different gene pools (probably because of the lack of geographic barriers) have had a significant impact on the shaping of the genetic diversity of this species. However, because evidence of germplasm exchange between Mesoamerica and the Andes has been documented (Gepts, 1988a), a strict relationship between the gene pools and the areas from which the common bean was introduced into Europe cannot be assumed; similarly, hybrids between gene pools could also have originated in the Americas and the progeny later introduced into the Old World. To obtain a comprehensive picture of the origins, levels, and structures of the common bean diversity in Europe, representative samples from different European and Mediterranean countries should be compared with an appropriate large sample from the Americas using different types of molecular markers.

Conclusions

We have shown how the advent of molecular techniques has greatly improved our ability to understand the complex evolutionary history of the common bean and how various evolutionary forces have contributed to the structure of its genetic diversity in the New World and, more recently, in the Old World. New molecular tools have been developed recently for the bean, and others are likely to become available in the near future (Broughton et al., 2003), which will expand our capacity for investigation. For instance, along with nuclear markers, the development in the bean of SSRs and sequence-tagged sites (STSs) specific for chloroplast DNA (Sicard et al., in press) and mitochondrial DNA (Arrieta-Montiel et al., 2001) could be of particular interest in tracking the migration pathways. Indeed, migration would be better studied using molecular markers that differ in their inheritance patterns (uniparental

vs. biparental; Provan et al., 2001). Moreover, we have shown how a combination of molecular maps and gene-tagging markers and neutral markers can distinguish the evolutionary role played by selection from that caused by drift and migration.

The relative roles of evolutionary forces should be resolved if it is possible to compare the information from the gene-tagging and neutral markers. As was first pointed out by Cavalli-Sforza (1966), whereas migration and drift affect loci similarly across the entire genome, selection affects only specific loci because of recombination. Today, readily available sequence information and genetic and physical maps open new perspectives for the possibility of tracking the signatures of evolutionary forces along the genome, even if several methodological problems remain to be resolved. The use of molecular markers tagging specific gene family domains, such as those that are AFLP derived and that have been developed to study wild bean populations (Neema et al., 2001), would also be particularly interesting, and they could also be developed for other gene families (van Tienderen et al., 2002). Similarly, SSRs and single nucleotide polymorphisms located in genic regions (Yu et al., 2000; Gaitán-Solís et al., 2002; McClean et al., 2002; Blair et al., 2003; Guerra-Sanz, 2004) and STSs linked to genes of interest (Murray et al., 2002; McClean et al., 2002; Erdmann et al., 2002) would be of particular interest when used in combination with putative neutral markers such as SSRs developed from genomic libraries (Gaitán-Solís et al., 2002). The development of gene-tagging markers for *Phaseolus* will also increase with the growing expressed sequence tag (EST) sequencing efforts (Broughton et al., 2003). These opportunities should be enhanced by the location of molecular markers and sequence data within genetic (Kelly et al., 2003; Broughton et al., 2003) and physical (Vanhouten and MacKenzie, 1999; Kami and Gepts, 2000; Melotto et al., 2004) maps.

As long as we are able to interpret the increasing amounts of data that are being generated, the development of genomics studies should allow not just the development of new research tools but also an improved understanding of the genome organization and structure and its evolution.

Acknowledgments

This work was partially supported by the Italian Government, Project MURST-Cofin2002, "Beans in Europe," # 2002075423.

References

Arrieta-Montiel, M., A. Lyznik, M. Woloszynska, H. Janska, J. Tohme, and S. Mackenzie. 2001. Tracing evolutionary and developmental implications of mitochondrial stoichiometric shifting in the common bean. *Genetics* 158(6): 851–64.

Becerra, V.L., and P. Gepts. 1994. RFLP diversity of common bean (*Phaseolus vulgaris*) in its centres of origin. *Genome* 37: 256–263.

