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Article

Crop wild relatives (CWR) priority in Italy: distribution, ecology, *in situ* and *ex situ* conservation and expected actions

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Abstract: The study presents an updated overview of the 14 non-endemic threatened Crop Wild Relatives (CWR) in Italy: *Aegilops biuncialis*, *Ae. uniaristata*, *Ae. ventricosa*, *Asparagus pastorianus*, *Beta macrocarpa*, *Brassica insularis*, *B. montana*, *Crambe hispanica* subsp. *hispanica*, *C. tataria* subsp. *tataria*, *Ipomoea sagittata*, *Lathyrus amphicarpos*, *L. palustris*, *Vicia cusnae* and *V. serinica*. Geographical distribution, ecology (with plant communities and habitat 92/43/EEC aspects), genetics (focused on gene pools), property, and *in situ* and *ex situ* conservation were analyzed. In addition, with the aim of their protection and valorization, specific actions are recommended.

Keywords: conservation; gene pool; geographical distribution; threatened; valorization.

1. Introduction

Crop Wild Relatives (CWR) are wild species closely related to crops, potential sources of important traits (such as pest or disease resistance), yield improvement and/or stability. It must also be considered that they are a critical component of plant genetic resources for food and agriculture (PGRFA), although they have been neglected for conservation purposes [1], and *in situ* and *ex situ* conservation approaches should be deployed to ensure their availability for use [2].

In monetary terms, the CWR have contributed significantly to the agricultural and horticultural industries, and to the world economy [3,4]. Pimentel et al. [5] estimated that wild relatives contribute approximately US$ 20 billion toward increased crop yields per year in the United States and US $115 billion worldwide. Phillips and Meilleur [6] noted that losses of rare wild plants represent a substantial economic loss to agriculture, estimating that the endangered food crop relatives have a worth of about US$ 10 billion annually in wholesale farm values. Although these studies show significant divergence, they highlight the major global economic value of CWR diversity to humanity.

The CWR following the definition of Maxted et al. [7] are *taxa* belonging to the same genus as the cultivated species. With this approach we would have about 80% of the European and Mediterranean flora species as CWR and important from a socioeconomic point of view [8]. However, a genetic other than taxonomic approach suggests that only those species able to interbreed with cultivated species in relation to their “gene pool” should be considered CWR. According to Harlan and de Wet [9], the gene pool represents a reservoir of diversity that can be tapped into by organisms to adapt to a changing environment, and breeders for crop improvement. Wild relatives of a given crop are thought to be in the same gene pool, and even when they appear to be taxonomically different, they can exchange genes with their related cultivated taxon. Unfortunately, not all wild relatives are equally ready to do this. For this reason, CWR have been classified into 3 groups (GP1, GP2, GP3) based on the ability to exchange genes with the cultivated species to which they are naturally related [9]. The primary gene pool (GP1) includes species that can be directly crossed with the cultivated species to produce fertile breeds. For example, it is easier for *Beta macrocarpa* Guss. (GP1) to interbreed with cultivated chard (*Beta vulgaris* L.) as they have a very good genetic affinity, than other species that are less related, and for that they belong to more distant gene pools (GP2 or GP3).

2. Materials and Methods

The study was planned starting from 43 Italian threatened CWR [10,11], according to the taxon group concept of CWR [7] and not at all of the gene pool concept [9], from which the 29 taxa endemic to Italy were excluded and will be treated in a separate work. Thus, the following 14 taxa were investigated: *Aegilops biuncialis* Vis., *Ae. uniaristata* Vis., *Ae. ventricosa* Tausch, *Asparagus pastorianus* Webb & Berthel, *Beta macrocarpa* Guss., *Brassica insularis* Moris, *B. montana* Pourr., *Crambe hispanica* L. subsp. *hispanica*, *C. tataria Sebeók* subsp. *tataria*, *Ipomoea sagittata* Poir., *Lathyrus amphicarpos* L., *L. palustris* L., *Vicia cusnae* Foggi & Ricceri and *V. serinica* R. Uechtr. et Huter.

The *taxa* at risk are those reported in Annex I of the International Treaty on Plant Genetic Resources for Food and Agriculture (ITPGRFA) [10], those mentioned by the Italian Institute of Statistics (ISTAT) for cultivated areas and yield between 2007 and 2012 [12], the policy species threatened and near threatened, the taxa included in Red Lists, at both Italian and European level [13-24], in the Bern Convention [25], and in the Annexes to the 92/43/EEC Directive [26] (Table 1).

For each of the 14 non-endemic wild relatives three levels of attention were considered regarding *ex situ* conservation with high priority (HP) for *taxa* present in the Italian RIBES (Rete Italiana delle Banche del Germoplasma) seed banks with zero accessions, normal priority (NP) for *taxa* present with less than five accessions (from 1 to 4) and zero priority (ZP) for those species present with five or more accessions (from 5 to 140), and one level of attention for the *in situ* conservation (A) that includes the native *taxa* related to a crop of worldwide and national importance for food and agriculture, which are included in (at least) the National and in the European Red Lists, and in the International Conventions and that need specific monitoring/protection measures (Table 1). No taxa belonging to the other two levels of attention for the *in situ* conservation have been identified, the level (B) that concerns the native taxa related to important crops, which on the basis of current knowledge, have no need of any immediate specific protection or monitoring measures, and the level (C) that includes the native taxa related to important crops, neither endemic nor subendemic to Italy, which, on the basis of current knowledge, have no need of any immediate specific protection or monitoring measures [1, 27]. For a better evaluation of *in situ* and *ex situ* conservation, vegetation and 92/43/EEC Habitat data have been included (Table 2).

Finally, the 14 non-endemic wild relatives were evaluated considering their gene pools (GP1, GP2 and GP3), according to the concept of Harlan and de Wet [9], by consulting the checklist www.cwrdiversity.org/checklist/ (accessed 2 November 2020) and Vincent et al. [28], and checking their *in situ* and *ex situ* conservation priorities (Table 3).

The results are shown in alphabetical order by genus and species and discussed individually with the evaluation of the following aspects: geographical distribution, reasons of threat and priorities for conservation (both *ex situ* and *in situ*), ecology, vegetation types (only those recognized from a phytosociological point of view) and/or habitat 92/43/EEC, properties, and a management indicator (called *expected actions*).

