

Genetic diversity of Sicilian common bean germplasm

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INTRODUCTION

In Italy, common bean is the major cultivated legume, with a dry beans production of 11176 tons and over 6001 cultivated hectares (1). Many Sicilian bean landraces have become obsolete due to the spread of commercial varieties and are endangered. Italian farmers preserved traditional local landraces, which usually have local names, are well adapted to specific pedo-climatic conditions and are appreciated by consumers valuing their better taste and higher nutritional properties (2, 3, 4). These landraces are poorly known but represent a genetic heritage to be preserved and enhanced. The biobanks playing a major role in preserving genetic resources and needs extensive characterization, including also the genomic and proteomic profiling (5).

"Sicilian Plant Germplasm Repository" of STEBICEF Department of Palermo University (SPGR/ PA) and "Living Plants Germplasm Bank" of Nebrodi Park (ME-Italy) stored a germplasm collection of fifty-seven Sicilian common bean (*Phaseolus vulgaris* L.) landraces. Seed morphological characterization has allowed to identify 37 morphotypes (6), highlighting redundancy of some accessions, supporting the hypothesis that the same cultivar is named in multiple way by local farmers. The present study reports the genetic characterization of common bean landraces with the aim to define the core collection and to develop a comprehensive strategy for the conservation of this crop genetic resource.

MATERIALS AND METHODS

57 cultivars of *Phaseolus vulgaris* L. (common bean), were collected in different areas of Sicily (Fig.1), where are local spread and often unknown at commercial level (Tab. 1). The *ex situ* field conservation was carried out in "Living Plants Germplasm Bank" of Ucria (ME-Italy), founded by the Nebrodi Regional Park, and in SPGR/PA.

Leaf materials of five plants/accession were lyophilized and used for DNA extraction (NucleoSpin Plant II Macherey Nagel). Eight SSR loci (BM159, BM160, BM172, BM210, GATS91, PV-ag001, PVBR25 and PVBR163), used in Italian common bean genetic studies, were selected based on their Polymorphic Information Content (PIC) values and dispersed genetic map locations (7-8). Standard check genotypes BAT93 (Mesoamerican gene pool) and JALOP558 and MIDAS (Andine gene pool) were included in this study. Data SSR fragment analysis was performed with GeneMapper software v3.7 (ABI) and converted to a binary data matrix to estimate the genetic relationships among the cultivars using PAST software (9). Cervus 3.0.7 (10) software was used to estimate allele frequencies, while GenAEx 6.51b2 (11, 12) to recovering private allele and the identity of individuals carrying them.

