



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



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# Crop wild relative

Issue 8 April 2012

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Conserving plant genetic resources

for use now and in the future



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### Editors:

Shelagh Kell  
Nigel Maxted

### Assistant editor:

Hannah Fielder

### Design and layout:

Shelagh Kell  
Hannah Fielder

### Front cover:

*Brassica villosa* subsp. *bivoniana* pictured with clip cage containing whiteflies in field trials for plant host resistance, Wageningen

### Photo:

Koen Pelgrom, Wageningen UR Plant Breeding, The Netherlands



### Correspondence address:

Shelagh Kell, School of Biosciences, University of Birmingham, Edgbaston, Birmingham, B15 2TT, UK  
Email: [s.kell@bham.ac.uk](mailto:s.kell@bham.ac.uk)

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We are pleased to welcome you to Issue 8 of *Crop wild relative*—the first issue to be published in the context of PGR Secure, a Collaborative Project funded under the EU Seventh Framework Programme. The PGR Secure project (full title: *Novel characterization of crop wild relative and landrace resources as a basis for improved crop breeding*) focuses on ensuring that the genetic diversity inherent in crop wild relative (CWR) and landrace (LR) populations and which is important for crop improvement—particularly to adapt crops to grow in our changing climate—is conserved and available for use by plant breeders. The emphasis lies with the end user; how can useful traits for crop improvement be identified in CWR and LR? How can material containing desirable traits be made available to the end user? How can we conserve CWR and LR both *in situ* and *ex situ* to ensure that material is maintained in the long term and is accessible for use in plant breeding programmes? It is fundamental that the link be made between CWR and LR conservation and use to underpin global food security—a key element of this project is that it bridges the ‘gap’ between the conservation and plant breeding communities.

Since the last issue of *Crop wild relative* was published, there have been a number of major steps forward in the conservation of European CWR. In 2009, work began on the assessment of nearly 600 priority European CWR species as part of the joint EC/IUCN-funded initiative to produce the first European Red List (<http://ec.europa.eu/environment/nature/conservation/species/redlist/>). The *European Red List of Vascular Plants* ([http://ec.europa.eu/environment/nature/conservation/species/redlist/downloads/European\\_vascular\\_plants.pdf](http://ec.europa.eu/environment/nature/conservation/species/redlist/downloads/European_vascular_plants.pdf)) was published in November 2011 and identified that at least 16 % of the CWR species assessed at European level for which sufficient data were available are threatened or are likely to become threatened in the near future and that many more are threatened at national level (see page 43). The inclusion of a significant number of CWR species in the European Red List, as well as the publication of assessments of 188 species endemic to Europe in the IUCN Red List of Threatened Species ([www.iucnredlist.org/](http://www.iucnredlist.org/)) is a major step forward in CWR conservation and will we hope go a long way towards raising the profile of CWR amongst the nature conservation community.

In September 2010, the joint AEGRO/ECPGR symposium *Towards the establishment of genetic reserves for crop wild relatives and landraces in Europe* ([www3.uma.pt/cem/aegro\\_ec\\_pgr\\_symp/index.html](http://www3.uma.pt/cem/aegro_ec_pgr_symp/index.html)) was held at the University of Madeira in Funchal, during which the results of the EC-funded AEGRO project (<http://aegro.jki.bund.de/aegro/>) were disseminated and invited speakers presented their work in the field of CWR and LR conservation. The symposium provided an opportunity to

# Editorial

convene the first meeting of the ECPGR *In Situ* and On-farm Conservation Network and to carry out a ‘horizon scanning’ exercise which set both short and long term priorities for

CWR and LR conservation in Europe (see page 8). The proceedings of the symposium, including the results of the horizon scanning exercise and additional contributions were published by CAB International this year under the title, *Agrobiodiversity Conservation: Securing the Diversity of Crop Wild Relatives and Landraces* (see page 43).

The PGR Secure project started on March 1st 2011 and significant progress has been made in the first year. You can gain an insight into the overall objectives and work programme on page 4 and in subsequent articles read more detailed information about the activities of the project, including: the identification of plant breeders’ needs and barriers to the use of CWR and LR in plant breeding (page 10); characterization of CWR and LR using phenomics and genomics approaches (pages 11 and 12); predictive characterization using the Focused Identification of Germplasm Strategy (FIGS) approach (page 14); development of CWR conservation strategies in Spain, Italy and the UK (pages 18, 22 and 24 respectively); and the development of an information system which will improve and facilitate access to conserved CWR and LR resources for use in crop improvement programmes (page 28).



Participants in the workshop, ‘Conservation strategies for European crop wild relative and landrace diversity’, Palanga, Lithuania, 7–9 September 2011 (Photo: Pavol Hauptvogel)

A major highlight of the first year was the staging of the joint PGR Secure/ECPGR workshop, *Conservation strategies for European crop wild relative and landrace diversity* ([www.pgrsecure.org/palanga\\_workshop](http://www.pgrsecure.org/palanga_workshop)) in Palanga, Lithuania, the goal of which was to discuss and agree a strategic approach to European and national CWR and LR conservation (see page 17). Ongoing support to assist national PGR programmes to develop national CWR and LR inventories and conservation strategies is being provided by the PGR Secure helpdesk ([www.pgrsecure.org/helpdesk](http://www.pgrsecure.org/helpdesk)).

*Crop wild relative* is not restricted to reporting research within the context of PGR Secure; we aim to incorporate news and research whether it be from within Europe or elsewhere. In this issue, research is presented from other projects, highlighting the necessity for both *in situ* (pages 31 and 33) and *ex situ* (page 34) conservation of CWR. The penultimate article gives an insight into the challenges facing CWR conservation in Papua New Guinea (page 39)—challenges which most certainly are not restricted to this country alone. The newsletter concludes with a case study of the Critically Endangered species *Lathyrus belinensis* (page 44).

We hope you find this issue informative and stimulating and we look forward to receiving your contributions for Issue 9 of *Crop wild relative* which is due to be published in spring 2013.

# PGR Secure: enhanced use of traits from crop wild relatives and landraces to help adapt crops to climate change

N. Maxted and S. Kell

School of Biosciences, University of Birmingham, Edgbaston, Birmingham B15 2TT, UK. Email: [s.kell@bham.ac.uk](mailto:s.kell@bham.ac.uk)

*Our food depends on the continued availability of novel sources of genes to breed new varieties of crops which will thrive in the rapidly evolving agri-environmental conditions we are now faced with as a result of climate change. Wild plant species closely related to crops (crop wild relatives) and traditional, locally adapted crop varieties (landraces) contain vital sources of such genes, yet these resources are themselves threatened by the effects of climate change, as well as by a range of other human-induced pressures and socio-economic changes. Further, while the value of crop wild relatives and landraces for food security is widely recognized, there is a lack of knowledge about the diversity that exists and precisely how that diversity may be used for crop improvement. PGR Secure aims to address these issues by: a) developing fast and economic methods to identify and make available genetic material that can be used by plant breeders to confer resistance to new strains of pests and diseases and tolerance to extreme environmental conditions such as drought, flooding and heat stress—the biotic and abiotic pressures which are rapidly evolving and having an increasingly detrimental effect on crop productivity; and b) developing a Europe-wide systematic strategy for the conservation of the highest priority crop wild relative and landrace resources to secure the genetic diversity needed for crop improvement.*

*Read on to learn more about the project.*

## **PGR Secure context: a call for a step change in agrobiodiversity conservation and use**

The EC Biodiversity Action Plan for Agriculture ([www.epbrs.org/PDF/EPBRS-IR2004-BAP%20Agriculture.pdf](http://www.epbrs.org/PDF/EPBRS-IR2004-BAP%20Agriculture.pdf)) highlighted the need for a step change in crop cultivar production in Europe because of rapidly changing consumer demands and the need to ensure food security across the continent; particularly in the light of the impacts of climate change. If these requirements are to be met, plant breeders need a broader pool of diversity to supply the necessary range of mitigating traits, as well as greater efficiency in characterization and evaluation techniques to locate the desired traits. The Action Plan also argued that maintaining the *status quo* for agrobiodiversity conservation and use is no longer tenable and that a step change in systematic conservation and use is required. The two major components of agrobiodiversity that offer the broadest range of diversity for breeders are crop wild relatives (CWR) and lan-



*Medicago falcata* (Photo: Pavol Eliás)

draces (LR), but there is currently a gap between their conservation and use and they remain under-exploited by the user community. In order to meet the needs of future generations, there are five key areas that need to be addressed:

1. *Climate change mitigation* – The adverse impacts of climate change (such as extreme weather events) on patterns of crop diversity and local cultivar adaptation are predicted to have a negative impact on crop yields. Breeders will be increasingly required to take adaptive action—breeding for example novel drought, pest and disease resistant cultivars—which will require extensive screening of genetic resources and use of adaptive traits in breeding resistant cultivars.
2. *Limited success of traditional characterization to meet breeders' needs* – Traditional phenotypic characterization and evaluation using field trials is resource intensive—thus,



Sugar beet, *Beta vulgaris* (Photo: Strube Research GmbH & Co KG)

the vast majority of conserved CWR and LR accessions remain uncharacterized and as a direct result largely unutilized. Novel approaches to characterization and evaluation beyond those previously applied are required to extend the use of CWR and LR diversity.

3. *Lack of systematic CWR and LR conservation* – Within European *ex situ* germplasm collections, only a very small percentage of germplasm holdings are CWR species and these are not a representative sample of the genetic diversity found in European wild populations. Although the numbers of gene bank holdings of LR are undoubtedly greater, without an inventory or conservation strategy it is unknown if these holdings truly reflect the diversity still maintained by farmers today in Europe. There is also currently no active *in situ* conservation of CWR in Europe as these species tend to fall between the priorities of the plant genetic resources for food and agriculture (PGRFA) conservation and nature conservation communities. Better systematic CWR and LR conservation and promoting their availability means that greater adaptive diversity could be made available to breeders.
4. *Threats facing CWR and LR diversity* – Worldwide, biodiversity is under severe threat from a range of deleterious factors (e.g., habitat destruction, degradation and fragmentation, over-exploitation, invasive alien species and changes in land management), but in the medium to long term climate change is predicted to be a degree of magnitude more catastrophic in terms of loss of species and genetic diversity. Recent research shows that at least 16 % of the high-



Peter Brinch, maintainer of landrace beetroot 'Cheltenham Green Top' (Photo: Pupak Haghighi-Brinch)



*Avena sterilis* subsp. *ludoviciana* (Photo: Richard Lia)

est priority CWR species in Europe are threatened (Critically Endangered, Endangered or Vulnerable) or Near Threatened ([http://ec.europa.eu/environment/nature/conservation/species/redlist/downloads/European\\_vascular\\_plants.pdf](http://ec.europa.eu/environment/nature/conservation/species/redlist/downloads/European_vascular_plants.pdf)); however, the threat to genetic diversity is even greater, meaning that the pool of locally adapted diversity required by breeders is decreasing. Landraces are under threat from agricultural intensification, market failure and socio-economic change. It is more difficult to quantify the loss of LR diversity because we do not yet have a comprehensive inventory of the diversity that exists; however, it is likely that LR are an even more threatened resource than CWR.

5. *Lack of plant genetic resource informatics cohesion* – In recent years there has been significant informatics development within the European PGRFA community. The *European Cooperative Programme for Plant Genetic Resources* (ECPGR – [www.ecpgr.cgiar.org](http://www.ecpgr.cgiar.org)) crop networks developed the *European Central Crop Databases* (ECCDB – [www.ecpgr.cgiar.org/germplasm\\_databases.html](http://www.ecpgr.cgiar.org/germplasm_databases.html)) that contain accession passport, characterization and evaluation data for major crop collections; the ECPGR *Documentation and Information Network* through the FP5 consortium EPGRIS developed the EURISCO web catalogue of European gene bank holdings (<http://eurisco.ecpgr.org/>); and the ECPGR *In Situ and On-farm Conservation Network* ([www.ecpgr.cgiar.org/networks/in\\_situ\\_and\\_on\\_farm.html](http://www.ecpgr.cgiar.org/networks/in_situ_and_on_farm.html)) through the FP5 consortium PGR Forum ([www.pgrforum.org](http://www.pgrforum.org)) created the Crop Wild Relative Catalogue for Europe and the Mediterranean (accessible via the Crop Wild Relative Information System, CWRIS – [www.pgrforum.org/cwr/cwrinfo.asp](http://www.pgrforum.org/cwr/cwrinfo.asp)) containing a nomenclatural checklist and occurrence data for European CWR species. However, each system currently stands alone and there is a need to link these systems into one comprehensive information portal for European PGRFA. Furthermore, vast quantities of data on gene sequences are continually expanding in world databases and transcriptomic information is close behind. Effective CWR and LR diversity conservation and use requires advanced informatic techniques to join up all these information systems and the data they contain.

