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RESEARCH ARTICLE

The World Saffron and *Crocus* collection: strategies for establishment, management, characterisation and utilisation

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Abstract Since 2007, the European Commission AGRI GEN RES 018 "*CROCUS*BANK" action has permitted the creation of the alleged World Saffron and *Crocus* Collection (WSCC), a unique collection which contains a representation of the genetic variability present in saffron crop and wild relatives at global scale. At present the germplasm collection, housed at the Bank of Plant Germplasm of Cuenca (BGV-CU, Spain), consists of 572 preserved accessions representing 47 different *Crocus* species (including saffron *Crocus*) and is expected to increase up to more than 600 accessions by the end of *CROCUS*BANK action (May 2011). The preserved biodiversity of saffron (*Crocus sativus* L.) covers a wide range of the genetic variability of the crop and

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currently consists of 220 accessions from 15 countries: 169 of these come from European cultivation countries, 18 from commercial areas in non EU countries, 26 from regions of minimal or relict production and/or from abandoned fields and 7 from commercial nurseries. The non-saffron *Crocus* collection currently comprises 352 accessions: 179 collected from the wild in 12 countries of natural distribution, 24 from donations of public and private institutions, 91 from commercial nurseries and 58 acquired from BGV-CU collection management. Here we provide a record of collections, activities concerns and current strategies for documentation, conservation, characterisation, and management of the collection as important tools for researchers with interest in these valuable genetic resources.

Keywords Genetic resources · Database · Crop diversity · *Ex situ* conservation

Introduction

One of the main goals of the Convention on Biological Diversity (CBD 1993) is to achieve a significant reduction of the current rate of biodiversity loss, including plant taxa more or less closely related with species of direct socio-economic importance such as food, medicinal, condiments, ornamental, etc. (Maxted et al. 2007). A major step towards achieving this goal is improving efforts for systematic conservation of plant genetic resources, to ensure adequate and representative diversity for future uses including breeding programmes.

Crocus L. belongs to the subfamily *Crocoideae*, the largest of the four subfamilies currently recognized in *Iridaceae* family (Goldblatt et al. 2006). The genus consists of 88 small, corm-bearing, perennial species distributed in Central and Southern Europe, North Africa, and from Southwest Asia to Western China (Mathew 1982; Petersen et al. 2008) and are highly prized as garden plants for their colourful flowers, horticultural varieties, for industrial applications and as unique collector's items (Rashed-Mohassel 2007; Petersen et al. 2008). The majority of taxa (species and subspecies) are restricted to Turkey and the Balkan Peninsula. Greece alone is homeland of ca. 40% of the world's wild *Crocus* diversity (Tsoktouridis et al. 2009) while a total of 32 species (18 of them being endemic) are included in Turkey′s

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flora (Arslan et al. 2007). Several countries have also representatives of some *Crocus* species including Italy (10 species), Spain (6 species), Hungary (6 species), and others.

The genus is primarily known by *C. sativus*, commercially cultivated for the production of the spice saffron (Fernández 2004). Saffron is a high-value, sustainable crop where improvement is possible through exploitation of biodiversity, and it contains many novel or poorly characterized bioactive molecules consistent with its use as a spice and medicinal supplement over thousands of years (Abdullaev 2002, 2004; Abdullaev and Espinosa-Aguirre 2004; Radjabian et al. 2009; Dalezis et al. 2009). Saffron spice is made from the dried stigmas of the saffron flower (*C. sativus*), a triploid sterile plant species that is vegetatively propagated by means of corms (vulgarly called bulbs or "onions"). Because corm multiplication does not induce genome variations with the exception of random mutations that in a triploid saffron population are not easily detectable, it is supposed that saffron material should be similar around the world. Although different commercial products are known that possibly suggests the existence of different saffron ecotypes or commercial varieties, nevertheless, the actual genetic variability present in *C. sativus* at worldwide scale still remains unknown. There is a general suspicion regarding the existence of scarce variation, but no serious effort has been carried out to ascertain this important issue until recently when the *CROCUS*BANK initiative arose (http://www.crocusbank.org).

The decrease of land surface dedicated to saffron crop in many areas has possibly resulted in corresponding genetic erosion that adds up to the limited genetic variation suspected for *C. sativus* due to its sterile habit. Thus, the situation seems dramatic at present time and compromises any attempt of genetic improvement regarding this highly-valued crop (Fernández 2004, 2007; De-Los-Mozos-Pascual et al. 2010a, b). Consequently, the creation of a germplasm bank of this species can be considered as a great achievement in the first place. In addition, the inclusion of wild species related have proved useful to saffron improvement as sources of a variety of valuable traits taking into account that the wild portion of a crop genepoll generally contains much greater genetic variation than that contained in the cultivated taxa (Khoury et al. 2010).