3. Results

According to the taxon group concept, the 14 non-endemic CWR priority in Italy belong to the Brassicaceae and Fabaceae families, each with 4 species, followed by Poaceae with 3 species and Asparagaceae, Chenopodiaceae and Convolvulaceae, each represented by only 1 species. The most represented genus is *Aegilops* L. with 3 species, followed by *Brassica* L., *Crambe* Vosmaer, *Lathyrus* Schinzand and *Vicia* L., each with two species, and finally the genera *Asparagus* L., *Beta* L. and *Ipomoea* L., with only one species (Table 1)

Table 1. Prioritized list of 14 (non-endemic) taxon group crop wild relatives, reasons of threatening, adapted and updated from Landucci et al. [27], Magrini et al. [11] and Perrino and Perrino [1]

Taxa	IT	IS	1	‡2	‡3	‡4	5	6	7	8	9	10
Aegilops biuncialis	X	X	LC	CR						VU[1]		
Aegilops uniaristata	X	X	LC	EN		DD	V	VU		VU[2]		
Aegilops ventricosa	X	X	LC							VU[3]		
Asparagus pastorianus	X	X	VU	VU	VU				NT[2]			
Beta macrocarpa	X	X	EN									
Brassica insularis	X	X	NT		EN			NT	NT[1]	NT[4]	X	X
Brassica montana	X	X	LC					VU	VU[2]			
Crambe hispanica subsp. *hispanica*	X		LC		EW					VU[5]		
Crambe tataria subsp. *tataria*	X		LC	VU			V	NT	NT[1]		X	
Ipomoea sagittata	X	X	VU	EN	EN		E					
Lathyrus amphicarpos	X	X	NT	LR	LR		R					
Lathyrus palustris	X	X						EN	EN[2]			
Vicia cusnae	X	X		LR			R	VU		VU[6]		
Vicia serinica	X	X		LR			R		EN[2]			

IT=ITPGRFA: taxa included in Annex I of the the International Treaty on Plant Genetic Resources for Food and Agriculture [10].
IS=ISTAT: taxa mentioned by the Italian Institute of Statistics (ISTAT) for cultivated areas and yield in the last 5 years before 2012 [12].
1 Bilz et al. [16] (European Red List): EN=Endangered, LC=Least concern, NT=Near Threatened, VU=Vulnerable.
‡ Conti et al. [14]; 2=Italy, 3=Sicily, 4=Sardinia: EW=Extinct in the wild, CR=Critically Endangered, EN=Endangered, VU=Vulnerable, LR=Lower risk, DD=Data deficient.
5 Conti et al. [13]; 5=National Red List: E=Endangered, V=Vulnerable, R=Rare.
6 Rossi et al. [17] (Italian Red List of Policy Species and other threatened species): EN=Endangered, VU=Vulnerable, NT=Near Threatened.
7 RED LIST OF THREATENED VASCULAR PLANTS IN ITALY: taxa included in [1]Rossi et al. [18] and in [2]Orsenigo et al. [24]: EN=Endangered, VU=Vulnerable, NT=Near Threatened.
8 OTHERS IUCN CARDs: taxa included in "Red List of Italian Vascular and Cryptogamic Flora cards" published since 2013 on Informatore Botanico Italiano become Italian Botanist; [1]Perrino and Wagensommer [20]; [2]Perrino and Wagensommer [19]; [3]Perrino and Wagensommer [21]; [4]Santo et al. [23]; [5]Perrino et al. [22]; [6]Foggi et al. [15]: VU=Vulnerable, NT=Near Threatened.
9 EUROPEAN COMMISSION: Annex II of the Directive 92/43/EEC [26].
10 BERN CONVENTION: Appendix I [25].

3.1. Ex situ and In situ conservation

3.1.1. Taxon group CWR with high priority (A) of conservation

Accordingly, we crossed the data of the 760 species selected for their need of *in situ* [27] and for their need of *ex situ* [11] conservation, with those of the 14 selected species, with the aim to compare the situation of the latter for their *in situ* and *ex situ* conservation. The results show that for *in situ* conservation all species (14 out of the 14) have the highest priority (A), while for *ex situ* conservation, 7 species have the highest priority (HP), 4 normal priority (NP), and 3 zero priority (ZP) (Table 2).

3.1.2. Relationship between *in situ* and *ex situ* conservation

Globally, all species (14 out of 14) have a highest priority for *in situ* (A) conservation, but only half of total (7 out of 14), *Asparagus pastorianus* Webb & Berthel, *Beta macrocarpa* Guss., *Ipomoea sagittata* Poir., *Lathyrus amphicarpos* L., *Lathyrus palustris* L., *Vicia cusnae* Foggi & Ricceri, and *Vicia serinica* R. Uechtr. et Huter, are in the worst situation, since they have the highest priority also for *ex situ* (HP) conservation. For the remaining species, the situation could be considered less hard, since high priority for *in situ* (A) is balanced by low (NP) or zero (ZP) priority for *ex situ* (only 3 taxa with

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zero priority). In conclusion, all species have high *in situ* priority (A), need to monitoring and updating, and should be considered at risk (Table 2).

Table 2. Prioritized list of 14 taxon group crop wild relatives (non-endemic), their status of *ex situ* and *in situ* conservation and relationships with plant communities and/or habitat 92/43 EEC

Taxa	*Ex situ* priority conservation			*In situ* priority conservation	*Syntaxon*/Habitat (code)
	HP	NP	ZP	A	
Aegilops biuncialis		X		X	(6220*)
Aegilops uniaristata		X		X	(6220*)
Aegilops ventricosa		X		X	(6220*)
Asparagus pastorianus	X			X	*Asparago pastoriani-Chamaeropetum humilis*
Beta macrocarpa	X			X	?
Brassica insularis			X	X	?
Brassica montana			X	X	*Reichardio maritimae-Brassicetum robertianae*
Crambe hispanica subsp. *hispanica*			X	X	*Crambetum hispanicae*
Crambe tataria subsp. *tataria*		X		X	*Centaureo-Globularietum cordifoliae* (62A0)
Ipomoea sagittata	X			X	*Calystegion sepium* (6430)
Lathyrus amphicarpos	X			X	?
Lathyrus palustris	X			X	*Molinio-Arrhenatheretea* (6410, 6420)
Vicia cusnae	X			X	*Thlaspion rotundifolii* (8210)
Vicia serinica	X			X	*Sideridenion italicae* (6210*)
TOTAL	7	4	3	14	

***Ex situ priority conservation*. HP**: taxa with high priority (zero accessions); **NP**: taxa with normal priority (1-4 accessions); **ZP**: taxa with no priority (5-140 accessions). Adapted and updated from Magrini et al. [11].
***In situ priority conservation*. A**: includes native taxa related to a crop of worldwide and national importance for food and agriculture, which are included as Threatened (EW, CR, EN, VU) or Near threatened in (at least) one of the following sources: IUCN (European Red List) [16]; Regional Red List (national catalogue and catalogue for Sicily and Sardinia) [14]; National Red List [13,15,17-23,29]. These taxa need specific protection and/or monitoring measures.
Vegetation type and/or Habitat 92/43 EEC (Italy). Code habitat [30]. Vegetation type (see reference in the text when discuss of the relative species).