**Box 1 PGR Secure work packages****Work package 1** – Phenomics, genomics and transcriptomics

- Demonstrate how novel phenomics, genomics and transcriptomics tools can be used to speed up plant breeding
- Insect resistance in brassica crops as a case study

**Work package 2** – Informatics

- Produce a web-based Trait Information Portal (TIP) to provide access to CWR and LR trait data
- Predictive characterization (using FIGS) to identify populations of CWR and LR with adaptive traits for pest and disease resistance and tolerance to environmental conditions (*Avena*, *Beta*, *Brassica* and *Medicago* as case studies)

**Work package 3** – Crop wild relative conservation

- Produce national and Europe-wide CWR inventories
- National CWR conservation strategy case studies for the UK, Finland, Italy and Spain
- Develop a European CWR conservation strategy for priority crop gene pools
- Produce a generic European CWR conservation strategy combining the regional and national approaches

**Work package 4** – Landrace conservation

- Gain an understanding of the diversity of European LR and their present conservation status
- Develop a systematic European LR conservation strategy to promote their use by breeders and by local communities and farmers

**Work package 5** – Engaging the user community

- Promote the use of CWR and LR in Europe
- Consultation with stakeholders (gene bank managers, breeding companies, public research bodies, NGOs), SWOT analysis to identify constraints in CWR and LR use
- Promote the flow of pre-breeding material and information gained in the project to stakeholders

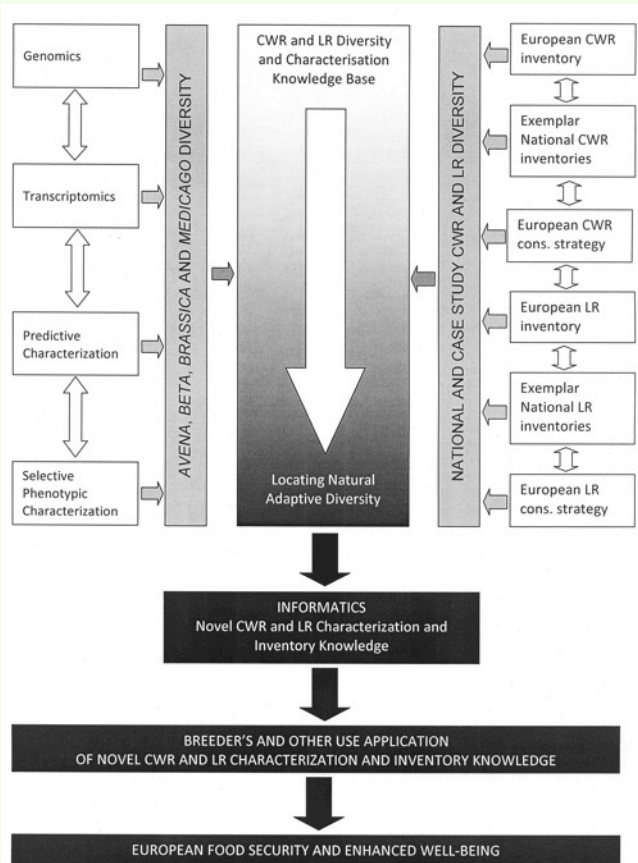
**Work package 6** – Dissemination and training

- Website, web-enabled inventories, TIP, publications, workshops, dissemination conference

**Work package 7** – Project management**PGR Secure: answering the call**

PGR Secure is a collaborative project funded under the EU's Framework 7 Programme and aims to address the issues outlined above by advancing CWR and LR diversity conservation and use. The goals of PGR Secure are to a) research novel characterization techniques for CWR and LR, b) develop conservation strategies for European CWR and LR diversity, and c) to enhance crop improvement by breeders as a means of underpinning European food security in the face of climate change. To achieve these goals PGR Secure has four research themes: 1) novel characterization techniques, 2) CWR and LR conservation, 3) improved use of CWR and LR by breeders, and 4) informatics (Fig. 1). Themes 1 and 3 address how to improve breeders' use of conserved CWR and LR diversity by applying novel characterization techniques such as genomics, transcriptomics, metabolomics, high-throughput phenotyping and GIS-based predictive characterization, as well as clarifying through dialogue exactly what breeders need to bridge the conservation–use gap and facilitating the flow of selected material and knowledge from the project to the breeder community. Theme 2 enhances CWR and LR species and genetic diversity conservation through development of CWR and LR inventories and systematic conservation strategies, while Theme 4 addresses the management and provision of access to CWR and LR trait and conservation information.

The project is implemented through seven work packages (WPs) (Box 1).



**Figure 1** Schematic diagram of interrelated PGR Secure project themes



Sugar beet, *Beta vulgaris* (Photo: Strube Research GmbH & Co KG)



Array machine and plate arrays (Photo: Sean May)

The expected impacts of these actions are:

- Enhanced techniques to identify useful adaptive traits and to accelerate plant breeding;
- Better access to and wider take-up of CWR and LR resources in plant breeding programmes;
- Increased capacity and options for crop improvement to support European farming;
- Improved conservation of European CWR and LR resources;
- Provision of a resource base and tools to back-stop food security in Europe;
- Enablement of coherent planning of plant breeding and agrobiodiversity conservation policy in Europe.

#### Who is involved?

The PGRFA user community in Europe is diverse; ranging from breeding companies, public research institutes, gene banks, non-governmental organizations (NGOs), universities and farmers; but it is use by plant breeders that has potentially the greatest economic and social benefit in Europe. FAO's Second Report on the State of the World's Plant Genetic Resources for Food and Agriculture ([www.fao.org/agriculture/crop/core-themes/theme/seeds-pgr/sow/sow2/en/](http://www.fao.org/agriculture/crop/core-themes/theme/seeds-pgr/sow/sow2/en/)) highlights that "Considerable opportunities exist for strengthening cooperation among those involved in the conservation and sustainable use of PGRFA, at all stages of the seed and food chain. Stronger links are needed, especially between plant breeders and those involved in the seed system, as well as between the public and private sectors". The PGR Secure project seeks to strengthen these links and involves collaboration between European policy, conservation and breeding sectors throughout Europe.

The project was initiated by and involves members of the *In Situ* and On-farm Conservation Network ([www.ecpgr.cgiar.org/networks/in\\_situ\\_and\\_on\\_farm.html](http://www.ecpgr.cgiar.org/networks/in_situ_and_on_farm.html)) of the European Cooperative Programme for Plant Genetic Resources (ECPGR) from throughout Europe. The Consortium itself comprises 11 partner institutes and includes both plant breeding and conservation research institutes, as well as Europe's primary plant breeding research network, the European Association for Research in Plant Breeding (EUCARPIA) (Box 2). The Consortium is supported by an External Advisory Board which in-

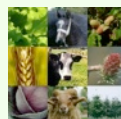
volves senior researchers in plant breeding and PGRFA conservation and policy, as well as a Breeders' Committee comprising plant breeders and pre-breeders of major European food crops.

Stakeholders in the project are Europe-wide and include: a) small and large plant breeding companies; b) scientists and policy-makers in public and private research institutes; c) farmers and others working in the agricultural sector; d) plant gene banks, protected areas and the broader conservation community; e) government agencies and non-governmental organizations involved in plant conservation, plant breeding and national or local nutrition and food supply issues; and f) the European Commission.

For further information, please visit the project website: [www.pgrsecure.org](http://www.pgrsecure.org) or contact the Project Manager, [s.kell@bham.ac.uk](mailto:s.kell@bham.ac.uk).

#### Box 2 The 11 partner institutes forming the PGR Secure Consortium

1. The University of Birmingham, UK (Coordinator)
2. Wageningen UR Plant Breeding and Centre for Genetic Resources, The Netherlands
3. Bioversity International
4. The University of Perugia, Italy
5. Julius Kühn-Institut, Federal Research Centre for Cultivated Plants, Germany
6. NordGen, Sweden
7. MTT Agrifood Research, Finland
8. The University of King Juan Carlos, Spain
9. ServiceXS BV, The Netherlands
10. The University of Nottingham, UK
11. European Association for Research on Plant Breeding, Hungary



# CWR horizon scanning: what are we doing and what should we be doing?

N. Maxted and S. Kell

School of Biosciences, University of Birmingham, Edgbaston, Birmingham B15 2TT, UK. Email: [s.kell@bham.ac.uk](mailto:s.kell@bham.ac.uk)

To define what future conservation actions are needed, we must first know what measures are currently in place. To this end, a horizon scanning exercise was undertaken involving delegates at the symposium 'Towards the establishment of genetic reserves for crop wild relatives and landraces in Europe' held in Funchal, Madeira, 13–16 September 2010. Contributors to this exercise included partners in the EC AGRI GENRES project 'An Integrated European *In Situ* Management Workplan: Implementing Genetic Reserve and On-farm Concepts' (AEGRO – <http://aegro.jki.bund.de/aegro/>), 53 members of the ECPGR *In Situ* and On-farm Conservation Network and others with an interest in crop wild relative (CWR) and landrace (LR) conservation (Fig. 1). Horizon scanning is a participatory approach to establishing future priorities and was in this case used to review the current status of European CWR and LR conservation and agree priorities for their conservation over the next 30 years. All conference delegates were invited to contribute suggestions for conservation actions and then each voted for those that they consider a priority.

The results of the survey of short-term CWR conservation priorities in 33 European countries indicate that nearly all countries have some *ex situ* conservation of CWR diversity and that most have some form of national CWR inventory and threat assessment. However, IUCN Red List assessments of CWR have only been undertaken as part of overall national threat assessment, so the assessments are coincidental and not specifically focused on CWR species. Most countries also reported some *ad hoc* use of CWR diversity in plant breeding and that there is some national public awareness of the value of CWR diversity. However, few countries have a prioritized list of national CWR species, have developed national CWR action plans, undertaken genetic gap analysis for even their most important CWR species, and it is rare to have specific mention of CWR conservation or protection in national conservation

legislation. Perhaps what is most surprising, given the increasing publicity afforded to *in situ* CWR conservation in the last 20 years, is the minimal progress in establishing working genetic reserves to conserve CWR diversity; although with the notable exceptions of Armenia, Azerbaijan, Israel, Turkey and Ukraine.

On the basis of the analysis of current conservation measures it was possible to identify gaps and the short-term actions needed. The five leading short-term priorities for CWR conservation to be achieved by 2020 are:

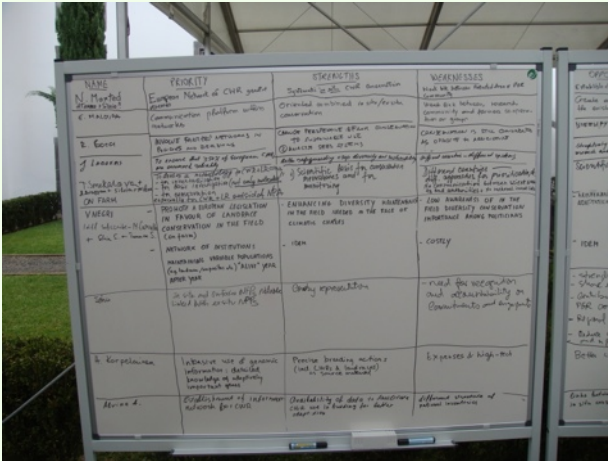
1. National genetic gap analysis of CWR species as a basis for the establishment of national CWR conservation action plans.
2. Systematic genetic reserve implementation to conserve *in situ* CWR diversity at both national and European scales. This should be accompanied by *ex situ* gap analysis and the establishment of backup collections of priority *in situ* populations.
3. Integration of *in situ* CWR conservation into on-farm initiatives, thus undertaking complementary CWR and landrace *in situ* conservation.
4. Incorporation of legislative protection for CWR species and genetic diversity at both national and European levels.
5. Enhancement of CWR utilization—as CWR use is likely to hold the key to sustainability of conservation, breeders' use of CWR diversity should be promoted.