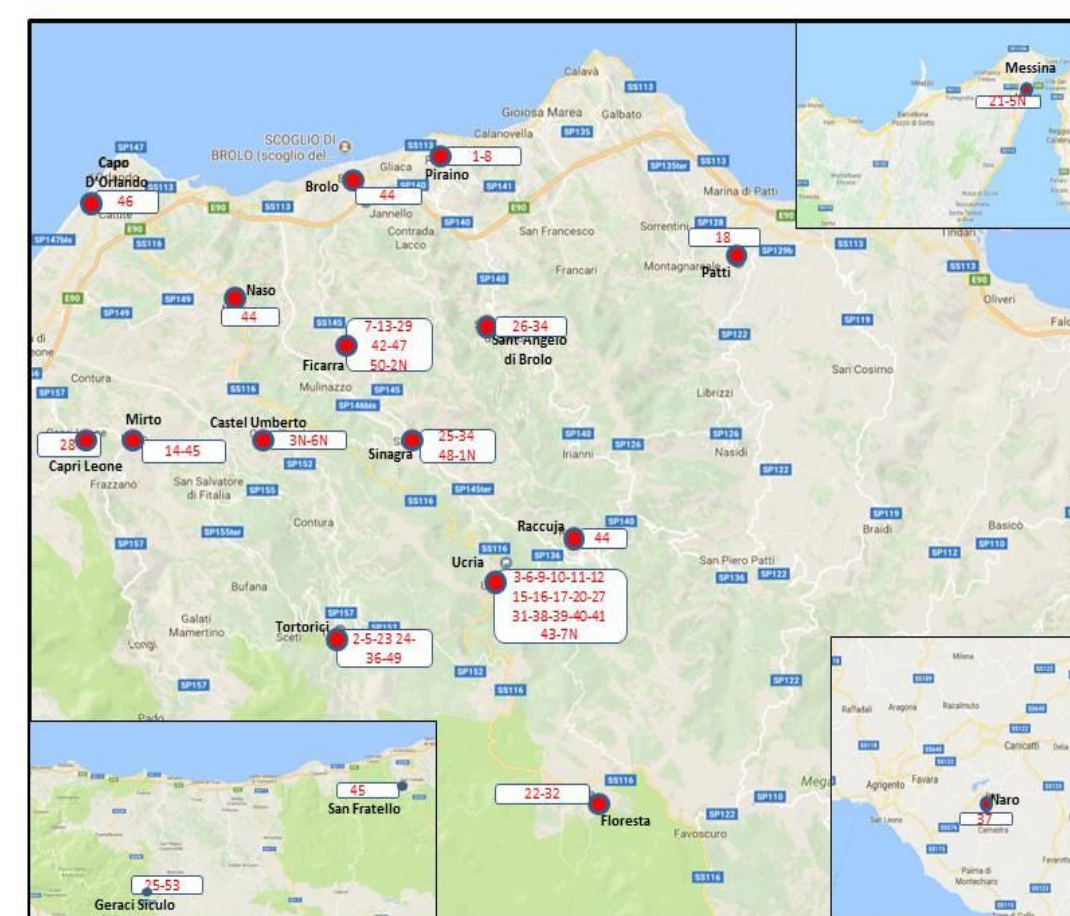


Fig.1: Map showing the geographic localization (left) of the 57 Sicilian common bean accessions and *ex situ* field conservation placed at Ucria (right).