3.1.3. The 14 taxon group CWR in the light of the gene pool concept

Since plant breeders would concentrate on wild relatives that may cross easily with crops, we have checked which ones of the 14 taxon group wild species belong to the three gene pools, foreseen by the Harlan and de Wet [9] concept. The results (Table 3) show that only 8 species out of the 14 belong to one or two gene pools. In particular, two species, *Beta macrocarpa* Guss. and *Crambe hispanica* L. subsp. *hispanica*, share only the primary gene pool (GP1), five species, *Aegilops biuncialis* Vis., *Aegilops uniaristata* Vis., *Aegilops ventricosa* Tausch, *Brassica insularis* Moris (Policy species), and *Brassica montana* Pourr., share the secondary and tertiary gene pools (GP2 and GP3), one, *Lathyrus amphicarpos* L., belongs only to the secondary gene pool (GP2), and not (GP3) as indicated by www.cwrdiversity.org/checklist/ (accessed 20 November 2020). In conclusion, two species belong only to GP1, one only to GP2, five share GP2 and GP3, while for the other 6 taxa, at the moment, to the best of our knowledge, there is no information.

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Table 3. Crop wild relatives (8 out of 14) belonging to at least one gene pool and their conservation prioritization updated from Landucci et al. [27] and Perrino and Perrino [1]

Taxa	Gene Pools (GP)			*Ex situ* priority conservation			*In situ* priority conservation
	GP1	GP2	GP3	HP	NP	ZP	A
Aegilops biuncialis		X	X		X		X
Aegilops uniaristata		X	X		X		X
Aegilops ventricosa		X	X		X		X
Beta macrocarpa	X			X			X
Brassica insularis		X	X			X	X
Brassica montana		X	X			X	X
Crambe hispanica subsp. *hispanica*	X					X	X
Lathyrus amphicarpos		X		X			X
TOTAL	2	6	5	2	3	3	8

Gene Pools (GP): taxa with a ascertain use in plant breeding belonging to the primary (GP1), secondary (GP2) and terciary GP (GP3).

***Ex situ* priority conservation**. HP: taxa with high priority; NP: taxa with normal priority; ZP: taxa with zero priority. Adapted and updated from Magrini et al. [11].

***In situ* priority conservation**. A: includes native taxa related to a crop of worldwide and national importance for food and agriculture, which are included as Threatened (EW, CR, EN, VU) or Near threatened in (at least) one of the following sources: IUCN (European Red List) [16]; Regional Red List (national catalogue and catalogue of Sicily and of Sardinia) [14]; National Red List [13,17-18,21,24,29]. These taxa need specific protection and/or monitoring measures.

4. Discussion

4.1. *Aegilops biuncialis* Vis., *Aegilops uniaristata* Vis. *and Aegilops ventricosa* Tausch

The genus *Aegilops* L. has been intensively studied due to its close relationship with cultivated wheats and related of their vast genetic diversity that represents a rich source of alleles of agronomic interest, which could be used to widen the wheat gene pool and improve tolerance to diseases, pests, drought, cold and other environmental stresses [31], and for improving micro-nutrient content (such as Fe and Zn) in wheat grains. About the last point, it should be noted that Zn deficiency affects 17.3% of the world population in Asia and Africa, leading to the death of over 400,000 children every year [32-34]. Wheat rich in micronutrients, that is, bio-fortified wheat, can improve the lives of these people. It is difficult to find germplasm with high Zn and Fe content in the wheat gene pool [35], although some *Ae.* show three to four times higher Zn and Fe grain content, such as *Ae. ventricosa* (genome DN) [36].

In Europe, wild wheat relatives of the *Triticum-Aegilops* complex grow in sympatry with cultivated bread wheat (*Triticum aestivum* L.) and spontaneous hybridization is known for most of the tetraploid *Ae.* species. The probability of gene transfer and gene retention in hybrid progenies is, however, higher when a gene is located on a shared genome, particularly on the D genome shared with *Ae. cylindrica* and *Ae. ventricosa*. Some studies have shown through optimized experimentation to support the hybridization (experimental soil layout, flowering synchrony) that the cross-pollination between the cultivated wheat and its relatives occur at a significant level as for *Ae. biuncialis* [37].

The chromosome number is 4n=28 in *Ae. biuncialis*, 2n=14 in *Ae. uniaristata*, and 4n=28 in *Ae. ventricosa* [38,39].

The species belonging to this genus are mainly distributed in Southwest and Central Asia and throughout the Mediterranean Basin [40,41]. In Italy their geographical distribution, ecology, vulnerability has recently been updated [42]. Among the priority CWR it is the most represented genus with 3 species [*Ae. biuncalis* (genome UM), *Ae. uniaristata* (genome N) and *Ae. ventricosa* (genome DN)], all listed as threatened in the red lists (Table 1), with high *in situ* priority (A), and normal *ex situ* priority (NP) (Table 2) and secondary and tertiary gene pools (GP2 and GP3) (Table 3).

The flowering time of *Ae.* in Italy is from April to June, depending on the species and its eco-geographical location [42], and partially meets the flowering of the cultivated wheat that starts in May and ends in June [43], phenological condition that would suggest an *in situ* crossbreeding experimentation.

The three *Ae.* mentioned grow in peculiar annual meadows of *Brachypodietalia distachyi* Rivas-Martínez 1978 order (subtype 3 of priority habitat 6220*) [42,44], but a specific classification framework from a phytosociological point of view is lacking.

4.1.1. *Expected actions*

- *In situ* and *ex situ* conservation to prevent the risk of extinction by increasing the number of individuals of existing wild populations.
- *In situ* translocation to the fields edge of cultivated ancestral wheat to verify and update the hybridization capacity, thanks to the comparable flowering periods.
- Starting cultivation in cooperation with local farmers especially of *Ae. ventricosa* that, unlike cultivated wheat varieties, has a higher quantity of microelements such as Fe and Zn. Then, verify the prospect of production and marketing of its flour and/or pasta as a natural alternative to conventional medicine, as helpful for people with Fe and Zn deficiency.
- Study plant communities of annual meadows to define their phytosociological framework.

4.2. *Asparagus pastorianus* Webb & Berthel

Several wild species of the genus *Asparagus* L. in the Italian Peninsula have long been the object of harvesting for food consumption and in the case of *A. officinalis* L. also of ancient domestication and cultivation. Since the Middle Ages the cultivated and wild species of this genus has always had an important place in the gastronomic culture. The young shoots of *A. pastorianus* are eaten in Morocco (vernacular name: sekoum), the stems and roots are used in popular medicine as aphrodisiac [45,46] in Canary Islands (vernacular names: "esparraguera de espinas" or "espina blanca") to produce smoke, prepare infusion, decoction with white wine as insect repellent and diuretic slimming. The bioactive phytochemicals are glycosides and sapogenins [47].

