National Strategy for the Conservation of Crop Wild Relatives of Spain

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PGR Secure: Novel characterization of crop wild relative and landrace resources as a basis for improved crop breeding

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COOPERATION

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Front Cover Picture: *Lupinus angustifolius* L., by Rubén Milla

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1 Introduction

In the past few years, many studies have reported the importance of Crop Wild Relatives (CWRs) and the need to preserve them, suggesting different criteria for their selection and protocols for their conservation (Heywood *et al*., 2007; Magos-Brehm *et al*., 2010; Patausso 2012). The relevance of CWRs essentially responds to the need to guarantee food security through the provision of a wide gene pool of potential gene donors (Maxted *et al*., 2012a). This is especially important considering that the predicted changes in temperature and precipitation patterns worldwide in a climate change context will affect crop production and quality. In many areas, strong variations in rainfall and temperatures are already being observed; so farmers are presently trying to adapt their crops to the changing environment (Turner & Meyer, 2011). However, highly productive cultivars are known to have a narrow genetic base and, in many cases, lack adaptation mechanisms to long-term extreme environmental conditions (Stamp & Visser, 2012). As CWRs have already been used and proved to be useful in crop breeding (Hajjar & Hodgkin, 2007), increasing knowledge on them and improving their conservation is becoming urgent nowadays.

A CWR can be defined as any wild species related to a crop that can contribute genetic material for plant breeding, but, unlike crop species, has not been domesticated (Heywood *et al*., 2007). In general terms, all species related to any crop of socio-economic importance or belonging to a genus that holds a crop species could be considered a crop wild relative (Kell *et al.,* 2008; Maxted *et al*., 2006). However, different interpretations are found when a more precise definition is needed for planning specific conservation actions. The key is to delimitate which species should be included in the so-called checklist of CWRs. According to Meilleur & Hodgkin (2004), it should include the "wild congeners or closely-related species of a domesticated crop or plant species, including relatives of species cultivated not just for food but also for medicinal, forestry, forage, ornamental or other reasons". Following this definition, the checklist of CWRs in some countries would be so large that it would be impossible to implement specific conservation actions for all of them. Consequently, there is a common agreement on limiting the number of CWR species in a prioritized checklist by applying criteria about relatedness, based on the gene pool concept (Harlan & de Wet, 1971) or the taxon group concept (Maxted *et al.* 2006), which allow researchers to classify plants in relation to their degree of relatedness to crops (Barazani *et al*., 2008; Flor *et al*., 2004; Maxted *et al*., 2006).

2 Prioritization of Crop Wild Relatives in Spain

2.1 Introduction

Spain is one of the countries with the greatest CWR diversity in Europe with more than 6500 taxa (Kell *et al*., 2008). However, the case of Spain is extremely complex due to the idiosyncrasy and high biodiversity of the country. In order to generate a prioritized checklist of CWR based on operable criteria, it was important to delimitate a clear subset of plant species that should be included in that prioritized CWR checklist for further action. Food, forage and fodder crops should clearly be included but, what about forestry species? What about relatives of promissory species in emerging markets which are currently not as important as major crops, the so-called neglected crops and those only produced in narrow geographic ranges? Should all species, native and introduced be included? What about relatives of ornamentals and crops used for industrial purposes?

Considering the administrative structure of genetic resources conservation in Spain, it was decided not to include forestry species in the CWR checklist. Forestry species are, indeed, of great socio-economic importance, and they are already protected by specific national forestry laws, are the object of specific national actions of biodiversity conservation (Mountain Law 10/2006) and are also the focus of specific actions at regional levels (e.g. Forestry Law 3/1993 in Valencia Autonomous Community, Forestry Law 15/2006 in Aragón Autonomous Community, Forestry Law 3/2008 in Castilla La Mancha Autonomous Community, etc.). Furthermore, the current draft of the Strategic Plan for Biodiversity Conservation developed under Law 42/2007 considers the creation of a specific National Inventory of Forestry species. Food and Forage & Fodder crop wild relatives were included in the final checklist, as they are clearly a "priority when prioritizing", as well as other crop genera and their relatives (e.g.: ornamental and industrial uses). This responds to the unquestionable need to preserve a wide range of biodiversity and conserve the complex culture of Spanish genetic resources.

Different approaches have been used regarding naturalized taxa. Some countries have included them in their prioritized national CWR checklists because of their importance in the national economy (e.g., Portugal, Magos-Brehm *et al*., 2008) or because they constitute potential sources of novel adaptations (e.g., United States, Khoury *et al*., 2013). However, other countries or regions have not included naturalized taxa (e.g., Europe, Kell *et al*., 2012 or United Kingdom, Maxted *et al*., 2007). We have only considered crop genera with species native to Spain, because of the need to prioritize and because introduced and naturalized species have their diversification centres in

other areas where they are expected to hold maximum diversity levels. Historically, their presence in Spain is likely to have taken place through a series of introduction events that may have significantly reduced their gene pool.

2.2 Methods

2.2.1 Generation and prioritization of the crop genera lists

The process for the generation of the Spanish Checklist of CWRs began with the prioritization of crops and the identification of the CWRs that relate to these crops. Genera were selected for the list of important crops based on their contribution to food security worldwide and their economic importance for the country. Therefore, the baseline list used for generating the prioritized National Crop list was the list in Annex 1 of the FAO International Treaty on Plant Genetic Resources for Food and Agriculture (FAO, 2001) and the crops included in the Spanish Annual Directory of Agricultural Statistics of 2010 (MAGRAMA, 2011). Ornamental genera were obtained by consulting the Annual Reports of the Community Plant Variety Office in Europe (2010) and selecting the ten most important ornamental species in Europe. The lists of the International Union for the protection of new varieties of plants (UPOV, 2010) (hereafter UPOV list) and the Germplasm Resources Information Network (GRIN) database of the United States Department of Agriculture (http://www.ars-grin.gov/cgi-bin/npgs/html/index.pl?language=es) were checked to add important genera due to other uses (aromatic or medicinal uses, for example). The latest available publications on trends in breeding were also consulted (Kole, 2011a; 2011b; 2011c; 2011d; 2011e), and the CWR inventories and prioritized lists of other countries were checked to compare and add genera of potential interest (e.g. *Iris* L. or *Jasminum* L.) (Berlingeri & Crespo, 2012; Barazani *et al*. 2008; Labokas *et al*. 2010; Magos-Brehm *et al.,* 2010 and Markkola, 2005).

Once the list of genera of the important crops was completed, information on the use of the crops, production data, number of registered varieties and the geographic distribution of wild species belonging to the genera was gathered (Table 1).

Table 1: Information gathered for each genus of selected crops during the generation of crop lists for its subsequent prioritization

Data field	Content
Main Use	Main use of the crop: 1) Food, 2) Fodder & forage, 3) Ornamental and 4) Industrial & other uses.
Secondary uses	Other uses of the crop in addition to the primary use.
FAO International Treaty	Indicates whether at least one crop belonging to the genus is included in Annex 1 of the FAO International Treaty on Plant Genetic Resources for Food and Agriculture (FAO, 2001)
Registered varieties in Spain	Informs if the genus contains any registered varieties in Spain (1973-2010)
Number of varieties registered	Number of varieties registered per genus in Spain from 1973 to 2010
Canary Islands	Genus contains at least one species that is found wild or naturalized in the Canary Islands according to the list of wild *Spermatophyta* species in the Canary Islands (Acebes Ginovés *et al*., 2010)
Iberian Peninsula and Balearic Islands	Genus contains at least one species that is found wild or naturalized in the Iberian Peninsula or Balearic Islands according to *Flora Iberica* (Castroviejo *et al.,* 1986-2012)
Natural Distribution	Genus contains at least one species that has part of its natural distribution in the Iberian Peninsula, Balearic Islands or Canary Islands according to *Flora Iberica* (Castroviejo *et al.,* 1986-2012) or the list of wild Spermatophyta species in the Canary Islands (Acebes-Ginovés *et al*., 2010).
Spanish Annual Directory of Agricultural Statistics	Depicts the presence of a crop belonging to the genus in the 2010 edition of the Spanish Annual Directory of Agricultural Statistics (MAGRAMA, 2011)
Production (Food)	Production in tons for food crops according to the Spanish Annual Directory of Agricultural Statistics (MAGRAMA, 2011)
Cultivated Area (Food)	Cultivated surface in ha for food crops according to the Spanish Annual Directory of Agricultural Statistics (MAGRAMA, 2011)
Production (Forage)	Production in tons for forage crops according to the Spanish Annual Directory of Agricultural Statistics (MAGRAMA, 2011)
Cultivated Area (Forage)	Cultivated surface in ha for forage crops according to the Spanish Annual Directory of Agricultural Statistics (MAGRAMA, 2011)
Production (Others)	Production in tons for other crops, including ornamentals according to the Spanish Annual Directory of Agricultural Statistics (MAGRAMA, 2011)
Cultivated Area (Others)	Cultivated surface in ha for other crops, including ornamentals according to the Spanish Annual Directory of Agricultural Statistics (MAGRAMA, 2011)
No. of species in the UPOV list	Number of species belonging to the genus that are found on the UPOV list (International Union for the Protection of New Varieties of Plants) (UPOV, 2010)

Table 1: Information gathered for each genus of selected crops during the generation of crop lists for its subsequent prioritization

Data field	Content
No. of infraspecific taxa in the UPOV list	Sum of the number of infraspecific *taxa* belonging to the genus that are found on the UPOV list (UPOV, 2010)
No. of hybrids in the UPOV list	Number of hybrids within the genus that are found on the UPOV list (UPOV, 2010)

The genera on the crop list were prioritized based on the following criteria:

1) The genus contains at least one wild species native to Spain, **and** 2) the genus is listed in Annex 1 of the FAO International Treaty on Plant Genetic Resources for Food and Agriculture (FAO, 2001) **or** contains a crop in the Spanish Annual Directory of Agricultural Statistics (MAGRAMA, 2011) **or** contains a crop that has at least one registered variety in Spain, as an unequivocal sign of economic concern to the country.

The resulting crop genera list based on these criteria was assessed by national experts from institutions dealing with the breeding of the most important food and fodder & forage crops in Spain. Dr. Mayor, who works on onion breeding in the Research Centre for Food and Agriculture of Aragon (Centro de Investigación y Tecnología Agroalimentaria - CITA, Aragón) and Dr. Díez Niclós, who is from the Centre for Conservation and Improvement of Agro-Biodiversity in Valencia (Instituto Universitario de Conservación y Mejora de la Agrodiversidad Valenciana - COMAV, Valencia) and an expert in horticultural species, were contacted through Dr. De la Rosa from the Spanish National Genebank for PGRFA (Centro de Recursos Fitogenéticos – Instituto Nacional de Investigación y Tecnología Agraria y Alimentación - INIA). Dr. Rubiales Olmedo from the Institute for Sustainable Agriculture - CSIC (Instituto de Agricultura Sostenible, Centro Superior de Investigaciones Científicas - CSIC) was contacted to check the leguminous species; Dr. Oliveira Prendes from the University of Oviedo was contacted regarding the fodder species and finally, Dr. Ordás and Dr. Cartea González, from 'Misión Biológica de Galicia' were contacted for the *Solanum* and *Brassica* crops. These assessments resulted in the inclusion of three new genera (*Deschampsia* P. Beauv., *Hedysarum* L. and *Ornithopus* L.) and their corresponding wild species which were not listed in any of the previously consulted sources. The national experts validated all of the species already included in the list.

2.2.2 Generation of the Spanish CWR Checklist

For each of the selected crop genera, all CWR species naturally occurring in Spain were identified using the national flora of reference *Flora Iberica* (Castroviejo *et al*., 1986-

2012), information resources like the Anthos Project (http://www.anthos.es) and other bibliography (Pascual, 2004; Romero Zarco, 1996; Killian *et al*. 2011). The list of wild animal and plant species in Canary Islands (Acebes Ginovés *et al*. 2010) was also consulted to include CWRs in the Canary Islands, as *Flora Iberica* only lists species in the Iberian Peninsula and Balearic Islands. The taxonomic level of reference for the CWR Checklist was set at the species level. Thus, infraspecific levels were not taken into account as separate entities.

A database was built in which information was collated for each CWR species on endemicity, threat status, genepool (Harlan & de Wet, 1971) and taxon group (Maxted *et al*., 2006), number of chromosomes, inclusion in the habitats directive, abundance and priority use of the related crop. In order to apply the genepool concept or the taxon group concept, a list of cultivated species was generated for each genus by consulting the Germplasm Resources Information Network (GRIN) (http://www.ars-grin.gov/cgi-bin/npgs/html/index.pl?language=es). Crop and vernacular names were linked to specific scientific names. These generated a list of 134 food crops (72 different species), 46 forage & fodder crops (46 species), 54 ornamental crops (44 species and 10 hybrids) and 9 crops of industrial uses (8 species and 1 hybrid) for a total of 243 priority crops.

The genepool concept always prevailed over the taxon group concept. After checking that genepool information was not available, the taxon group concept was applied according to the taxonomic classification following *Flora Iberica (*Castroviejo *et al*., 1986-2012*)*. This checklist together with all this information constitutes the Annotated Checklist of Spanish CWRs. The data fields with the compiled information are shown in Table 2.

Table 2: Information gathered for each CWR species listed for the generation of the Annotated Checklist of Spanish CWRs.

Data field	Content	Source
Priority use of the crop which the wild species is related to	Use of the main crop of the genus	Annex 1 of the FAO International Treaty on Plant Genetic Resources for Food and Agriculture (FAO, 2001); Spanish Annual Directory of Agricultural Statistics (MAGRAMA, 2011); GRIN world Economic Plant Database (www.ars-**grin**.gov/cgi-bin/npgs/html/index.pl?language=es)
Taxonomic information	Classification of the taxon in the following taxonomic levels: Family, Subfamily, Tribe, Subtribe, Genus,	*Flora Iberica* (Castroviejo *et al.,* 1986-2012*)*; Anthos project (www.anthos.es); GRIN Taxonomy for Plants (www.ars-**grin**.gov/cgi-

Table 2: Information gathered for each CWR species listed for the generation of the Annotated Checklist of Spanish CWRs.

Data field	Content	Source
	Subgenus, Section, Species	bin/npgs/html/index.pl?language=es); additional references*
Habitats directive	Presence of the species in the EU Habitats Directive	Habitat directive 92-43 EEC; Habitat directive 2007 EEC; Summary of amendments to annex I after the inclusion of Romania and Bulgaria
IUCN Red List	Presence of the species in the Red List of Spanish Vascular Flora	Red List of Spanish Vascular Flora (Moreno, 2008)
IUCN Red List Category	Classification of threat according to the Red List	Red List of Spanish Vascular Flora (Moreno, 2008)
Number of subspecies on Red List	Number of infraspecific taxa belonging to the species included in the Red List of Spanish Vascular Flora	Red List of Spanish Vascular Flora (Moreno, 2008)
Endemicity	Indicates whether the species is endemic to Spain (Iberian Peninsula, Balearic Islands or Canary Islands)	Flora Iberica (Castroviejo *et al.*, 1986-2012) and additional references*.
Gene Pool/ Taxon Group concepts	Gene Pool considered as a possibility of gene transfer (Primary, Secondary, Tertiary (Harlan & de Wet, 1971)). Taxon Group considered in terms of hierarchical taxonomy (Maxted *et al.*, 2006)	Crop Wild Relatives and Climate Change (2012) Online resource. (www.cwrdiversity.org). *Flora Iberica* (Castroviejo *et al*., 1986 - 2012). Additional references*.
Abundance	Number of populations known to be found in Spain	GBIF data portal (data.gbif.org)
Chromosome number	Chromosome number of the species	*Flora Iberica* (Castroviejo *et al*., 1986 - 2012); Anthos project (www.anthos.org); Additional references*.

* *All additional references are listed at* http://pgrsecurespain.weebly.com/spanish-proposal-for-the-national-strategy---european-deliverable-d-32.html

2.2.3 Prioritization of the Spanish CWR Checklist – Spanish National Inventory of CWRs

The Spanish CWR Checklist obtained contains a large number of CWR species. It is objectively unrealistic to perform an analysis of the conservation status of all the species on this list, as it would be impossible to implement management activities on

all of the CWR species in need of conservation. Therefore, the Spanish CWR checklist was subject to an additional prioritization process with the purpose of obtaining a shorter list of species for which a conservation status analysis would be carried out and specific recommendations for implementing conservation actions could be formulated. The prioritized Spanish CWR Checklist along with the database resulting from the information generated from the analysis of the conservation status of these species constitute the Spanish National Inventory of CWR.

Once information was compiled for all the species on the Spanish CWR Checklist, the checklist was prioritized using the criteria presented in Table 3. CWR species related to crops used for Food or Fodder & Forage species (Plant Genetic Resources for Food and Agriculture (PGRFA)) that met any of the three criteria were selected. For CWR species related to Ornamental and Industrial & Other Uses, only the criterion related to gene pool and taxon group was used.

Table 3. Criteria applied for the prioritization of CWRs on the Spanish Checklist.

Criterion	Selected values	CWR use categories to which the criterion is applied
Gene pool	Categories 1 and 2	Food, Forage & Fodder, Ornamental, and Industrial & other uses
Taxon group	Concepts 1, 2 and 3	
Threatened and near threatened taxa according to IUCN criteria	CR, EN, VU, NT	Food, Forage & Fodder
Endemicity	Spain	Food, Forage & Fodder

In the Ornamental and Industrial & Other Uses groups, the criteria linked to threat and endemicity were not applied because the use of this additional criteria would have selected a very high number of species (i.e. taxonomy of some of the included genera, such as *Limonium*, renders a great number of endemic microtaxa, many of which are under threat due to their narrow distribution). Furthermore, the food security precautionary principle is not applicable to these groups, so the selection of species was stricter.