The results of the horizon scanning exercise also identify long-term CWR conservation actions in European countries that require implementation between 2020 and 2030. Although already identified as a short-term priority, the extension of national and European genetic reserves to form a coherent European network that systematically conserves the highest priority CWR diversity—particularly that associated with crop Gene Pools 1b or 2, or Taxon Groups 1b, 2 or 3 (Maxted *et al.*,



**Figure 1** Delegates at the 'Towards the establishment of genetic reserves for crop wild relatives and landraces in Europe' symposium, Funchal, Madeira, 13–16 September 2010





**Figure 2** White boards used for horizon scanning exercise, Madeira symposium, September 2010

2006)—is the highest priority. Second, the establishment of closer links between the agrobiodiversity and biodiversity conservation communities to help raise awareness of the need for both CWR species and intra-specific diversity conservation; ideally in association with the stakeholders in the Natura 2000 Network ([www.natura.org/](http://www.natura.org/)). Third is the need for systematic threat assessment of priority CWR species using the IUCN Red List Criteria, and as an adjunct to this, the requirement to develop a set of criteria to assess threat to intra-specific genetic diversity as an ‘extension’ to the current IUCN Red List Criteria. The fourth priority is the need to rethink management and monitoring models for CWR conservation to take greater account of the role of local communities in genetic reserve conservation, in order to recognize the importance of partnerships between local communities and conservationists. The fifth priority is associated with the intrinsic value of CWR, which is to improve the identification and use of adaptive traits in crop improvement. Steps towards achieving this goal are: a) making sure countries have national CWR inventories, b) that the inventory is prioritized on the potential of CWR species to contribute economically valuable traits, and c) that as a CWR community we engage in a dialogue with breeders to understand their changing demands. Given that CWR conservation has a real cost and the fact that CWR diversity is currently neither systematically or effectively conserved *ex situ* or *in situ*, it can be argued that more efficient utilization (or the option value associated with more efficient utilization), is the primary justification for the resources required to conserve CWR diversity.

It is also worth noting that implementation of the five key short-term and long-term priorities will help ensure that  $\geq 70\%$  of European CWR are conserved reliably, which is directly linked to Target 9 of the Global Strategy for Plant Conservation (CBD, 2010) which is “70 per cent of the genetic diversity of crops and other socio-economically valuable plant species conserved, while respecting, preserving and maintaining associated indigenous and local knowledge”. More explicitly within the European context, the European Strategy for Plant Conservation (Planta Europa, 2008) calls for the establishment of 25 CWR genetic reserves in Europe, along with undertaking gap analysis of current *ex situ* CWR holdings, followed by the systematic filling of diversity gaps. Methodologies to achieve these goals are available, but the constraint to their implementa-

tion over the next 30 years is likely to be the availability of resources.

It is anticipated that the results of the horizon scanning exercise will be used by policy-makers to prioritize CWR conservation action in Europe and to help ensure the plant genetic resource action (PGR) research community engage in a more coordinated and strategic approach to future CWR research, while at the same time acting as a model for participatory decision-making in other areas of PGR conservation, research and priority-setting inside and outside of Europe. The exercise involved a range of stakeholders with responsibility for CWR conservation within 28 European countries and the European Union; therefore, the resultant issues identified and the priorities formulated are authoritative and have ‘buy-in’ from the European PGR community—they now need to be fully enacted!

“five key short-term and long-term priorities will help ensure that  $\geq 70\%$  of European CWR are conserved reliably”

A summary of the results of the horizon scanning exercise are available on the PGR Secure website ([www.pgrsecure.org/](http://www.pgrsecure.org/)) and it is intended that the Current Status Table will be updated periodically to provide a tool to compare progress across Europe. For the full analysis and results, see Maxted *et al.* (2012).

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# PGR Secure: Engaging the user community

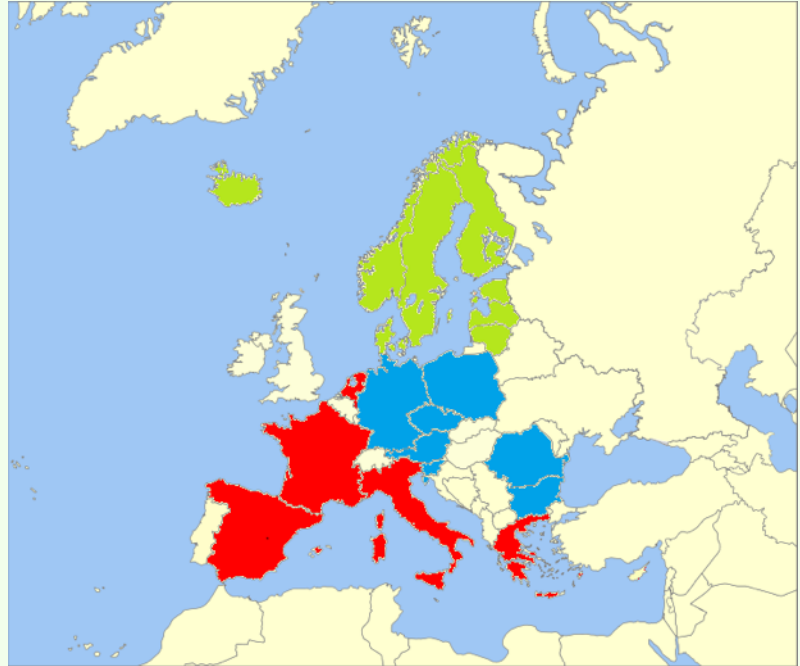
C. Kik<sup>1</sup>, G. Poulsen<sup>2</sup>, G. Neuhaus<sup>3</sup> and L. Frese<sup>3</sup>.

<sup>1</sup>Centre for Genetic Resources, the Netherlands (CGN), Droevendaalsesteeg 1, 6708 PB Wageningen, the Netherlands. Email: [chris.kik@wur.nl](mailto:chris.kik@wur.nl)

<sup>2</sup>Nordic Genetic Resource Center, Smedjevägen 3, SE-230 53 Alnarp, Sweden. Email: [gert.poulsen@nordgen.org](mailto:gert.poulsen@nordgen.org)

<sup>3</sup>Julius Kühn-Institut (JKI), Federal Research Centre for Cultivated Plants, Erwin-Baur-Str. 27, D-06484 Quedlinburg, Germany. Email: [lothar.frese@jki.bund.de](mailto:lothar.frese@jki.bund.de), [gisela.neuhaus@jki.bund.de](mailto:gisela.neuhaus@jki.bund.de)

The extent to which plant genetic resources (PGR)—in particular landraces and CWR—managed by collection holders in Europe are utilized for research and breeding or in crop production directly is not clear. However, the full use of the PGR available is important for sustainable European agriculture. In order to obtain more clarity on this issue in the context of the PGR Secure project, PGR networks in various European countries are being analysed to identify whether problems occur in the transfer of germplasm from collection holders to users and if they do occur, how they can be solved. The different types of stakeholders identified are government, genebanks, research institutes, commercial breeders and agro-NGOs. The approach that has been chosen is firstly to interview the different stakeholders in various European countries and secondly to send an online questionnaire to all stakeholders identified. On the basis of the outcome of both studies, a report will be written on the state of art of PGR utilization in Europe which will be used as an input for a European conference on this issue.



**Figure 1** European countries selected for interviewing. The countries in light green are covered by NordGen, countries in light blue by JKI, and countries in red by CGN.

Since the start of the PGR Secure project, a number of activities have taken place. Firstly, European countries were selected where interview activities would take place. As well as covering its own country, NordGen covers northern Europe,

JKI middle Europe and CGN southern Europe. The countries that were selected for interviewing are depicted in Figure 1.

Secondly questionnaires were developed for semi-structured interviews in the various countries. Thirdly, country key persons were identified. The country key persons prepared a list of stakeholders, selected one to three organizations per stakeholder group to be interviewed and arranged the logistics in their country. A typical interview lasted one to two hours (Fig. 2) and a country visit lasted one week. The interviews were usually transposed directly after the interview took place and were sent to be checked by the country key person and the person interviewed. On the basis of the interviews, a country report (including a SWOT analysis and actions required) was written together with the country key person.



**Figure 2** Interviewing in Greece; from left to right: D. Bladenopoulos, G. Eugenides, (Cereal Institute, Thessaloniki), A. Katsiotis (Agricultural University of Athens; country key person) and C. Kik (CGN) (Photo: Chris Kik, CGN, Wageningen, the Netherlands)

The next step in our research will be to develop an online questionnaire which will be sent to all stakeholders in the countries that were previously selected during 2012. The idea behind the online questionnaire is that it will give quantitative data on the functioning of PGR networks in the various countries selected, and in this way will substantiate the observations gained during the interviews which were earlier held.

# A phenomics and genomics approach to the use of landraces and crop wild relatives for crop improvement

B. Vosman

Wageningen UR Plant Breeding, P.O. Box, 16 6700AA Wageningen. Email: [ben.vosman@wur.nl](mailto:ben.vosman@wur.nl)

At present it is not possible to feed the world population without the application of insecticides. Worldwide yield losses caused by insects would be at least 30–50 % when no insecticides are used. However, the use of pesticides is hazardous to the environment and usually not very durable as insects may develop resistance against pesticides very rapidly. Additionally, on the 13th January 2009 the European Parliament proposed legislation placing controls on crop spraying and banning 22 pesticide chemicals. Therefore, crop production is in need of alternative control measures of which the use of resistant varieties is the most promising. To be able to develop such varieties one first needs to identify resistant sources. Often such resistances are found in crop wild relatives (CWR) and landraces (LR). Once the accessions containing the resistance traits are known, the chromosomal region or preferably the genes involved need to be identified to facilitate transfer to the crop species using molecular markers. Novel phenomics, genomics and transcriptomics technologies can speed up the identification of such markers (Broekgaarden *et al.*, 2011).



**Figure 1** Kale plant heavily infested with cabbage whitefly (Source: Greet Steenhuis of Plant Research International)

In the PGR Secure project we focus on the identification of resistance factors against the cabbage aphid (*Brevicoryne brassicae*) and the cabbage whitefly (*Aleyrodes proletella*), which are both specialist insects that feed only on members of the Cruciferae family, to which *Brassica oleracea* varieties and their wild relatives belong. These two insect species are phloem feeding and can cause serious problems in cultivation of *B. oleracea* crops in Europe. In particular, Brussels sprouts, kale and Savoy cabbage can be heavily infested by these herbivorous insects (Fig. 1). Aphid feeding causes chlorosis and leaf curling, whereas whitefly females lay eggs in circular patterns that are visible as white patches. Besides this cosmetic damage, both insects excrete a sugary substance (honeydew) that allows the growth of sooty mould. Both types of damage seriously reduce the marketability of the crop.

Plants can defend themselves against herbivores through physical and chemical barriers that can be constitutively present (i.e. present regardless of attack and forming a first line of defence) or induced upon herbivore attack (Schoonhoven *et al.*, 2005; Alvarez *et al.*, 2006). Plant morphological features, such as a wax layer or leaf toughness, form a first line of defence by preventing herbivores from settling or feeding on a plant. In addition, plants can deter herbivores through the production of repellent volatile secondary metabolites and defensive compounds or the production of proteins that directly affect herbivore performance. Glucosinolates, a group of second-

dary metabolites that are almost exclusively found in *Brassica* species, are well studied defensive compounds (Hopkins *et al.*, 2009). When plant cells are disrupted, glucosinolates are hydrolyzed by the enzyme myrosinase resulting in the formation of a variety of toxic compounds such as isothiocyanates (Halkier and Gershenzon, 2006). However, most specialist insects have evolved enzymes to detoxify glucosinolates and/or their breakdown products (Ratzka *et al.*, 2002). Therefore, plant resistance towards specialist herbivores is probably based on defensive compounds or proteins other than glucosinolates.

Several proteins/compounds and the genes encoding them have been shown to play an important role in plant resistance towards herbivores. To elucidate the resistance mechanisms present, the electrical penetration graph (EPG) technology can be used, which allows a close analysis of the detailed mechanisms of resistance to sap-feeding pests (Alvarez *et al.*, 2006). Such information complements the analysis of secondary metabolites and in combination with the gene expression data (Couldridge *et al.*, 2007) allows informed hypotheses of gene function to be generated.

## General approach in PGR Secure

Phenotyping is time and space consuming, which is a big problem when large collections have to be evaluated for a particular trait. This is especially true for the evaluation of plant material for insect resistance. Starting from a collection of about 3700 *Brassica* accessions in BrasEDB, a selection of around 400 has been made for the phenotyping (Pelgrom *et al.*, this issue). From these, some 125 accessions will be selected for further analysis using metabolomics and a further subset of these will be assessed in terms of resistance/susceptibility using the EPG. This will determine the underlying mechanisms of resistance by measuring insect feeding behaviour, as we have already done for rice (Bahagia *et al.*, 2009a). Based on the resistant and susceptible subsets that are identified, next generation sequencing technologies will be used to access the total gene transcriptome content of around 15 accessions of *Brassica* CWR and LR which will allow the identification of novel genes (and allelic variation) in this plant material—again as already shown in rice (Bahagia *et al.*, 2009b). Transcriptomics will be carried out using Affymetrix *Arabidopsis* genechips to assess gene expression variation across different populations of the accessions to determine within species variation and response to insect attack. In a second line we will develop segregating populations derived from crosses between plant material that is resistant and susceptible towards cabbage aphid and the cabbage whitefly. These populations will be used for quantitative trait loci (QTL) analy-

sis. A much smaller study on *Medicago sativa* populations/accessions as proof of applicability of the technologies to CWR other than *Brassica* species will be carried out.