Materials and methods

Collecting expeditions

In order to gather a good representation of the plausible genetic variability present in the saffron crop, the collecting places were subdivided in (a) zones of commercial cultivation of saffron (EU countries and outside EU) including Protected Denomination of Origin (DOP) and (b) worldwide zones of remaining yields of saffron spice including several abandoned fields. To collect samples of saffron allies (*Crocus* spp.) four sources of germplasm were exploited: (a) from the wild (natural populations), (b) from botanical gardens, (c) from commercial nurseries and (d) from BGV-CU collection management (seeds).

Conservation strategies

Experimental farm

The collection is housed at the Bank of Plant Germplasm of Cuenca (BGV-CU, Spain). The experimental farm is located in the latitude and longitude 40°04′08.17″ N and 2°11′57.07″W, respectively, with altitude between 950 and 1,000 m, average annual temperature of 11.5°C, and precipitation ranging from 550 to 600 mm mainly concentrated in spring and autumn. The edaphic characteristics in the plot are typical of the area with sandy loams, alkaline pH (7.6–8.4), normal electric conductivity (<400 nmhos/cm) and low content in organic matter (1–2.5%).

Ex situ conservation and multiplication

Genetic materials are preserved in the form of corms (saffron and wild relatives) and seeds (only wild species). General conservation and multiplication strategies for the collections are the standards for international genebanks outlined by Engels and Visser (2003). In addition, specific strategies and bank design for the *ex situ* conservation were established mainly based on information regarding the source of the species, reproductive biology, mode of multiplication, sample type and objectives of the collection. The design comprises three main collections:

1. Reserve Vegetative Collection (saffron and allies): 10 corms of each accession were sown in special flower-pots with substrate of the collecting zone and/or specific mixture (soil enriched with organic matter mixed with sand) and placed in a greenhouse with semi-controlled conditions. Irrigation and weeding were done by hand when necessary.
2. Exchange vegetative Collection (saffron and allies): The accessions (40 corms each) were sown in the experimental farm (field conditions) in a 10–15 cm furrow, with 15 cm among plants and 50 cm among furrows. Conventional labouring was used to prepare soil for seeding and previous fertilization applying N (80 UF), P (100 UF) and K (100 UF) was carried out. Irrigation schedule was applied depending on the climatic conditions.
3. Seed collection (wild *Crocus*): The acquired seeds both from the wild and from seed harvesting in BGV-CU were placed in hermetic jars including silica gel inside and stored in a refrigerated chamber at 4°C and 30% relative humidity.

Documentation

Descriptors definitions

In order to facilitate retrieval and updating of information, descriptors were grouped attending to the categories proposed by Bioversity (Bioversity International 2007): passport (accession, collecting, ethnobotanical data), management, site and environment and characterisation. Passport, site and environment descriptors were adapted from the multicrop passport descriptors (MCPD) proposed by Bioversity with the inclusion of three specific descriptors: registry of income (hyperlink field), intermediary (in the acquisitions process) and photo (Ole object). Two groups of descriptors for management, seed management descriptors and vegetative management descriptors, were adapted and/or newly defined. In both cases (saffron and its wild relatives), passport information was gathered through formal Collecting Form Sheet accompanying each accession during the acquisitions process (collecting, donations, etc.) and available for download from http://www.crocusbank.org.

Central information system for Crocus germplasm

Documentation and management of the collection have been running through the Central Information System (CIS), a comprehensive electronic platform that provides an interactive documentation system including relational database and a web interface. The CIS has been developed using *html* and *asp* pages (Macromedia Dreamweaver© CS4) as a front-end application and MS-Access© 2007 as a back-end application.

Characterisation and evaluation

First, we defined a list of descriptors valid for the genus *Crocus*, based on the revision of taxonomic criteria for the genus (Mathew 1977, 1982), as well as our experience of research team involved and the preliminary characterisation assays carried out (De-Los-Mozos-Pascual et al. 2010c). We also took into account the categories present in other Bioversity lists of descriptors (http://www.bioversityinternational.org).

Regarding to characterisation, we have currently evaluated 100 accessions, including 66 saffron accessions from different origins (Azerbaijan, France, India, Iran, Italy, Morocco, New Zealand, Spain and Turkey) and 34 other *Crocus* accessions from 21 different species. For these purposes, a randomized block design, with three blocks and ten corms per accession and block has been followed in the facilities of BGV-CU.