2.3 Results and Discussion

2.3.1 Generation and prioritization of the crop genera lists

After compiling the genera listed in the Annex 1 of the International Plant Treaty, the Spanish Annual Directory of Agricultural Statistics, Annual Reports of the Community

Plant Variety Office in Europe, UPOV list, GRIN World Economic Plant Database, other inventories, information on trends in breeding and experts' suggestions, a final list of 203 genera containing crops of importance was obtained. This list of genera with the associated information for each genus is available at http://pgrsecurespain.weebly.com/important-crops-for-spain-and-europe---crop-lists.html. The application of criteria for the prioritization of the genera on the crop list generated a list of 60 genera (Table 4).

After structuring the genera by main use, the Food category comprises 33 genera in 13 families; the Fodder & Forage category encompasses 12 genera in 2 families; the Ornamental category holds 5 genera in 5 families and the Industrial & Other Uses category has 10 genera in 7 families. *Fabaceae* and *Poaceae* are the two most important families with 11 genera each, followed by *Brassicaceae* with 7 genera. Figure 1 shows the relative importance of the contribution of each family to the prioritized crop list.

Table 4: List of prioritized genera obtained for the generation of the Spanish Checklist of CWRs. Information on families, use categories to which they have been assigned, and the reason for their inclusion on the list.

Genus	Family	Use category	Reason for inclusion
Aegilops	Poaceae	Food	Experts in agrobiodiversity
Allium	Liliaceae	Food	Spanish Annual Directory / Registered varieties
Apium	Apiaceae	Food	Spanish Annual Directory / Registered varieties
Asparagus	Liliaceae	Food	Annex 1 / Spanish Annual Directory / Registered varieties
Avena	Poaceae	Food	Annex 1 / Spanish Annual Directory / Registered varieties
Beta	Chenopodiaceae	Food	Annex 1 / Spanish Annual Directory / Registered varieties
Borago	Boraginaceae	Food	Spanish Annual Directory / Registered varieties
Brassica	Brassicaceae	Food	Annex 1 / Spanish Annual Directory / Registered varieties
Capsella	Brassicaceae	Food	Use in breeding
Cicer	Fabaceae	Food	Annex 1 / Spanish Annual Directory / Registered varieties
Cichorium	Asteraeceae	Food	Spanish Annual Directory / Registered varieties
Cynara	Asteraceae	Food	Spanish Annual Directory / Registered varieties
Daucus	Apiaceae	Food	Annex 1 / Spanish Annual Directory / Registered varieties
Diplotaxis	Brassicaceae	Food	Use in breeding / Annex 1
Erucastrum	Brassicaceae	Food	Use in breeding
Fragaria	Rosaceae	Food	Annex 1 / Spanish Annual Directory / Registered varieties

Table 4: List of prioritized genera obtained for the generation of the Spanish Checklist of CWRs. Information on families, use categories to which they have been assigned, and the reason for their inclusion on the list.

Genus	Family	Use category	Reason for inclusion
Hordeum	Poaceae	Food	Annex 1 / Spanish Annual Directory / Registered varieties
Lactuca	Asteraceae	Food	Spanish Annual Directory / Registered varieties
Lathyrus	Fabaceae	Food	Annex 1 / Spanish Annual Directory / Registered varieties
Lens	Fabaceae	Food	Annex 1 / Spanish Annual Directory / Registered varieties
Malus	Rosaceae	Food	Annex 1 / Spanish Annual Directory / Registered varieties
Moricandia	Brassicaceae	Food	Use in breeding
Olea	Oleaceae	Food	Spanish Annual Directory / Registered varieties
Patellifolia	Chenopodiaceae	Food	Experts in agrobiodiversity
Pisum	Fabaceae	Food	Annex 1 / Spanish Annual Directory / Registered varieties
Prunus	Rosaceae	Food	Spanish Annual Directory / Registered varieties
Pyrus	Rosaceae	Food	Spanish Annual Directory / Registered varieties
Raphanus	Brassicaceae	Food	Use in breeding // Annex 1
Secale	Poaceae	Food	Annex 1 / Spanish Annual Directory / Registered varieties
Sinapis	Brassicaceae	Food	Use in breeding / Annex 1
Solanum	Solanaceae	Food	Annex 1 / Spanish Annual Directory / Registered varieties
Vicia	Fabaceae	Food	Annex 1 / Spanish Annual Directory / Registered varieties
Vitis	Vitaceae	Food	Spanish Annual Directory / Registered varieties
Agrostis	Poaceae	Forage & fodder	Annex 1
Astragalus	Fabaceae	Forage & fodder	Annex 1
Dactylis	Poaceae	Forage & fodder	Annex 1 / Registered varieties
Deschampsia	Poaceae	Forage & fodder	Experts in agrobiodiversity
Festuca	Poaceae	Forage & fodder	Annex 1 / Registered varieties
Hedysarum	Fabaceae	Forage & fodder	Experts in agrobiodiversity / Annex 1 / Spanish Annual
Lolium	Poaceae	Forage & fodder	Annex 1 / Spanish Annual Directory / Registered varieties
Lupinus	Fabaceae	Forage & fodder	Annex 1 / Spanish Annual Directory / Registered varieties
Medicago	Fabaceae	Forage & fodder	Annex 1 / Spanish Annual Directory / Registered varieties
Ornithopus	Fabaceae	Forage & fodder	Experts in agrobiodiversity / Annex 1
Poa	Poaceae	Forage & fodder	Annex 1 / Registered varieties
Trifolium	Fabaceae	Forage & fodder	Annex 1 / Spanish Annual Directory / Registered varieties

Table 4: List of prioritized genera obtained for the generation of the Spanish Checklist of CWRs. Information on families, use categories to which they have been assigned, and the reason for their inclusion on the list.

Genus	Family	Use category	Reason for inclusion
Argyranthemum	Asteraceae	Ornamental	Registered varieties
Dianthus	Caryophyllaceae	Ornamental	Spanish Annual Directory / Registered varieties
Rosa	Rosaceae	Ornamental	Spanish Annual Directory / Registered varieties
Limonium	Plumbaginaceae	Ornamental	Experts in agrobiodiversity
Narcissus	Amaryllidaceae	Ornamental	Experts in agrobiodiversity / Registered varieties
Carthamus	Asteraceae	Industrial	Spanish Annual Directory / Registered varieties
Linum	Linaceae	Industrial	Experts in agrobiodiversity
Papaver	Papaveraceae	Industrial	Registered varieties
Genciana	Gentianaceae	Industrial	Experts in agrobiodiversity
Lavandula	Lamiaceae	Industrial	Experts in agrobiodiversity / Spanish Annual Directory
Hypericum	Guttiferae	Industrial	Experts in agrobiodiversity
Brachypodium	Poaceae	Industrial	Experts in agrobiodiversity
Thymus	Lamiaceae	Industrial	Experts in agrobiodiversity
Salvia	Lamiaceae	Industrial	Experts in agrobiodiversity
Sideritis	Lamiaceae	Industrial	Experts in agrobiodiversity

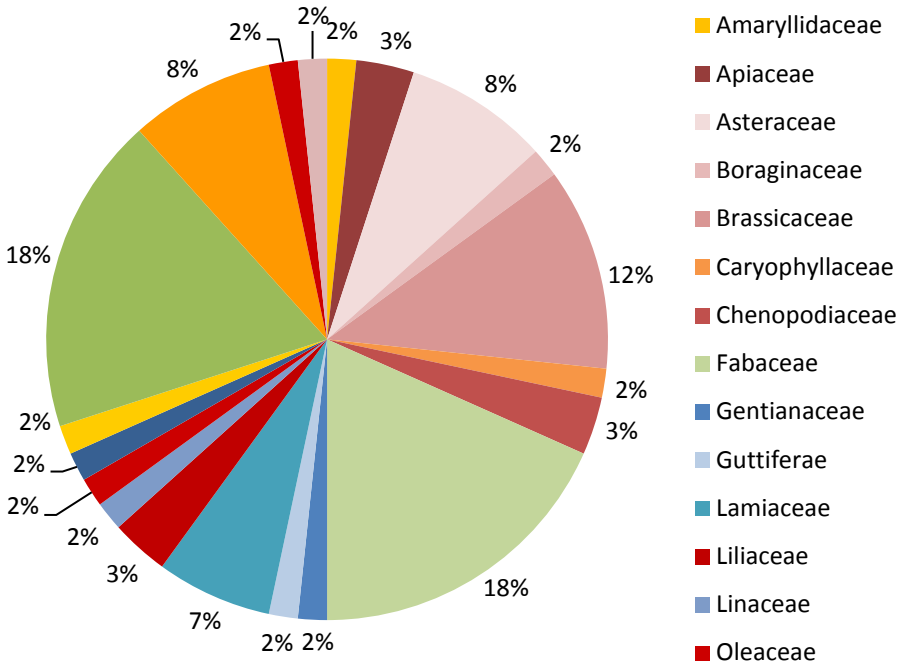


Figure 1: Relative importance of the families contributing to the prioritized crop genera list.

2.3.2 Generation of the Spanish CWR Checklist

The total number of CWR species composing the prioritized genera list extracted from the different consulted sources was 1050. Once naturalized species were eliminated, a total of 941 species constituted the Spanish Checklist of CWRs. This list is available at http://pgrsecurespain.weebly.com/crop-wild-relatives-in-spain-ndash-spanish-checklist-of-cwr.html. The list, classified according to the four main use categories, contains 224 species related to food crops (24%), 270 species related to fodder & forage crops (29%), 240 species related to ornamental crops (25%) and 207 species related to industrial & other uses crops (22%) (Fig. 2). The Spanish annotated checklist of CWRs integrated by these species and information regarding their gene pool and taxon group concept, threat status, endemicity and rest of information presented in Table 2 above, is available at http://pgrsecurespain.weebly.com/crop-wild-relatives-in-spain-ndash-spanish-checklist-of-cwr.html.

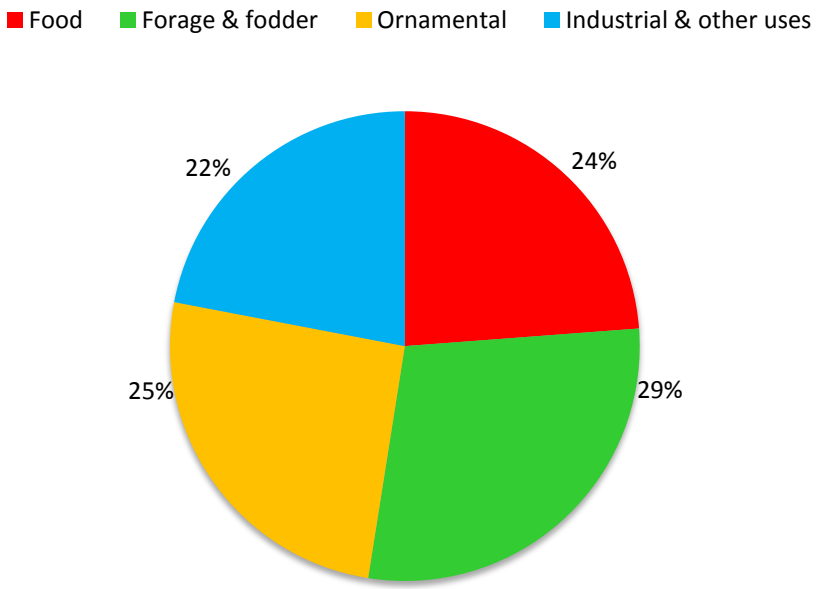


Figure 2: Distribution of the Spanish Checklist of CWR by use categories

2.3.3 Prioritization of the Spanish CWR Checklist – Spanish National Inventory of CWRs

The application of the criteria proposed for the prioritization of the Spanish Checklist of CWRs significantly reduced the list. Thus, the prioritized checklist contains 580 species: 140 species related to food crops, 184 species related to fodder & forage

crops, 161 species related to ornamental crops and 95 species related to industrial & other uses crops. This entails a reduction of around 38% in relation to the original CWR checklist.

Overall, most selected species were classified as primary or secondary relatives of the selected crop according to the genepool concept (Harlan & de Wet, 1971) or classified in taxon group 2 (same section or subsection as the reference crop) (Maxted *et al.*, 2006) (Fig.3).

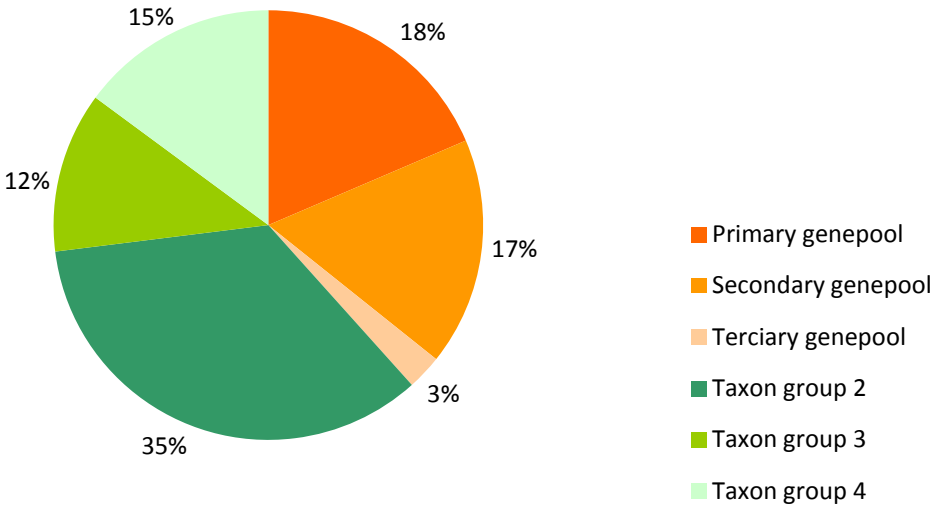


Figure 3: Classification of prioritized species according to the different genepool or taxon group categories coined by Harlan & de Wet (1971) and Maxted *et al*., (2006).

Approximately 24% of the species were classified in one of the IUCN threat categories (including NT). Of these species 25.5%, 21.3%, 42.6% and 9.2% were classified as CR, EN, VU and NT respectively; two of the species (*Astragalus algerianus* E. Sheld. and *Astragalus baionensis* Loisel.) are considered to be extinct (1.4%) (Fig. 4). In the European CWR threat assessment (Bilz *et al*., 2011) 11.5% of the species under study (66 CWR species out of 571 species) were classified under some category of threat. Nevertheless, in the European assessment only CWR for human or animal food crops were considered. If only these CWRs are taken into consideration, the Spanish threat status improves considerably, and reaches similar levels to those found in Europe (10.3%). Special emphasis must be placed on the 9 species threatened at both Spanish and European levels (Table 5).

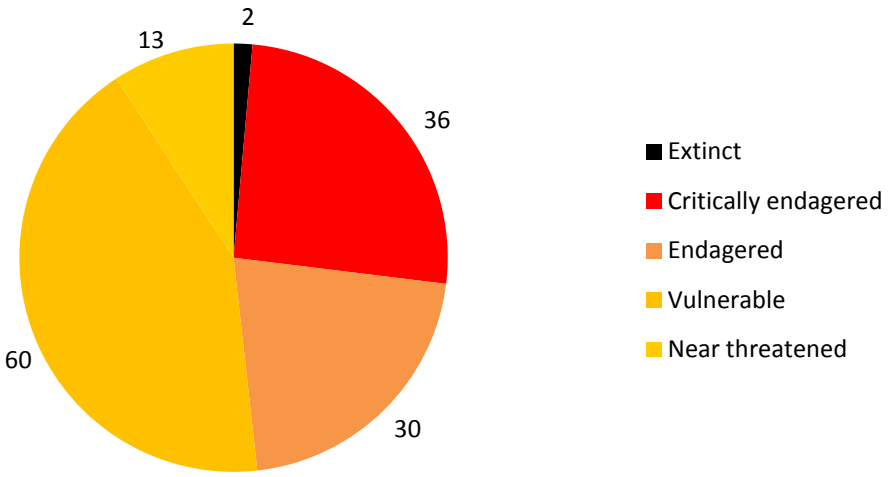


Figure 4: Number of CWR species on the Spanish prioritized checklist classified in the different IUCN Red List categories according to the Spanish Red List of Vascular Plants (Moreno, 2008)

Table 5: CWR species that are threatened at the European level and corresponding status at the Spanish level. (CR: critically endangered; EN=endangered; VU=vulnerable; NT=near threatened). From Bilz *et al*. (2011) and completed with the Spanish Red List of Vascular Flora (Moreno, 2008)

Family	Species	Red List European Status	Red List Spanish Status
Liliaceae	*Allium pyrenaicum*	VU	NT
Liliaceae	*Allium schmitzii*	VU	VU
Liliaceae	*Asparagus arborescens*	VU	Not Assessed
Liliaceae	*Asparagus fallax*	EN	EN
Liliaceae	*Asparagus nesiotes*	EN	EN
Liliaceae	*Asparagus pastorianus*	VU	Not Assessed
Liliaceae	*Asparagus plocamoides*	VU	Not Assessed
Poaceae	*Avena murphyi*	EN	EN
Chenopodiaceae	*Beta macrocarpa*	EN	Not Assessed
Fabaceae	*Cicer canariense*	EN	EN

Table 5: CWR species that are threatened at the European level and corresponding status at the Spanish level. (CR: critically endangered; EN=endangered; VU=vulnerable; NT=near threatened). From Bilz *et al*. (2011) and completed with the Spanish Red List of Vascular Flora (Moreno, 2008)

Family	Species	Red List European Status	Red List Spanish Status
Asteraceae	*Lactuca singularis*	VU	Not Assessed
Fabaceae	*Medicago citrina*	CR	CR
Chenopodiaceae	*Patellifolia webbiana*	CR	Not Assessed
Rosaceae	*Prunus lusitanica*	VU	VU
Rosaceae	*Prunus ramburii*	VU	VU

About 39.7 % of the species on the prioritized checklist were endemic of Spain or the Iberian Peninsula. This is noticeably higher than the percentage of endemics in the Portuguese CWR inventory (6.1%) (Magos-Brehm *et al*., 2008), the UK inventory (2.3%) (Maxted *et al*., 2007) in Europe, or even than that found in tropical countries with high biodiversity such as Venezuela (12%) (Berlingeri & Crespo, 2012). The importance of the Spanish CWR diversity and its preservation is underscored by these results which give relevance to the unique value of the South Mediterranean biodiversity.