#### Partners in the project and contribution:

- DLO, Wageningen, the Netherlands: resistance screens, metabolomics, QTL analysis
- University of Birmingham, UK: resistance screens, EPG, gene expression
- University of Nottingham, UK: gene expression
- ServiceXS, Leiden, the Netherlands: sequencing, genotyping

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## Looking for resistance to phloem feeders in *Brassica oleracea*

K. Pelgrom<sup>1</sup>, G. Sharma<sup>2</sup>, C. Broekgaarden<sup>1</sup>, R. Voorrips<sup>1</sup>, N. Bas<sup>3</sup>, J. Pritchard<sup>2</sup>, B. Ford-Lloyd<sup>2</sup> and B. Vosman<sup>1</sup>

<sup>1</sup> Wageningen UR Plant Breeding, Wageningen University & Research Centre, PO Box 386, 6700 AJ Wageningen, The Netherlands. Email: [koen.pelgrom@wur.nl](mailto:koen.pelgrom@wur.nl)

<sup>2</sup> School of Biosciences, University of Birmingham, Birmingham B15 2TT, UK. Email: [j.pritchard@bham.ac.uk](mailto:j.pritchard@bham.ac.uk)

<sup>3</sup> Centre for Genetic Resources, The Netherlands (CGN), Wageningen, The Netherlands. Email: [noortje.bas@wur.nl](mailto:noortje.bas@wur.nl)

There are several ways to protect crops against insects. One of them is to look for natural resistance in crop wild relatives (CWR) and landraces (LR). Because it is not possible to test all CWR and LR that are known, we made use of the core collection that was established by Boukema *et al.* (1997). This collection contains representatives of each *Brassica* crop type and from different ecogeographical origins. In total, 434 accessions (105 CWR and 329 LR) were screened for resistance against the cabbage aphid (*Brevicoryne brassicae*) and the cabbage whitefly (*Aleyrodes proletella*); both phloem feeding insects (Table 1). The field experiment of 2011 was conducted at two different locations: one in Wageningen, the Netherlands (Fig. 1) and the other in Stratton Audley near Bicester (Fig. 2), run by Oxford Agricultural Trials, in the United Kingdom. At both locations five week old plants were transplanted in the field. Plant growth and natural infestation (a choice test) of cabbage aphids and whiteflies were monitored.



**Figure 1** Cabbage field near Wageningen. Plants were planted in a complete randomized block order with ten replications.

“105 CWR and 329 LR were screened for resistance against the cabbage aphid (*Brevicoryne brassicae*) and the cabbage whitefly (*Aleyrodes proletella*)”

#### Wageningen field experiment, the Netherlands

In Wageningen, the complete set of 434 accessions was planted. In this field experiment the focus was on cabbage whitefly, of which the natural infection was very low, probably due to the wet and cold summer of 2011 in the Netherlands. It was not possible to distinguish susceptible from resistant accessions; therefore, no-choice tests with clip cages containing five female whiteflies were performed on the field grown cab-



**Figure 2** Cabbage field in Stratton Audley near Bicester. Plots were scored for the presence of whitefly and aphids.

bage plants using whiteflies obtained from a rearing at WUR Plant breeding (Broekgaarden *et al.*, 2012).

Two preliminary tests were conducted on all 434 accessions to eliminate a large part of the most susceptible accessions. After seven days the clip cages were removed and the number of whitefly (alive/dead) and the number of eggs were counted. We used two selection criteria to select our candidate resistant lines. All accessions with zero adult survival and/or accessions with an oviposition rate (eggs/female/day) lower than one (normal range between one and four) were selected. This brought the total number of candidates down to 100. A third test with four biological replicates was carried out on the 100 selected accessions and ten controls. For several reasons, such as uneven leaf surfaces that allowed whiteflies to escape from the clip cages, it was not possible to obtain a complete dataset for all accessions tested. Therefore, the analysis had to be restricted to 77 accessions of eight different species (21 CWR, 49 LR and 7 controls) (Table 1).

The results showed that resistance to cabbage whitefly can be found among *Brassica oleracea* subsp. *capitata* landraces. This is similar to the resistance that was found in a commercial white cabbage F<sub>1</sub> hybrid variety by Broekgaarden *et al.* (2012). The highest level of resistance in the CWR accessions was found in *B. villosa*. All eight *B. villosa* accessions tested in the field experiment passed the preliminary selection and six yielded sufficient data for the statistical analysis. Four *B. villosa* accessions are represented in the most significant group. This confirms the resistance earlier found by Ramsey *et al.* (1996) in *B. villosa*. In the field experiment one *B. incana* accession was found with almost complete resistance against the cabbage whitefly when considering survival and oviposition rate. Conversely, susceptible *B. incana* accessions were also found. This opens the possibility for studying the genetics of whitefly resistance using an intraspecific *B. incana* cross population.

### Bicester field experiment, United Kingdom

In contrast to the field experiment in Wageningen, where the complete set of 434 *Brassica* accessions were planted, the field experiment in Bicester screened a subset of 200 that were then selected for study—including all 105 CWR accessions and a random selection of LR (Table 1). Accessions were planted in a 24 x 40 m grid. There were three replicate blocks each with the 200 accessions selected from the larger Netherlands panel of accessions. Each of the selected accessions was planted randomly in a grid of 200 cells, with four seeds drilled in each cell. All three plots were surrounded by a set of guard plants. The staff at Oxford Agricultural monitored the plants over the summer, but they were unable to report any infestation of aphids or whitefly until 23rd August. Staff advised that scoring should commence three to four weeks after this date when both aphid and whitefly infestation had increased sufficiently.

Each of the three plots was scored for the presence of whitefly and aphids by staff of the University of Birmingham on September 29th. Aphids were scored on a scale from zero to four with zero being complete absence and four being heavy infestation (Fig. 3). Since infestation had occurred late in the season, a lot of plants were large, requiring much searching to check for the one potential aphid. In addition, the long growth period led to a difference in developmental state between accessions—some were in the late stages of flowering while others remained still fully vegetative. Large differences in phenotype were seen, such as those with very hairy leaves and others less hirsute, but also remaining uninfested by aphids, making us hopeful that the basis of their resistance may be revealed by electrical penetration graph (EPG).

**Table 1** *Brassica* species—CWR and LR—that were screened for resistance against cabbage aphids and whitefly

Species	No. of accessions tested for whitefly resistance	No. of accessions tested for aphid resistance
<i>B. bourgeau</i>	2	2
<i>B. balearica</i>	0	2
<i>B. cretica</i>	10	12
<i>B. fruticulosa</i>	15	16
<i>B. incana</i>	10	10
<i>B. insularis</i>	1	2
<i>B. macrocarpa</i>	2	3
<i>B. maurorum</i>	2	2
<i>B. montana</i>	7	7
<i>B. oleracea</i>	370	130
<i>B. rapa</i>	3	0
<i>B. rupestris</i>	3	3
<i>B. bioniana</i>	0	1
<i>B. rubertina</i>	0	1
<i>B. spinescens</i>	1	1
<i>B. villosa</i>	8	8
<b>Totals</b>	<b>434</b>	<b>200</b>

Comparison of the data on the three replicate plots revealed 17 resistant accessions (nine CWR and eight LR), which showed no aphid infestation in any of the three sub-plots. These resistant plants were mainly *Brassica oleracea*, with two *B. incana* and one *B. villosa*. Fourteen susceptible accessions were identified, with the *B. oleracea* dominance broken by a

single *B. cretica* and a single *B. villosa*, but split equally between seven CWR and seven LR. With the addition of seven accessions identified in the Netherlands this currently leaves us with 38 candidates. We are also currently analysing the data for any correlation between aphid and whitefly resistance and although this is not yet complete, initial impressions suggest that there is no clear correlation, which may indicate that different mechanisms operate against the two pests.



**Figure 3** Susceptible *Brassica* leaf with heavy infestation of cabbage aphid

#### Further research

Currently in consultation, the University of Birmingham and Wageningen UR Plant Breeding are confirming their choice of genotypes through no-choice greenhouse experiments and are securing seed stocks. The no-choice greenhouse experiments are being conducted to confirm the resistance to cabbage aphid and cabbage whitefly that was found in the field experiments of 2011.

The selected resistant material will be the starting point for a segregating population that can be used in further research by the Netherlands group. Once resistant genotypes are confirmed, the University of Birmingham will begin a rolling program of EPG screening of this interesting material which will complement the subsequent transcriptomic analysis.

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## Applying FIGS to crop wild relatives and landraces in Europe

I. Thormann

Biodiversity International, Via dei Tre Denari 472/a, 00057 Maccarese, Rome, Italy. Email: [i.thormann@cgiar.org](mailto:i.thormann@cgiar.org)

The Focused Identification of Germplasm Strategy (FIGS) (Mackay and Street, 2004) emerged as an approach to target accessions more likely to possess specific genetic variation sought by breeders. FIGS was a collaborative development involving researchers from the International Centre for Agricultural Research in the Dry Areas (ICARDA), the Australian Winter Cereals Collection and the Russian N.I. Vavilov Research Institute of Plant Industry. It involves gathering available information and knowledge to facilitate the identification of candidate accessions. Geographic information system (GIS) tools and statistical and modelling techniques can then be used to select the candidate accessions for evaluation, based on an understanding of relationships between traits and the environment.

The rationale behind FIGS is that plants survive in harsh environments because they develop adaptive traits that allow them to cope with environmental stresses, and that the environment influences the geographic distribution of plants. If researchers are interested in genotypes with a specific adaptive trait, they should first find environments where that particular trait is advantageous. Accession level information can then be matched to climate and environmental information to identify accessions that originate in those environments associated with specific adaptive traits. This FIGS approach has so far been successfully applied to barley (Endresen, 2010; Endresen *et al.*, 2011)

and wheat collections; for example to identify subsets of accessions with potential resistance to stem rust (Bari *et al.*, 2012), Russian wheat aphid (El Bouhssini *et al.*, 2011), or Sunn pest (El Bouhssini *et al.*, 2009).

“plants survive in harsh environments because they develop adaptive traits that allow them to cope with environmental stresses”

The PGR Secure project represents the framework for making one of the first attempts to apply FIGS to other crops as well as to wild species. The project is testing the FIGS approach on landraces (LR) and crop wild relatives (CWR) from the project's four target genera, *Avena* L. (oats), *Beta* L. (beet), *Brassica* L. (cabbages and mustard), and *Medicago* L. (Medics including alfalfa and lucerne), to identify potential accessions and *in situ* populations that might harbour abiotic and biotic resistance traits of interest to breeders as well as conservationists.

A predictive computer modelling method is used to identify material with potential abiotic or biotic resistance traits. This method is based on and demands *a priori* known trait evalua-

tion data in addition to climatic and environmental data. The specific biotic resistance trait of interest to the PGR Secure project is resistance to sap feeding insects. Abiotic stress resistance traits that are of interest in the four target genera are aluminium toxicity in *Avena*, drought tolerance in *Beta*, salinity and drought tolerance in *Brassica*, and frost, drought, and aluminium toxicity tolerance in *Medicago*. The FIGS study addressing these abiotic traits is based on collecting information on the environmental conditions most likely to support the adaptive development of these target traits; we call it the 'ecogeographic method'.

### The basis for the FIGS study – our data

An essential first step in FIGS studies is to compile all necessary datasets, such as passport data, ecogeographic data and evaluation data. The main sources from which data were collated are the European Catalogue of *ex situ* collections, EURISCO (<http://eurisco.ecpgr.org/>), the European Central Crop Databases (ECCDBs) ([www.ecpgr.cgiar.org/germplasm\\_databases.html](http://www.ecpgr.cgiar.org/germplasm_databases.html)), the Global Biodiversity Information Facility (GBIF) portal ([www.gbif.org/](http://www.gbif.org/)), the System-wide Information Network for Genetic Resources (SINGER) of the Consultative Group on International Agricultural Research (CGIAR) (<http://singer.cgiar.org/>) and the United States Department of Agriculture's (USDA) Germplasm Resources Information Network (GRIN) ([www.ars-grin.gov/npgs/searchgrin.html](http://www.ars-grin.gov/npgs/searchgrin.html)).