Present germplasm activities and procedures

In agreement with Khoury et al. (2010), among the most important current issues regarding the conservation and use of plant genetic resources in the BGV-CU are: acquisition of germplasm (collecting), multiplication and conservation, documentation (information systems), definition of crop descriptors, characterisation and user priorities (supplying). A view of the main activities carried out in the BGV-CU is shown in Fig. 1.

Germplasm acquisition

The development of efficient collecting strategies depends on the extent of the information of the type of genetic variation in target populations of taxa and their distribution in the target geographical region (Upadhyaya et al. 2006, 2008). Therefore, the genetic diversity of the saffron crop in the farmer field, relict

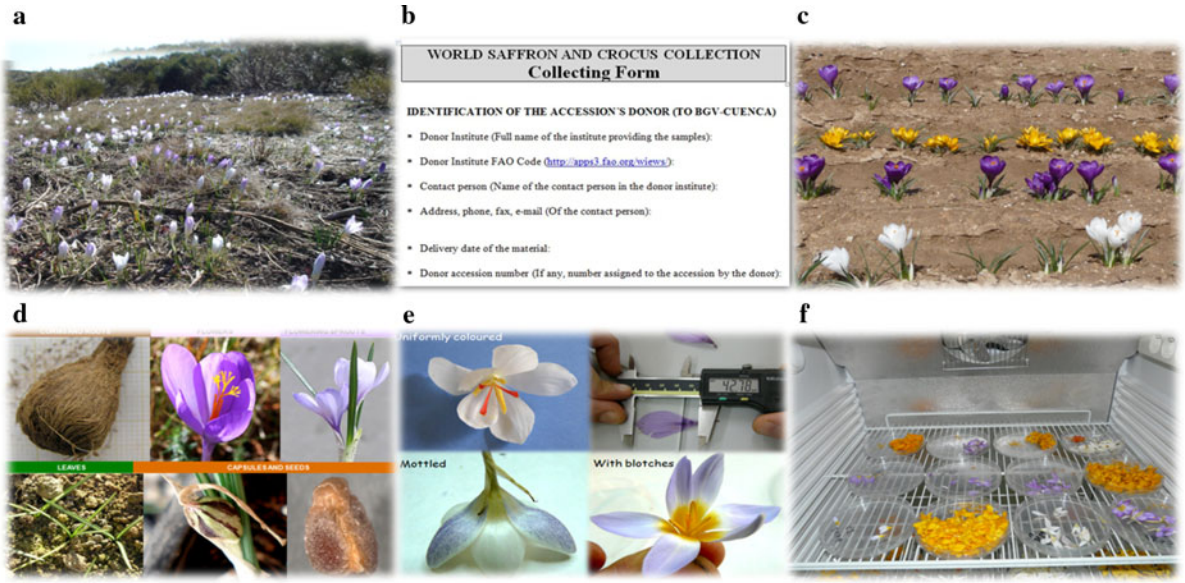

Fig. 1 Major activities at BGV-CU: **a** Collecting, **b** Documentation through the Collecting Form and the Central Information System (http://www.crocusbank.org), **c** *Ex situ* conservation and multiplication **d** Descriptors definition, **e** Characterisation, and **f** Management of germplasm materials for supplying

zones or abandoned fields has been complemented with the diversity present in the relatives of the crop (*Crocus* spp., see Fig. 2).

Collecting expeditions

Since 2006, eighty-one (81) acquisition trips have been made to collect saffron accessions (Table 1). The results of exploration and collection have yielded saffron germplasm from commercial areas in EU countries (5 countries, 169 accessions), from commercial areas in other non-EU countries (7 countries, 18 accessions) and from zones of remaining minimal or relict productions (5 countries, 26 accessions). The acquired materials cover a wide range of the genetic variability of the crop. The maintenance of accessions from relict areas is an effort to rescue the germplasm that has been lost due to the reduction of land surface dedicated to the saffron crop in the last decades.

In addition, as a result of a large-scale international collecting effort, during the period 2006–2009, one hundred and one (101) expeditions, lasting from 1 to 4 days each, were carried out covering phytogeographical regions of 10 countries where wild *Crocus* taxa have been documented (Table 2). Two hundred and forty six (246) accessions, belonging to thirty-one (31) different *Crocus* taxa (only species) were acquired as the result of the mentioned effort. At this stage, it is reasonable to indicate that germplasm collection has a good representation of the wild *Crocus* germplasm of plausible utility in saffron breeding. However, wild relatives are increasingly valued and the consciousness of the need for more extensive collecting must be a permanent goal (Khoury et al. 2010).