Concerning the CWRs related to Food crops, 6 species met the three selection criteria used: *Allium schmitzii* Cout., *Avena murphyi* Ladiz., *Brassica bourgeaui* (Webb ex Christ) Kuntze, *Cynara alba* Boiss. ex DC., *Prunus ramburii* Boiss and *Solanum lidii* Sunding. Regarding the gene pool and taxon group concepts, most selected species were wild forms of the crops of reference (GP-1B) or classified in GP-2 (with possibility of natural gene transfer) (21.4% and 17.9%, respectively).Twenty-five percent of the species are classified in one of the IUCN Red List categories (22.9% as critically endangered, 20% as endangered, 45.7% as vulnerable and 11.4% as near threatened) (Fig. 5), 41 species are endemic of Spain, and seven are Iberian endemics. Ten of the prioritized species have confirmed potential use or are already used in crop breeding and are considered useful sources of genes for crops of worldwide importance.

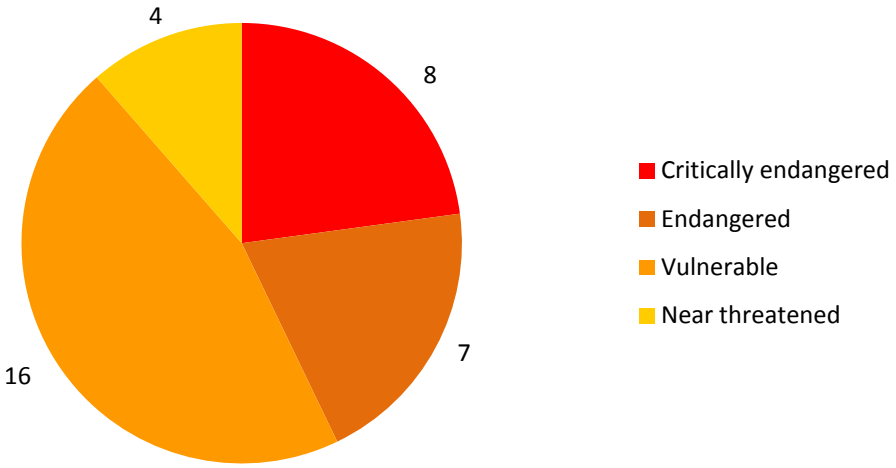


Figure 5: Number of Food-related species in the Prioritized Spanish CWR checklist classified under any of the IUCN Red List categories (critically endangered, endangered, vulnerable and near threatened).

Regarding the Forage & Fodder group, three species met the three prioritization criteria (*Astragalus cavanillesii* Podlech; *Astragalus tremolsianus* Pau; *Medicago citrina* (Font Quer) Greuter). In relation to the genepool and taxon group concept, 26.1% and 1.6% are classified into GP-1 and GP-2, respectively. *Medicago hybrida* (Pourr.) Trautv. has previously been reported to have potential use in breeding (http://www.ars-grin.gov/cgi-bin/npgs/html/taxon.pl?314561), even though it is found in GP3 http://www.cwrdiversity.org/checklist/cwr-details.php?specie_id=300. Fourteen percent of the species are classified in one of the IUCN Red List categories (8% Extinct, 24% critically endangered, 24% endangered, 40% vulnerable and near threatened 4%) (Fig. 6), and 48 species are endemic to Spain or to the Iberian Peninsula.

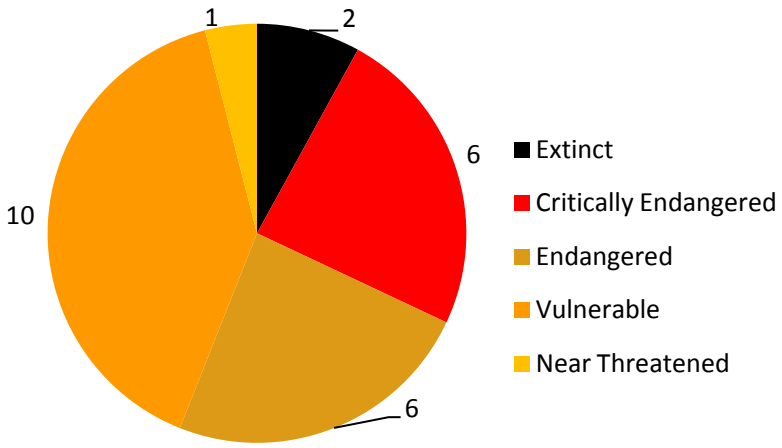


Figure 6: Number of Fodder & Forage related species on the Prioritized Spanish CWR checklist classified under any of the IUCN Red List categories (extinct, critically endangered, endangered, vulnerable and near threatened).

Concerning the CWRs related to the Ornamental group and the Industrial group, all of the prioritized species were selected according to the genepool and taxon group criterion.

In the Ornamental category, 11.8% belong to the GP-1 category and 16.8 % to the GP-2 category. Nearly 42% of the selected species are classified under the IUCN Red List categories (22% critically endangered, 27% endangered, 43% vulnerable and 8% near threatened) (Fig. 7), and 93 species are endemic to Spain or to the Iberian Peninsula (88 and 5 respectively).

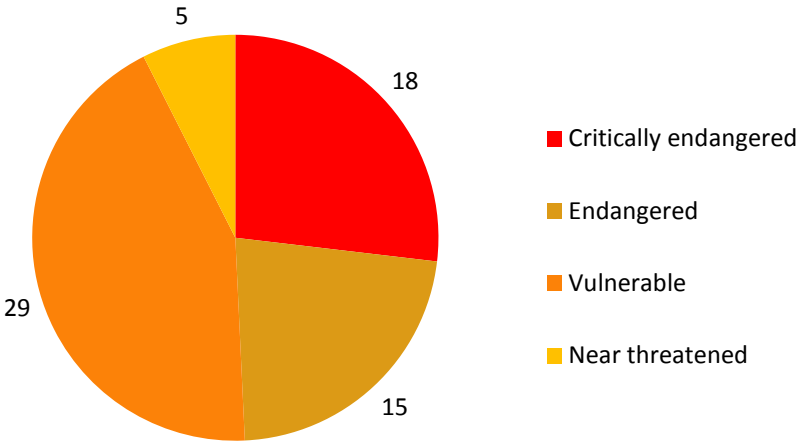


Figure 7: Number of ornamental related CWR species on the Prioritized Spanish CWR checklist classified under any of the IUCN Red List categories (critically endangered, endangered, vulnerable and near threatened).

In the Industrial category, 15.8% are classified in the GP-1 category and 45.3 % in the GP-2 category; 14.8% of the selected species are classified under the IUCN Red List categories (28.6% critically endangered, 14.3% endangered, 35.7% vulnerable and 21.4% near threatened) (Fig. 8) and 50 species are endemic to Spain or to the Iberian Peninsula (46 and 4 respectively).

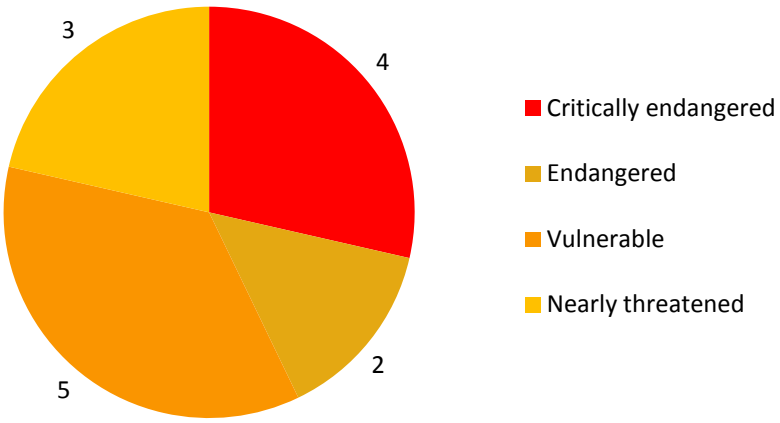


Figure 8: Number of industrial related CWR species on the Prioritized Spanish CWR checklist classified under any of the IUCN Red List categories (critically endangered, endangered, vulnerable and near threatened).

The annotated prioritized checklist is available at http://pgrsecurespain.weebly.com/crop-wild-relatives-in-spain---prioritization-of-the-checklist.html.

Similar processes for the generation of National Inventories have been followed in other countries such as United Kingdom, Portugal, Venzuela or the United States (Maxted *et al*., 2007; Magos-Brehm *et al*., 2008; Berlingeri & Crespo, 2011; Khoury *et al*., 2013). The list from the PGR Forum Crop Wild Relative Catalogue for Europe and the Mediterranean (Kell *et al*., 2005) served as a starting basis for the Portuguese and UK inventories (Maxted *et al*., 2007; Magos-Brehm *et al*., 2008). A floristic approach (Maxted *et al*., 2012b) was also followed in the cases of Venezuela and USA (Berlingeri & Crespo, 2012; Khoury *et al*., 2013). In contrast, in the Spanish case, a previous prioritization of socio-economically important crops and the identification of only those CWR related to the corresponding genera was carried out. This greatly simplified the process helped to focus on the most important CWR. The different national inventories produced have considered similar use categories. Food and Forage & Fodder categories have been included in all inventories and other uses such as

medicinal, ornamental, environmental or industrial uses (among others) have been reflected on all of them except for the Venezuelan case. As well as in the other inventories, close genetic relatives of the selected crops have been prioritized. When this information was not available, genetic relatedness was inferred from taxonomic information (Taxon Group concept by Maxted *et al*. (2007)). It is worthy to note that the National Inventories of Portugal, UK and Venezuela (but not the UK's) also included non-native species, in response to the importance of introduced plants in the economy of their countries (Magos-Brehm *et al*., 2008). In the Spanish case, the large number of native CWRs and the fact that introduced species have their diversification centres elsewhere and are likely to have reduced genetic diversity in Spain inclined the decision towards the exclusion of non-native species.

3 Conservation assessment of prioritized CWR

3.1 Introduction

The next step in the development of the national strategy for CWR conservation was the evaluation of the *in situ* and *ex situ* conservation status of the prioritized CWR taxa. In order to evaluate the *in situ* conservation status, a 'gap analysis' approach (Scott *et al*., 1993) was followed, as previously used and recommended in similar instances (Markkola, 2005; Heywood *et al.* 2007; Maxted *et al.* 2007; Maxted *et al*. 2008a; Maxted *et al*. 2011; Scholten *et al*. 2011; Green *et al.* 2012; Maxted *et al*., 2012b). Gap analysis consists of finding and identifying gaps in conservation by comparing *in situ* or *ex situ* conserved populations to all known populations for each target species. Gap analysis can be conducted at different geographic levels, allowing different scales to be covered depending on the focus of the study. In order to perform a gap analysis, a Geographic Information System (GIS) needs to be established with all the necessary information. GIS has been used in the last few years to help managing Plant Genetic Resources (PGR). Thus, it has proved to be useful in identifying the best locations for the establishment of genetic reserves and selecting the populations that should be monitored (Guarino *et al*. 2002). Furthermore, GIS together with ecogeographic land characterization maps (ELC maps) developed for Spain and the Balearic Islands (Parra-Quijano *et al*., 2012a) have been recently used for identifying spatial and ecogeographical gaps to improve *ex situ* collections (Parra-Quijano *et al*. 2012b). The use of ELC maps is a helpful tool when no information on genetic diversity is available, considering that the ecogeographic diversity (ED) found in a species' distribution can be used as a proxy for genetic diversity (GD) (Maxted *et al*., 2012a). Taking this into account, the combined use of gap analysis techniques and ELC maps makes it possible to assess the ratio of conserved populations and estimate how well they represent the overall GD of the species. This is helpful for setting conservation priorities among the selected species, pointing out those which are more threatened or vulnerable in relation to their conservation status (Maxted *et al*. 2008b).

On the other hand, the *ex situ* conservation of plant species ensures that natural populations have a representative backup sample preserved in germplasm banks. This preserved material can then be used for breeding purposes as it is easily accessible to breeders. Thus, the evaluation of the conservation status of prioritized CWR taxa in *ex situ* collections is also an important step in developing conservation strategies.

3.2 Material and Methods

Both *in situ* and *ex situ* conservation assessments of CWR species are recommended when planning conservation actions (Maxted *et al*., 2012b) as they will provide a joint vision of gaps in conservation. For the evaluation of the *in situ* conservation status, three objectives were set: 1) to estimate the ratio of population occurrences in protected areas over total number of occurrences, 2) to assess the ratio of number of ecogeographic units covered by protected areas over total number of ecogeographic units where the species occurs and, 3) to evaluate the number and distribution of these ecogeographic units for each CWR to provide a perspective on the different degrees of habitat specialization occurring in the different CWRs.

3.2.1 In situ gap analysis

a) Species distribution data

Distribution data for 139 species out of 140 prioritized species in the food group, 167 out of the 184 prioritized species in the forage & fodder group, 154 species out of 168 prioritized in the ornamental group and 93 species out of 95 prioritized species in the industrial group were downloaded from the data portal of the Global Biodiversity Information Facility (data.gbif.org). Distribution data for the rest of the species were not available or did not appear in the GBIF data portal. Data were downloaded, species by species, taking into account possible mistakes in spelling and synonymy. The filters used to select the data to download were species' name (with the above-mentioned considerations) and country of origin (Spain).

Once all data were available, taxonomic harmonization was carried out following *Flora Iberica* (Castroviejo *et al*. 1986–2012) or the Anthos project (www.anthos.es). When any inconsistency was found between these sources, the Anthos project taxonomy prevailed over *Flora Iberica* taxonomy because the former is an online resource that is more updated than *Flora Iberica*.

The resulting taxonomically harmonized data were very heterogeneous in terms of the quality of georreferencing data. Therefore, to properly conduct the gap analysis, data records were selected by their quality according to the following criteria: a) Data records should have geographic coordinates that, expressed in decimal degrees, hold at least two decimal digits. This provides a resolution of less than 1 km; b) Data records should have available information on locality description to allow for an evaluation of the quality of their geographic coordinates. Therefore, those occurrences with the above-mentioned coordinate accuracy but no locality description were eliminated.

Based on the geographic coordinates, duplicate data were eliminated. Furthermore, when possible, discrimination between wild and cultivated forms of the species was made (e.g., in *Vitis* and *Brassica* species), and the latter were eliminated.

b) Gap analysis

All data were included into a GIS using ArcGIS software version 10.0 (ESRI 2010). Separate shapefiles for each use category (Food, Forage & Fodder, Ornamental and Industrial & Other Uses) were created. A layer with the political boundaries of Spain was used to select those populations within the limits of the country and eliminate those records with erroneous coordinates (e.g., falling in the sea or in neighbouring countries). The ELC map for the Iberian Peninsula and Balearic Islands (Fig. 9) developed by Parra–Quijano *et al*. (2012a) was overlaid, and the value corresponding to the ecogeographical unit was added to each record of all the populations. It was not possible to generate a similar map for the ecogeographical analysis of the populations occurring in the Canary Islands due to the insular character of this territory, rugged topography and the scarcity of meteorological stations.

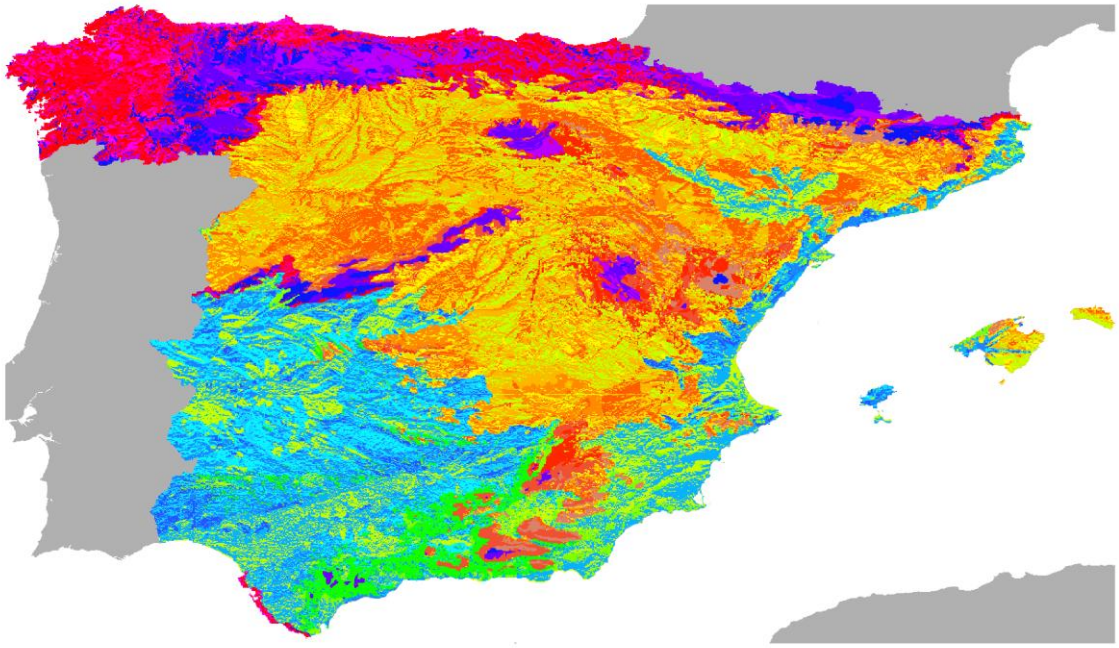


Figure 9: Ecogeographical Land Charaterization (ELC) map developed by Parra-Quijano *et al*. (2012a) for the Iberian Peninsula and Balearic Islands. Each color represents one of the 27 ecogeographic units found in the map, with a 1x1 km resolution.