Beebe, S., J. Rengifo, E. Gaitan, M.C. Duque, and J. Tohme. 2001. Diversity and origin of Andean landraces of common bean. *Crop Science* 41: 854–862.

Beebe, S., P.W. Skroch, J. Tohme, M.C. Duque, F. Pedraza, and J. Nienhuis. 2000. Structure of genetic diversity between common bean land races of Middle American origin based on correspondence analysis of RAPD. *Crop Science* 40: 264–273.

Beebe, S., O. Toro, A.V. González, M.I. Chacon, and D.G. Debouck. 1997. Wild-weed–crop complexes of common bean (*Phaseolus vulgaris* L, Fabaceae) in the Andes of Peru and Colombia, and their implications for conservation and breeding. *Genetic Resources and Crop Evolution* 44: 73–91.

Birri, F. and C. Coco. 2000. *Cade a Fagiolo. Dal mondo antico alla nostra tavola. Storia e miti della carne dei poveri.* Marsilio, Venice, IT.

Blair, M.W., F. Pedraza, H.F. Buendia, E. Gaitán-Solís, S.E. Beebe, P. Gepts, and J. Thome. 2003. Development of a genome-wide anchored microsatellite map of common bean (*Phaseolus vulgaris* L.). *Theoretical and Applied Genetics* 107: 1362–1374.

Broughton, W.J., G. Hernandez, M. Blair, S. Beebe, P. Geptsand, and J. Vanderleyden. 2003. Beans (*Phaseolus* spp.): Model food legumes. *Plant and Soil* 252: 55–128.

Brown, J.W.S., F.A. Bliss, and T.C. Hall. 1982. Genetic variation in the subunits of globulin-1 storage protein of French bean. *Theoretical and Applied Genetics* 59: 83–88.

Brucher, B. and H. Brucher. 1976. The South American wild bean (*Phaseolus aborigenus* Burk.) as an ancestor of the common bean. *Economic Botany* 30: 257–272.

Caicedo, A.L., E. Gaitan, M.C. Duque, O. Toro Chica, D.G. Debouck, and J. Tohme. 2000. AFLP fingerprinting of *Phaseolus lunatus* L. and related wild species from South America. *Crop Science* 39: 1497–1507.

Cattan-Toupance, I., Y. Michalakis, and C. Neema. 1998. Genetic structure of wild bean populations in their South-Andean centre of diversity. *Theoretical and Applied Genetics* 96: 844–851.

Cavalli-Sforza, L.L. 1966. Population structure and human evolution. *Proceedings of the Royal Society, London* Ser. B 164: 362–379.

Clark, R.M., E. Linton, J. Messing, and J.F. Doebley. 2003. Pattern of diversity in the genomic region near the maize domestication gene *tb1*. *Proceedings of the National Academy of Sciences (USA)* 101: 700–707.

Debouck, D.G. 1991. Systematics and morphology. In A. van Schoonhoven and O. Voysest (eds.), *Common Beans, Research for Crop Improvement,* 55–118. CAB International, Oxon, UK.

Debouck, D.G. 1999. Biodiversity, ecology and genetic resources of *Phaseolus* beans: Seven answered and unanswered questions. In *Wild Legumes.* National Institute of Agrobiological Resources Tsukuba, Ibaraki, Japan.

Debouck, D.G., J.H. Linan Jara, S.A. Campana, and R.J.H. de la Cruz. 1987. Observations on the domestication of *Phaseolus lunatus* L. *FAO/IBPGR Plant Genetic Resources Newsletters* 70: 26–32.

Delgado-Salinas, A. 1985. Systematics of the genus *Phaseolus* (Leguminosae) in North and Central America. Ph.D. dissertation, University of Texas, Austin, TX, USA.

Delgado-Salinas, A., A. Bruneau, and J. J. Doyle. 1993. Chloroplast DNA phylogenetic studies in New World Phaseolinae (Leguminosae: Papilionoideae: Phaseoleae). *Systematic Botany* 18: 1, 6–17.