Germplasm donations

Twenty-four (24) accessions corresponding to 13 *Crocus* species (subspecific taxonomic categories and commercial cultivars are not included in the statistics) have been acquired through donations from cultivated samples in botanical gardens. Three target *Crocus taxa* missed in collecting expeditions (*C. kotschyanus*, *C. ochroleucus* and *C. cancellatus*) were recently included in the collection through donations from botanic gardens (Table 3). Additional efforts such as contacting with private companies, traditional cultivators and producers, rendered donations of another 11 saffron accessions (Table 4).

Acquisition from nurseries

A total of ninety-one (91) accessions from the most reputed nurseries worldwide have been acquired. As

Fig. 2 Some of the wild *Crocus* preserved: **a** *C. sativus* L., **b** *C. sativus* L. var. *cashmeriensis*, **c** *C. cartwrightianus* Herb. cv. Albus, **d** *C. cartwrightianus* Herb., **e** *C. hadriaticus* Herb., **f** *C. mathewii* Kerndorff et Pasche, **g** *C. asumaniae* Mathew et Baytop, **h** *C. pallasii* Goldb., **i** *C. thomasii* Ten., **j** *C. kotschyanus* Koch, **k** *C. ochroleucus* Boiss. et Gaill., **l** *C. imperati* Ten., **m** *C. minimus* DC., **n** *C. corsicus* Maw, **o** *C. serotinus* Salisb., **p** *C. nevadensis* Amo, **q** *C. carpetanus* Boiss. et Reut., **r** *C. fleischeri* Gay

a result of these efforts, nineteen (19) new species (subspecific taxonomic categories and commercial cultivars are not included in the statistics), that failed to be acquired through collecting expeditions and donations, were included in the collection (Table 5). Taking into account the importance of the genus as a

Table 1 Saffron (*C. sativus* L.) germplasm acquisition trips

Year	Country	Collecting objectives	Principals	Expeditions
2006	Morocco	Commercial area of *Talliouine*	UCLM (Spain)	1
2006	Iran	Commercial areas of *Khorasan*	TJMT (France)	4
2006–2008	Spain	Commercial areas including DOP "*Azafrán de La-Mancha*"	UCLM, UPVLC, JCCM (Spain)	32
2006–2007	Spain	Commercial areas of *Teruel* including DOPs (in course) "*Azafranes del Jiloca*" and "*Azafranes de Campo Bello*"	UCLM, UPVLC (Spain)	5
2007–2010	Spain	Zones of minimal or relict production and/or abandoned fields in Spain	UCLM, UPVLC, JCCM (Spain)	13
2007	Italy	Commercial areas of *Perugia* and *L'Aquila* including DOP "*Zafferano dell'Aquila*"	UNICIT (Italy)	3
2007	Italy	Commercial areas of *Tuscany* and *Abruzzo* including DOP "*Zafferano di San Gimignano*"	UCLM (Spain)	2
2007	Hungary	Relict Hungarian saffron from *Csongrad* and *Pest megye*	UD (Hungary)	2
2008	Azerbaijan	Collection in relict areas of *Aspheron*	ANAS (Azerbaijan)	2
2008	Greece	Commercial areas including DOP "*Krokos Kozani*"	NAGREF (Greece)	1
2008	Turkey	Traditional areas of *Safranbolu*	GU (Turkey)	2
2008–2009	France	Commercial areas of *Quercy* and *Gâtinais*	INPT, TJMT (France)	10
2009	Italy	Commercial areas of *Medio Campidiano* including DOP "*Zafferano di Sardegna*"	UNICIT (Italy)	1
2010	Greece	Commercial areas including DOP "*Krokos Kozani*"	NAGREF, AUA (Greece)	2
2010	Morocco	Traditional cultivation area of *Ourika* Valley	UCLM (Spain)	1

source of ornamental plants we included some of the most priced cultivars grown as garden plants.

Germplasm *ex situ* conservation concerns and strategies

At present, the collection contains 572 accessions representing 47 *Crocus* species (subspecific taxonomic categories and commercial cultivars are not included in the statistics) from 18 countries. In this connection, the BGV-CU has two main objectives: the first involves material conserved for the medium and long term, with the aim of preserving the genetic information in the accession, and the second concern material currently in use or about to be used. The current status of the collection is summarized in Table 6.