In the *in situ* gap analysis, a layer with the polygons of the sites of community interest (SCI) in the Natura 2000 network of Spain was used to select all occurrences for each category falling within protected areas. This step was replicated with each genus using the SCI layer for the Canary Islands. The results obtained in both processes were

exported and merged into a single Excel file, containing all occurrences inside SCI per genus. Subsequently, the ratio of number of populations within protected areas over total number of populations was calculated for each species.

c) Areas of high richness of prioritized CWRs

To identify high-richness or hotspot areas where high concentrations of CWRs are found, four shapefiles containing all CWR species of each group (Food, Fodder & forage, Ornamental and Industrial & other uses categories) was created. Additionally, a joint shapefile for all CWRs under study was used for the overall evaluation of the complete list of CWR species under study. Using DIVA-GIS version 7.5 a 10x10 km grid was created and the resulting 10x10 km squares were hierarchically classified according to the number of species they held.

d) Complementarity analysis for the selection of areas

A complementarity analysis was performed in DIVA-GIS version 7.5 to select and order areas which contribute species to the network. This analysis first selects the location with the highest species richness; subsequently it selects an additional location containing the highest species richness after excluding those species already present in previously selected locations from the analysis. This process continues until all species under study are contained at least in one of the cells (Rebelo, 1994). Once again, the analysis was performed using a 10x10 km grid. This complementarity analysis allows the efficiency of an *in situ* conservation network to be maximized by gathering the highest number of focal CWR species in the lowest number of locations.

This complementarity analysis was carried out both for all four categories separately and with all CWR species with high quality occurrence data.

To complete the analysis, a gap analysis with the Sites of Community Interest in the Natura 2000 network was conducted to assess how many of the selected complementarity areas were already subject to passive protection.

3.2.2 *Ex situ* gap analysis

a) Seed accession data

In order to evaluate how well represented the prioritized CWRs are in genebanks, all existing national genebanks were contacted. These genebanks were the Spanish National Genebank for PGRFA (Centro de Recursos Fitogenéticos – INIA), which is the national reference seedbank for crops, landraces and CWR, the Andalusian Plant Germplasm Bank (BGVA) (Córdoba); the Atlantic Botanical Garden (BG-JBA) (Oviedo); the Botanical Garden of Barcelona (Barcelona); the Botanical Garden of Castilla La Mancha (BGV-JBCLM) (Albacete); the Botanical Garden of Soller (Mallorca); the Botanical Garden of the University of Valencia (Valencia), the Botanical Garden Viera y

Clavijo (JBVC) (Las Palmas de Gran Canaria); the César Gómez Campo germplasm bank of the Universidad Politénica de Madrid (BGV-UPM) (Madrid); the Marimurtra Botanical Garden (Gerona); the Royal Botanical Garden Juan Carlos I (Madrid); the Royal Botanical Garden of Madrid (RJBM) (Madrid); and the seedbank of the University Rey Juan Carlos (BG-URJC) (Madrid). In addition, data from the Germplasm Resources Information Network (GRIN) of the United States Department of Agriculture (USDA-GRIN, http://www.ars-grin.gov/) and the EURISCO catalogue (http://eurisco.ecpgr.org/home_page.html), a web-based catalogue encouraged by the European Cooperative Programme for Plant Genetic Resources (ECPGR) (http://www.ecpgr.cgiar.org) and managed by Bioversity International (http://www.bioversityinternational.org), were also collected and used.

All institutions were asked to collaborate with information on number of accessions native to Spain for the selected species and the coordinates of the collecting sites. This information was obtained only for the species in the Plant Genetic Resources for Food and Agriculture group (Food and Fodder & Forage categories) which hold 324 species.

3.3 Results and discussion

3.3.1 *In situ* gap analysis

a) Distribution data

The selection of distribution data records according to the established standards of quality resulted in a reduction of data occurrences and the number of species under analysis. In any case, data were obtained for 87.9% of the prioritized species (Table 6). A comprehensive list with the number of data records initially downloaded for each species, the final number of records for each species as well as the species with no data is accessible at http://pgrsecurespain.weebly.com/crop-wild-relatives-in-spain-ndash-gap-analysis-for-the-in-situ-conservation-assessment.html.

Although it was possible to obtain occurrence data for a high percentage of the focal CWR species, 22% (117 out of 515) of these species had fewer than 10 localities with high-quality occurrence data. This threshold of 10 localities has been identified by Gotelli & Ellison (2004) as the minimum number of locations needed to ensure a reliable assessment. These results suggest that further work in the inventorying and georeferencing of Spanish plant biodiversity is needed. To get to know which species are found in a given territory is essential for the implementation of effective and coherent conservation policies. Fortunately, some steps in that direction are foreseen for the coming years (MAGRAMA, 2013). In any case, it must be taken into account that in some cases the low number of occurrences truly represents the current distribution of the species, as they are narrowly distributed endemics. This is the case

of *Lupinus mariae-josephae* H. Pascual, *Festuca curvifolia* Lag. ex Lange or *Argyranthemum spp.*, among others.

Table 6: Number of prioritized Spanish CWR species and number of prioritized Spanish CWR species with high-quality occurrence data classified by use category.

Group	Number of prioritized CWR species	Number of prioritized CWR species with high-quality occurrence data
Food	140	123
Forage & Fodder	184	162
Ornamental	161	138
Industrial	95	87
TOTAL	580	510

b) Gap analysis

b.1) Ratio of number of CWR populations occurring in protected areas to number of populations in the overall distribution of prioritized CWRs

Overall 42±24% (Mean±Standard Deviation) of the populations of species in the Spanish National Inventory under analysis is located in Sites of Community Interest (SCI) belonging to the Natura 2000 network. Results grouped by use categories show that the ratio of CWR population occurrences in protected areas (PA) over total occurrences is 37±24% (M±SD) in the Food category, 47±23% (M±SD) in the Fodder & Forage category, 39±26% (M±SD) in the Ornamental category and 41 ± 21% (M±SD) in the Industrial category (Fig. 10).

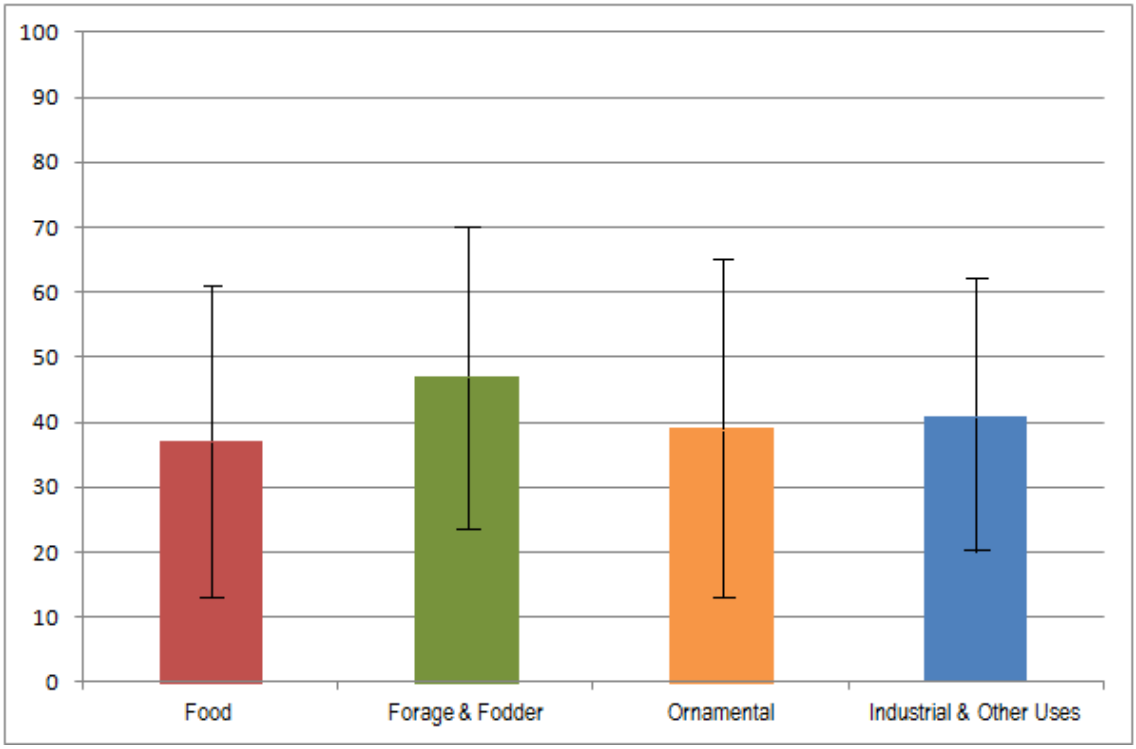


Figure 10: Percentage of prioritized CWR populations occurring in protected areas (Sites of community interest in the Natura 2000 network of Spain) per use category (mean values ± standard deviations).

We found that 38 species in the National Inventory did not have even a single population occurring within a protected area (12 in the Food category, 5 in the Fodder & Forage category, 18 species in the Ornamental category and 3 species in the Industrial category) (Table 7). On the contrary, all recorded populations of 18 species fell within protected areas (5 species in the Food category, 10 in the Fodder & Forage category and 5 in the Ornamental category). It should be noted that only three out of these 55 (37+18) species (*Festuca pseudeskia* Boiss., *Narcissus cyclamineus* DC.and *Sideritis reverchonii* Willk.) have more than 10 recorded populations in our analysis, 25 species have 6 or fewer data occurrences and 27 species have just one recorded population.

Table 7: Prioritized CWR species of Spain that have no known populations in Protected Areas (PA) using as a reference base the Sites of Community Interest of the Natura 2000 network.

Category	*SPECIES*	*Number of georeferenced populations*
Food	*Allium pruinatum*	1
Food	*Allium schmitzii*	2
Food	*Allium stearnii*	1
Food	*Asparagus fallax*	1
Food	*Asparagus nesiotes*	2
Food	*Asparagus pastorianus*	6

Table 7: Prioritized CWR species of Spain that have no known populations in Protected Areas (PA) using as a reference base the Sites of Community Interest of the Natura 2000 network.

Category	*SPECIES*	*Number of georeferenced populations*
Food	*Asparagus plocamoides*	2
Food	*Cyanara tournefortii*	5
Food	*Lactuca livida*	1
Food	*Patellifolia procumbens*	2
Food	*Vicia argentea*	4
Food	*Vicia scandens*	1
Forage & Fodder	*Astragalus cavanillessii*	3
Forage & Fodder	*Festuca agustinii*	1
Forage & Fodder	*Festuca graniticola*	1
Forage & Fodder	*Festuca longiauriculata*	1
Forage & Fodder	*Trifolium lucanicum*	2
Ornamental	*Argyranthemum broussonetii*	3
Ornamental	*Argyranthemum foeniculaceum*	1
Ornamental	*Argyranthemum winteri*	1
Ornamental	*Limonium arborescens*	1
Ornamental	*Limonium auriculae-ursifolium*	1
Ornamental	*Limonium bourgeaui*	3
Ornamental	*Limonium carthaginense*	2
Ornamental	*Limonium dodartii*	1
Ornamental	*Limonium fruticans*	2
Ornamental	*Limonium hibericum*	1
Ornamental	*Limonium pectinatum*	5
Ornamental	*Limonium puberulum*	1
Ornamental	*Limonium revolutum*	1
Ornamental	*Limonium subglabrum*	5
Ornamental	*Limonium tuberculatum*	2
Ornamental	*Narcissus cyclamineus*	12
Ornamental	*Narcissus elegans*	2
Ornamental	*Narcissus perez-chiscanoi*	2
Industrial & Other Uses	*Sideritis lotsyi*	1
Industrial & Other Uses	*Sideritis reverchonii*	17
Industrial & Other Uses	*Sideritis serrata*	6

If we were to consider that a species has adequate *in situ* passive protection when at least 75% of the known populations occur inside a protected area, 51 species would meet this threshold (11 species in the Food category, 19 in the Fodder & Forage category, 12 in the Ornamental group and nine in the Industrial & Other Uses group). Once again, only 21 of these species have more than 10 population occurrences under analysis.

A species-by-species analysis of these results for all use categories can be found at http://pgrsecurespain.weebly.com/crop-wild-relatives-in-spain-ndash-gap-analysis-for-the-in-situ-conservation-assessment.html.

Overall, the high mean value of populations lying in protected areas (42%) suggests that a good representation of the Spanish CWR is passively protected in protected areas. Nevertheless, the high variability obtained in this parameter depending on the species (represented by the high standard deviation values) and the uncertainty generated by the lack of high-quality occurrence data suggests that these results should be considered cautiously and that more detailed studies should be encouraged for those species with a low number of recorded populations. Specifically, species with five or fewer populations under study should be prioritized for individual analysis of their conservation status, as they are more susceptible to anthropic actions or stochastic events in a short term period (Maxted *et al.*, 2008b)

b.2) Ratio of number of ecogeographic units in populations occurring in protected areas to number of ecogeographic units in the overall distribution of prioritized CWRs

A total of 66±27% (M±SD) of the ecogeographic units, where the prioritized CWR species of the Iberian Peninsula and Balearic Islands are found, are represented in the populations that occur in protected areas in the Natura 2000 network. Results per use categories show that the ratio of ecogeographic units represented in populations in protected areas is 65±27% (M±SD) for the Food category, 69±26% (M±SD) for the Fodder & Forage category, 60±29% (M±SD) for the Ornamental category and 69±22% (M±SD) for the Industrial category (Fig. 11).

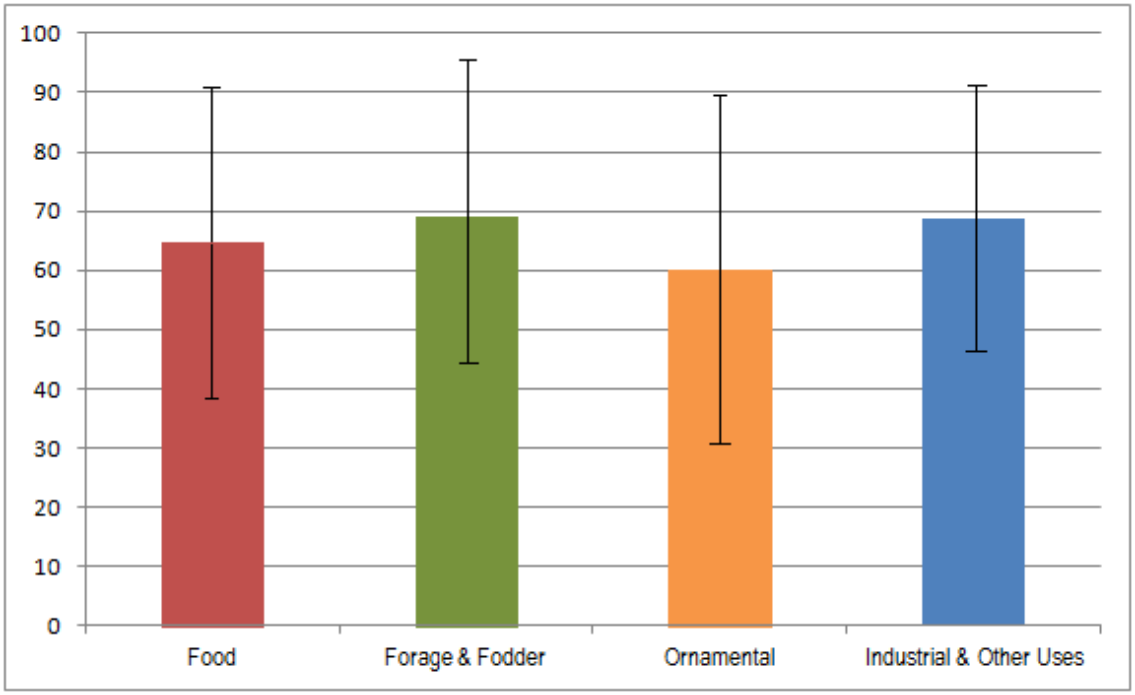


Figure 11: Percentage of ecogeographic units present in populations occurring in protected areas in relation to those in the overall distribution of prioritized CWRs, structured by use categories (mean values ± standard deviations).

All of the ecogeographic units of 70 species are represented in populations occurring in protected areas (16 species in the Food category, 29 species in the Fodder & Forage category, 16 species in the Ornamental category and 9 species in the Industrial category).

If we were to consider that the genetic diversity of a CWR species is well conserved when at least 75% of the ecogeographic units of the target CWR species is represented by populations occurring in protected areas, 215 species would meet this threshold (51 species in the Food category, 81 in the Fodder & Forage category, 45 in the Ornamental category and 38 in the Industrial category).