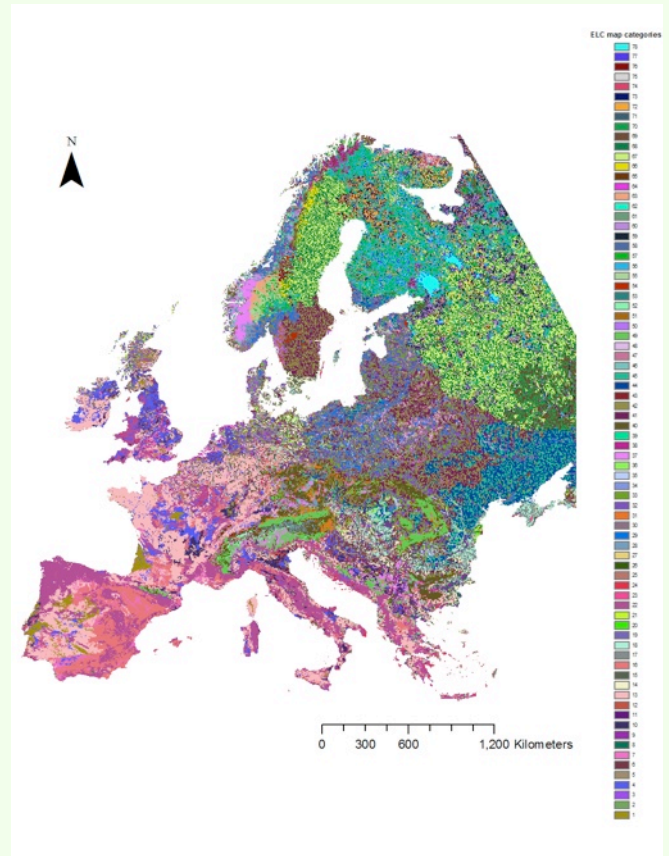
The application of FIGS requires the availability of passport data that include geographical coordinates of the collecting sites or at least location information sufficiently detailed to allow georeferencing. We collected presence data records for occurrences of all four genera within Europe. The quality of existing geographic coordinates was assessed and records with missing coordinates were georeferenced using GeoLocate ([www.museum.tulane.edu/geolocate/](http://www.museum.tulane.edu/geolocate/)). The resulting dataset contained approximately 33,000 presence points. A quality threshold was applied to exclude records with very low quality georeferences. Approximately 18,000 of the occurrences were considered to have an acceptable georeferenced coordinate quality.

The following ecogeographic data were extracted which yielded 108 ecogeographical variables for the presence points:

- Geophysical data from Digital Elevation Model (Rabus *et al.*, 2003)
- Soil data from harmonized World database ([www.iiasa.ac.at/Research/LUC/External-World-soil-database/HTML/](http://www.iiasa.ac.at/Research/LUC/External-World-soil-database/HTML/))
- Climate data from Worldclim ([www.worldclim.org/](http://www.worldclim.org/))

In order to create baseline datasets for the application of the FIGS methodology, the occurrence data were joined with the ecogeographic data. The resulting number of records for each of the four genera, separated into LR and CWR, were as follows: *Avena* LR (3855 records), *Avena* CWR (3900 records), *Beta* LR (1614 records), *Beta* CWR (1596 records), *Brassica* LR (3606 records), *Brassica* CWR (886 records), *Medicago* LR (149 records) and *Medicago* CWR (2153 records).

The ecogeographic method demands the production of ecogeographic land characterization (ELC) maps, which delineate areas with similar environmental characteristics (Fig. 1). These were created for each genus based on the variables that most



**Figure 1** An ecogeographic land characterization (ELC) map for *Avena sativa*

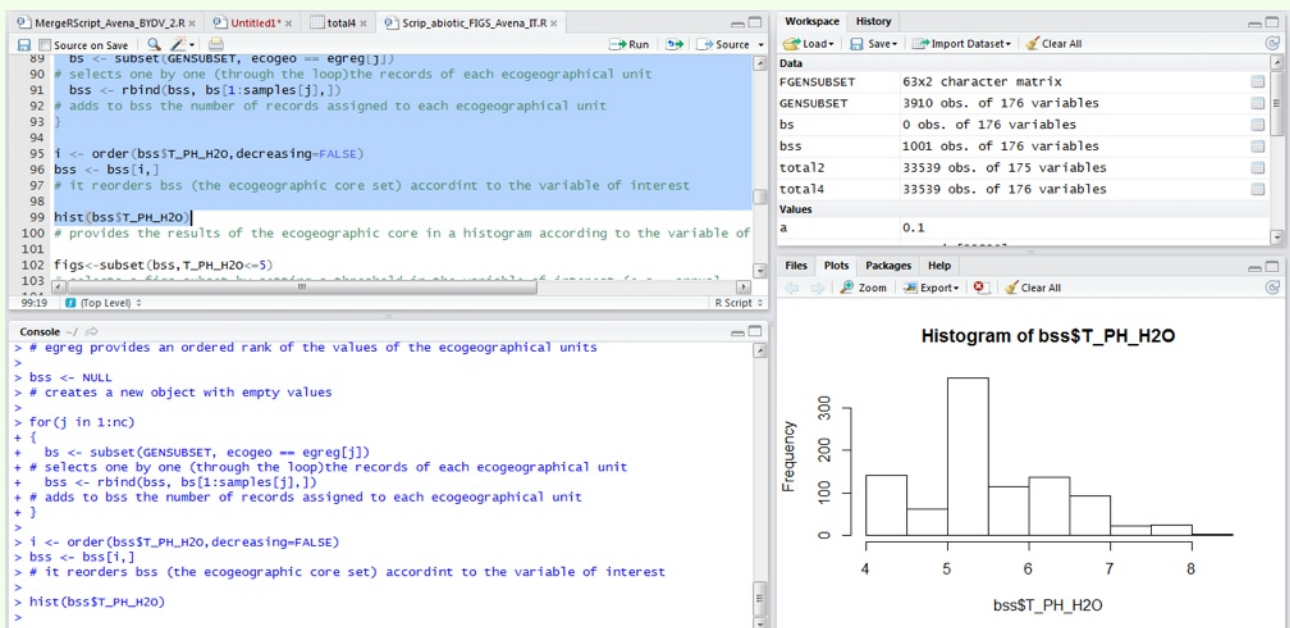
influence the distribution of the genus across Europe (Parra-Quijano *et al.*, 2012).

Trait evaluation data for abiotic and biotic stresses for the four genera were extracted from GRIN and the ECCDBs.

### The two FIGS methods

The ecogeographic method first generates an optimized ecogeographic core subset using the respective ELC map of the genus. When we have a large number of accessions or population occurrences, the core subset is composed of records that are representative of the different ecogeographic units of the ELC within the distribution of the genus. Within each ecogeographic unit, accessions are selected that are better ranked according to the variable used to look for adaptation for a particular abiotic stress tolerance. R, a free software environment for statistical computing and graphics ([www.r-project.org](http://www.r-project.org)) is used for this purpose and specific R scripts have been developed to carry out this selection (Fig. 2). To obtain the final subset of interest, the ecogeographical variable of interest is used to rank the optimized ecogeographic core subset and select the records with the highest or lowest values of the variable of interest. An R script can be used to select the best of a previously specified number of accessions. Alternatively, all accessions that comply with a particular threshold can be selected.

The modelling method, based on trait evaluation data, aims to calibrate a predictive computer model with R to identify gene bank accessions and populations with a higher density of ge-



**Figure 2** Running the R script to select the *Avena* subset for aluminum toxicity tolerance

netic variation for a target trait. It uses known trait data as a training set to calibrate the model. Climatic data and other environmental data layers serve as the explanatory or independent multivariate variables.

While we have sufficiently large datasets with occurrence data for the ecogeographic method, the quantity of available evaluation data results are a major limitation. The typical number of trait evaluation data points per species extracted from the data sources was fewer than ten, although some of the species have a few hundred trait data points. However, when matching the germplasm occurrences and accessions with trait data points to the occurrences with acceptable georeferenced coordinates, the number of data points per species dropped considerably and left—even in the best cases—less than 50 records per species.

The predictive computer models were intended to be calibrated using the Random Forest algorithm (Breiman, 2001), one of the methods that had performed best previously. Due to the low number of trait evaluation data in the final datasets, these sets did not succeed in calibrating any predictive models. The method of predictive modelling with FIGS for R was however refined using a dataset for stem rust on wheat made available by the USDA Agricultural Research Service (ARS), which had already been explored in previous FIGS studies (Endresen *et al.*, 2011; Bari *et al.*, 2012). In order to apply the method to the PGR Secure target genera, availability of additional evaluation data are required to have a sufficiently large number of records.

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# The Palanga workshop: European PGRFA experts convene to develop national strategy protocols for crop wild relative and landrace diversity conservation

S. Kell and N. Maxted

School of Biosciences, University of Birmingham, Edgbaston, Birmingham, B15 2TT, UK. Email: [s.kell@bham.ac.uk](mailto:s.kell@bham.ac.uk)

One of the goals of the PGR Secure project is to assist national PGR programmes to generate and implement conservation strategies for national crop wild relative (CWR) and landrace (LR) diversity. In collaboration with the European Cooperative Programme for Plant Genetic Resources (ECPGR), PGR Secure convened the workshop, 'Conservation strategies for European crop wild relative and landrace diversity' in Palanga, Lithuania from 7–9 September 2011. The aim of the workshop was to discuss and agree a strategic approach to European and national CWR and LR conservation that will result in their systematic conservation throughout Europe—the first time a systematic approach has been taken to CWR and LR conservation at a continental level.

The workshop, organized by the University of Birmingham, University of Perugia, ECPGR and the Nature Research Centre (NRC), Lithuania and hosted by the NRC, was attended by 101 delegates from 38 European countries and one delegate (Dr. John Wiersema—a member of the PGR Secure External Advisory Board) from the Agricultural Research Service of the United States Department of Agriculture. National representatives were nominated to attend the workshop by ECPGR National Coordinators. Thirty-six members of the Wild Species Conservation in Genetic Reserves Working Group and 32 members of the On-farm Conservation Working Group of the ECPGR *In Situ* and On-farm Conservation Network were nominated, along with 30 *In Situ* National Focal Points (NFPs) and 31 On-Farm NFPs<sup>1</sup> (associated with the ECPGR Documentation and Information Network).



**Figure 1** Participants take notes during a workshop plenary session (Photo: Juozapas Labokas)



**Figure 2** José Iriondo presenting an introduction to ecogeographic data analysis in the CWR working group session, with Mora Aronsson in the Chair (Photo: Juozapas Labokas)

The main topics discussed at the workshop were:

- How to create national CWR and LR inventories;
- CWR and LR prioritization, diversity analysis and threat assessment;
- Data collection, management and exchange;
- Linking conservation to use;
- Development and implementation of national CWR/LR conservation strategies by the ECPGR Network members.

**“A key issue arising from the workshop is the need to design national strategies to suit individual countries”**

The workshop comprised a series of presentations on the current state of the art of CWR and LR conservation in Europe, available approaches and methods for CWR and LR conservation illustrated with case studies, and discussion sessions on the practical application of the approaches and methods, during which participants shared knowledge on current national activities and agreed on the way forward (Figs. 1 and 2). The programme consisted of three plenary sessions and three parallel working group sessions: 1) CWR conservation, 2) LR conservation and 3) information management.

<sup>1</sup> Some participants are members of more than one working group.

Participants agreed on the general approach to the development of national CWR and LR conservation strategies and the issue of funding the planning stages of the strategies was debated. PGR Secure, via the project's helpdesk ([www.pgrsecure.org](http://www.pgrsecure.org)), agreed to make available a list of policy drivers that will help persuade governments to provide the money needed, along with guidance on identifying relevant national obligations and how to take them forward. A key issue arising from the workshop is the need to design national strategies to suit individual countries, adapting the models discussed according to available resources.

After the workshop, participants joined an excursion to the Curonian Spit—a 98 km long, thin, curved sand dune spit that

separates the Curonian Lagoon from the Baltic Sea coast (Figs. 3 and 4). This UNESCO World Heritage Site stretches from Kaliningrad Oblast, Russia in the south to southwestern Lithuania in the north. The Curonian Spit National Park occupies all the land of the Spit and nearby strips of the coastal waters on both sides. There are two strict nature reserves as well as forest seed stands and several landscape reserves established in the National Park. For further information visit [www.visitneringa.com/en](http://www.visitneringa.com/en).

For a full report of the workshop, visit [www.pgrsecure.org/palanga\\_workshop](http://www.pgrsecure.org/palanga_workshop).