A. pastorianus is a perennial shrub that grows in the garrigues near the sea, and have a south-western Mediterranean-Macaronesian distribution. In Italy, the species grows only in a restricted area of Sicilian Region [48]. On the southern coast of Sicily, between Selinunte and the mouth of the Verdura River, on Pleistocene deposits consisting of a succession of calcarenites and sandy clays, grows a peculiar low shrubby plant community association characterized by *A. pastorianus*, described as *Asparago pastoriani-Chamaeropetum humilis* Raimondo & Bazan 2008 [49], included in the alliance *Oleo-Ceratonion* Br.-Bl. ex Guinochet & Drouineau 1944 em. Rivas-Martínez 1975.

The dispersion of seeds in Canary Islands (Lanzarote, Fuerteventura, Gran Canaria, Tenerife, La Gomera) occurs through small mammals (squirrel) [50] and birds (Shrikes and Kestrels) [51], while there are no available data from the Sicilian population.

The chromosome number is 2n=40 [52] (material from Santa Lucía-Gran Canaria, cultivated in Botanical Garden of Oslo). Although the gene pool is unknown, among the conservation priorities, *A. pastorianus* is one of the most important for conservation interest because it is listed in the red list with VU category in Europe [16] and NT category in Italy [24] (Table 1), resulting in a high *in situ* (A) and *ex situ* (HP) priority (Table 2).

4.2.1. *Expected actions*

- Monitoring of the few known sites of coastal area in southern Sicily, well preserved and for which *in situ* conservation actions would be appropriate, because of the following potential threats: a) policies development that aim at the tourist exploitation [49]; b) potential negative effect of mammals, especially rodents and lagomorphs on seed germination, as already observed in Canary Islands [53].
- Targeted actions for the collection of germplasm to *ex situ* conservation as the species has zero accessions in the RIBES seed banks.

- Research activities to verify the gene pool through crossing with other species of the same genus and any differences with the populations of the Canary Islands and Morocco.
- Verify the seeds dispersal system in Sicily as it was done in the Canary Islands.
- Evaluate the enhancement of the Sicily populations for the production of their use for medicinal purposes.

4.3. Beta macrocarpa Guss.

The genus *Beta* L. is divided into two sections: *Beta* and *Corollinae* [54]. The section *Beta* includes five taxa, *B. vulgaris* L. subsp. *maritima* (L.) Arcang. (the sea beet), which is considered as the wild ancestor of all cultivated beets, the different forms of cultivated beets (*B. vulgaris* L. subsp. *vulgaris*), *B. macrocarpa*, which is an annual self-compatible plant thought to reproduce predominantly by autogamy, *B. patula* Aiton, which is endemic to two small islets of Madeira Archipelago [55], and finally *B. vulgaris* L. subsp. *adanensis* (Pamukç.) Ford-Lloyd & J.T. Williams, that grows in some Eastern Mediterranean areas of Greece and Turkey [55-57]. Therefore, in the western Mediterranean area, only two species of the section *Beta* can be found in coastal and inland ruderal habitats: *B. vulgaris* subsp. *maritima* and *B. macrocarpa* [58]. *B. macrocarpa* is located in inland or coastal habitats in western and eastern Mediterranean areas [59] and in Italy it grows in uncultivated clayey soils [60] in Campania, Basilicata and Sicily, while its occurrence is doubtful in Sardinia and Trentino Alto Adige [48], always rare in each region.

The chromosome number of *B. macrocarpa* is 2n=36 (from accessions of the Canary Islands) [61]. *B. macrocarpa* is closely related to *B. vulgaris* L. subsp. *maritima* (L.) Arcang. by genetic structure. In particular, *B. macrocarpa* has a genotypic structure and a high level of genetic differentiation indicative for selfing (it is an extreme degree of inbreeding) [58]. In fact, the two species can spontaneously hybridize [61-63], sharing the primary gene pool (GP1) (Table 3).

This species is listed in the European red list [16] with EN category (Table 1), shows high *in situ* priority (A) and high *ex situ* priority (HP) (Table 2), and primary gene pool (GP1) (Table 3). It is worthy to note that the wild taxa of section *Beta*, except *B. vulgaris* subsp. *maritima*, are all listed in IUCN Red List, as VU (*B. vulgaris* subsp. *adanensis*) [64], EN (*B. macrocarpa*) [65] and CR (*B. patula*) [66]. For threatened therophytes, such as *B. macrocarpa*, it is important to consider that natural phenomena can cause considerable fluctuations in the number of individuals, and that therefore repeated counts in subsequent years are necessary for a correct estimate of the population size [67].

A specific study on herbaceous vegetation useful for animals on wetland environments in Tunisia, shows a high concentration of minerals in *B. macrocarpa*, in particular the highest ones compared to all the other herbaceous species, locally sampled, on K (15,4g kg^{-1} dry matter), Ca (31,2g kg^{-1} dry matter), Mg (15,1g kg^{-1} dry matter) and although high on average, the lowest NaCl (54,3g kg^{-1} dry matter) content among the Chenopodiaceae family, in addition to a high concentration value of phenols (30,1g kg^{-1} dry matter) and oxalate (64,6g kg^{-1} dry matter) [68].

4.3.1. *Expected actions*

- Cultivation, in cooperation with local farmers and both plant and animal breeders, to test the commercial product, thanks to the high contents of minerals, important for food and feed.
- Monitoring of populations in known areas and field surveys to find new sites.
- Targeted actions for the collection of germplasm to *ex situ* conservation, as there are no accessions in the RIBES seed banks.
- *In situ* and *ex situ* crossing test with *Beta vulgaris* subsp. *maritima*, due to the sharing of GP1.
- Phytosociological studies in regions where it grows for vegetation, habitat and ecological evaluation.

4.4. Brassica insularis Moris (Policy species), *Brassica montana* Pourr.

Wild taxa in *B. oleracea* L. play an important role to improve cultivated crops, but the genomic relationships between wild and cultivated forms have not been well clarified [69]. *B. insularis* and *B.*

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montana belong to *B.* sect. *Brassica*, which encloses the *taxa* with the same C genome (n=9) of *B. oleracea* crops [70-72], and the crossing experiments have confirmed that they are closely related [73].

B. insularis is an endemic Mediterranean member of the *B. oleracea* group which occurs only in France (Corsica), Italy (Sardinia and Pantelleria), Tunisia (La Galite, Zembra and Zembretta) and Algeria (Kabylie) [23,74], while *B. montana* is widespread along the coasts of the northern Mediterranean Sea, from north-eastern Spain to south-western Italy [75]. In Italy, *B. insularis* grows only in the two islands (Sicily and Sardinia), and its occurrence is doubtful in Tuscany, while *B. montana* has a fragmentary distribution [Liguria (very common along the coast), Emilia Romagna, Tuscany, Marche, Latium, Campania, Basilicata and Calabria] [48], probably due to the relict origin [76].