The high mean value (66%) of the ratio of number of ecogeographic units in populations occurring in protected areas to number of ecogeographic units in the overall distribution of prioritized CWRs shows that an acceptable representation of the genetic diversity of Spanish CWRs is well covered by the SCI in the Natura 2000 network. Once again, the high variability in this parameter depending on the species under consideration and the uncertainty generated by the lack of high-quality occurrence data indicate that these results should be interpreted with caution and that additional efforts should be made to improve the quantity and quality of occurrence data. The ecogeographical map used in this study for the Spanish CWR conservation assessment has proved to be an effective indicator of phenotypic diversity and

adaptation to different environmental conditions in other plant species (Parra-Quijano *et al.*, 2012a) and has been used in the generation of optimized seed collecting strategies (Parra-Quijano *et al.*, 2012b). Similarly, this map can be applied in the selection of populations that represent the genetic diversity of the species for *in situ* conservation actions.

b.3) Number and distribution of ecogeographic units in prioritized CWRs

The number of ecogeographic units in which a species is distributed and the number of populations found in each type of ecogeographic unit provides information on the species' distribution and the different habitats in which the species can be found. In total, 198 CWR species of the National Inventory are found in 15 or more different types of ecogeographic units. Of these species, 54 have a completely generalist distribution and are found in all 27 ecogeographic units in which the Iberian Peninsula and Balearic Islands are distributed (Fig. 12).

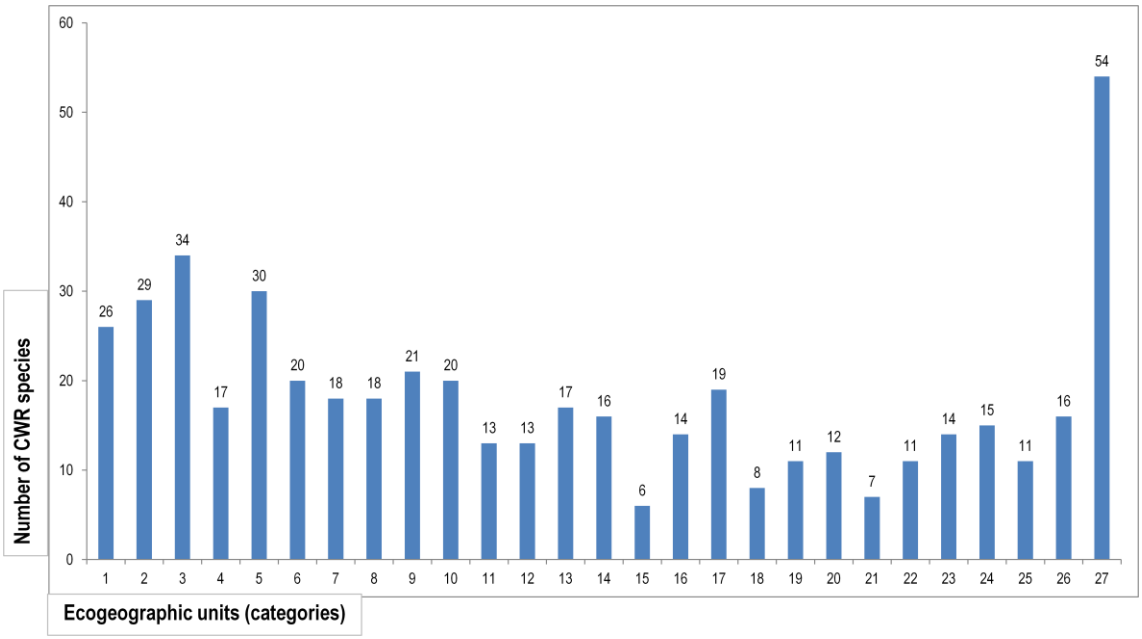


Figure 12: Histogram of the number of ecogeographic units found in the distribution of prioritized CWR species, based on an Ecogeographic Land Characterization map of the Iberian Peninsula and Balearic Islands composed of 27 ecogeographic units (Parra-Quijano *et al.*, 2012a).

The results obtained per species can be found at http://pgrsecurespain.weebly.com/crop-wild-relatives-in-spain-ndash-gap-analysis-for-the-in-situ-conservation-assessment.html.

The use of ELC maps for the generation of detailed information on the genetic representativeness of each CWR species in a given territory, such as the Iberian Peninsula, allows us to identify the most common and rarest environments where the

taxa can be found. Populations occurring in the most common environments are the most representative of the species, whereas populations occurring in rare environments may provide valuable genetic diversity in terms of adaptation to different and extraordinary environmental conditions (Sexton *et al*., 2011). However, populations occurring in rare habitats may be located at the edge-of-range distribution of the species and may have lower population densities (Sagarin & Gaines, 2002), and thus, reduced genetic diversity according to the abundant-center distribution concept (Eckert *et al*., 2008).

c) Areas of high richness of prioritized CWRs

The richness analysis was completed separately for each of the four use categories (Food, Forage & Fodder, Ornamental and Industrial & Other Uses) to focus on specific groups to cater to breeders' needs. On the other hand, the joint evaluation of all CWRs in the National Inventory provides a general overview of the distribution of CWR diversity in Spain which is helpful when proposing conservation actions involving national or regional governments.

The global richness analysis, using the joint shapefile for all prioritized CWRs, identified fourteen 10x10 km areas as the richest in number of species. Fourteen areas were selected as hostspots of CWR diversity. The two locations with the highest species richness were found in the province of Navarra (79 species each). Seventy-eight species were found in one location in Girona province and 77 in one location in Córdoba province (Fig. 13). Although other richness areas are located in East Spain and South West Spain, generally associated to mountainous systems, the highest richness areas are located in the Northern and Northeastern regions of the Iberian Peninsula.

A general overview of distribution data and CWR richness areas situates most of the diversity in mountainous systems but specially in the Pyrenees and in Navarra region, in North Spain. This partly coincides with some of the areas where higher plant richness is expected in the Mediterranean region locating in Spain four important hotspots of diversity (Baetic and Sub-baetic mountains, Canary Islands, Pyrenees and Balearic Islands (Médail & Quézel, 1999). However, the higher diversity found in Northern Spain may also respond to the greater investments in the inventorying of flora carried out in these regions.

When the analysis was carried out separately for the CWRs belonging to the Food category, the area with the greatest species richness is in the Navarra region in Northern Spain with 30 species, followed by a location in the province of Cordoba with 29 species. Finally, two additional areas have 25 species each and are located in Navarra and in the province of Girona (Fig. 14).

Regarding the Fodder & Forage category, the richness analysis selected 14 locations (10x10 km) as hotspots of CWR diversity. The area with the greatest species richness is in the province of Girona in Catalonia with 40 species. The next four areas encompass 37 species each and are located in the provinces of Barcelona, Cáceres, Badajoz and Huelva (Fig. 15).

In the Ornamental category, ten 10x10 km areas had the highest species richness (between 12 and 14 species). The higher densities (14 species) are found in the provinces of Navarra, and Lleida. The next five areas hold 13 species each in the provinces of León, Álava, Huesca, Lleida and Barcelona (Fig. 16).

In the Industrial category, 22 areas (10x10 km grid) had the highest richness, holding between 14 and 16 species. Five areas have 16 species in the provinces of Lleida, Málaga, Granada (two), whereas ten areas hold 15 species in the provinces of Barcelona (two), Girona, Guadalajara, Cuenca, Córdoba, Málaga (two) and Granada (two) (Fig. 17).

All this information is available at: http://pgrsecurespain.weebly.com/crop-wild-relatives-in-spain-ndash-richness--complementarity-analysis-for-the-in-situ-conservation-assessment.html.

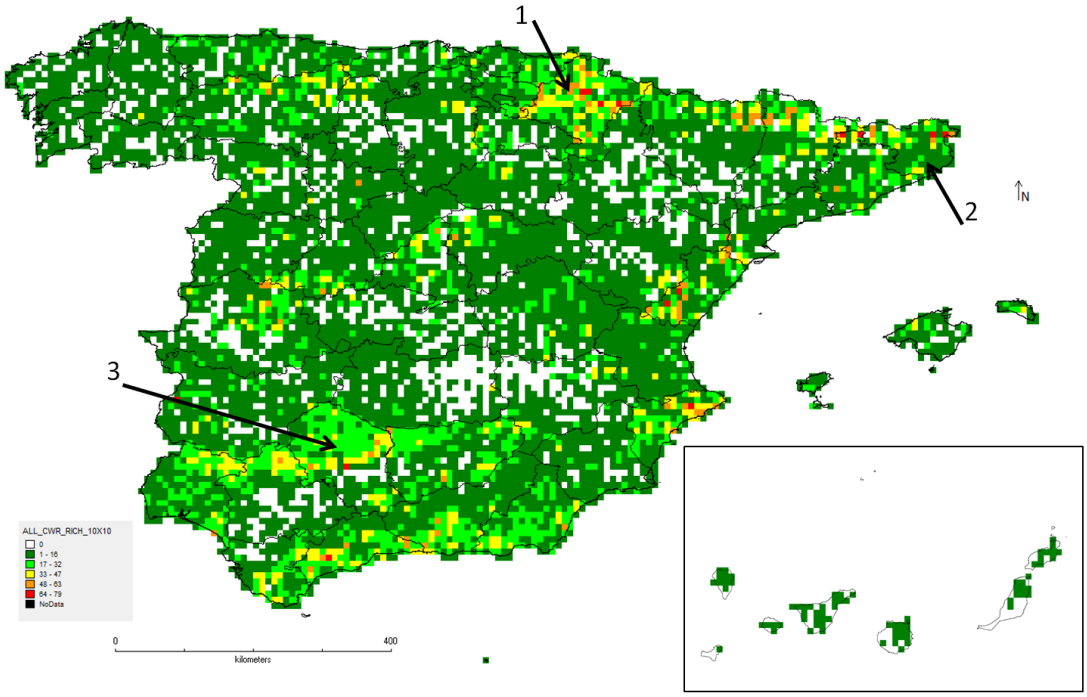


Figure 13: Hotspot areas (10 x 10 km) for all CWR species in the Spanish National Inventory. Red areas encompass the highest number of CWR species. The numbers point to locations where the highest number of species is found (Provinces where locations are found: 1=Navarra, 2=Girona, 3=Córdoba).

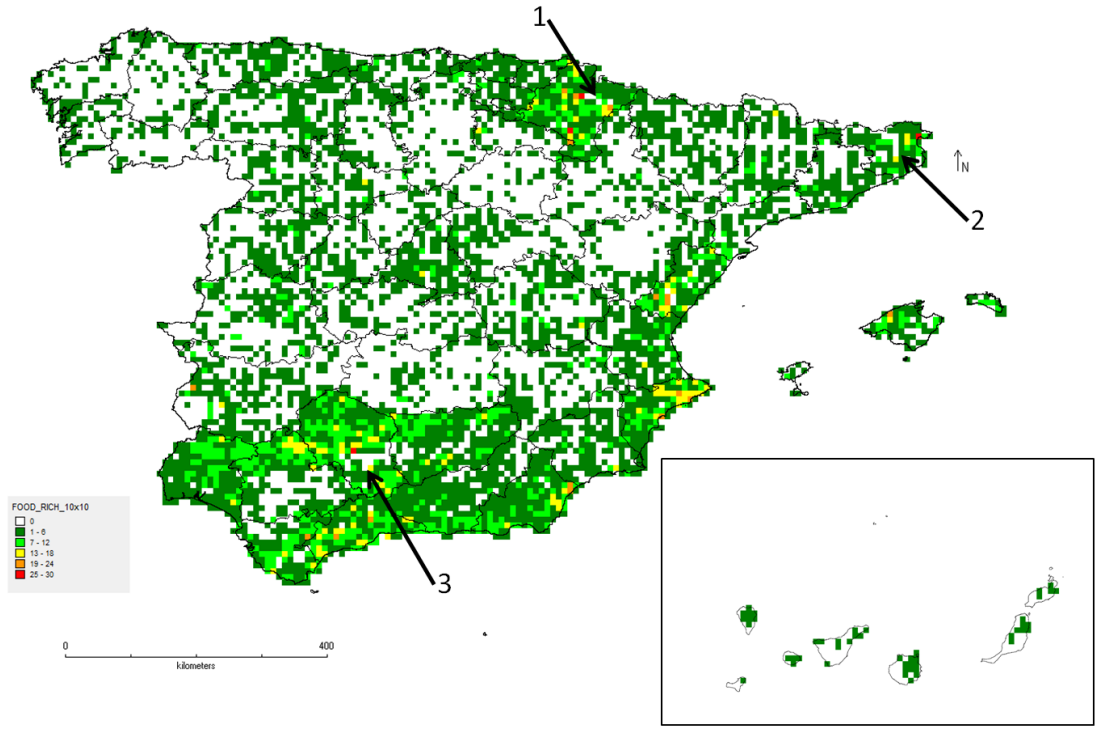


Figure 14: Hotspot areas (10 x 10 km) for the FOOD crops related species in the National Inventory. Red areas encompass the highest number of CWR species. The numbers point to locations where the highest number of species is found (Provinces where locations are found: 1=Navarra, 2=Girona, 3=Córdoba).

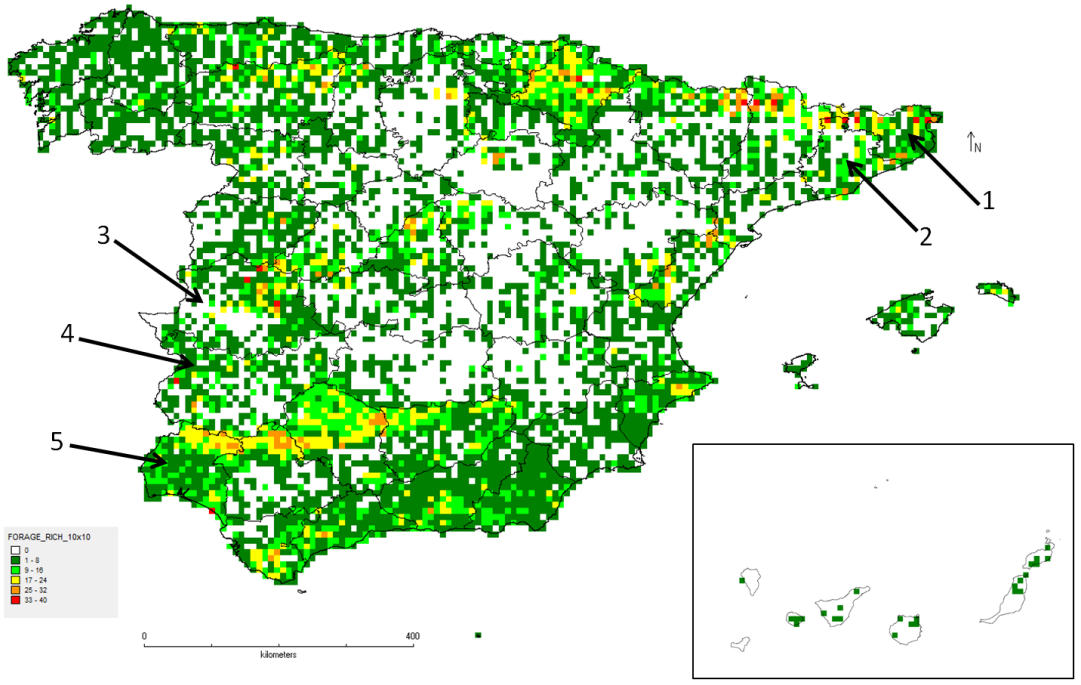


Figure 15: Hotspot areas (10 x 10 km) for the Fodder & Forage crops related species in the National Inventory. Red areas encompass the highest number of CWR species. The numbers point to locations where the highest number of species is found (Provinces where locations are found: 1=Girona, 2=Barcelona, 3=Cáceres, 4=Badajoz and 5=Huelva)

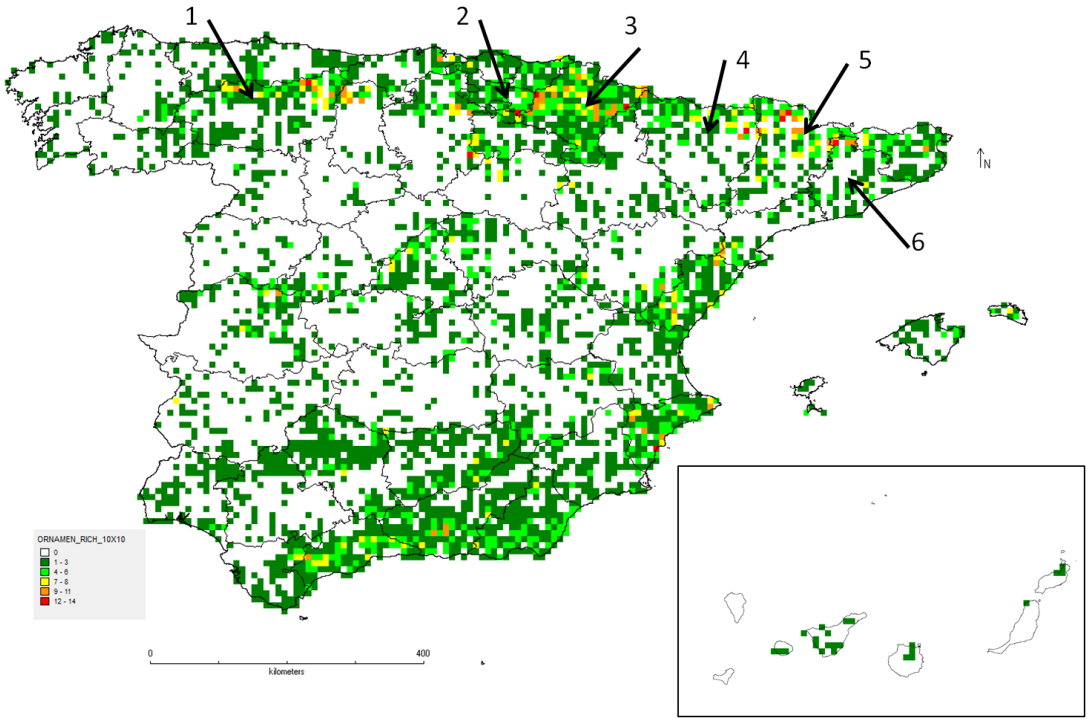


Figure 16: Hotspot areas (10 x 10 km) for the Ornamental crop related species in the National Inventory. Red areas encompass the highest number of CWR species. The numbers point to locations where the highest number of species is found (Provinces where locations are found: 1=León, 2=Álava, 3=Navarra, 4=Huesca, 5=Girona and 6=Barcelona).