Delgado-Salinas, A., T. Turley, A. Richman, and M. Lavin. 1999. Phylogenetic analysis of the cultivated and wild species of *Phaseolus* (Fabaceae). *Systematic Botany* 24: 438–460.

de Meaux, J., I. Cattan-Toupance, C. Lavigne, T. Langin, and C. Neema. 2003. Polymorphism of a complex resistance gene candidate family in wild populations of common bean (*Phaseolus vulgaris*) in Argentina: Comparison with phenotypic resistance polymorphism. *Journal of Molecular Evolution* 12: 263–273.

de Meaux, J. and T. Mitchell-Olds. 2003. Evolution of plant resistance at the molecular level: Ecological context of species interactions. *Heredity* 91: 345–352.

de Meaux, J. and C. Neema. 2003. Spatial patterns of the putative recognition domain of resistance gene candidates in wild bean populations. *Journal of Molecular Evolution* 57: S90–S102.

Doebley, J., A. Stec, J. Wendel, and M. Edwards. 1990. Genetic and morphological analysis of a maize–teosinte F_2 population: Implications for the origin of maize. *Proceedings of the National Academy of Sciences (USA)* 87: 9888–9892.

Ellstrand, N., H. Prentice, and J. Hancock. 1999. Gene flow and introgression from domesticated plants into their wild relatives. *Annual Review of Ecology and Systematics* 30: 539–563.

Erdmann, P. M., R. K. Lee, M. J. Bassett, and P. E. McClean. 2002. A molecular marker tightly linked to P, a gene required for flower and seedcoat color in common bean (*Phaseolus vulgaris* L.), contains the *Tyj*–gypsy retrotransposon *Tpv3g*. *Genome* 45: 728–736.

Escribano, M. R., M. Santalla, P. A. Casquero, and A. M. De Ron. 1998. Patterns of genetic diversity in landraces of common bean (*Phaseolus vulgaris* L.) from Galicia. *Plant Breeding* 117: 49–56.

Evans, A. M. 1973. Plant architecture and physiological efficiency in the field bean. In D. Wall (ed.), *Potentials of Field Bean and Other Food Legumes in Latin America,* 279–284. CIAT, Cali, Colombia.

Evans, A. M. 1980. Structure, variation, evolution and classification in *Phaseolus*. In R. J. Summerfield and A. H. Bunting (eds.), *Advances in Legume Science,* 337–347. Royal Botanic Gardens, Kew, UK.

Ferreira, J. J., E. Alvarez, M. A. Fueyo, A. Roca, and R. Giraldez. 2000. Determination of the outcrossing rate of *Phaseolus vulgaris* L. using seed protein markers. *Euphytica* 113: 259–263.

Ferrier-Cana, E., V. Geffroy, C. Macadre, F. Creusot, P. Imbert-Bollore, M. Sevignac, and T. Langin. 2003. Characterization of expressed NBS–LRR resistance gene candidates from common bean. *Theoretical and Applied Genetics* 106: 251–261.

Fofana, B., J. P. Baudoin, X. Vekemans, D. G. Debouck, and P. du Jardin. 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.

Free, J. B. 1966. The pollination of the beans *Phaseolus multiflorus* and *Phaseolus vulgaris* by honeybees. *Journal of Apicultural Research* 5: 87–91.

Freyre, R., R. Ríos, L. Guzmán, D. Debouck, and P. Gepts. 1996. Ecogeographic distribution of *Phaseolus* spp. (Fabaceae) in Bolivia. *Economic Botany* 50: 195–215.

Freyre, R., P. Skeoch, V. Geffroy, A.-F. Adam-Blondon, A. Shirmohamadali, W. Johnson, V. Llaca, R. Nodari, P. Pereira, S.-M. Tsai, J. Tohme, M. Dron, J. Nienhuis, C. Vallejos, and P. Gepts. 1998. Towards an integrated linkage map of common bean. 4. Development of a core map and alignment of RFLP maps. *Theoretical and Applied Genetics* 97: 847–856.