**Figure 3** Information boards at the Curonian Spit National Park (Photo: Pavol Hauptvogel)



**Figure 4** View of the Curonian Spit and Lagoon (Photo: Hannah Fielder)

## Prioritization of crop wild relatives in Spain

M.L. Rubio Teso<sup>1</sup>, M.E. Torres<sup>2</sup>, M. Parra-Quijano<sup>2</sup> and J.M. Iriondo<sup>1</sup>

<sup>1</sup> Departamento de Biología y Geología, Área de Biodiversidad y Conservación, Universidad Rey Juan Carlos, C/ Tulipán, s/n. E-28933 Móstoles (Madrid) Spain. Email: [marialuisa.rubio@urjc.es](mailto:marialuisa.rubio@urjc.es)

<sup>2</sup> Departamento de Biología Vegetal, Universidad Politécnica de Madrid, Ciudad Universitaria, Avda. Complutense, s/n. E-28040 Madrid, Spain. Email: [elena.torres@upm.es](mailto:elena.torres@upm.es)

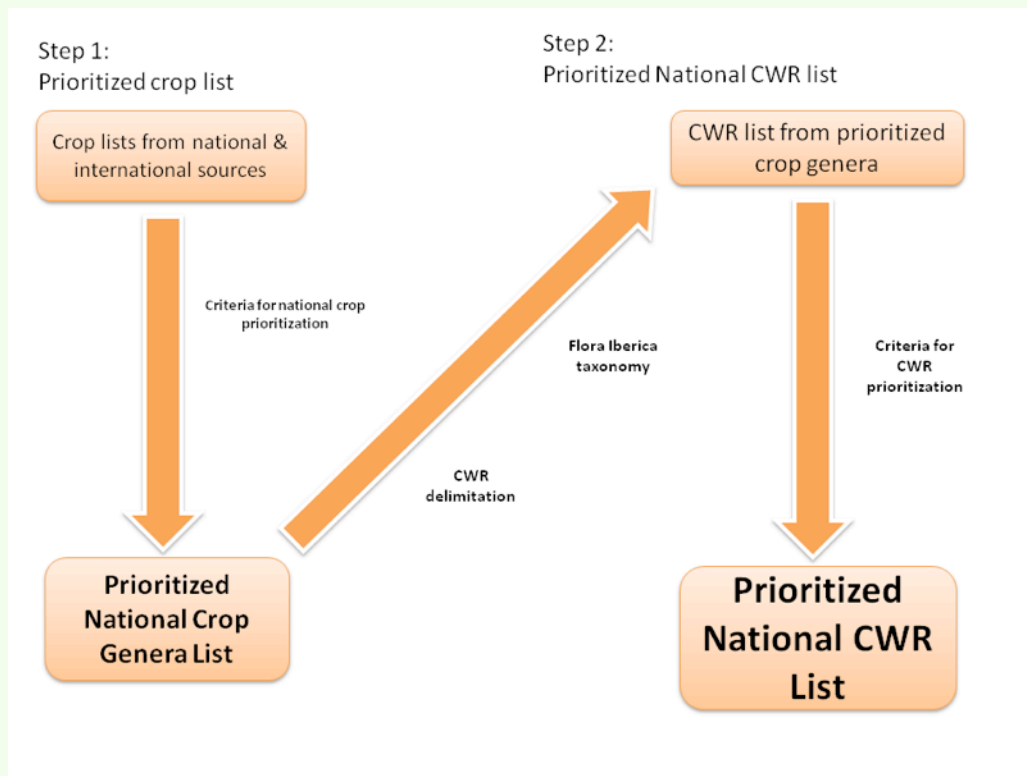
The relevance of crop wild relatives (CWR) essentially responds to the necessity of guaranteeing food security through the provision of a wide gene pool of potential gene donors. The key matter is to delimitate which species are to be included in the so-called group of CWR. Taking into consideration the idiosyncrasy of each country it is important to be able to apply a precise definition of the concept of CWR in order to determine a clear subset of plant species that can be thought of for further action. The case of Spain, one of the countries with the greatest CWR diversity in Europe with well over 6000 species (Kell *et al.*, 2008), constitutes a challenge for deciding how many and which CWR should be included in the final prioritized list.

There should not be discussion about the inclusion of CWR of major food and forage and fodder crops but, should relatives of promissory species in emerging markets which currently are

not as important as major crops, those so-called marginalized crops or those only produced in narrow geographic ranges be considered? Should non-native or naturalized species be included? What about ornamentals and crops utilized for purposes other than food? These questions and many similar others must be answered when planning CWR prioritization actions.

### Setting the CWR groups: delimitation of the crop wild relative concept

Assuming that food and forage CWR are unequivocally a 'priority when prioritizing', some decisions about the inclusion of other groups were made. For instance, although forestry species are indeed of great socio-economic importance and could be viewed as CWR, it was decided to not consider them as candidates for inclusion in the Spanish national CWR inventory. The reason for this decision was simple: they are already



**Figure 1** Step by step process followed in the strategy to produce the Spanish national CWR inventory

protected by specific national forestry laws and are the object of specific national and regional actions of conservation of biodiversity and fight against climate change.

Other groups such as those classified as 'Other uses' (including industrial, textile, medicinal, aromatic, etc.) and 'Ornamental' were included in the target group, answering to the unquestionable necessity of preserving a large range of biodiversity uses and conserving the overall complex Spanish genetic resources breadth.

Concerning the relatives of promissory species, marginalized crops and crops from narrow geographic ranges, they were not given a specific weight and were included only if they were related to the stated groups.

As a result of this, CWR were finally categorized in four groups: 1) food, 2) fodder and forage, 3) ornamental, and 4) other uses.

#### Generating initial lists

Given the large number of CWR species present in Spain, instead of starting from the Spanish subset of the CWR Catalogue for Europe and the Mediterranean generated by the PGR Forum project (Kell *et al.*, 2005), the initial list was generated from a process that started with the prioritization of crops and then identified the CWR related to these crops (Fig.1).

According to this approach, a comprehensive list of genera of important crops was generated taking as a baseline the crops included in Annex 1 of the FAO International Treaty on Plant Genetic Resources for Food and Agriculture (FAO, 2001) and

the Spanish Annuary of Agriculture Statistics (Ministerio de Medio Ambiente, Medio Rural y Marino, 2010). Subsequently, additional genera were included after consulting the Annual Report of the Community Plant Variety Office in Europe (2010), and the list of the International Union for the Protection of New Varieties of Plants (UPOV) (2010). Furthermore, experts on agrobiodiversity were asked for advice and additional proposals, and bibliographic resources related to trends in breeding were also consulted (Kole, 2011a,b,c,d,e). Finally, other national inventories and prioritized lists were checked to evaluate their approaches and include additional genera of potential interest (Markkola, 2005; Barazani *et al.*, 2008; Labokas *et al.*, 2010; Magos Brehm *et al.*, 2010; Berlinger and Crespo, 2011).

#### Setting criteria for prioritization

As the generated list of crop genera was too extensive to prioritize all the wild relatives associated with them, some criteria were set for selecting the most important crops for Spain and for world food security as follows:

1. The genus must be listed in Annex 1 of the International Treaty or in the Spanish Annuary of Agricultural Statistics; and
2. at least one of the species in the genus is native to Spain; and
3. it has registered varieties in Spain.

Some additional genera were also considered based on expert advice due to their socio-economic importance for the country. From this point, following the scheme depicted in Figure 1, genera were classified into the four categories of CWR considered, and all their corresponding species were compiled from Flora Iberica (Castroviejo *et al.*, 1986–2011) or the An-

thos project (Anthos, 2011) when the genus was not yet included in Flora Iberica. In the case of the Canary Islands the List of Wild Animal and Plant Species of the Canary Islands (Acebes Ginovés *et al.*, 2010) was consulted.

Once the CWR list was generated, additional criteria for prioritization were set. In this case, only species native to Spain were taken into consideration, following similar approaches followed in other countries (Maxted *et al.*, 2007; Kell *et al.*, 2012). Naturalized taxa, although considered in some neighbouring countries because of their importance to the development of the national economy (Magos Brehm *et al.*, 2008), were not selected as target species. We considered that the genetic diversity of wild populations of naturalized taxa is probably not great and less important than that of native taxa.

In addition to focusing on native taxa, the additional criteria adopted for prioritizing CWR are listed below:

1. Taxon must be in Gene Pools 1B and 2, or classified into Taxon Groups 1B, 2 or 3; or
2. be a threatened or near threatened taxon according to IUCN Red List Categories (CR – Critically Endangered, EN – Endangered, VU – Vulnerable, NT – Near Threatened), or
3. be endemic to Spain.

Gene Pool classifications were established according to the Gene Pool concept proposed by Harlan and de Wet (1971). For this aim the gene pool database created by Vincent *et al.* (2012) was consulted. The Taxon Group concept followed the definition given by Maxted *et al.* (2006), and was applied to those taxa which have not yet been classified according to the Gene Pool concept. Endemism was evaluated following Flora Iberica and the List of Wild Animal and Plant Species of the Canary Islands (Acebes Ginovés *et al.*, 2010).

Using the gene pool information, some species from genera that had not previously been prioritized were included, as they have been confirmed of potential use, or are already being used in breeding for the previously selected genera (e.g., *Diplotaxis*, *Moricandia* or *Capsella* for brassica crops).

**Table 1** Prioritized crop genera classified into the four use categories

Food genera		Fodder and forage genera	Ornamental genera	Other uses
<i>Aegilops</i>	<i>Lactuca</i>	<i>Agrostis</i>	<i>Argyranthemum</i>	<i>Carthamus</i>
<i>Allium</i>	<i>Lathyrus</i>	<i>Dactylis</i>	<i>Dianthus</i>	<i>Gentiana</i>
<i>Apium</i>	<i>Lens</i>	<i>Festuca</i>	<i>Limonium</i>	<i>Hypericum</i>
<i>Asparagus</i>	<i>Malus</i>	<i>Lolium</i>	<i>Narcissus</i>	<i>Lavandula</i>
<i>Avena</i>	<i>Moricandia</i>	<i>Lupinus</i>	<i>Rosa</i>	<i>Linum</i>
<i>Beta</i>	<i>Olea</i>	<i>Medicago</i>		<i>Papaver</i>
<i>Borago</i>	<i>Patellifolia</i>	<i>Poa</i>		
<i>Brassica</i>	<i>Pisum</i>	<i>Trifolium</i>		
<i>Capsella</i>	<i>Prunus</i>	<i>Astragalus</i>		
<i>Cicer</i>	<i>Pyrus</i>	<i>Brachypodium</i>		
<i>Cichorium</i>	<i>Raphanus</i>			
<i>Cyanara</i>	<i>Secale</i>			
<i>Daucus</i>	<i>Sinapis</i>			
<i>Diplotaxis</i>	<i>Solanum</i>			
<i>Erucastrum</i>	<i>Vicia</i>			
<i>Fragaria</i>	<i>Vitis</i>			
<i>Hordeum</i>				



*Lupinus angustifolius* L. (Photo: Rubén Milla)

## Results

After compiling and adding all genera included in the above-mentioned lists and inventories, the initial list of genera of important crops for Spain and world food security amounted to 202 genera. The application of the criteria for prioritizing crop genera led to a list of 54 genera. This list comprised 33 food related genera in 12 families, 10 fodder and forage related genera in two families, five ornamental related genera and six genera related to 'other uses' (Table 1). The family with the most genera was Fabaceae, with 10 genera, followed by Poaceae and Brassicaceae with nine and seven respectively.

Out of these 54 genera, 33 of them, as mentioned before, are related to major food crops and have led to a final prioritized list of 149 food-related CWR species. Results are as follows:

- a) Number of criteria fulfilled: 110 species accomplish one of the criteria; 33 species meet two of the requirements; and six species out of the 149 fit all criteria.
- b) Gene Pool and Taxon Group concepts: 22 of the selected species are wild forms of the crops of reference (GP1B); 25 of them are classified into GP2 (with possibility of natural gene transfer); 16 have been classified in TG2 (same series or section as the crop) and finally, 10 species have been assigned to TG3 (same subgenus as the crop).
- c) Threat assessment: 42 out of the 149 species are considered as threatened or near threatened, under the CR, EN, VU or NT categories set by IUCN.
- d) Endemism: 48 species of the complete list are endemic to Spain. Among these, seven are endemic to the Iberian Peninsula.
- e) Uses in breeding: 19 species have confirmed potential use or are already used in crop breeding and are considered useful sources of genes for crops of importance worldwide.



Detail of *Brassica barrelieri* (L.) Janka, one of the CWR prioritized for Spain (Photo: Julia Chacón Labella)

### Next steps

The same process already achieved with the food group is being applied to the other three groups. Taking into consideration the results obtained until now, we expect to end up with a final list of about 400 taxa, which would represent about 7 % of the total national CWR flora of Spain and would constitute the prioritized Spanish national CWR inventory.

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# Floristic and vegetation databases as tools for CWR surveys: a case study from Central Italy

F. Landucci, L. Panella, D. Gigante, D. Donnini, R. Venanzoni, R. Torricelli and V. Negri  
 Department of Applied Biology, University of Perugia (UNIPG), Borgo XX Giugno 74, I-06121 (Perugia) Italy.  
 Email: [flavia.landucci@gmail.com](mailto:flavia.landucci@gmail.com)

Our work in PGR Secure is dedicated to the national CWR inventory and development of CWR conservation strategies. Currently we are operating at both national and regional scale, building a national CWR checklist, while at the same time concentrating our research on a small group of CWR important for their economic and conservation value, such as species included in the genera *Apium*, *Avena*, *Beta*, *Brassica* and *Prunus*. Our intent is to improve the knowledge on the ecology and distribution of these taxa to plan specific monitoring and conservation strategies.