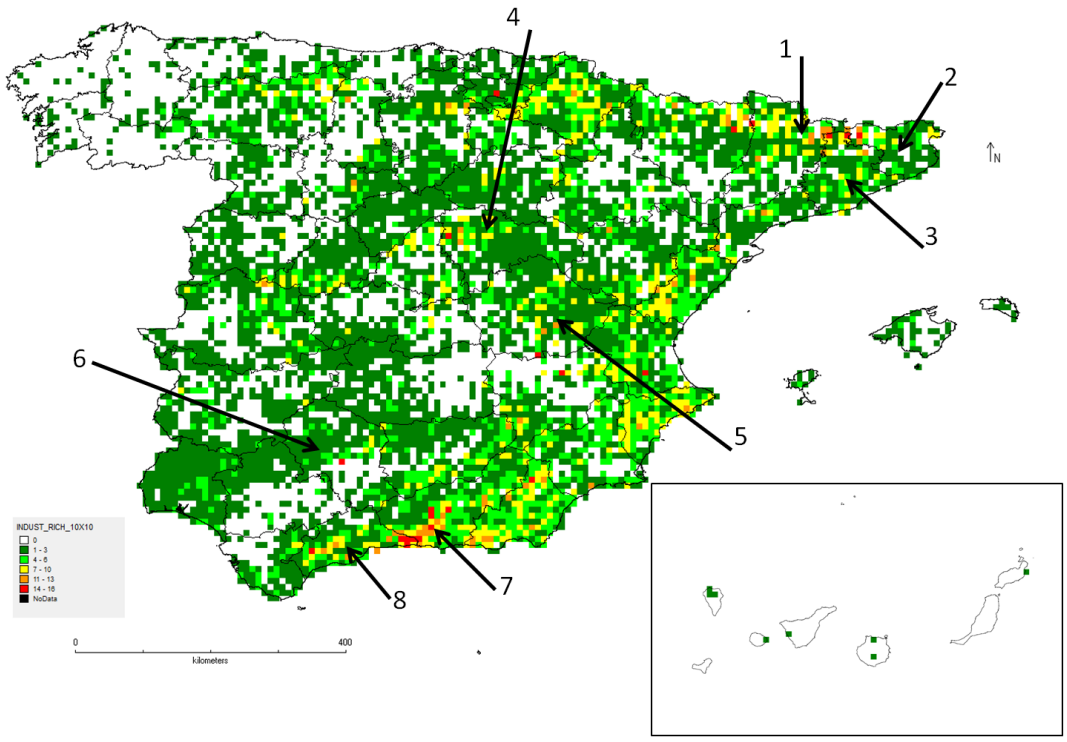


Figure 17: Hotspot areas (10 x 10 km) for the Industrial crop related species in the National Inventory. Red areas encompass the highest number of CWR species. The numbers point to locations where the highest number of species is found (Provinces where locations are found: 1=Lleida, 2=Girona, 3=Barcelona, 4=Guadalajara, 5=Cuenca, 6=Córdoba, 7=Granada and 8=Málaga).

d) Complementarity analysis for the selection of priority areas for conservation

The global complementarity analysis carried out for all category uses showed that 122 sites (10x10 km grid) would be required to protect all 508 CWR species under analysis (Fig. 18). This result is far from being a reasonable number of places to be proposed as genetic reserves, both economically and geographically speaking. If we considered that preserving two thirds of the prioritized CWR species *in situ* is a suitable objective for the short medium term (Maxted *et al*., 2007), only 20 sites would be required (Fig. 19). Each of these 20 sites contributes at least five species to the network up to a total of 333 species. This result is in concordance with the one obtained by Maxted *et al*. (2007) in which 17 sites were proposed as genetic reserves to cover two thirds of their CWR species. Five of these 20 sites are found inside Sites of Community Interest (SCI) of the Natura 2000 network and the 83 CWRs occurring there are hence, under passive protection. Furthermore, 7 additional sites are partially included in SCIs, and thus, easily assimilable to the protection network. Finally, three additional sites are outside SCI but located close to them (less than 10 km away) (Table 8). If all these 15 sites were provided protection through the Natura 2000 network, 210 CWR species would be covered by them (about 41.3% of the prioritized CWR species). Special emphasis on the creation of a genetic reserve must be put in the case of the first selected area in the complementarity analysis; this area is located in Navarra province, being the richest 10x10 km area in Spain with 79 species, but outside the SCIs in the Natura 2000 network. The implementation of this genetic reserve, together with the adhesion of the sites mentioned above would provide passive protection to 289 CWR (≈60% of the CWR diversity). In a similar work carried out in UK, nine of the 17 areas selected (≈52.9%) through the complementarity analysis were already under passive protection. Still very few references regarding this kind of analysis are available for CWR inventories in other countries although many studies are currently being performed in this direction. Current recommendations for the *in situ* conservation assessment and the design of protection plans for CWR are, indeed, directing steps in this way (Maxted *et al*., 2012a; 2012b).

Table 8: Priority ranking of the selected complementarity areas that are found totally or partially inside Sites of Community Interest of the Natura 2000 Network or in their surroundings. Name of the Site of Community Interest, site code, bioregion and coverage of the priority CWR is shown.

Priority ranking	Name of SCI	Site code	Bioregion	Coverage
2	Sierra de las Nieves	ES6170006	MED	Inside SCI
3	Prepirineu Central català	ES0000018	ALP/MED	Less than 10 km away
	Montgrony	ES5120024	ALP	
4	Aiguamolls de l'Alt	ES0000019	MED	Partially inside

Table 8: Priority ranking of the selected complementarity areas that are found totally or partially inside Sites of Community Interest of the Natura 2000 Network or in their surroundings. Name of the Site of Community Interest, site code, bioregion and coverage of the priority CWR is shown.

Priority ranking	Name of SCI	Site code	Bioregion	Coverage
	Empordà			
6	Sierras Almagrera, de los Pinos y el Aguilón	ES6110012	MED	Partially inside
	Sierra del Alto Almagro	ES6110011	MED	
7	Cuenca del Río Lozoya y Sierra Norte	ES3110002	MED	Inside SCI
8	Sierra de Salinas	ES5213039	MED	Less than 10 km away
	Saleo y Cabecicos Villena	ES5212007	MED	
10	Valle de San Emiliano	ES4130035	ATL	Inside SCI
	Alto Sil	ES0000210	ATL/MED	
11	Sierra Nevada	ES6140004	MED	Partially inside
12	Sierras de Gador y Enix	ES6110008	MED	Inside SCI
13	Jaizkibel	ES2120017	ATL	Partially inside
	Txingudi-Bidasoa	ES2120018	ATL	
	Aiako Harria	ES2120016	ATL	
14	Ríos Alagón y Jerte	ES4320071	MED	Less than 10 km away
	Sierra de Gredos y Valle del Jerte	ES4320038	MED	
16	Larrondo-Lakartxela	ES2200009	ALP	Partially inside
	Larra-Aztaparreta	ES0000123	ALP	
18	Aigüestortes	ES0000022	ALP/MED	Inside SCI
19	Desierto de Tabernas	ES0000047	MED	Partially inside
	Ramblas del Gergal, Tabernas y Sur de Sierra Alhamilla	ES6110006	MED	
20	Sierra de Camarolos	ES6170012	MED	Partially inside
	Río Guadalmedina	ES6170028	MED	

As with the richness analysis, separate analyses for each CWR use category were carried out in order to facilitate the access to specific genetic resources according to the breeders' needs.

In the Food category, the complementarity analysis selected 42 areas (10x10 km grid) that encompassed all 125 species under study. Out of these, the first 15 sites held 78% of the species, and each site contributed 3 or more additional species. Six of these areas were located in protected areas, five areas were located partially inside of a PA, and one area (number eight in the sequence contributing with three species) is located

very close to one SCI. The first and the second selected areas contributed 30 and 15 additional species, respectively (Figs. 20a). Again, the first selected area is not located in a PA. Thus its inclusion in the network or the creation of a genetic reserve for its preservation is highly recommended as it represents a valuable source of plant genetic resources for human food crops. The second selected area is partially overlapping to a PA and the extension of it would provide with passive protection to another 15 species. If these two new areas were designated, the passive protection of at least 60 species would be guaranteed.

In the Fodder & Forage category, 35 areas (10x10 km grid) encompass the 162 species under analysis. The first 14 sites contribute 3 or more additional species to the network and hold 82% of the species. Six of these 14 sites are located in protected areas, three are partially covered by the network. Finally, four areas are located less than 10 km away of PA, so they could be easily annexed to the network. The first four selected areas contribute more than ten additional species to the network (40, 24, 14 and 10, respectively) (Fig. 20b). Except for the third selected area which is not covered by any PA, the rest of them are total or partially in the Natura 2000 network. If genetic reserves were established in these areas and in the one which is not still protected, 82% of the prioritized CWR species of Forage and Fodder crops would be protected.

In the Ornamental category, 61 areas (10x10 km grid) encompass the 138 species under analysis. The first 13 sites contribute 3 or more additional species and hold 53% of the species. Six of these 13 areas are in protected areas, two areas are partially covered by the network, and, finally, three other areas are located less than 10 km away of PA, so they could be easily annexed to the network. The first two selected areas contribute more than ten additional species to the network (14 and 11, respectively) and are both partially covered by SCIs (Fig. 21a).

In the Industrial & other uses category, 29 areas (10x10 km grid) encompass the 87 species under analysis. The first 10 sites contribute 3 or more species and hold 37% of the species. Four of these 10 sites are located in protected areas and two sites are located less than 10 km away from a PA so they could be easily annexed to the network. The first two selected areas contribute more than ten additional species to the network (16 and 14, respectively) (Fig. 21b).

Complementarity areas identified for specific use categories have some coincident points with the global complementarity areas analysis, but each has a quite distinct pattern. The structuring of the complementarity analysis into specific use categories reduces the global efficiency of the proposed areas (more areas are needed to preserve a similar number of CWR), but accommodates the profile for the possibility of private companies engaging in the funding and sustainment of genetic reserves

oriented to the conservation of specific use categories of CWR. Thus, human food multinational companies might be interested in supporting genetic reserves specialized in the conservation of prioritized CWR belonging to the Food category.

All this information is available at: http://pgrsecurespain.weebly.com/crop-wild-relatives-in-spain-ndash-richness--complementarity-analysis-for-the-in-situ-conservation-assessment.html.

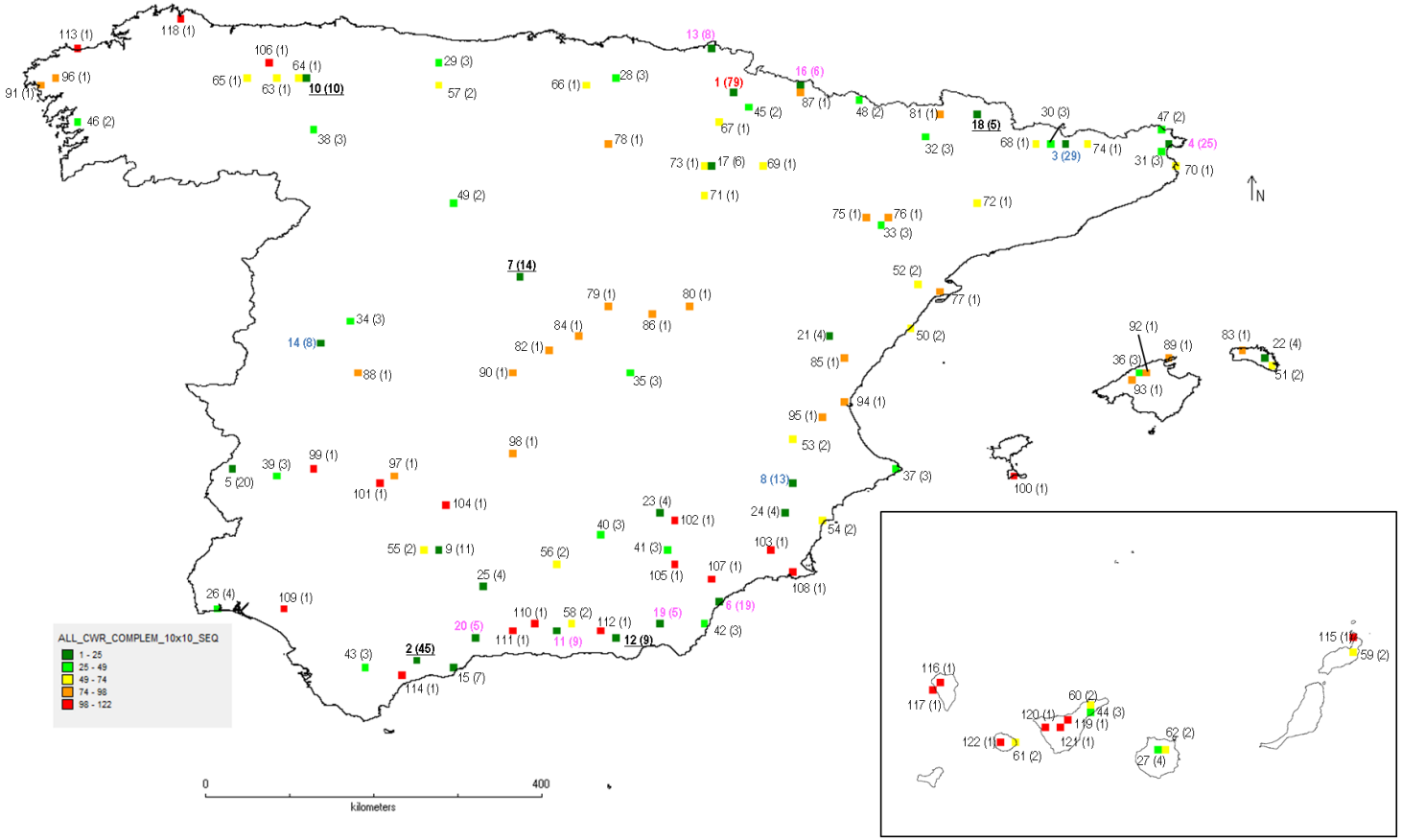


Figure 18: Complementarity areas needed to cover all CWR species in the Spanish National Inventory for protection. Selection ranking order and number of additional species added to the network (in brackets) are presented for each site. Underlined and bold numbers tag those areas that are inside protected areas (PA). Number in red points out the first area selected according to the number of CWR species added to the network, but outside the Natura 2000 network of SCI. Pink numbers show areas partially overlapping with a PA and blue numbers point out areas less than 10 km away from a PA in Natura 2000 network. Selection ranking order is also described by the color of the square pointing to the location of the site according to the information indicated in the legend.