Both taxa are listed in red lists (Table 1), with high *in situ* priority (A), and zero *ex situ* priority (ZP) (Table 2) due to the 27 accessions present in the seed-banks of Sardinia and Perugia [11], and secondary and tertiary gene pools (GP2, GP3) (Table 3). *B. insularis* is also listed in Annex II of the Habitat Directive 92/43/EEC [26] and under Appendix I of the Berna Convention [25].

B. insularis is a perennial rupestrian, xerophilous species that grows under the influence of wet marine flows with high soil salinity and marine aerosols, while it is less frequent in inland areas, on slopes, cliffs and vertical walls, at altitudes from 0 to 1200 m a.s.l. [23,77], with a flowering period that extends from March to May and with only a small proportion of individuals flowering in any given month [78].

B. montana grows in habitats influenced by human activities, for instance quarries, roadsides and building grounds [79], and the flowering period is from March to April. On Monte Conero (Marche) *B. montana* is common in two types of plant communities [76]: a) the *Reichardio maritimae-Brassicetum robertianae* Biondi 1982 in rocky crevices of lightly elevated calcareous walls, reached directly by marine aerosol and occasionally by waves; and b) the *B. montana* and *Matthiola incana* (L.) W. T. Aiton community in the most elevated sectors of rocky walls reached by winds with low salt concentration.

Several studies testify that wild forms can be considered as potential resources to improve the current *B. oleracea* crops, especially when some favorable traits have been identified in wild types of *B. oleracea* such as resistance against *Sclerotinia sclerotiorum* [80], blackleg (*Peronospora parasitica*) [81,82], cabbage white fly (*Aleyrodes proletella*) [83], and cabbage root fly (*Delia radicum*) [84]. *B. insularis* showed seed sinigrin content, with unusual glucosinolate pattern, low progoitrin and high gluconasturtiin levels, and benzyl glucosinolates traces [85], while *B. montana* shows a high seed glucosinolate content that could be used for increasing the total content of specific glucosinolate profiles for improving biocidal and anticarcinogenic activity in cultivated *Brassica* [86].

4.4.1. *Expected actions*

- All Italian *taxa* of genus *Brassica* may be used as genetic resources, as potential host valuable traits that could be transferred to the respective cultivated crops (cabbage, cauliflower, broccoli, etc.) [75], starting from the places with greater ecological affinities and closest to the known localities where the wild species grows.
- *In situ* experiments in cooperation with local growers, thanks to their high potential agronomic value and high tolerance to drought, insects, and high content of glucosinolate [87,88].
- I*n situ* and *ex situ* crosses with cultivated *B. oleracea*, although some preliminary studies have shown low fertility values of crosses between *B. montana* and cultivated forms of *B. oleracea* [79]. The wild populations could be maintained with low on-site management because they grow on cliff sites, and suffer especially for availability of nutrients.
- *Ex situ* conservation of wild populations is necessary, especially to avoid species extinction or further genetic erosion after ecological changes [89], and can be realized by plant conservation in botanical gardens and seed-banks, the latter started by Gomez-Campo and Gustafsson in 1986 [74].
- Low levels of observed heterozygosity in natural populations of *B. insularis* document the importance of developing conservation guidelines appropriate for the populations of this species [74]. Geographical variation studies might be further investigated with physiological analyses [90].

- Monitoring of known populations and field surveys to find new sites (especially for *B. insularis*). Despite the restriction on collecting *B. insularis* from the known sites, the cleaning of cliffs to create suitable climbing areas could be a problem. Preventing access to the *B. insularis* populations appear to be the most suitable conservation measure with the support of protection policies how it was done for the Corsica populations [74].
- Ecological studies are needed to determine the role of grazing (especially by goats) on population maintenance [91].
- Phytosociological studies in Italy, where it occurs, to evaluate vegetation, habitat, ecology and biodiversity, especially for *B. insularis* for which there is lack of data.

4.5. Crambe hispanica L. subsp. *hispanica*, *Crambe tataria* Sebeók subsp. *tataria* (Policy species)

Many species of the genus *Crambe* L. are considered industrial crops [92]. For instance *C. tataria* could be used for paper production when mixed with long fibrous materials [93], to obtain higher oil and erucic acid yield [94]; *C. hispanica* was used for the production of special lubricants, in industrial vulcanization processes and in those that lead to erucamide from erucic acid [95-97], biodiesel, meal and husk for animal feed [98]. For this purpose, in 1975 *C. hispanica* seed samples were collected in Apulia (Gargano) and Sardinia by a team of Breeders from California (USA) and agronomists from the Germoplasm Istitute of Bari (CNR) [99], for its cultivation as a new alternative crop to other industrial crops [100,101].

The genus *Crambe* has an extensive area of distribution that goes from the Macaronesian archipelagoes to the West of China and North of India and from the Arctic Polar Circle on the Scandinavian Peninsula to 5° Latitude South in the North of Tanzania. It is well represented in the Macaronesian, Euro-Siberian, Mediterranean, Sindico-Saharan, Irano-Turkish and Sudan-Zambezian (Ethiopia and Tanzania) regions [95] and includes more than 35 species [102,103]. Based mainly on the dimensions and shape of the proximal joint of the fruit, the genus is divided in three sections (*Crambe*, *Dendrocrambe* and *Leptocrambe*) that closely correspond to the geographical areas of distribution (104,105). In Italy, only two species of *Crambe* are present [48], both considered CWR [1]: *C. hispanica* subsp. *hispanica* (section Leptocrambe) and *C. tataria* subsp. *tataria* (section *Crambe*).

C. hispanica subsp. *hispanica* is a South-Mediterranean-Turanian entity reported in northern Ethiopia [95], Morocco, Portugal, Spain, former Yugoslavia (Serbia and Montenegro), Greece, Cyprus, Lebanon, Syria, Israel, Jordan, Italy [106], Turkey [107] and Albania [108]. The Italian distribution sites are synthesized in the study of Perrino et al. [22] and concern well-defined areas of Apulia, Calabria, Sicily and Sardinia regions, while the presence in Basilicata is doubtful [48].

C. tataria subsp. *tataria* is endemic to the Pontic-Pannonian region, with a strong disjunction from its main distributional range in Friuli-Venezia Giulia Region, the only site in Italy [109].

C. hispanica subsp. *hispanica* is a sub-nitrophilic-synanthropic species, which in Italy grows on calcareous soils, on sandy soils of volcanic origin and on brown soils, exclusively in habitats subject to anthropic disturbance and semi-rupestrian environments. It can be located at the edge of abandoned olive groves, near lake basins, along dry stone walls, in the shade of isolated trees, often of *Quercus trojana* Webb ascribed to the *Crambetum hispanicae* Perrino, Tomaselli, Signorile, Angiulli, Silletti 2011 association [110], along the banks of rivers [111], in aspects of shrub vegetation dominated by *Cytisus villosus* Pourr. and *Spartium junceum* L., to margins of thermophil woods, in uncultivated arid areas [112], and also in correspondence of road bumps. In the Apulian populations it grows on calcareous substrate, with a certain enrichment, never very intense, in soil nutrients, while in Sicily the species prefers humid environments and on Etna, where the best preserved Sicilian populations occur, it grows on shallow and very humified soils [113]. The flowering time of *C. hispanica* subsp. *hispanica* in Italy is from March to April.