For many crops, but particularly the vegetatively propagated species, the development of new strategies, or extension of strategies to more genotypes (e.g. wild accessions) is needed for better conservation systems (Khoury et al. 2010). The experience gained during 3 years of conservation has guaranteed the representativeness of accessions through the different collections in the bank design as a good strategy for further exploitation of the collection (Table 7).

Reserve vegetative collection

This collection has been created for the long-term storage and preservation, the recovery of missing accessions during the multiplication process and the preservation of the germplasm at BGV-CU. No distribution or exchange of material is achieved from this collection. At present, all the collection in vegetative form (corms) is represented in the RSVC (see Table 7) including saffron (220 accessions) and other *Crocus* (251 accessions).

Exchange vegetative collection

It refers to the collection kept for medium-term preservation and it is responsible for implementing distribution and utilisation (characterisation and evaluation). Ideally, the collection must be maintained in sufficient quantities to be available to the users. Currently around 100% of saffron accessions

Table 2 Wild *Crocus* germplasm acquisition trips, nomenclature according to Mathew (1982)

Year	Country	Collecting objectives	Principals	Expeditions
2006–2010	Spain	*C. serotinus* Salisb. ssp. *clusii* (Gay) Mathew & ssp. *salzmanii* (Gay) Mathew, *C. carpetanus* Boiss. et Reut., *C. nevadensis* Amo, *C. nudiflorus* Sm. *C. vernus* (L.) Hill ssp. *albiflorus* (Kit.) Asch. et Graebner, *C. cambessedessi* Gay	UPVLC, UCLM, JCCM (Spain)	40
2006–2007	Romania	*C. vernus* (L.) Hill ssp. *vernus* (=*C. heuffelianus* Herb.), *C. banaticus* Gay	UD (Hungary)	7
2006–2007	Ukraine	*C. vernus* (L.) Hill ssp. *vernus* (=*C. heuffelianus* Herb.), *C. banaticus* Gay	UD (Hungary)	2
2006 and 2008	Turkey	*C. ancyrensis* (Herb.) Maw, *C. speciosus* Bieb.	GU (Turkey)	2
2007	Slovaquia	*C. vernus* (L.) Hill ssp. *vernus* (=*C. scepusiensis* Rehm. et Wol.)	UD (Hungary)	1
2007	Hungary	*C. vernus* (L.) Hill ssp. *vernus* (=*C. vittatus* Schloss et Vukot), *C. tommasinianus* Herb., *C. reticulatus* Stev. ex Adams ssp. *reticulatus*, *C. vernus* (L.) Hill ssp. *vernus* (=*C. heuffelianus* Herb.)	UD (Hungary)	4
2007–2009	Italy	*C. thomasii* Ten., *C. vernus* (L.) Hill ssp. *vernus* et ssp. *albiflorus* (Kit.) Asch. et Graebner, *C. imperati* Ten. ssp. *imperati*, *C. longiflorus* Raf., *C. biflorus* Mill. ssp. *biflorus* & ssp. *weldenii* (Hoppe et Fürnr.) Mathew, *C. ligusticus* Mariotti, *C. etruscus* Parl., *C. versicolor* Ker Gawl.	UNICIT (Italy)	16
2008	Azerbaijan	*C. biflorus* Mill. ssp. *adamii* (Gay) Mathew, *C. caspius* Fisch. et Mey, *C. speciosus* Bieb. ssp. *speciosus*	ANAS (Azerbaijan)	4
2008	Greece (Cyprus)	*C. cyprius* Boiss. et Kotschy, *C. hartmannianus* Holmboe	NAGREF (Greece)	1
2008–2010	Greece	*C. hadriaticus* Herb. ssp. *hadriaticus* & *parnassicus* (Mathew) Mathew, *C. cartwrightianus* Herb., *C. sieberi* Gay ssp. *sublimis* (Herb.) Mathew & *atticus* Boiss. et Orph., *C. pulchellus* Herb., *C. veluchensis* Herb., *C. goulimyi* Turril ssp. *goulimyi* & ssp. *leucanthus* (Mathew) Mathew, *C. boryi* Gay, *C. cvijicii* Košanin, *C. tourneforti* Gay, *C. niveus* Bowles, *C. olivieri* Gay	NAGREF (Greece)	24

and 77% of other *Crocus* accessions are represented in EXVC (Table 7).

Seed collection

Due to the biological characteristics of *Crocus* species, the collection is maintained and reproduced basically in a vegetative way. However, we have preserved seeds of 101 accessions from 20 different *Crocus* taxa (Table 7).