## Floristic and vegetation databases are useful tools in nature conservation

During the last two decades, modern information technologies have facilitated the development of electronic databases on various aspects of biodiversity (Michener and Brunt, 2000). Worldwide, many different databases for botanical data were developed in the context of plant biodiversity and vegetation science. Some of these databases are freely consultable in the web, such as 'FloraWeb' (<http://floraweb.de>), 'VegBank' ([www.vegbank.org](http://www.vegbank.org)), 'SOPHY' (<http://sophy.u-3mrs.fr>), 'SIVIM' ([www.sivim.info/sivi](http://www.sivim.info/sivi)), while others are available at private or public institutions such as 'BASECO' (Gachet *et al.*, 2005) or the 'Czech National Phytosociological Database' (Chytrý and Rafajová, 2003). Metadata about major vegetation-plot databases of the world are registered and available at the 'Global Index of Vegetation-Plot Databases' (GIVD - [www.givd.info](http://www.givd.info), Dengler and GIVD Steering committee, 2010).

Each database was born with the intent of storing, easily managing and analysing floristic and vegetation data (herbarium specimens or phytosociological relevés). These types of databases are important tools for nature conservation because they contain data such as plant traits, taxonomic, geographi-



**Figure 1** Spikelet of *Avena barbata* Pott ex Link (Photo: Flavia Landucci)

cal, temporal and ecological information. Such data could be used for identifying biodiversity hot spots, spatial and diachronic analysis of plant diversity or simply for synthesizing and reviewing the knowledge about single species, groups of species, vegetation types or particular habitats. Information about the status, distribution and ecology of CWR can be directly or indirectly obtained by these archives.

## The 'anArchive' project

Among the available floristic and vegetation databases, 'anArchive' ([www.anarchive.it](http://www.anarchive.it)) is an open-source web database to store, retrieve and analyse botanical data, which involves several Italian universities. Currently the database hosts several regional projects and the national project VegItaly (Venanzoni *et al.*, in press). The taxonomical nomenclature adopted by the database is largely in accordance with 'An Annotated Checklist of the Italian Vascular Flora' (Conti *et al.*, 2005, 2007) and the main national and international taxonomic revisions. Floristic and vegetation data from the national literature, herbaria specimens and unpublished phytosociological relevés are stored in the database.

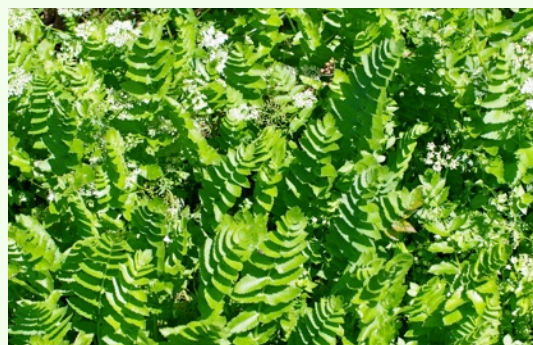
## A case study at regional scale

'anArchive' has been initially used to investigate the status and the distribution of celery, beet, oat, cabbage, cherry and plum crops and their CWR at regional scale (Umbria, Central Italy). The CWR under study belong to the genera *Aegopodium*, *Apium*, *Avena* (Fig. 1), *Beta* (Fig. 2), *Brassica*, *Helosciadium* (Fig. 3), *Petroselinum*, *Prunus* and *Smyrniunum*.

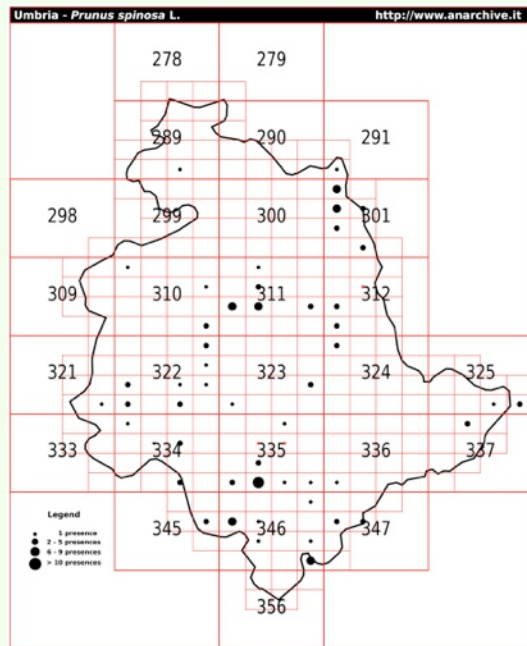
According to the reviewed literature and data stored in the database, one species of *Aegopodium*, one of *Apium*, one of *Helosciadium*, one of *Petroselinum*, one of *Smyrniunum*, four of *Avena*, two of *Beta*, six of *Brassica* and nine of *Prunus* are recorded in Umbria. The less mentioned taxa are *Brassica*



**Figure 2** *Beta vulgaris* L. subsp. *maritima* (L.) Arcang. in a clay gully (Photo: Flavia Landucci)



**Figure 3** Community of *Helosciadium nodiflorum* (L.) W.D.J. Koch growing inside a channel (Photo: Roberto Venanzoni)



**Figure 4** Distribution map of *Prunus spinosa* L. as extracted from the anArchive database

*gravinae* Ten., *B. rapa* L., *Prunus armeniaca* L., *P. domestica* L. subsp. *domestica* and *P. domestica* L. subsp. *insititia* Bonnier et Layens, while the most represented species are *Avena barbata* Pott ex Link, *A. fatua* L., *Prunus avium* L., *P. mahaleb* L. and *P. spinosa* (L.) Batsch. For each investigated taxon a distribution map was constructed as shown in Figure 4.

A few records are available for cultivated plants such as *Apium graveolens* L., *Beta vulgaris* L., *Brassica nigra* (L.) Koch, *B. napus* L., *Prunus dulcis* L. or *P. persica* (L.) Batsch (Table 1). It seems that the scientific interest for cultivated taxa has been decreasing in the last century. Such taxonomic entities are in fact recorded only by few authors in the recent literature, while herbaria specimens of these plants date back to the 19th century. Most records are instead available for wild taxa such as *Prunus spinosa* L., *Avena barbata* Pott ex Link or *Helosciadium nodiflorum* (L.) W.D.J. Koch (Fig. 4) which often occur in vegetation plots and relevés, both within and outside protected areas. The distribution and abundance of most of the taxa are probably underestimated in Umbria, because different parts of the region were explored with different accuracy. The most studied localities are usually included in protected or important areas for

some conservation aspects.

This initial work showed that a poor level of information exists about CWR in Umbria, as probably in the whole of Italy (Table 1). The next step will then consist of field surveys to integrate the existing knowledge. A second phase will be to stratify the territorial grid according to the species record density and to overlay these data with the land use, in order to outline a model which can point out the most CWR-rich landscapes.

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**Table 1** Status of the knowledge and distribution of the investigated taxa (data extracted from the 'anArchive' database)

Taxon	Cultivation status	Status of knowledge	Distribution
<i>Aegopodium podagraria</i> L.	Wild	Good	widespread
<i>Apium graveolens</i> L.	Escaped from cultivation	Not sufficient	localized
<i>Avena barbata</i> Pott ex Link	Wild	Good	widespread
<i>Avena fatua</i> L.	Wild	Good	widespread
<i>Avena sativa</i> L.	Escaped from cultivation	Not sufficient	widespread
<i>Avena sterilis</i> L.	Wild	Sufficient	localized
<i>Beta trygina</i> Watsd. e Kit	Wild	To be confirmed	localized
<i>Beta vulgaris</i> L.	Escaped from cultivation	Not sufficient	localized
<i>Beta vulgaris</i> L. subsp. <i>maritima</i> (L.) Arcang.	Wild	Recently recorded	localized
<i>Brassica fruticulosa</i> L.	Wild	Erroneously recorded	localized
<i>Brassica gravinae</i> Ten.	Wild	Not sufficient	localized
<i>Brassica napus</i> L.	Escaped from cultivation	Not sufficient	localized
<i>Brassica nigra</i> (L.) Koch	Escaped from cultivation	Not sufficient	localized
<i>Brassica oleracea</i> L.	Escaped from cultivation	Not sufficient	localized
<i>Brassica rapa</i> L.	Escaped from cultivation	Not sufficient	localized
<i>Helosciadium nodiflorum</i> (L.) W.D.J. Koch	Wild	Not sufficient	widespread
<i>Petroselinum crispum</i> (Mill.) Fuss	Escaped from cultivation	Not sufficient	localized
<i>Prunus armeniaca</i> L.	Escaped from cultivation	Not sufficient	localized
<i>Prunus avium</i> L.	Escaped from cultivation	Good	widespread
<i>Prunus cerasus</i> L.	Escaped from cultivation	Not sufficient	localized
<i>Prunus domestica</i> L. subsp. <i>domestica</i>	Escaped from cultivation	Not sufficient	localized
<i>Prunus domestica</i> L. subsp. <i>insititia</i> Bonnier et Layens	Escaped from cultivation	Not sufficient	localized
<i>Prunus dulcis</i> L.	Escaped from cultivation	Not sufficient	localized
<i>Prunus mahaleb</i> L.	Wild	Good	widespread
<i>Prunus persica</i> (L.) Batsch.	Escaped from cultivation	Not sufficient	localized
<i>Prunus spinosa</i> L.	Wild	Good	widespread
<i>Smyrnium olusatrum</i> L.	Wild	Good	localized

# UK wild species to underpin food security: species selection, genetic reserves and targeted collection

H. Fielder<sup>1</sup>, J. Hopkins<sup>2</sup>, C. Smith<sup>3</sup>, S. Kell<sup>1</sup>, B. Ford-Lloyd<sup>1</sup> and N. Maxted<sup>1</sup>

<sup>1</sup> School of Biosciences, University of Birmingham, Edgbaston, Birmingham, B15 2TT, UK. Email: [hvf708@bham.ac.uk](mailto:hvf708@bham.ac.uk)

<sup>2</sup> Natural England, 3rd Floor, Touthill Close, City Road, Peterborough, PE1 1UA. Email: [John.Hopkins@naturalengland.org.uk](mailto:John.Hopkins@naturalengland.org.uk)

<sup>3</sup> Scottish National Heritage, Caspian House, Mariner Court, Clydebank Business Park, Clydebank, G81 2NR. Email: [cecile.smith@snh.gov.uk](mailto:cecile.smith@snh.gov.uk)

As a result of the domestication process, crop plants have often experienced a genetic bottleneck which has left them with limited genetic diversity (Tanksley and McCouch, 1997); it has been estimated that during the last century 75 % of crop genetic diversity has been lost (FAO, 1998). This is due to the cultivation of highly bred crop monocultures with a narrow genetic base which as a consequence are poorly equipped to respond to the changing environment. On the other hand, the wild relatives of crops may harbour much higher levels of genetic diversity and can be used to donate beneficial genes to crops for their improvement. The conservation of crop wild relatives (CWR) can therefore preserve a wider gene pool that can be used to improve future food security.

Kell *et al.* (2008) identified that 8 % of European CWR can be found in the UK; among the most economically notable in a UK context are perhaps *Beta vulgaris* subsp. *maritima* (Fig.1) and wild *Brassica* species, the wild relatives of sugar beet (*Beta vulgaris* L.) and cultivated brassicas (e.g., *Brassica oleracea* L.), respectively. For there to be an adequate global response to the loss of crop genetic diversity, the conservation of CWR taxa in all parts of their range is required. To this end, and as part of the PGR Secure project, preparatory scientific work for a conservation strategy for UK CWR is being carried out, and the first stages of this work are reported here.

This project is particularly timely as, at a strategic level, conservation of plant genetic resources for food and agriculture (PGRFA) is becoming a higher priority internationally and in the UK. There is a mandate to conserve CWR via a number of global and European policy instruments such as the Global Strategy for Plant Conservation (GSPC) (CBD, 2010) as part of the Convention on Biological Diversity (CBD) (CBD, 1992), the International Treaty on Plant Genetic Resources for Food and Agriculture (ITPGRFA) (FAO, 2001) and the European Strategy for Plant Conservation (ESPC) (Planta Europa, 2008). Recently, a report on UK CWR conservation by Hopkins



Figure 1 *Beta vulgaris* subsp. *maritima* (Photo: Nigel Maxted)

and Maxted (2011) was published in order to raise the profile of the importance of this work. Subsequently, the Department for Environment, Food and Rural Affairs (Defra) has developed a national conservation strategy for England, *Biodiversity 2020* (Defra, 2011), which includes the objective to conserve “agricultural genetic diversity in cultivated plants, farmed animals and wild relatives”.