C. tataria subsp. *tataria* is reported on steppes and hills rich in clay and limestone from Eastern Europe to the Caucasus [105]. In Italy it grows on extensive deep beds of alluvial, calcareous gravel deposited by the rivers Cellina and Meduna that characterize the ‘’magredi’’ landscape. The use of land for military purposes in a sparsely inhabited area has somehow helped in the preservation of

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''magredi'' fragments, where *C. tataria* is one of the most typical elements of this characteristic grassland formation [109] referred to *Centaureo dichroanthae-Globularietum cordifoliae* Pignatti 1953 association [114] which is considered habitat 92/43/EEC "Eastern sub-mediterranean dry grasslands (*Scorzoneretalia villosae*)" (code 62A0). The flowering time of *C. tataria* subsp. *tataria* in Italy is from May to June.

The chromosome number is 2n=60 in *C. hispanica* subsp. *hispanica* [95] and 2n=30 in *C. tataria* subsp. *tataria* [115].

C. hispanica subsp. *hispanica* and *C. tataria* subsp. *tataria* are both reported as threatened in Italy, while they are considered LC in European Red List (Table 1). Both species have a high *in situ* priority (A). As for *ex situ* conservation, *C. tataria* subsp. *tataria* with 1 accession has normal priority (NP) and *C. hispanica* subsp. *hispanica* with 5 accessions has no priority (ZP) [11] (Table 2), but the number of accessions is still low. In addition, *C. hispanica* subsp. *hispanica* is important for its primary gene pool (GP1) (Table 3). However, independently from the gene pool, *C. tataria* also needs widespread protection since it is rare throughout the global range and its habitats are often destroyed, and in fact the taxon is also reported in the Red Books of the USSR and Kazakhstan [116].

4.5.1. *Expected actions*

- *In situ* and *ex situ* conservation to prevent the risk of extinction by increasing the number of individuals in wild populations. For *C. hispanica* subsp. *hispanica*, an *ex situ* conservation strategy is strongly needed to support the industrial purposes as an oil plant, through on farm conservation, while as for *in situ* conservation research to learn more about the breeding system and the vertical pollen transfer [113].
- *In situ* conservation is the most appropriate strategy for *C. tataria* subsp. *tataria*, although recent in vitro regeneration studies [109] suggest a possible long-term conservation of plant tissue by *ex situ* strategies.
- Habitat conservation at global level is very helpful because *C. tataria* grows in different habitats of 92/43/EEC Directive of several countries where it is reported, as Italy [Eastern sub-mediterranean dry grasslands (*Scorzoneretalia villosae*) (code 62A0)], Romania [Sub-pannonic steppic grasslands (6240*) and Ponto-Sarmatic steppes (code 62C0*)] [117], and Kazakhstan [Semi-natural dry grasslands and scrubland facies on calcareous substrates (*Festuco-Brometalia*) (code 6210*)] [118].
- The man-made summer fires with the aim to clean the soil in the habitat of *C. hispanica* subsp. *hispanica* (*Crambetum hispanicae*), when not avoidable, must be targeted to the dry component of the plant and carried out from the end of July, after the period of seed dissemination (May-June), otherwise the plants could be irreversibly damaged. As for the populations growing along the road, it is crucial that bodies responsible of road management be informed about the presence of threatened species. The species in question is linked to abandoned or extensively managed agricultural areas, which have a good naturalness (defined HNVF= High Nature Value Farmland) due to the immediate proximity of shrubland or woodland vegetation. Therefore, one of the main threats to the survival of the species and its plant community is the current trend to use high impact agriculture, including chemical input, such as herbicides.
- Monitoring of known populations for both species.
- Maintenance of *C. hispanica* subsp. *hispanica* in Botanical Gardens for educational purposes also.
- Genetic research (*ad hoc*) to define the gene pools of *C. tataria* subsp. *tataria*.

4.6 Ipomoea sagittata Poir.

The genus *Ipomoea* L. has an amphi-Atlantic distribution, and probably it arrived in Europe only after contact with the new world [119], with 18 species [120]. *I. sagittata* is known in the eastern Atlantic and the Mediterranean region from Algeria, the Balearic Islands, Corsica, Cyprus, Greece, Lebanon, Italy, Malta, Portugal, Sicily, Spain, Syria, Tunisia, and Turkey [120]. Austin [121] suggests that *I. sagittata* is native to the circum-Caribbean region of the Americas and it probably arrived in Europe for the first time in Greece or France by sea with maritime trade and then it spread to other Mediterranean territories. Other authors [122] believe that the way had been the seed dispersion mediated through ocean currents and this would explain probably why *I. sagittata* grows in the salt marsh, and made it to Europe in prehistoric times. Thus, we can conclude that due to the large disjunction in its distribution it is a controversial species since it probably is, even if introduced a long time ago, an exotic wild species [123]. So that, its nativity needs to be re-evaluated.

In Italy, *I. sagittata* grows with other nine species of the same genus: *I. stolonifera* (Cyr.) J.F.Gmel. [48] and eight exotic taxa [*I. batatas* (L.) Lam., *I. cairica* (L.) Sweet, *I. coccinea* L., *I. indica* (Burm.) Merr., *I. pandurata* (L.) G.Mey., *I. purpurea* (L.) Roth, *I. tricolor* Cav., *I. triloba* L.] [124]. Italian distribution of *I. sagittata* include Latium, Apulia, Calabria and Sicily regions [48].

I. sagittata is a rhizomatous geophyte flowering from June to September, typical of coastal marshes, wet brackish muds and banks [125] always very localized in Italy and threatened by the rarity and vulnerability of the environments in which it grows as shown by its disappearance in historic sites such as those of the coast of Mondello (Palermo - Sicily), "Pantano del Taro" (Taranto - Apulia) and on the islet of "S. Nicolicchio" (Taranto - Apulia) for reasons related to human activity [125]. Fortunately, the botanical explorations have made it possible to discover new stations in the Salento peninsula (Apulia Region) such as those at "Le Cesine" [126] "Palude di Rauccio" (Lecce) [127], "Laghi Alimini" [128], "Torre Rinalda", Basins of Ugento, at "Punta Prosciutto" in the "Palude del Conte" [125], and "Torre Chianca" [129], in many localities in Province of Trapani (Sicily), such as "Isola Grande dello Stagnone" [130], "Santa Ninfa" [131], and "Petrosino" along the drainage canals [132]. It is confirmed along the southern edges of "Lago Fondi" and "Canale S. Anastasia" into Regional Natural Park of "Monti Ausoni e Lago di Fondi" in Lazio Region [133].