Germplasm documentation

Documentation of information on Plant Genetic Resources (PGR) is imperative for planning and implementing activities related to their conservation, sustainable utilisation and sharing of benefits accrued from their use (Agrawal et al. 2007). The need of maintaining and exchanging such information is specifically recognized in Articles 7d and 17 of the Convention of Biological Diversity (CBD 1993). The design of global information database, are of extreme importance to the conservation and use of collections (Wallace et al. 2008; Pandey et al. 2008; Ravisankar et al. 2008; Upadhyaya et al. 2008; Khoury et al. 2010). The conversion of off-line database to a searchable on-line database is a high priority on acting to build better information systems (Khoury et al. 2010).

Database implementation and descriptions (back-end application)

A specific relational database with 28 related tables, 20 data entry forms, 2 reports, and 60 queries based on the passport and management descriptors

Table 3 Germplasm donations from botanical gardens recorded since 2006 in the BGV-CU, Spain, nomenclature according to Mathew (1982)

Institution	Germplasm	Accessions
Conservatoire et Jardin Botaniques de la Ville Genéve (Switzerland)	*C. vernus* (L.) Hill. ssp. *albiflorus* (Kit.) Asch. et Graebner	1
Nationale Plantentuin van België (Belgium)	*C. kotschyanus* Koch. ssp. *kotschyanus*, *C. flavus* Weston	2
Botanic Garden Utrecht University (Netherlands)	C. *ochroleucus* Boiss. et Gaill., *C. vernus* (L.) Hill, *C. speciosus* Bieb., ssp. *speciosus*, *C. banaticus* Gay	3
Jardin des Plantes Médicinales et Aromatiques. Marie de Chemille (France)	*C. sativus* L.	1
Conservatoire Botanique National de Brest (France)	*C. goulimyi* Turrill	1
Niels Jacobsen. The Royal Veterinary and Agricultural University (Denmark)	*C. cartwrightianus* Herb., *C. hadriaticus* Herb., *C. goulimyi* Turrill, *C. cancellatus* Herb. ssp. *mazziaricus* (Herb.) Mathew	12
Ljubljana University Botanic Garden (Slovenia)	*C. pulchellus* Herb.	1
Jardin alpin du Lautaret, Université Joseph Fourier, Grenoble I (France)	*C. vernus* (L.) Hill	2
Herbario SANT (Spain)	*C. serotinus* Salisb. ssp. *salzmanii* (Gay) Mathew	1

Table 4 Donations of saffron germplasm from private companies, producers and/or traditional farmers

Year	Country	Donation objectives	Intermediaries	Accessions
2006	New Zealand	Micro-scale productions of *Rangiora*	UCLM (Spain)	1
2006	Argentina	Commercial areas of *Atos-Pampa*, *Córdoba*	UCLM (Spain)	1
2006	United Kingdom	Zones of minimal relic production in Cambridge	UCLM (Spain)	2
2007	Italy	Commercial areas including DOP "*Zafferano di Sardegna*"	UCLM (Spain)	1
2007	India	Commercial areas of Jammu and Kashmir	UCLM (Spain)	1
2008	Afghanistan	Commercial areas of *Herat*	UCLM (Spain)	1
2009	Switzerland	Ancient productions of *Mund* including DOP "*Munder Safran*"	UCLM (Spain)	6
2010	France	Saffron commercial lands of *Limousin*	UCLM (Spain)	1

mentioned above has been prepared. A control panel (main menu) with a series of user-friendly menus was designed in order to facilitate the accurate updating of database and to provide easy access to stored information. A link to photographic documentation and phenology behaviour of each accession has been included.

The database comprises the following categories:

a) Accession passport descriptors: 25 descriptors including identification data related to the registration of the sample at the germplasm bank.

b) Collecting passport descriptors: 26 descriptors providing detailed information regarding the conditions prevailing at the species-specific habitat in the natural environment.

c) Management: 20 descriptors are being recorded providing detailed information regarding the current status of conservation and multiplication of each accession through the different collections.

d) Characterisation/Evaluation: A database structure with specific fields is being prepared to include characterisation data after publication.