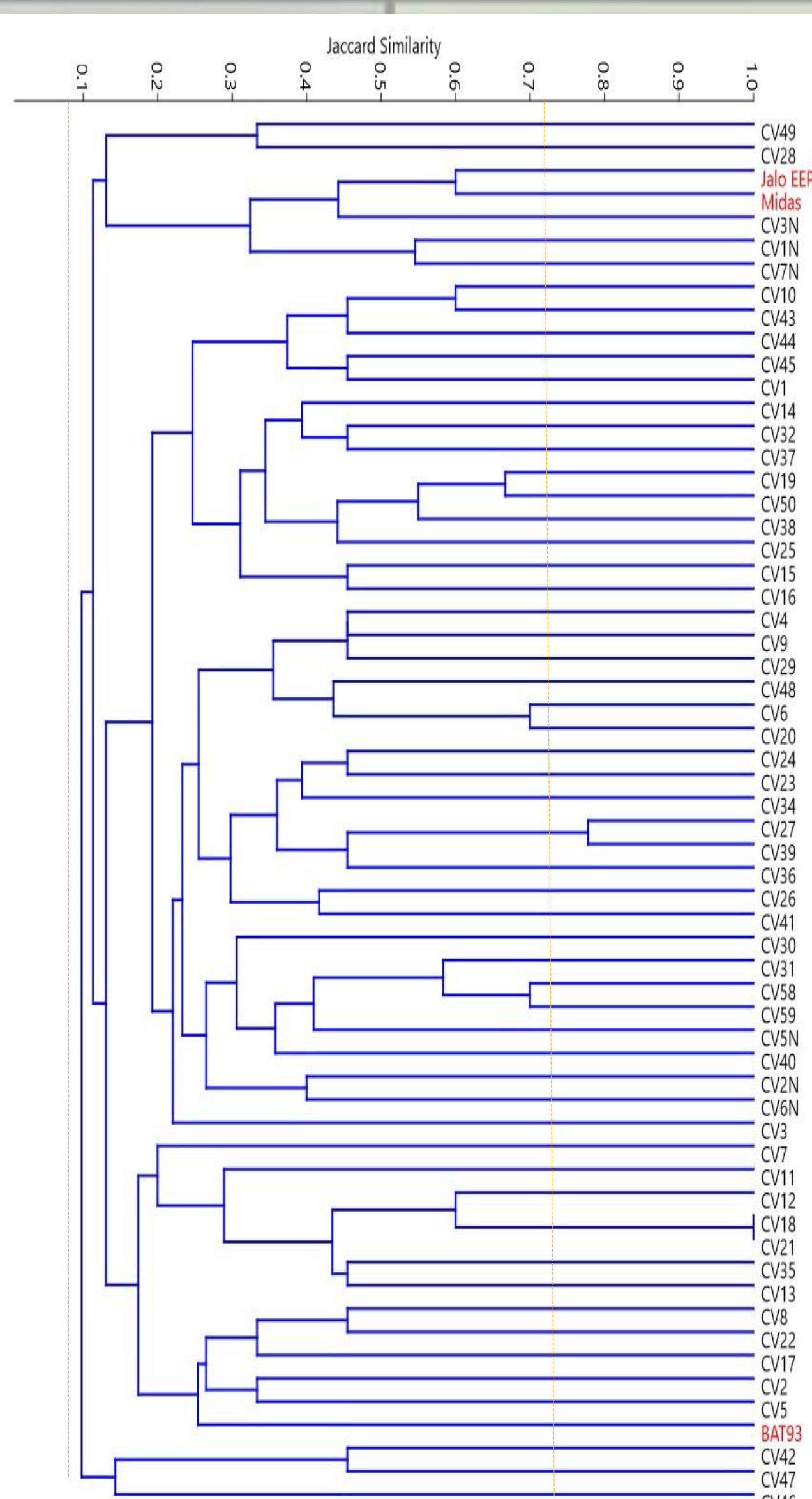


Fig.3: Dendrogram representation of genetic relationships among 57 Sicilian common bean accessions and 3 standard check genotypes

Accession number	Local name	Accession number	Local name
CV1	'Badda (o del prete)'	CV30	'Lumachella chiara'
CV2	'Carrubara baccello rugoso'	CV31	'Carruzzu di Ucria'
CV3	'Buttuna di gaddu'	CV32	'Crucchittu di Floresta'
CV4	'Curnittu calabrisi'	CV34	'Occhittu di pircici'
CV5	'Carrubaru niuru'	CV35	'Sanfratellano'
CV6	'Carruzzu criotu'	CV36	'Crucchittu rummuletta'
CV7	'Calabrese baccello dritto'	CV37	'Rosa tunnu'
CV8	'Vasolu calabrisi biancu'	CV38	'Carruzzu di Piano Campo'
CV9	'Carruzzu viola'	CV39	'Carruzzu niuru pi siccaru'
CV10	'Bavaluciaru di tri voti'	CV40	'Disio'
CV11	'Rosa lungo'	CV41	'Carruzzu nustrali'
CV12	'Carruzzu di maisa'	CV42	'Niuoro senza sfiazzi'
CV13	'Vasolu di pasta biancu'	CV43	'Vasolu di Padre Bernardino'
CV14	'Santagasi'	CV44	'Virdi lungarinu'
CV15	'Carruzzu rosa'	CV45	'Pinittanu'
CV16	'Vasolu di petra'	CV46	'Rampicante nerella'
CV17	'Mignacca'	CV47	'Vasolu lanchittu ma beddu'
CV18	'Calabrisi'	CV48	'Zicca'
CV19	'Crucchittu settembrinu'	CV49	'Occhittu rosa'
CV20	'Carruzzu du miricanu'	CV50	'Massaru'
CV21	'Tignusu calabrese'	CV58	'Munachedda'
CV22	'Ucchittu di zappa'	CV59	'Facigghiu'
CV23	'Vanadia di Tortoricu'	CV1N	'Nanu palermitanu'
CV24	'Carruzzu pi siccaru'	CV2N	'Nanu niuru'
CV25	'Setticanni'	CV3N	'Nanu virdi'
CV26	'Ucchittu santanglisi'	CV5N	'Nanu calabrisi'
CV27	'Crucchittu criotu'	CV6N	'Nanu di castania'
CV28	'Carrubara baccello liscio'	CV7N	'Nanu carruzzu'
CV29	'Vasolu di caminu'		