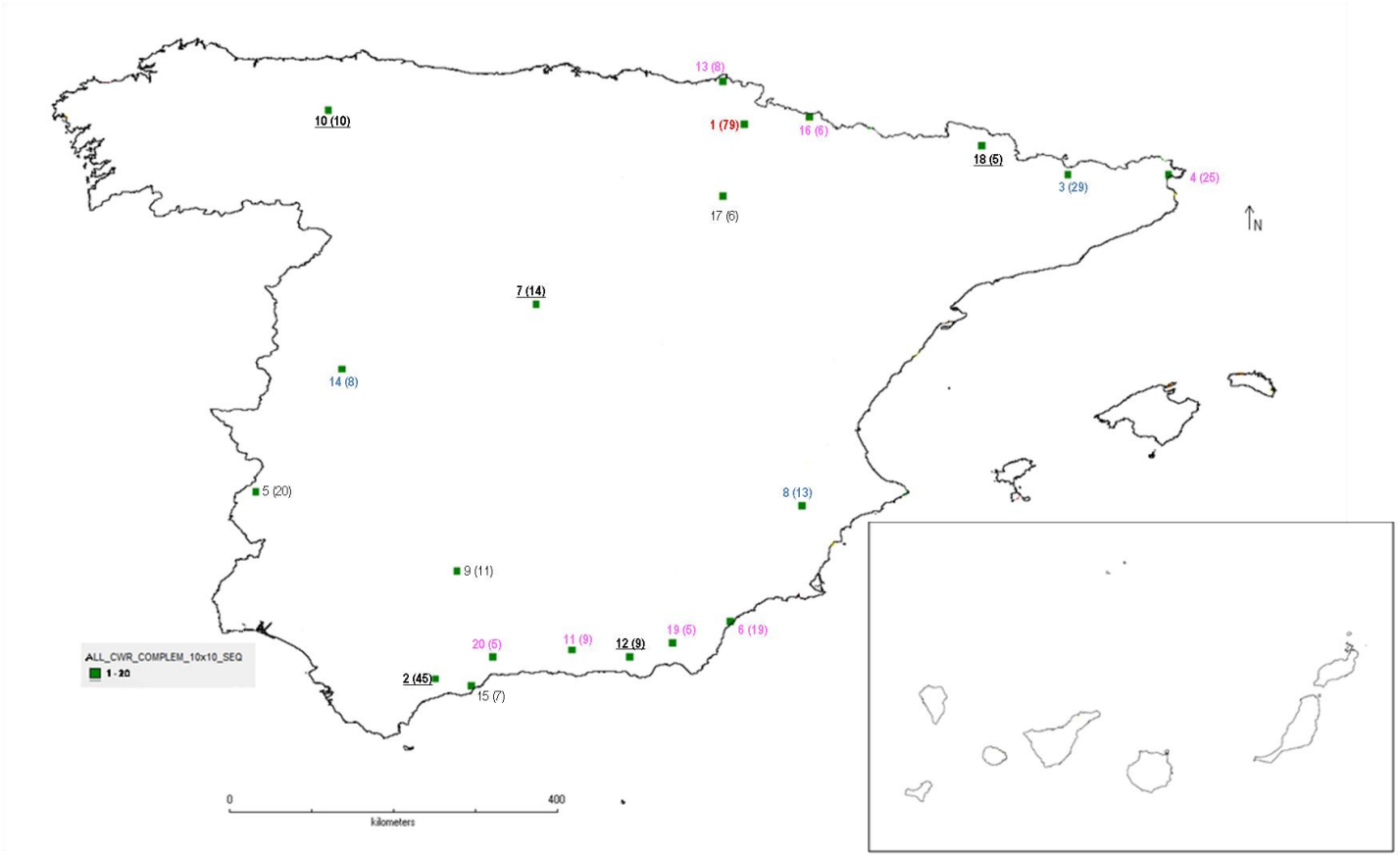


Figure 19: Complementarity areas selected to cover two thirds of all CWR species in the Spanish National Inventory for protection. Selection ranking order and number of additional species added to the network (in brackets) are presented for each site. Underlined and bold numbers tag those areas that are inside protected areas (PA). Number in red points out the first area selected according to the number of CWR species added to the network, but outside the Natura 2000 network of SCI. Pink numbers show areas partially overlapping with a PA and blue numbers point out areas less than 10 km away from a PA in Natura 2000 network.

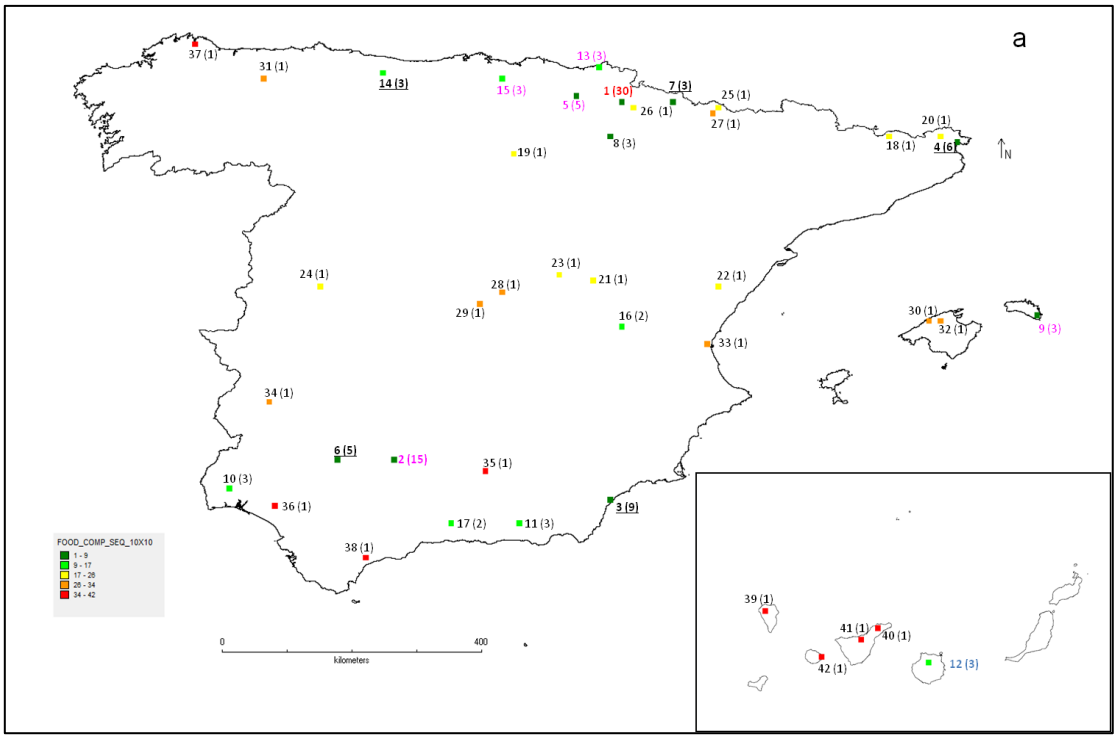
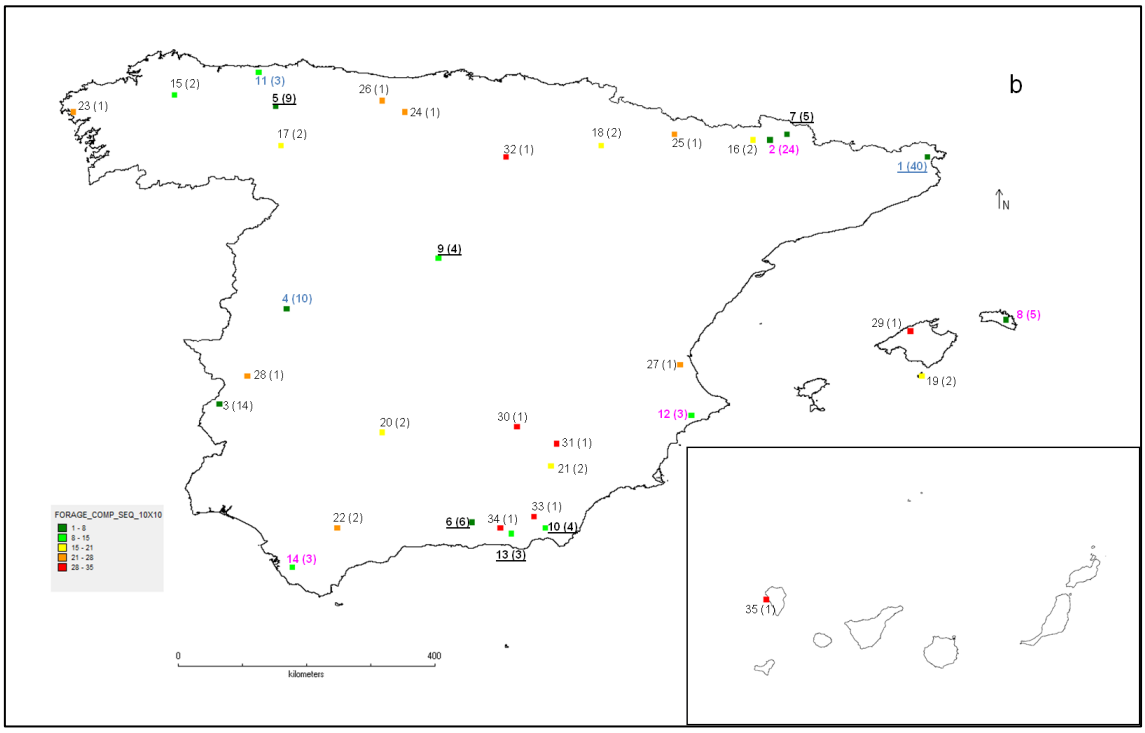


Figure 20: Complementarity areas selected to cover (a) Food and (b) Forage and Fodder species in the Spanish National Inventory for protection. Selection ranking order and number of additional species added to the network (in brackets) are presented for each site. Underlined and bold numbers tag those areas that are inside protected areas (PA). Pink numbers show areas partially overlapping with a PA and blue numbers point out areas less than 10 km away from a PA in Natura 2000 network. Selection ranking order is also described by the color of the square pointing to the location of the site according to the information indicated in the legend.



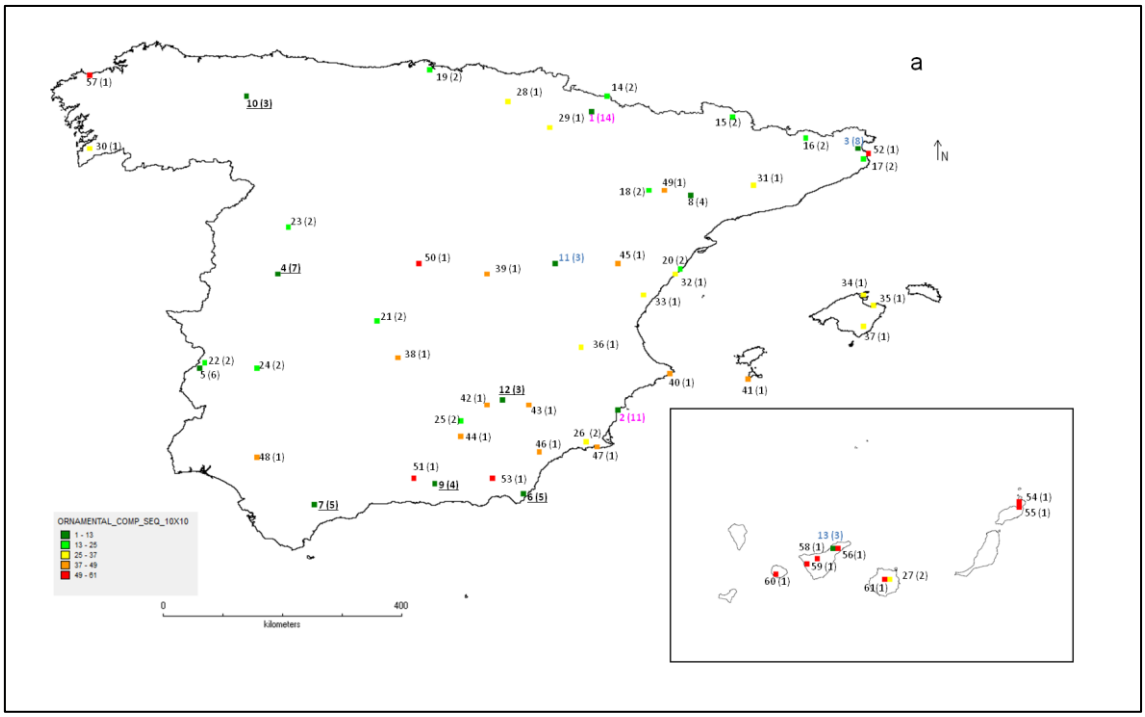
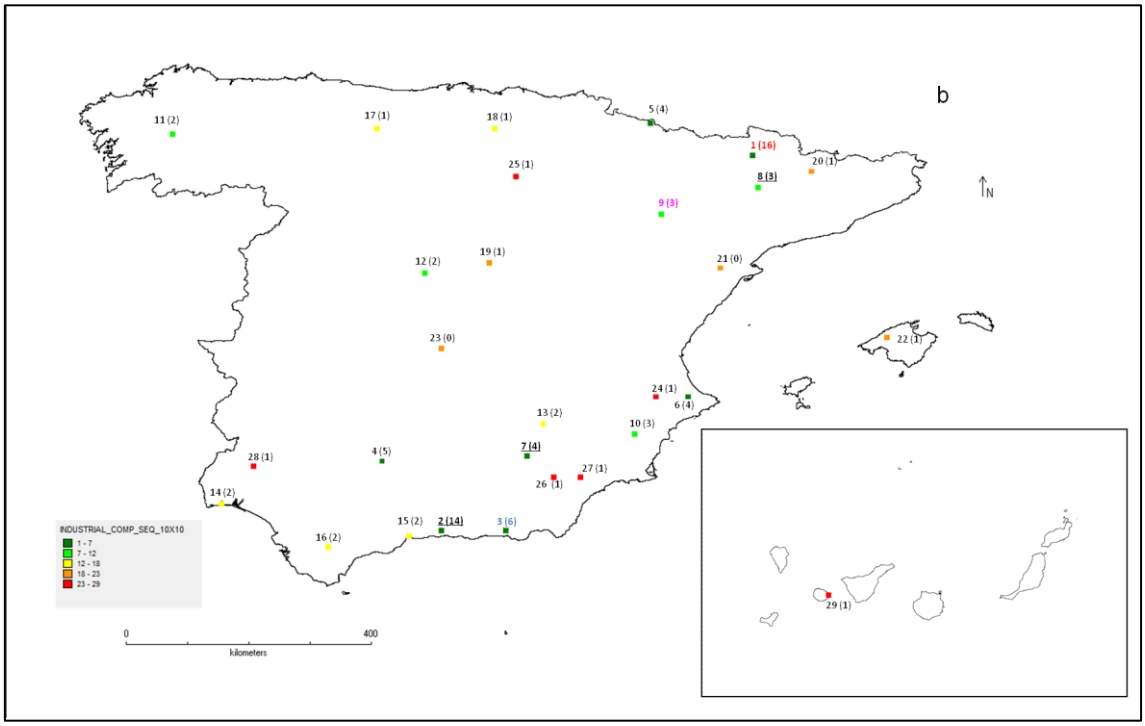


Figure 21: Complementarity areas selected to cover (a) Ornamental and (b) Industrial species in the Spanish National Inventory for protection. Selection ranking order and number of additional species added to the network (in brackets) are presented for each site. Underlined and bold numbers tag those areas that are inside protected areas (PA). Pink numbers show areas partially overlapping with a PA and blue numbers point out areas less than 10 km away from a PA in Natura 2000 network. Selection ranking order is also described by the color of the square pointing to the location of the site according to the information indicated in the legend.



Supplementary information on the presence of complementarity areas in existing protected areas is available at http://pgrsecurespain.weebly.com/crop-wild-relatives-in-spain-ndash-richness--complementarity-analysis-for-the-in-situ-conservation-assessment.html.

3.3.2 *Ex situ* gap analysis

a) Seed accession data

Data from germplasm banks and contacted institutions were collated and ordered in a joint file which can be found at http://pgrsecurespain.weebly.com/crop-wild-relatives-in-spain-ndash-ex-situ-assessement.html.These results show that 71% of prioritized PGRFA CWR species is represented in the network of Spanish genebanks.

Ninety-five of 324 PGRFA CWR species have no accessions preserved in any of the germplasm banks and institutions contacted. Furthermore, 100 of the 229 species that are represented in the germplasm banks have between 1 and 5 accessions preserved in banks (table 9), whereas just 60 have 20 or more accessions conserved in *ex situ* collections.

Considering that five populations is the minimum number of different original populations that should be represented in a genebank for each CWR species to adequately cover the genetic diversity of the species (Maxted *et al*., 2008b), results show the necessity to improve the status of CWR germplasm collections, as only 160 CWR species of the PGRFA group have more than 5 accessions in germplasm collections. It must be noted also that coordinates of these accessions have not been studied and are not available in many cases, so duplicates of the same accession may be expected, which would reduce the effective number of accessions in genebank collections.

One of the objectives of *ex situ* conservation is to preserve all different genetic adaptation patterns of a given species (Parra-Quijano *et al*., 2012a). Thus the incorporation of geographic and ecogeographic information to the seed collections is highly desirable. The inclusion of this information in databases of germplasm banks would greatly facilitate the design of sampling and collecting expeditions to improve the genetic diversity of existing collections.

Table 9: List of PGRFA CWR species with five or less accessions preserved in germplasm banks.

GROUP	FAMILY	SPECIES	NUMBER OF ACCESIONS
FOOD	Asteraceae	*Cynara alba*	5
FOOD	Chenopodiaceae	*Beta maritima*	5
FORAGE & FODDER	Fabaceae	*Astragalus glycyphyllos*	5

Table 9: List of PGRFA CWR species with five or less accessions preserved in germplasm banks.

GROUP	FAMILY	SPECIES	NUMBER OF ACCESIONS
FORAGE & FODDER	Fabaceae	*Astragalus granatensis*	5
FORAGE & FODDER	Fabaceae	*Astragalus longidentatus*	5
FORAGE & FODDER	Fabaceae	*Trifolium boissieri*	5
FORAGE & FODDER	Fabaceae	*Trifolium squamosum*	5
FOOD	Fabaceae	*Vicia filicaulis*	5
FOOD	Fabaceae	*Vicia pyrenaica*	5
FOOD	Liliaceae	*Allium grosii*	5
FOOD	Liliaceae	*Asparagus officinalis*	5
FORAGE & FODDER	Poaceae	*Agrostis curtisii*	5
FOOD	Poaceae	*Hordeum bulbosum*	5
FORAGE & FODDER	Poaceae	*Poa pratensis*	5
FOOD	Rosaceae	*Malus sylvestris*	5
FOOD	Rosaceae	*Prunus lusitanica*	5
FOOD	Brassicaceae	*Erucastrum canariense*	4
FOOD	Chenopodiaceae	*Patellifolia procumbens*	4
FORAGE & FODDER	Fabaceae	*Astragalus clusianus*	4
FORAGE & FODDER	Fabaceae	*Astragalus hispanicus*	4
FORAGE & FODDER	Fabaceae	*Astragalus oxyglottis*	4
FORAGE & FODDER	Fabaceae	*Medicago murex*	4
FORAGE & FODDER	Fabaceae	*Trifolium vessiculosum*	4
FOOD	Liliaceae	*Allium rouyi*	4
FOOD	Liliaceae	*Asparagus arborescens*	4
FOOD	Liliaceae	*Asparagus stipularis*	4
FORAGE & FODDER	Poaceae	*Festuca elegans*	4
FOOD	Apiaceae	*Apium graveolens*	3
FOOD	Chenopodiaceae	*Patellifolia webbiana*	3
FORAGE & FODDER	Fabaceae	*Medicago turbinata*	3
FORAGE & FODDER	Fabaceae	*Trifolium hirtum*	3
FORAGE & FODDER	Fabaceae	*Trifolium incarnatum*	3
FORAGE & FODDER	Fabaceae	*Trifolium lappaceum*	3
FORAGE & FODDER	Fabaceae	*Trifolium thalii*	3
FOOD	Fabaceae	*Vicia cordata*	3
FOOD	Fabaceae	*Vicia glauca*	3
FOOD	Liliaceae	*Asparagus nesiotes*	3
FORAGE & FODDER	Poaceae	*Festuca iberica*	3
FORAGE & FODDER	Poaceae	*Poa alpina*	3
FORAGE & FODDER	Poaceae	*Poa bulbosa*	3
FOOD	Rosaceae	*Pyrus spinosa*	3
FOOD	Asteraceae	*Cichorium spinosum*	2

Table 9: List of PGRFA CWR species with five or less accessions preserved in germplasm banks.