Fuchs, L. 1543. *Von Welschen Bonen. Cap.* CCLXIX *in Neu Kreuternuch.* Michal Isengrin, Basel. Reprinted in 1964. K. Kölb, Munich, Germany.

Gaitán-Solís, E., M. C. Duque, K. J. Edwards, and J. Thome. 2002. Microsatellite repeats in common bean (*Phaseolus vulgaris*): Isolation, characterization, and cross-species amplification in *Phaseolus* ssp. *Crop Science* 97: 847–856.

Geffroy, V., D. Sicard, J. C. F. De Oliveira, M. Sevignac, S. Cohen, P. Gepts, C. Neema, T. Langin, and M. Dron. 1999. Identification of an ancestral resistance gene cluster involved in the coevolution process between *P. vulgaris* and its fungal pathogen *Colletotrichum lindemuthianum*. *Molecular Plant–Microbe Interactions* 12: 774–784.

Gepts, P. 1988a. A Middle American and Andean gene pool. In P. Gepts (ed.), *Genetic Resources of* Phaseolus *Beans,* 375–390. Kluwer Academic Publishers, Dordrecht, The Netherlands.

Gepts, P. 1988b. Phaseolin as an evolutionary marker. In P. Gepts (ed.), *Genetic Resources of* Phaseolus *Beans,* 215–241. Kluwer Academic Publishers, Dordrecht, The Netherlands.

Gepts, P. 1996. Origin and evolution of cultivated *Phaseolus* species. In B. Pickersgill and J. M. Lock (eds.), *Advances in Legume Systematics 8: Legumes of Economic Importance,* 65–74. Royal Botanic Gardens, Kew, UK.

Gepts, P. 1999. Development of an integrated genetic linkage map in common bean (*Phaseolus vulgaris* L.) and its use. In S. Singh (ed.), *Bean Breeding for the 21st Century,* 53–91, 389–400. Kluwer, Dordrecht, The Netherlands.

Gepts, P. 2002. A comparison between crop domestication, classical plant breeding, and genetic engineering. *Crop Science* 42: 1780–1790.

Gepts, P. and F. A. Bliss. 1988. Dissemination pathways of common bean (*Phaseolus vulgaris,* Fabaceae) deduced from phaseolin electrophoretic variability. II. Europe and Africa. *Economic Botany* 42: 86–104.

Gepts, P., T. C. Osborne, K. Rashka, and F. A. Bliss. 1986. Electrophoretic analysis of phaseolin protein variability in wild forms and landraces of the common bean, *Phaseolus vulgaris* L.: Evidence for two centers of domestications. *Economic Botany* 40: 451–468.

Gepts, P. and R. Papa. 2002. Evolution during domestication. In *Encyclopedia of Life Science.* Macmillan, Nature Publishing Group, London.

González, A., A. Wong, A. Delgado-Salinas, R. Papa, and P. Gepts. 2005. Assessment of inter simple sequence repeat markers to differentiate sympatric wild and domesticated populations of common bean (*Phaseolus vulgaris* L.). *Crop Science* 45: 606–615.

Guerra-Sanz, J. M. 2004. New SSR markers of *Phaseolus vulgaris* from sequence databases. *Plant Breeding* 123: 87–89.

Guzman, P., R. L. Gilbertson, R. Nodori, W. C. Johnson, S. R. Temple, D. Mandala, A. B. C. Mkandawire, and P. Gepts. 1995. Characterization of variability in the fungus *Phaeoisariopsis griseola* suggests coevolution with the common bean (*Phaseolus vulgaris* L.). *Phytopathology* 85: 600–607.

Hamann, A., D. Zink, and W. Nagl. 1995. Microsatellite fingerprinting in the genus *Phaseolus*. *Genome* 38: 507–515.