The chromosome number is 2n=30 (from accessions of Pali district in India) [134] where it has been reported with a synonym (=*I. sagittifolia* Ker Gawl.) and with an interesting information on 77% of pollen fertility.

This species is listed as VU in the European Red List and as EN in the Italian National Red List (Table 1), with high priority *in situ* (A) and *ex situ* (HP) (Table 2), while there are no data about gene pool (Table 3). It is worthy to note that despite its limited Italian distribution, the species grows in several wetland types of vegetation as companion species, i.e. *Spartino-Juncetum maritimae* O. Bolòs 1962, *Soncho maritimi-Cladietum marisci* (Br.-Bl. & O. de Bolòs 1957) Cirujano 1980 [135], *Rubo ulmifolii-Myrtetum communis* Biondi & Bagella 2005, *Schoeno nigricantis-Plantaginetum crassifoliae* Br.-Bl. in Br.-Bl.., Roussine & Nègre 1952, *Schoeno nigricantis-Erianthetum ravennae* Pignatti 1953 [136] all observed in the Salento peninsula, and *Ranunculetum peltati* Sauer 1947 at Anguillara (Trapani) in Sicily [137]. It is also considered a diagnostic *taxon* of the alliance *Calystegion sepium* Tüxen ex Oberdorfer 1957 nom. mut. propos. Rivas-Martínez, T.E. Díaz, Fernandez-Gonzales, Izco, Loidi, Lousã & Penas 2002, that encloses the nitrophilous tall-herb communities that develop in humid, periodically inundated, habitats that are subjected to long periods of drainage and occasionally with a moderate salinity [138]. This peculiar type of vegetation is reported in Annex I of Directive Habitat 92/43/ECC as "Hydrophilous tall herb fringe communities of plains and of the montane to alpine levels" (code 6430) (Table 2).

It has been observed that in North-America *I. sagittata* can be harmful for agricultural purposes because harbor an insect "the weevil" that can infest crop potato, though found only in limited sections of the sweet potato-growing areas, mostly in the coastal and tide-marsh margins [139]. There are no recorded medicinal uses on *I. sagittata* in the Old World [121], and there are doubts that it is a real CWR, as indicated by Bilz et al. [16].

4.6.1. *Expected actions*

- Clarify with *ad hoc* studies and researchers (including historical ones) if the *taxon* is exotic or native in Europe.
- Monitoring of populations in known areas and field surveys to find new sites, especially in Lazio Region, where there are few sites.
- *Ex situ* germplasm conservation is needed because it has zero accessions in the RIBES seed banks, but *in situ* conservation actions are also important especially where it is highly threatened.
- Researches on gene pools and the content of microelements are welcome.

- Interviews, where the taxon is more widespread (e.g. in Salento), to check possible uses.

4.7 Lathyrus amphicarpos L., *Lathyrus palustris* L.

Some species of genus *Lathyrus* L. play an important role in the improvement of cultivated crops. According to the first results of interspecific hybridization, the closest allies of *L. sativus* Sibth. & Sm. ex Steud. (grasspea) were *L. amphicarpos* and *L. cicera* L. recorded in the secondary gene pool (GP2) of the grasspea [140,141]. Heywood et al. [142] extended the secondary gene pool also to *L. chrysanthus* Boiss., *L. gorgoni* Parl., *L. marmoratus* Boiss. & Balansa ex Boiss. and *L. pseudocicera* Pamp., with which *L. sativus* can cross and produce ovules, and only more remotely to *L. amphicarpos*, *L. blepharicarpus* Boiss., *L. chloranthus* Boiss. & Balansa, *L. cicera*, *L. hierosolymitanus* Boiss. and *L. hirsutus* L., with which *L. sativus* can cross to form pods. *L. gorgoni* and *L. hirsutus* are also reported in Italy [48]. The remaining species of the genus can be considered members of the tertiary gene pool (GP3) [143]. The results of electrophoretic comparative analysis of seed albumins and globulins showed *L. sativus* to be considerably different from the allied species. Consequently, exploitation of the germplasm resources in the breeding improvement programmes of the grasspea should be concentrated on the primary gene pool (GP1), as suggested by Yunus and Jackson [141].

L. palustris is a perennial plant and natural autohexaploid, having 2n=6x=42 [144], while *L. amphicarpos* has 2n=14 [145].

L. amphicarpos is a Mediterranean *taxon* which occurs in Algeria, Balearic Islands, France (Corsica), Greece, Crete, Italy, Morocco, Portugal and Spain, while *L. palustris* has a wider distribution being a circumboreal taxon [146]. In Italy, *L. amphicarpos* grows only in Sicily, Apulia and Latium Regions, while *L. palustris* has a fragmentary distribution in the northern regions and is absent in the Center-South of the Italian Peninsula [48]. *L. amphicarpos* grows in Latium on arid meadows and garrigues from 250 up to 600 m a.s.l. in the Ausoni chain on Mt. Leano [147], Mt. Cucca, M. Cavallo Bianco and M. Saiano [148], in the Natural Reserve “Pizzo Cane, Pizzo Trigna and Grotta Mazzamuto” (north-west of Sicily) [149], in other sites of Palermo municipality, at Monte Sparacio (Trapani) on Nebrodi mountains (Messina) in Sicily [150], and in the southern sector of Daunia Mts. in Apulia [151].

Among wild species of genus *Lathyrus*, *L. amphicarpos* showed the best results as antioxidant activity in seed methanolic extracts [152] and higher total saturated fatty acids. This data combined with the benefits attributable to the secondary metabolites (polyphenol contents), suggests the use of the genus *Lathyrus*, and in particular of *L. amphicarpos* in human and animal diet [153]. *L. palustris* has also favorable histological characteristics for use as fodder crop [145,154].

L. amphicarpos is listed as NT in the European Red List and as LR in the Italian National Red List (Table 1), with high *in situ* (A) and high *ex situ* (HP) priority (Table 2), as secondary gene pool (GP2) (Table 3) giving rise viable hybrids in crosses with *L. sativus* [141,155], while *L. palustris* is listed as EN in the Italian Red List (Table 1), with high *in situ* (A) and *ex situ* priority (HP) (Table 2) and with no information on gene pool (Table 3).

L. amphicarpos is an annual plant with an elongated flower axis, often without leaves, that flowers from March to April, from sea level to 600 m of altitude. The only ecological information in Italy comes from Sicily where the species is found in limestone and stony ground with sparse vegetation of annual species located in degraded *Ampelodesmos mauritanicus* (Poir.) T. Durand & Schinz grasslands subjected to the action of fire and grazing, in stations exposed to the action of atmospheric elements that cause soil erosion. In these habitats the species spreads its slender roots among the stones, developing the phenomenon of amphicarpy, a typical adaption in acid habitats, subject to fire [150].