GROUP	FAMILY	SPECIES	NUMBER OF ACCESIONS
FOOD	Asteraceae	*Lactuca perennis*	2
FOOD	Brassicaceae	*Diplotaxis muralis*	2
FORAGE & FODDER	Fabaceae	*Astragalus tremolsianus*	2
FOOD	Fabaceae	*Lathyrus tuberosus*	2
FOOD	Fabaceae	*Lens ervoides*	2
FORAGE & FODDER	Fabaceae	*Trifolium spumosum*	2
FOOD	Fabaceae	*Vicia angustifolia*	2
FOOD	Fabaceae	*Vicia argentea*	2
FOOD	Fabaceae	*Vicia bithynica*	2
FOOD	Fabaceae	*Vicia hybrida*	2
FOOD	Fabaceae	*Vicia sepium*	2
FORAGE & FODDER	Liliaceae	*Asparagus albus*	2
FORAGE & FODDER	Poaceae	*Agrostis rupestris*	2
FORAGE & FODDER	Poaceae	*Agrostis schleicheri*	2
FOOD	Poaceae	*Avena canariensis*	2
FORAGE & FODDER	Poaceae	*Deschampsia cespitosa*	2
FORAGE & FODDER	Poaceae	*Festuca burnatii*	2
FORAGE & FODDER	Poaceae	*Festuca frigida*	2
FORAGE & FODDER	Poaceae	*Festuca glacialis*	2
FORAGE & FODDER	Poaceae	*Festuca pratensis*	2
FORAGE & FODDER	Poaceae	*Festuca pseudeskia*	2
FOOD	Poaceae	*Secale montanum*	2
FOOD	Asteraceae	*Cynara scolymus*	1
FORAGE & FODDER	Fabaceae	*Astragalus alpinus*	1
FORAGE & FODDER	Fabaceae	*Astragalus australis*	1
FORAGE & FODDER	Fabaceae	*Astragalus bourgaeanus*	1
FORAGE & FODDER	Fabaceae	*Astragalus depressus*	1
FORAGE & FODDER	Fabaceae	*Astragalus mareoticus*	1
FORAGE & FODDER	Fabaceae	*Astragalus penduliflorus*	1
FORAGE & FODDER	Fabaceae	*Hedysarum flexuosum*	1
FORAGE & FODDER	Fabaceae	*Hedysarum spinossisimum*	1
FOOD	Fabaceae	*Lathyrus bauhini*	1
FORAGE & FODDER	Fabaceae	*Medicago disciformis*	1
FORAGE & FODDER	Fabaceae	*Medicago intertexta*	1
FORAGE & FODDER	Fabaceae	*Medicago soleirollii*	1
FORAGE & FODDER	Fabaceae	*Ornithopus sativus*	1
FORAGE & FODDER	Fabaceae	*Trifolium isthmocarpum*	1
FORAGE & FODDER	Fabaceae	*Trifolium ligusticum*	1
FORAGE & FODDER	Fabaceae	*Trifolium montanum*	1

Table 9: List of PGRFA CWR species with five or less accessions preserved in germplasm banks.

GROUP	FAMILY	SPECIES	NUMBER OF ACCESIONS
FORAGE & FODDER	Fabaceae	*Trifolium mutabile*	1
FORAGE & FODDER	Fabaceae	*Trifolium nigrescens*	1
FORAGE & FODDER	Fabaceae	*Trifolium pallidum*	1
FORAGE & FODDER	Fabaceae	*Trifolium retusum*	1
FORAGE & FODDER	Fabaceae	*Trifolium squarrosum*	1
FOOD	Fabaceae	*Vicia altissima*	1
FOOD	Fabaceae	*Vicia amphicarpa*	1
FOOD	Liliaceae	*Allium commutatum*	1
FOOD	Liliaceae	*Asparagus aphyllus*	1
FOOD	Liliaceae	*Asparagus fallax*	1
FORAGE & FODDER	Poaceae	*Agrostis stolonifera*	1
FOOD	Poaceae	*Avena fatua*	1
FORAGE & FODDER	Poaceae	*Dactylis smithii*	1
FORAGE & FODDER	Poaceae	*Deschampsia setacea*	1
FORAGE & FODDER	Poaceae	*Festuca brigantina*	1
FORAGE & FODDER	Poaceae	*Festuca rothmaleri*	1
FORAGE & FODDER	Poaceae	*Lolium saxatile*	1
FORAGE & FODDER	Poaceae	*Poa compressa*	1
FORAGE & FODDER	Poaceae	*Poa glauca*	1
FOOD	Apiaceae	*Daucus arcanus*	0
FOOD	Asteraceae	*Lactuca livida*	0
FOOD	Asteraceae	*Lactuca singularis*	0
FOOD	Brassicaceae	*Erucastrum gallicum*	0
FORAGE & FODDER	Fabaceae	*Astragalus algerianus*	0
FORAGE & FODDER	Fabaceae	*Astragalus baionensis*	0
FORAGE & FODDER	Fabaceae	*Astragalus cavanillesii*	0
FORAGE & FODDER	Fabaceae	*Astragalus danicus*	0
FORAGE & FODDER	Fabaceae	*Astragalus ginez-lopezii*	0
FORAGE & FODDER	Fabaceae	*Astragalus glaux*	0
FORAGE & FODDER	Fabaceae	*Astragalus hypoglottis*	0
FORAGE & FODDER	Fabaceae	*Astragalus nitidiflorus*	0
FORAGE & FODDER	Fabaceae	*Astragalus turolensis*	0
FORAGE & FODDER	Fabaceae	*Hedysarum glomeratum*	0
FOOD	Fabaceae	*Lathyrus cirrhosus*	0
FOOD	Fabaceae	*Lathyrus nudicaulis*	0
FOOD	Fabaceae	*Lathyrus pisiformis*	0
FOOD	Fabaceae	*Lathyrus pulcher*	0
FOOD	Fabaceae	*Lathyrus sativus*	0
FOOD	Fabaceae	*Lathyrus vivantii*	0

Table 9: List of PGRFA CWR species with five or less accessions preserved in germplasm banks.

GROUP	FAMILY	SPECIES	NUMBER OF ACCESIONS
FORAGE & FODDER	Fabaceae	*Lupinus pilosus*	0
FORAGE & FODDER	Fabaceae	*Medicago coronata*	0
FORAGE & FODDER	Fabaceae	*Medicago falcata*	0
FORAGE & FODDER	Fabaceae	*Medicago hybrida*	0
FORAGE & FODDER	Fabaceae	*Medicago italica*	0
FORAGE & FODDER	Fabaceae	*Medicago praecox*	0
FORAGE & FODDER	Fabaceae	*Medicago scutellata*	0
FORAGE & FODDER	Fabaceae	*Medicago secundiflora*	0
FORAGE & FODDER	Fabaceae	*Ornithopus perpusillus*	0
FORAGE & FODDER	Fabaceae	*Trifolium aureum*	0
FORAGE & FODDER	Fabaceae	*Trifolium badium*	0
FORAGE & FODDER	Fabaceae	*Trifolium cernuum*	0
FORAGE & FODDER	Fabaceae	*Trifolium diffusum*	0
FORAGE & FODDER	Fabaceae	*Trifolium dubium*	0
FORAGE & FODDER	Fabaceae	*Trifolium fragiferum*	0
FORAGE & FODDER	Fabaceae	*Trifolium hybridum*	0
FORAGE & FODDER	Fabaceae	*Trifolium leucanthum*	0
FORAGE & FODDER	Fabaceae	*Trifolium lucanicum*	0
FORAGE & FODDER	Fabaceae	*Trifolium medium*	0
FORAGE & FODDER	Fabaceae	*Trifolium michelianum*	0
FORAGE & FODDER	Fabaceae	*Trifolium micranthum*	0
FORAGE & FODDER	Fabaceae	*Trifolium obscurum*	0
FORAGE & FODDER	Fabaceae	*Trifolium occidentale*	0
FORAGE & FODDER	Fabaceae	*Trifolium ochroleucon*	0
FORAGE & FODDER	Fabaceae	*Trifolium ornithopodioides*	0
FORAGE & FODDER	Fabaceae	*Trifolium pallescens*	0
FORAGE & FODDER	Fabaceae	*Trifolium patens*	0
FORAGE & FODDER	Fabaceae	*Trifolium phleoides*	0
FORAGE & FODDER	Fabaceae	*Trifolium physodes*	0
FORAGE & FODDER	Fabaceae	*Trifolium rubens*	0
FORAGE & FODDER	Fabaceae	*Trifolium spadiceum*	0
FORAGE & FODDER	Fabaceae	*Trifolium suffocatum*	0
FORAGE & FODDER	Fabaceae	*Trifolium sylvaticum*	0
FOOD	Fabaceae	*Vicia chaetocalyx*	0
FOOD	Fabaceae	*Vicia lathyroides*	0
FOOD	Fabaceae	*Vicia leucantha*	0
FOOD	Fabaceae	*Vicia nataliae*	0
FOOD	Liliaceae	*Allium melananthum*	0
FOOD	Liliaceae	*Allium palentinum*	0

Table 9: List of PGRFA CWR species with five or less accessions preserved in germplasm banks.

GROUP	FAMILY	SPECIES	NUMBER OF ACCESIONS
FOOD	Liliaceae	*Allium schmitzii*	0
FOOD	Liliaceae	*Allium stearnii*	0
FOOD	Liliaceae	*Allium subhirsutum*	0
FOOD	Liliaceae	*Asparagus maritimus*	0
FORAGE & FODDER	Poaceae	*Agrostis canina*	0
FORAGE & FODDER	Poaceae	*Agrostis hesperica*	0
FORAGE & FODDER	Poaceae	*Agrostis pourreti*	0
FORAGE & FODDER	Poaceae	*Agrostis tenerrima*	0
FORAGE & FODDER	Poaceae	*Agrostis tileni*	0
FOOD	Poaceae	*Avena lusitanica*	0
FORAGE & FODDER	Poaceae	*Dactylis metlesicsii*	0
FORAGE & FODDER	Poaceae	*Festuca altopyrenaica*	0
FORAGE & FODDER	Poaceae	*Festuca aragonensis*	0
FORAGE & FODDER	Poaceae	*Festuca borderi*	0
FORAGE & FODDER	Poaceae	*Festuca cordubensis*	0
FORAGE & FODDER	Poaceae	*Festuca gigantea*	0
FORAGE & FODDER	Poaceae	*Festuca graniticola*	0
FORAGE & FODDER	Poaceae	*Festuca lasto*	0
FORAGE & FODDER	Poaceae	*Festuca longiauriculata*	0
FORAGE & FODDER	Poaceae	*Festuca paucispicula*	0
FORAGE & FODDER	Poaceae	*Festuca picoeuropeana*	0
FORAGE & FODDER	Poaceae	*Festuca quadrifolia*	0
FORAGE & FODDER	Poaceae	*Festuca querana*	0
FORAGE & FODDER	Poaceae	*Festuca reverchonii*	0
FORAGE & FODDER	Poaceae	*Festuca rivas-martinezii*	0
FORAGE & FODDER	Poaceae	*Festuca segimonensis*	0
FORAGE & FODDER	Poaceae	*Festuca summilusitana*	0
FORAGE & FODDER	Poaceae	*Festuca vettonica*	0
FOOD	Poaceae	*Hordeum distichon*	0
FOOD	Poaceae	*Hordeum zeocriton*	0
FORAGE & FODDER	Poaceae	*Lolium edwardii*	0
FORAGE & FODDER	Poaceae	*Poa angustifolia*	0
FORAGE & FODDER	Poaceae	*Poa laxa*	0
FORAGE & FODDER	Poaceae	*Poa pitardiana*	0
FORAGE & FODDER	Poaceae	*Poa supina*	0
FOOD	Rosaceae	*Pyrus cordata*	0

4. Recommendations

Considering all the information gathered in this study and the results of the *in situ* and *ex situ* conservation assessment the following recommendations are presented:

1.- There is an urgent need to improve the coverage and accuracy of chorological data of Spanish vascular plants in general and CWRs in particular. Although in the last decades a considerable effort has been made to digitalize existing chorological information and a relevant number of regional, national and international databases exist that provide chorological data on Spanish vascular flora there is an urgent need to collect chorological data on the current distribution of plants in a systematic way covering the entire territory with homogeneous search intensity. Recently created and increasingly popular networks of volunteers that record species occurrences in the territory, such as Biodiversidad Virtual (www.biodiversidadvirtual.org) can play a decisive role in this process. This will allow a better knowledge of the current distribution of CWR species and thus, better conservation action planning. Special priority should be given to the 81 prioritized CWR species which currently lack any high-quality georeferenced distribution data and to the 117 prioritized CWR species that have less than 10 high-quality georeferenced chorological records.

2.- Threatened Spanish CWR species should be prioritized for conservation actions focusing on those that currently lack the implementation of a recovery plan. It is especially important to design and implement a management plan on the nine CWR species that are threatened at both the Spanish and European level: *Allium pyrenaicum, Allium schmitzii, Asparagus fallax, Asparagus nesiotes, Avena murphyi, Cicer canariense, Medicago citrina, Prunus lusitanica* and *Prunus ramburii.*

3.- Regardless of the threat status, narrow endemics and species with poor representation in Spain of the prioritized CWR list of this study should be identified from the list of species that have few reported locations and ecogeographic units, and studied with particular care. These species should also be prioritized for conservation actions.

4. In average, 42% of the populations of the prioritized CWR species are passively protected and 66% of the ecogeographic units where the prioritized CWR species are found are represented in the populations that occur in these protected areas. Particular attention should be placed to the 37 species of the prioritized CWR list that have not a single population in a protected area and to the species that have a poor representation of the different ecogeographic units in which they occur.

5.- Genetic reserves should be established in the 12 global complementarity areas identified that are totally or partially in Sites of Community Interest of the Natura 2000 network (Table 8). The establishment of genetic reserves in existing protected areas has lower economic cost than the creation *de novo* in non-protected areas and management synergies with management actions already implemented in these sites can be generated (Maxted et al., 2007).

6. At least one microrreserve should be established in in the municipalities of Ezcabarte, Egüés, Burlada, Villava, Esteríbar and Pamplona in Navarra, where the richest prioritized CWR area is found (79 species); this will provide to this area a figure of protection and allow the establishment of a genetic reserve. Microrreserves are probably the most appropriate area protection figure in Spain for the establishment of genetic reserves in locations that currently do not have a protected area status, following the successful experiences obtained regarding the conservation of endemic and threatened flora in Valencia region (http://www.cma.gva.es/web/indice.aspx?nodo=2684&idioma=C). In order to do this, a detailed *in situ* study characterizing the natural prioritized CWR populations occurring at the site should be undertaken to design the limits and extension of the microrreserve. It is also recommended to establish additional microrreserves in Castellar de Nuch and, Palau Saverdera and Castelló de Empúries to fully cover the complementarity areas 3 and 4, respectively, very close or partially covered by protected areas.

7. Private companies, NGOs, associations, organizations and specific interest groups may wish to get involved in the support of *in situ* conservation actions of prioritized CWR. The analyses structured by CWR use categories carried out in this study open the way to the implementation of specific *in situ* conservation actions for the Food, Fodder & Forage, Ornamental and Industrial and other uses categories.

8.- The *ex situ* collections of the prioritized Spanish CWR should be greatly improved. With previous coordination among the different Spanish genebanks, a series of seed collecting expeditions should be organized to collect seeds of the 95 prioritized PGRFA CWR species identified in this study that are not represented in the network of Spanish genebanks. The representation of the ornamental and industrial prioritized CWR should also be completed In a second phase, a second series of optimized seed collections should be organized to adequately represent the genetic diversity of each species. Tentatively, Spanish genebanks should contain at least 5 accessions of each prioritized CWR species representing the most frequent ecogeographic units in which the species is found. The inclusion of ecogeographic information in the design of the sampling strategy will allow optimizing the range of genetic variation found in a given species (Parra-Quijano *et al*., 2012a). Collections should be carried out following a

sampling methodology that guarantees a proper representation of the within-population genetic variation. All new seed collections should provide high-quality georeferencing data. Spanish genebanks should also make an effort to improve the georeferencing data of the seed accessions that are currently stored.

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