Hammer, K. 1984. Das Domestikations syndrome. *Kulturpflanze* 32: 11–34.

Harlan, J. R. and J. M. J. de Wet. 1971. Towards a rational classification of cultivated plants. *Taxon* 20: 509–517.

Hernandez-Xolocotzi, E., S. Miranda Colin, and C. Prwyer. 1959. El origen de *Phaseolus coccineus* L. *darwinianus* Hernandez X. & Miranda C. subspecies nova. *Revista de la Sociedad Mexicana de Historia Natural* 20: 99–121.

Ibarra-Pérez, F., B. Ehadaie, and G. Waines. 1997. Estimation of outcrossing rate in common bean. *Crop Science* 37: 60–65.

Jaaska, V. 1996. Isoenzyme diversity and phylogenetic affinities among the *Phaseolus* beans (Fabaceae). *Plant Systematics and Evolution* 200: 3–4, 233–252.

Kami, J., V. Becerra-Velasquez, D. G. Debouck, and P. Gepts. 1995. Identification of presumed ancestral DNA sequences of phaseolin *Phaseolus vulgaris. Proceedings of the National Academy of Sciences (USA)* 92: 1101–1104.

Kami, J. and P. Gepts. 2000. Development of a BAC library in common bean genotype BAT93. *Annual Report Bean Improvement Cooperative* 43: 208–209.

Kaplan, N., R. Hudson, and C. Langley. 1989. The "hitchhiking" effect revisited. *Genetics* 123: 887–899.

Kelly, J. D., P. Gepts, P. N. Miklas, and D. P. Coyne. 2003. Tagging and mapping of genes and QTL and molecular marker–assisted selection for traits of economic importance in bean and cowpea. *Field Crops Research* 82: 135–154.

Khairallah, M. M., B. B. Sears, and M. W. Adams. 1992. Mitochondrial restriction fragment length polymorphisms in wild *Phaseolus vulgaris* L.: Insights on the domestication of the common bean. *Theoretical and Applied Genetics* 84: 915–922.

Kloz, J. 1971. Serology of the Leguminosae. In J. P. Harborne, D. Boulter, and B. L. Turner (eds.), *Chemotaxonomy of the Leguminosae,* 309–365. Academic Press, London, UK.

Koenig, R. and P. Gepts. 1989. Allozyme diversity in wild *Phaseolus vulgaris:* Further evidence for two major centers of genetic diversity. *Theoretical and Applied Genetics* 78: 809–817.

Koenig, R., S. P. Singh, and P. Gepts. 1990. Novel phaseolin types in wild and cultivated common bean (*P. vulgaris,* Fabaceae). *Economic Botany* 44: 50–60.

Koinange, E. M. K., S. P. Singh, and P. Gepts. 1996. Genetic control of the domestication syndrome in common bean. *Crop Science* 36: 1037–1045.

Lackey, J. A. 1981. Phaseolae. In R. M. Polhill and P. H. Raven (eds.), *Advances in Legume Systematics,* Part 2, 301–327. Royal Botanic Gardens, Kew, UK.

Lackey, J. A. 1983. A review of generic concepts in American Phaseolinae (Fabaceae, Faboideae). *Iselya* 2: 21–64.

Ladizinsky, G. 1998. *Plant Evolution and Origin of Crop Species.* Prentice Hall, Englewood Cliffs, NJ, USA.

Lioi, L. 1989. Geographical variation of phaseolin patterns in an old world collection of *Phaseolus vulgaris. Seed Science & Technology* 17: 317–324.

Llaca, V., S. A. Delgado, and P. Gepts. 1994. Chloroplast DNA as an evolutionary marker in the *Phaseolus vulgaris* complex. *Theoretical and Applied Genetics* 88: 646–652.

Maquet, A. and J. P. Baudoin. 1996. New lights on phyletic relations in the genus *Phaseolus. Annual Report Bean Improvement Cooperative* 39: 203–204.