Web interface (front-end application)

Documentation systems through internet enable rapid dissemination of the database information to users. In this connection, we developed and currently manage the CIS (Santana et al. 2010). The web interface

Table 5 New *Crocus* species incorporated in the BGV-CU from nurseries, nomenclature according to Mathew (1982)

Species	Accessions acquired	Nursery	Principals
C. asumaniae Mathew et Baytop	2	JW Dix Export (The Netherlands)	ULEIST (United Kingdom)
C. corsicus Vanucci	1	Pottertons Nursery (United Kingdom)	UCLM (Spain)
C. dalmaticus Vis.	1	Pottertons Nursery (United Kingdom)	UCLM (Spain)
C. fleischeri Gay	1	Pottertons Nursery (United Kingdom)	UCLM (Spain)
C. korolkowii Regel ex Maw	1	Pottertons Nursery (United Kingdom)	UCLM (Spain)
C. kosaninii Pulević	1	Pottertons Nursery (United Kingdom)	UCLM (Spain)
C. kotschyanus Koch.	1	Broadleigh Gardens (United Kingdom)	UCLM (Spain)
C. laevigatus Bory et Chaub.	1	Pottertons Nursery (United Kingdom)	UCLM (Spain)
C. malyi Vis.	1	Pottertons Nursery (United Kingdom)	UCLM (Spain)
C. mathewii Kerndorff et Pasche	1	Pottertons Nursery (United Kingdom)	UCLM (Spain)
C. medius Balb.	1	Broadleigh Gardens (United Kingdom)	UCLM (Spain)
C. oreocreticus Burtt	2	JW Dix Export (The Netherlands)	ULEIST (United Kingdom)
C. pallasii Goldb.	5	JW Dix Export (The Netherlands)	ULEIST (United Kingdom)
C. pestalozzae Boiss.	1	Pottertons Nursery (United Kingdom)	UCLM (Spain)
C. tournefortii Gay	1	Pottertons Nursery (United Kingdom)	UCLM (Spain)
C. vitellinus Wahlenb.	1	Pottertons Nursery (United Kingdom)	UCLM (Spain)

Table 6 Current status of the WSCC at the BGV-CU, Cuenca, Spain

Genetic Material	Accessions acquired[a]	Accessions Preserved[a]	Countries of origin
Saffron (*C. sativus*)	229	220	Afghanistan, Argentina, Azerbaijan, Spain, France, Greece, Hungary, India, Iran, Italy, Morocco, New Zealand, United Kingdom, Switzerland, Turkey
Other *Crocus* (47 species confirmed)	420	352	Azerbaijan, Slovakia, Spain, France, Greece, Hungary, Italy, United Kingdom, Romania, Switzerland, Turkey, Ukraine
Total	649	572	18 countries

[a] Differences between acquired and preserved accessions are due to losses during acquisition and/or management the materials

Table 7 Summarized *ex situ* conservation design and distribution strategy of the germplasm accessions at BGV-CU

	RSVC	EXVC	SC (wild taxa)
Objectives	Long-term conservation	Multiplication and distribution	Medium-term conservation
Conservation conditions	Greenhouse	In farm	Hermetic jars
Optimum quantity	10 corms	40 corms	>1,000 seeds
Saffron accessions	220	220	–
Other *Crocus* accessions	251	192	101

(http://www.crocusbank.org/Information%20System.html) comprises all the BGV-CU services subdivided in two main subsections:

a) Connections to a database in order to provide:

- Information related to the passport descriptors executing queries through direct links and/or searching in the database (http://www.crocusbank.org/Database/ buscador.asp),
- Information about the management of descriptors as the basis for further queries in order to inform on the availability of preserved materials and future request of accessions (http://www.crocusbank.org/Database/ConsultBulbsAvailability.asp),

- A summary of the characterisation descriptors recorded in the collection (will be available on Internet after publication).

b) Other BGV-CU services to the final users, including methodologies and protocols, interactive maps of current saffron producing areas and distribution of taxa in the genus *Crocus* (http://www.crocusbank.org/ammap_2.1.0/examples/drill_down/SaffronAreasMapNew.html), a review of the literature about saffron and other *Crocus* (http://www.crocusbank.org/Login.asp), documents related to the management of genetic resources and a photographic documentation of activities and actions made so far (http://www.crocusbank.org/GGallery.html).

Database on-line searches

Through the path: http://www.crocusbank.org/Database/buscador.asp, users can search all the information available from the CIS by identifying one or more of about 26 passport data of the accessions preserved. The text input files are set by default for an exact match (without accents, swung dash or other special characters). These searches can be modified to find records containing any part of the text the user needs to find. Complex queries can be executed integrating data from multiple passport data (Santana et al. 2010).