Tab. 1: List of samples analyzed, their accession number and local name

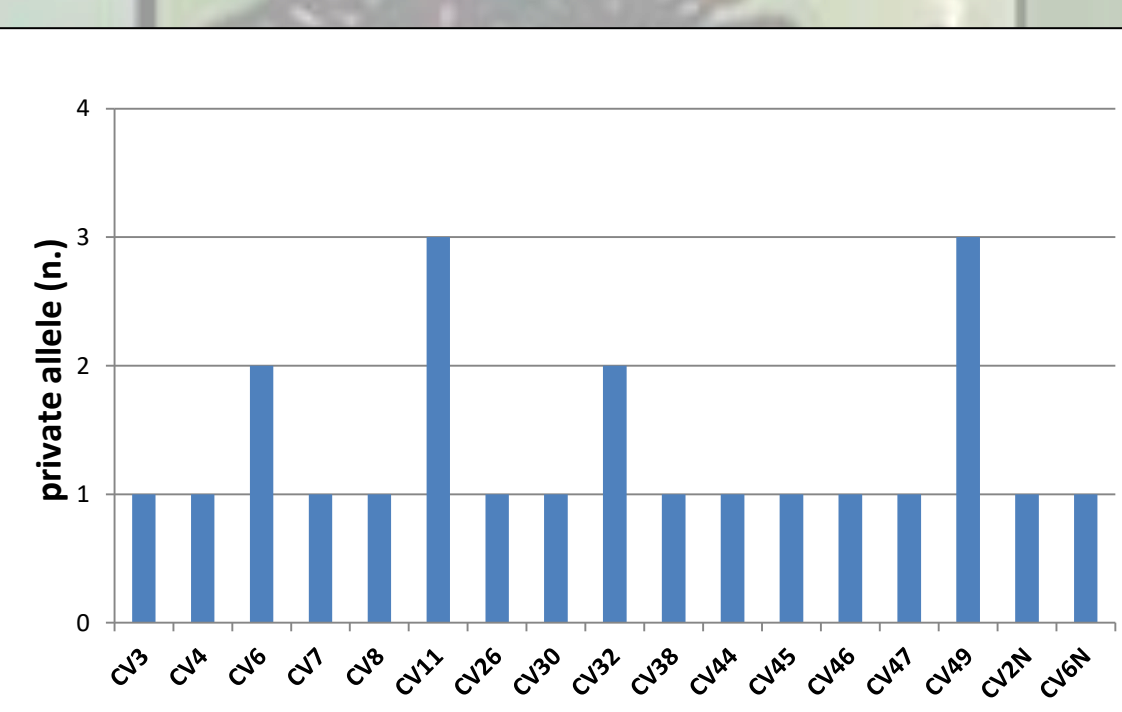


Fig.2: Distribution of private alleles in each accession

RESULTS

A total of 75 alleles were recorded using 8 SSR markers, and the number of alleles per locus ranged from 5 (BM159) to 13 (PVBR25). The lowest PIC values was 0.463 (PV-ag001) and the highest 0.853 (BM160). A total of 23 private alleles were recorded in 17 cultivars (Fig. 3). Cultivar CV49 (*Occhittu rosa*) and CV11 (*Rosa lungo*) have the highest number of private alleles, 3 alleles in 3 loci and 3 alleles in 2 loci respectively, followed by CV6 (*Carruzzu criotu*) and CV22 (*Crucchittu di Floresta*) with 2 alleles in 1 locus.

The UPGMA dendrogram (Fig.3) showed high level of genetic diversity among the accessions (14%-78%) and only two cultivars (CV18 and CV21) were with 100% of genetic similarity.

Three determined accessions (CV1N, CV3N and CV7N) showed 32% of similarity with Mesoamerican genotypes (MIDAS and Jalo EEP558), while Andine genotype (BAT93) clustered with five accessions at 25% of similarity. All the other accession differed more than 85% from the standard gene pool genotypes.

CONCLUSIONS

The high level of genetic diversity of Sicilian common bean collection evidence the importance of genetic characterization to define the core collection, confirming the weakness of morphological characterization and highlighting high intrinsic value as accurate criterion for planning and setting-up *ex situ* collections. Indeed, genetic markers should be used to define more accurately the range of inter- and intra-landraces diversity.

Besides playing a major role in distributing biospecimens and associated data for research purposes (13), biorepositories promote conservation policies and the sustainable use of intrinsic crop diversity. Community-based conservation can and should be shared with local farmers, who can directly benefit from these fundamental research to curb biodiversity loss in rural and protected areas.

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