Maréchal, R., J. Mascherpa, and F. Stanier. 1978. Etude taxonomique d'un groupe complexe d'espèces des genres *Phaseolus* et *Vigna* (Papilonaceae) sur la base de donées morphologiques, traitées par l'analyse informatique. *Boissiera* 28: 1–273.

Maynard Smith, J. and J. Haigh. 1974. The hitch-hiking effect of a favourable gene. *Genetical Research* 23: 23–35.

McClean, P. E., R. K. Lee, and P. N. Miklas. 2004. Sequence diversity analysis of dihydroflavonol 4-reductase intron 1 in common bean. *Genome* 47: 266–280.

McClean, P. E., R. Lee, C. Otto, P. Gepts, and M. J. Bassett. 2002. STS and RAPD mapping of genes controlling seed coat color and patterning in common bean. *Journal of Heredity* 93: 148–152.

Melotto, M., M. F. Coelho, A. Pedrosa-Harand, J. D. Kelly, and L. E. A. Camargo. 2004. The anthracnose resistance locus Co-4 of common bean is located on chromosome 3 and contains putative disease resistance–related genes. *Theoretical and Applied Genetics* 109: 690–699.

Murray, J., J. Larsen, T. E. Michaels, A. Schaafsma, C. E. Vallejos, and K. P. Pauls. 2002. Identification of putative genes in bean (*Phaseolus vulgaris*) genomic (Bng) RFLP clones and their conversion to STSs. *Genome* 45:1013–1024.

Neema, C., C. Lavignes, J. De Meaux, I. Cattan-Toupance, J. F. de Oliviera, A. Deville, and T. Langin. 2001. Spatial pattern for resistance to a pathogen. Theoretical approach and empirical approach at the phenotypic and molecular level. *Genetic Selection Evolution* 33: S3–S21.

Ocampo, C. H., J. P. Martin, J. M. Ortiz, M. D. Sanchez-Yelamo, O. Toro, and D. Debouck. 2002. Possible origins of common bean (*Phaseolus vulgaris* L.) cultivated in Spain in relation to the wild genetic pools of Americas. *Annual Report Bean Improvement Coop* 45: 236–237.

Ortwin-Sauer, C. 1966. *The Early Spanish Man.* University of California Press, Berkeley, CA, USA.

Papa, R., J. Acosta, A. Delgado-Salinas, and P. Gepts. In press. A genome-wide analysis of differentiation between wild and domesticated *Phaseolus vulgaris* from Mesoamerica. *Theoretical and Applied Genetics.*

Papa, R. and P. Gepts. 2003. Asymmetry of gene flow and differential geographical structure of molecular diversity in wild and domesticated common bean (*Phaseolus vulgaris* L.) from Mesoamerica. *Theoretical and Applied Genetics* 106: 239–250.

Payró de la Cruz, E., P. Gepts, P. Colunga Garcia-Marín, and D. Zizumbo Villareal. In press. Spatial distribution of genetic diversity in wild populations of *Phaseolus vulgaris* L. from Guanajuato and Michoacán, México. *Genetic Resources and Crop Evolution.*

Piergiovanni, A. R., D. Cerbino, and M. Brandi. 2000a. The common bean populations from Basilicata (southern Italy). An evaluation of their variation. *Genetic Resources & Crop Evolution* 489–495.

Piergiovanni, A. R., G. Taranto, and D. Pignone. 2000b. Diversity among common bean populations from the Abruzzo region (central Italy): A preliminary inquiry. *Genetic Resources & Crop Evolution* 47: 467–470.

Piñero, D. and L. Eguiarte. 1988. The origin and biosystematic status of *Phaseolus coccineus* subsp. *polyanthus:* Electrophoretic evidence. *Euphytica* 37: 199–203.