Germplasm distribution

A basic criterion for supplying plant materials including dates of request and sending has been established based on phenology of the materials and facilities at the BGV-CU. At the short time the provision of genetic materials (corms, leaves, styles, anthers and seeds) is addressed to carry out the complete characterisation/evaluation of the collection. From the next 2 years users can consult the availability of materials (through the CIS or by contacting the curator) in order to request accessions. Before the user receives the materials a Material Transfer Agreement between donor and recipient must be signed.

Germplasm characterisation

Descriptor definition

A descriptor list for whole characterisation and evaluation of the genus has been defined and improved during the last 3 years (unpublished data). It includes characterization descriptors (95 traits), evaluation descriptors (47 traits) and descriptors based on genetic markers technologies and cytological characters (14 traits), and embraces a diverse set of data (morphological, phenological, agronomical, phytochemical, molecular, etc.), with the aim of being a useful tool for the description of the genetic variation in the genus *Crocus.*

Characterisation/evaluation

Germplasm characterisation is an important operation for a genebank since the value of the germplasm collection depends on the availability and quality of the information relative to the preserved accessions. Therefore, one of the main goals of *CROCUS*BANK action is to strategically characterize and evaluate germplasm of saffron and allies at different levels. A partial characterisation/evaluation of the collection have been developed during last years taking into account morphological, phenological, agronomical, resistance to salt stress, phytochemical, and molecular characters.

Sixty-six (66) saffron accessions have been characterised/evaluated for morphological, phenological and agronomical traits, and the existence of variability has been observed, suggesting the existence of genetic differences among the accessions related to the geographic origin of the materials (unpublished data). These preliminary results are being confirmed by the data obtained in other approaches. A different sensitiveness to saline stress has been recorded among some of the above mentioned saffron accessions in relation to genotypes origin (unpublished data). The phytochemical analysis using gas chromatography (GC-MS) and/or spectroscopic methods (FT-IR, Raman) also indicates that potential variability occurs among saffron accessions (unpublished data). In the same way genomic AFLP and SNPs markers have been identified in a subset of accessions, showing clearly the existence of genetic variation in saffron crop (unpublished data). In addition, these genetic markers provide a specific genetic fingerprint that could be very useful for the rationalisation of the bank.

The significant genetic variability found in saffron, evidenced with the on-going characterisation/evaluation studies, opens the door to unravel the

Table 8 Priority actions and future prospects in the WSCC

Action category	Future prospect
Collection	High priority should be given on obtaining some *Crocus* taxa not represented in the WSCC or missed in previous acquisitions
Preservation	Efforts need to be focused on management of *ex situ* conservation and multiplication of crops, wild relatives and local varieties (e.g. in vitro conservation)
Characterisation	An extensive characterisation of the WSCC materials including agro-morphological, phenological, chemical, molecular and cytogenetical descriptors will allow performing analyses regarding the variability among and within populations
Evaluation and genetic enhancement	A coordinated and integrated effort to evaluate the WSCC is needed to identify useful genotypes (e.g. sources of biotic and abiotic tolerance, genotypes with optimal production of secondary metabolites, genotypes interesting for improving saffron yield, etc.)
Rationalization of the collection	The reduction of the collection size, without loss in the genetic variability is a very desirable approach in order to preserve the collection and to promote its utilisation (e.g. creation of a core collection)
Utilisation and dissemination	A new more dynamic portal exclusively for the WSCC utilisation and disseminations is being designed. It's based on Joomla CMS (www.joomla.org) and MySQL database with allows large numbers of people to contribute and share stored data and to improve communication between users through blogs, forums, etc.

peculiarities of "land varieties" of this minor but highly appreciated Mediterranean crop. Accordingly, these results scientifically support the importance of conserving the local and precious cultivated germplasm worldwide.

Similar studies have been programmed or are being developed indeed for other *Crocus* species integrated in the WSCC, however, the shortage of materials in most accessions is by the moment a limiting factor to develop more extensive studies. Anyway, preliminary trials considering different kind of traits (mainly morphological, phenological and molecular, but also salt stress resistance, and phytochemical in a lesser extent), have revealed both, interspecific and intraspecific variability, in 34 accessions belonging to 21 species (unpublished data). That information may be of interest for different purposes (commercial gardening, bank rationalisation, taxonomic or evolution studies, etc.), although much work remains to be done in the future with these materials.

Future actions and prospects on the WSCC

The WSCC has already a wide representation of the *Crocus* germplasm of plausible utility in saffron breeding which has never been achieved before. Additionally, for the first time worldwide it has been created a unique collection which contains a large part of the variability of the saffron crop and wild relatives at global scale for common use. Therefore, priority actions to make useful the genetic resources to potential users are needed as outlined in Table 8.

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