L. palustris is a perennial plant that flowers from June to August, from sea level to 800 m of altitude, for which few ecological data are available, especially about the vegetation in which it grows. In Alto Adige it is observed in the humid, uncultivated grasslands of the *Molinion caeruleae* Koch 1926 alliance [156], attributable to the habitat 92/43/EEC "*Molinia* meadows on calcareous, peaty or clayey-silty soils (*Molinion caeruleae*)" (code 6410), though it is a diagnostic of the

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"Mediterranean high and humid herbaceous grasslands of *Molinio-Holoschoenion*" habitat (code 6420) [157]. However, there is a gap in phytosociological studies for this species.

4.7.1. *Expected actions*

- *In situ* and *ex situ* conservation to prevent the risk of extinction by increasing the number of existing wild populations for both *Lathyrus* species.
- Use in the human diet of *L. amphicarpos* due to a good antioxidant activity present in their seeds and for the presence of high content of saturated fatty acids [153], but only after research and breeding with the aim to turn saturated to unsaturated fat acid content.
- Monitoring the known populations of *L. amphicarpos* and field surveys to find new sites because of the earlier confusion with *L. cicera*, which is very similar in morphology, though much more widespread in Italy.
- Specific ecological studies on *L. palustris* wetlands habitats, which could provide information on factors related to maintenance of wetlands and their conservation, considering the potential benefit of use as grazing fodder, especially for buffalo [158].
- Ecological and phytosociological studies for *L. amphicarpos*, since literature is poor.

4.8 Vicia cusnae Foggi & Ricceri , *Vicia serinica* R. Uechtr. et Huter

V. cusnae and *V. serinica* are two orophytes, systematically closed and belonging to a group formed also by *V. canescens* Labill. (Makmel massif in Lebanon), *V. variegata* Willd. (Erzerum mountains in Turkish Armenia) and *V. argentea* Lapeyr. (Central Pyrenees) [159], and can be considered geographic vicariates among themselves [160], although there are different readings such as that of Davis [161] which consider *V. serinica* and *V. variegata* as a subspecies *of V. canescens*. There are several morphological differences between the species of the group that concern microcharacters located in different parts of the plant [160].

V. cusnae is reported in three circumscribed sites, two in Italy, in the National Park of the Tuscan-Emilian Apennines (Emilia Romagna Region), at M. Cusna [160] and at Rio Re at M. Prado [162], and finally thanks to Philippe Küpfer in France in the Aurouze Massif [15]. *V. serinica* is reported in southern Italy and in northern Greece [163]. In Greece it was collected by Gustavsson in several mountains at Sterea Ellas, by Aldén from Mount Kakarditsa in Pindhos [164], and by Strid and Papanicolaou [165] on Mt. Belles (Kerkini), NE of the village of Ano Poroia. In Italy, it occurs only in a very confined area of Basilicata region, while it was reported by mistake in Campania [48]. In Basilicata it grows in only four stations of Sirino-Papa Massif [166,167], in the municipality of Potenza (recently confirmed by E.V. Perrino, unpubl. data).

V. cusnae and *V. serinica* have both the same chromosomal number (2n=10) just like the other three species of the group [160]. This datum can be interpreted as schizoendemisms with puntiform distribution and geographically isolated [168] that makes highly improbable genetic exchanges between the populations of these two species with the other conspecific populations.

V. cusnae is an alpine glareicole taxon that flowers from July to August on detrital soils of sedimentary rocks with southern exposure and in xerothermic conditions, from 1800 to 2100 m of altitude [15]. It reproduces mainly by vegetative parts, thanks to the presence of short underground stolons, which issue new closely shoots [15], as was also observed for *V. serinica* at M. Sirino (observed by E.V. Perrino, unpubl. data), rather than by seed dispersal [169]. It covers large areas in which it is a dominant taxon and it is referable to *Thlaspion rotundifolii* Jenny-Lips 1930 alliance [170] and habitat 92/43/EEC "Calcareous rocky slopes with chasmophytic vegetation" (code 8210).

V. serinica flowers in July on soils similar to those of *V. cusnae*, from 1500 to 1850 m of altitude, colonizing peculiar niches reserved for highly specialized species that are able to grow in extreme environmental conditions. The soil has a copious skeleton in the superficial horizons and a high sand content in all layers. The annual average precipitation is about 1400 mm, while the bioclimate is oceanic temperate of the humid supratemperate type [167]. The vegetation of *V. serinica* is to be referred to *Sideridenion italicae* Biondi et al. 1995 corr. Biondi, Allegrezza & Zuccarello 2005 sub-alliance, with a conspicuous number of species of the *Thlaspietalia rotundifolii* Br.-Bl. In Br.-Bl. et

Jenny 1926 order [167], to be related to the priority habitat 92/43/EEC "Semi-natural dry grasslands and scrubland facies on calcareous substrates (*Festuco-Brometalia*) (*important orchid sites)" (code 6210*) [30].

V. cusnae and *V. serinica* are listed respectively as VU [17] and EN [24] in the Italian Red List (Table 1), with high *in situ* (A) and *ex situ* priority (HP) (Table 2). There are no available data on their gene pools (Table 3).

4.8.1. *Expected actions*

- Research *in situ* with monitoring programs to better understand the reproductive biology and ecology of the species and the populations trends.
- Evaluate ecological and genetic affinities between the different populations of both species.
- Phytosociological studies to define the phytosociological association and discover why similar environments produce different types of vegetation and habitats.
- *Ex situ* conservation for both species. For *V. cusnae* it is possible, since it is an orthodox species, which means that it tolerates seed drying with high levels of germination (80%) after scarification, at 21 °C [171]. The only germplasm accessions of *V. cusnae* are preserved at the Millennium Seed Bank of the Royal Botanic Gardens in Kew (U.K.) and those of *V. serinica* in the seed bank collections of the Institute of Biosciences and Bioresources (IBBR - CNR) of Bari, but both are absent in RIBES seedbanks.
- Start crossbreeding studies with *V. sativa* L., whose seeds are consumed by birds and often used as forage, to test their gene pools and to check their taxonomy and systematics.

5. Conclusions

In Italy, according to the taxon group concept there are 43 CWR at risk for not adequate conservation either *in situ* or *ex situ*. However, disregarding the species endemic to Italy, the number of 43 falls down to 14. Further, according to the gene pool concept, which is more important from a plant breeding point of view, the number of 14 comes down to 8. For these latter species this paper provides a picture as complete as possible about their geographical distribution, level of protection, ecology (including vegetation and habitat 92/43 EEC), properties, gene pools and actions to avoid further genetic erosion, to improve *in situ* and *ex situ* conservation of the species and habitats, with the final goal of enhancing genetic resources management and their use both in plant breeding and to promote sustainable agriculture and environmental conservation through *ad hoc* research, suggested for each of the 14 CWR considered at risk.

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