Provan, J., W. Powell, and P. M. Hollingsworth. 2001. Chloroplast microsatellites: New tools for studies in plant ecology and evolution. *Trends in Ecology & Evolution* 16: 142–147.

Pueyo, J. J. and A. Delgado-Salinas. 1997. Presence of α-amylase inhibitor in some members of the subtribe Phaseolinae (Phaseolae: Fabaceae). *American Journal of Botany* 84: 79–84.

Rodiño, A. P., M. Santalla, A. M. De Ron, and S. P. Singh. 2003. A core collection of common bean from the Iberian peninsula. *Euphytica* 131: 165–175.

Rodiño, A. P., M. Santalla, I. Montero, P. A. Casquero, and A. M. De Ron. 2001. Diversity in common bean (*Phaseolus vulgaris* L.) germplasm from Portugal. *Genetic Resources & Crop Evolution* 48: 409–417.

Santalla, M., A. P. Rodiño, and A. M. De Ron. 2002. Allozyme evidence supporting southwestern Europe as a secondary center of genetic diversity for common bean. *Theoretical and Applied Genetics* 104: 934–944.

Schmit, V., P. du Jardin, J. P. Baudoin, and D. G. Debouck. 1993. Use of chloroplast DNA polymorphism for the phylogenetic study of seven *Phaseolus* taxa including *P. vulgaris* and *P. coccineus. Theoretical and Applied Genetics* 87: 506–516.

Seo, Y. S., P. Gepts, and R. L. Gilbertson. 2004. Genetics of resistance to the geminivirus, bean dwarf mosaic virus, and the role of the hypersensitive response in common bean. *Theoretical and Applied Genetics* 108: 786–793.

Sicard, D., Y. Michalakis, M. Dron, and C. Neema. 1997a. Population structure of *C. lindemuthianum* in the three centres of diversity of the common bean. *Phytopathology* 87: 807–813.

Sicard, D., Y. Michalakis, and C. Neema. 1997b. Diversity of *Colletotrichum lindemuthianum* in wild common bean populations. *Plant Pathology* 46: 355–365.

Sicard, D., L. Nanni, O. Porfiri, D. Bulfon, and R. Papa. In press. Genetic diversity of *Phaseolus vulgaris* L. and *P. coccineus* L. landraces in central Italy. *Plant Breeding.*

Simmonds, N.W. 1976. *Evolution of Crop Plants.* Longman, London, UK.

Singh, S.P. 2001. Broadening the genetic base of common bean cultivars: A review. *Crop Science* 41: 1659–1675.

Singh, S.P., R. Nodari, and P. Gepts. 1991. Genetic diversity in cultivated common bean. I. Allozymes. *Crop Science* 31:19–23.

Snoeck, C., J. Vanderleyden, and S. Beebe. 2003. Strategies for genetic improvement of common bean and rhizobia towards efficient interactions. *Plant Breeding Reviews* 23: 21–72.

Sonnante, G., T. Stockton, R.O. Nodari, V.L. Becerra Velásquez, and P. Gepts. 1994. Evolution of genetic diversity during the domestication of common bean (*Phaseolus vulgaris* L.). *Theoretical and Applied Genetics* 89: 629–635.

Sousa, M. and A. Delgado-Salinas. 1993. Mexican Leguminosae: Phytogeography, endemism, and origin. In T.P. Ramamoorthy, R. Bye, A. Lot, and J. Fa (eds.), *Biological Diversity of Mexico: Origins and Distribution,* 459–511. Oxford University Press, Oxford, UK.

Steadman, J.R., J. Beaver, M. Boudreau, D. Coyne, J. Groth, J. Elly, M. McMillan, R. McMillan, P. Miklas, M. Pastor Corrales, H. Schwartz, and J. Stavely. 1995. Progress reported at the 2nd International Bean Rust Workshop. *Annual Report of Bean Improvement Cooperative* 38: 1–10.

Sullivan, J.G. and G. Freytag. 1986. Predicting interspecific compatibilities in beans (*Phaseolus*) by seed protein electrophoresis. *Euphytica* 35: 201–209.

Tenaillon, M.I., C.M. Sawkins, A.D. Long, R.L. Gaut, J.F. Doebley, and B.S. Gaut. 2001. Patterns of DNA sequence polymorphism along chromosome 1 of maize (*Zea mays* ssp. *mays* L.). *Proceedings of the National Academy of Science (USA)* 98: 9161–9166.

Van der Groen, J. 1669. *Den Nederlandtsen Hovenier.* Reprint 1988. Sichting Matrijs, Utrecht, The Netherlands.

Vanhouten, W. and S. MacKenzie. 1999. Construction and characterization of a common bean bacterial artificial chromosome library. *Plant Molecular Biology* 40: 977–983.

Van Tienderen, P.H., A.A. Van Haan, C.G. Van der Linden, and B. Vosman. 2002. Biodiversity assessment using markers for ecologically important traits. *Trends in Ecology & Evolution* 17: 577–582.

Vekemans, X., O. Hardy, B. Berken, B. Fofana, and J.P. Baudoin. 1998. Use of PCR-RFLP on chloroplast DNA to investigate phylogenetic relationships in the genus *Phaseolus. Biotechnology, Agronomy, Society and Environment* 2: 128–134.

Verdecourt, B. 1970. Studies in the Leguminosae–Papilionoideae for the flora of tropical East Africa: IV. *Kew Bulletin* 24: 507–569.

Wang, R.L., A. Stec, J. Hey, L. Lukens, and J. Doebley. 1999. The limits of selection during maize domestication. *Nature* 398: 236–239.

Wang, R.L., A. Stec, J. Hey, L. Lukens, and J. Doebley. 2001. Correction: The limits of selection during maize domestication. *Nature* 410: 718.

Yu, K., S.J. Park, V. Poysa, and P. Gepts. 2000. Integration of simple sequence repeat (SSR) markers into a molecular linkage map of common bean (*Phaseolus vulgaris* L.). *Journal of Heredity* 91: 429–434.

Mallikarjuna K. Aradhya,
Daniel Potter, and Charles J. Simon

CHAPTER 7

Cladistic Biogeography of *Juglans* (Juglandaceae) Based on Chloroplast DNA Intergenic Spacer Sequences

Juglans L. is principally a New World genus within the tribe Juglandeae of the family Juglandaceae, comprising about 21 extant deciduous tree species occurring from North and South America, the West Indies, and southeastern Europe to eastern Asia and Japan (Manning, 1978). It is one of the approximately 65 genera that are known to exhibit a disjunct distributional pattern between eastern Asia and eastern North America (Manchester, 1987; Wen, 1999; Qian, 2002; figure 7.1). Four sections are commonly recognized within *Juglans,* based mainly on fruit morphology, wood anatomy, and leaf architecture (Dode, 1909a, 1909b; Manning, 1978). Section *Rhysocaryon* (black walnuts), which is endemic to the New World, comprises five North American temperate taxa: *J. californica* S. Wats., *J. hindsii* (Jeps.) Rehder, *J. nigra* L., *J. major* (Torr. ex Sitgr.) Heller, and *J. microcarpa* Berl.; three Central American subtropical taxa: *J. mollis* Engelm., *J. olanchana* Stadl. & I.O. Williams, and *J. guatemalensis* Mann.; and two South American tropical taxa, *J. neotropica* Diels and *J. australis* Griesb, mainly occurring in the highlands. They typically bear nuts that are four-chambered with thick nutshells and septa. Section *Cardiocaryon* (Asian butternuts) contains four taxa: *J. hopeiensis* Hu, *J. ailantifolia* Carr., *J. mandshurica* Maxim., and *J. cathayensis* Dode, all native to East Asia, and section *Trachycaryon* consists of the only North American butternut taxon, *J. cinerea* L. Both Asian and