“at a strategic level, conservation of plant genetic resources for food and agriculture (PGRFA) is becoming a higher priority”

## Species selection

To systematically conserve CWR across the UK, and to subsequently explore conservation options more practically in England and Scotland, we have taken the UK checklist of CWR species as a starting point (Maxted *et al.*, 2007). This checklist was derived from the CWR Catalogue for Europe and the Mediterranean (Kell *et al.*, 2005) and harmonized with accepted taxonomic classifications used by UK experts. The final checklist for the UK as a whole contains 1524 CWR species.

With an aim to develop management actions to improve the conservation of UK CWR genetic diversity—both *in situ* and *ex situ*—it is first necessary to select which species most require conservation attention. Limited funds and resources mean that it is impossible to conserve all species and so a selection process has been developed to create CWR inventories for each component country which identifies and prioritizes those species most in need of protection and management. The process involves the selection of criteria on which to base the prioritization of species requiring conservation attention, and the selection of priority species based on these criteria.

Through consultation with Natural England, the UK government’s conservation agency for England, four criteria were selected as a basis for prioritization of CWR in England, as outlined below (consultation with experts from Scottish National Heritage is yet to begin).

1. Use of the related crop  
The decision was taken to assign priority only to those CWR species that are related to crops used as a human food source or for animal feed (forage/fodder). Use data were extracted from GRIN Taxonomy for Plants (USDA, ARS, National Genetic Resources Program, 2012).
2. Economic value of the related crop  
The primary use of CWR is by definition genetic improvement of crop plants, therefore the value of any particular



crop will infer the current relative value of its wild relatives (Maxted *et al.*, 1997; Ford-Lloyd *et al.*, 2008; Maxted and Kell, 2009; Magos Brehm *et al.*, 2010; Kell *et al.* 2012). Taking a lead from the method used by Kell *et al.* (2012), we will use production quantity data at the global, European and national levels from FAOSTAT (2012), along with monetary value data from Eurostat (2012) as well as production quantity and monetary value data at the UK level to infer economic value.

### 3. Genetic closeness to a crop

CWR vary in their genetic closeness to their related crop and both the Gene Pool and Taxon Group concepts have been used to define this genetic relationship. It is seen as a reasonable assumption that the closer the CWR is to the crop, the higher the priority for conservation due to greater ease of gene transfer for crop improvement. Harlan and de Wet (1971) devised the Gene Pool (GP) concept which classifies cultivated plants and their wild relatives into: GP1A which contains the cultivated forms of the crop and GP1B the con-specific wild and weedy forms of the crop; GP2 which contains closely related species from which gene transfer is possible but may be difficult; and GP3 which contains species from which gene transfer to the crop is not possible or requires techniques such as embryo rescue, somatic fusion or genetic engineering. Taking this into consideration, priority will be given to wild relatives from GP1B and GP2. Where gene pool data are lacking the Taxon Group concept has been applied to infer genetic relatedness between a crop and its wild relatives using the taxonomic hierarchy, prioritizing Taxon Groups 1B, 2 and 3 (Maxted *et al.*, 2006). Data have been extracted from the 'global checklist of priority CWR' which lists the Gene Pool and Taxon Group concepts for over 1400 taxa, including 173 crops (Vincent *et al.*, 2012a,b).

### 4. Change in population range

By considering changes in population ranges over time it is possible to identify CWR taxa whose populations are in decline and priority can be assigned to these populations.

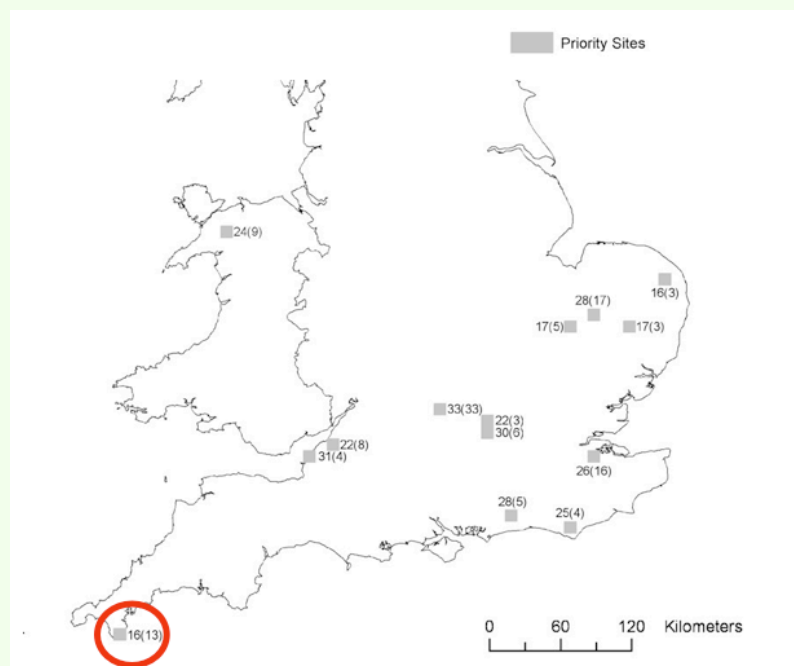
The occurrence and abundance of plant species in Britain were surveyed in both 1987 and 2004, and the changes in their population ranges between these years have been reported (Braithwaite *et al.*, 2006). Using these data we are able to identify the population trends of CWR in the UK.

Applying the use criterion, the total list of 1524 CWR species was significantly reduced to a provisional list of 314 CWR species that are related to crops cultivated specifically for human food or animal forage or fodder in England. Three of the 23 plant families included in the partially prioritized list of 314 CWR species contain a notably high number of CWR genera: Poaceae (27 genera), Brassicaceae (13 genera) and Fabaceae (12 genera).

We are currently in the process of assigning economic values (based on production quantity and monetary values) to each species. Once this is complete, the next steps will be to select the priority species for England to peer review and to conduct a GIS gap analysis to determine the current conservation status of each priority species. This process will also be carried out for CWR in Scotland. We hope to then develop management actions for the identified priority CWR species, thus ensuring they are actively managed and systematically conserved.

### Local CWR genetic conservation: The Lizard Peninsula, Cornwall

Maxted *et al.* (2007) analysed CWR distribution data from the Botanical Society of the British Isles (BSBI) to identify the most species rich areas in terms of CWR in the UK. The idea behind this was to locate areas of the UK which are most suitable for the establishment of protected areas for CWR conservation. They concluded that 17 sites would be sufficient to conserve two thirds of CWR species in the UK (Fig. 2). One of the 17 CWR rich sites identified was the Lizard Peninsula in Cornwall.



**Figure 2** Fourteen of the 17 priority sites identified for *in situ* CWR conservation in the UK, including the Lizard peninsula (circled in red) Source: Maxted *et al.* (2007)

The Lizard Peninsula (Fig. 3) is located in the southwest of England and includes Lizard Point, the most southerly point of Great Britain. It is a site which potentially harbours large amounts of genetic diversity due to i) its complex igneous, metamorphic and sedimentary geology, ii) varied land use pattern and iii) a warmer climate than much of the UK, with complex microclimatic variation related to topography and degree of maritime exposure.

In spring 2012 we will carry out field work in this area, collecting material from CWR populations in order to conduct a genetic assessment. This assessment will use AFLP markers to analyse genetic diversity within and between populations of the CWR on the Lizard. This will enable us to determine the patterns of genetic diversity across the Lizard site.



**Figure 3** An area of the Lizard peninsula in Cornwall, southwest England (Photo: Hannah Fielder)

Eight species related to some of the most economically valuable crops within the UK have been selected for this study: *Allium ampeloprasum* L. var. *babingtonii* (Borrer) Syme, *A. schoenoprasum* L., *Asparagus officinalis* L. subsp. *prostratus* (Dumort.) Corb., *Beta vulgaris* L. subsp. *maritima* (L.) Arcang., *Daucus carota* L. subsp. *gummifer* (Syme) Hook. f., *Linum bienne* Mill., *Trifolium occidentale* Coomb and *T. repens* L.

In conjunction with the Lizard genetic assessment, we will undertake a genetic assessment of the same species across the rest of the UK; the rationale behind this being that we can then compare the patterns of genetic diversity found on the Lizard to those found in the rest of the UK. Target 9.1 of the ESPC states the requirement for the “establishment of 25 European crop wild relative genetic reserves covering the major hotspots of species and genetic diversity” (Planta Europa, 2008). If the Lizard contains particularly high levels of or unique genetic diversity when compared to the rest of the UK, we can show that it is a fitting candidate for establishment of a CWR genetic reserve. Further, the genetic analysis is intended to explore the number and local distribution of populations required to conserve this genetic diversity.

#### Targeted collection

CWR are poorly represented in *ex situ* collections. Dias and Gaiji (2005) found that within European collections only 4 % of accessions were CWR. In addition, Maxted *et al.* (2008) showed that the ratio of cultivated species to wild relative accessions recorded in EURISCO was 12:1; the authors noted that this seems counterintuitive considering that wider genetic diversity is contained within CWR than in the crops themselves.

It is clear that vast gaps exist in current *ex situ* collections of CWR. In order to underpin future food security it is vital that these gaps are filled through further collection and storage of material. However, any additional collection in the field should not be random but should be specifically targeted to ensure the maximum range of genetic diversity can be collected and maintained within gene banks.

Genetic distance between accessions already conserved within gene banks can be measured to identify where genetic gaps exist. This can then help to identify where new collections should be sought. Put simply, if three accessions from three ecogeographically distinct areas were found to be genetically distinct from each other, it is highly likely that through further collection in different ecogeographic areas, more genetic diversity will be identified and can be

collected and stored (Fig. 4a). A wider range of genetic diversity can be collected using this method. In contrast, if two of the three accessions were genetically close to each other, then there would be no genetic gap between them and no need to carry out further collection (Fig. 4b).

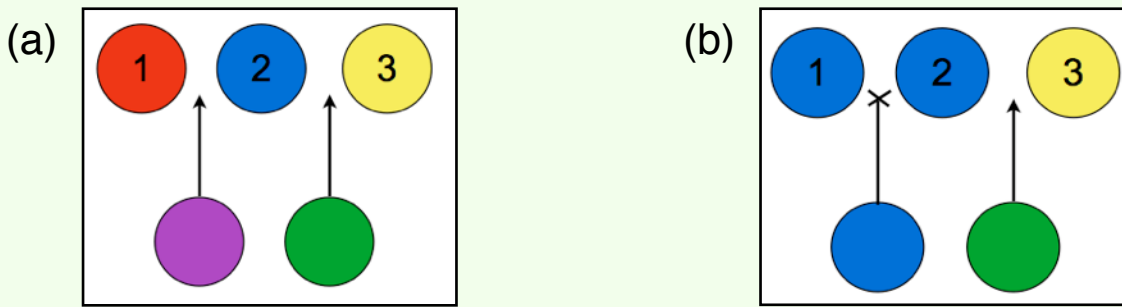
We are looking to use next generation sequencing (NGS) as a means of accurately quantifying levels of genetic distance between *ex situ* accessions to allow efficient targeting of collection in the field. Thus far, NGS technology has primarily been used within medical research; for example, Walsh (2010)

### “vast gaps exist in current *ex situ* collections of CWR”

found the technology to be a cheaper, more accessible and personalized method of genetic testing for mutations in breast cancer genes. However, we hope to be able to demonstrate a new application for NGS technology in the field of conservation. Through targeting collection of material we can maintain more comprehensive *ex situ* collections of CWR, covering a much wider range of genetic variation.

#### Conclusion

By developing CWR conservation actions for priority species in national inventories for England and Scotland we hope to improve the management of priority CWR populations throughout these countries. Furthermore, through the establishment of CWR genetic reserves, *in situ* conservation can be targeted in those areas containing particularly high levels of genetic diversity. These two approaches will help to promote active and systematic conservation of CWR populations *in situ*. *In situ* conservation must however be complemented by *ex situ* con-



**Figure 4** Schematic diagram showing (a) three genetically distinct accessions (1, 2 and 3) between which exist genetic gaps which must be filled through targeted collection and (b) a situation where two out of three accessions are genetically similar (1 and 2) so no further collection of material is required from these locations

ervation. We hope to demonstrate how NGS technology can be used to achieve targeted collection in order to maintain more comprehensive *ex situ* collections of CWR accessions. With complementary *in situ* and *ex situ* conservation across the UK we can take essential steps forward in helping to safeguard PGRFA for their use in crop improvement to meet the needs of future generations.

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## Pieces of the puzzle—Trait Information Portal

S. Dias

Biodiversity International, Rome, Italy. Email: [s.dias@cgiar.org](mailto:s.dias@cgiar.org)

The need to have a portal to access complementary data on traits is essential and a novelty arising from the PGR Secure project. Further, until now we have been able to access online many resources for *ex situ* information, but very few for *in situ* and on-farm information. This article focuses on one of the tasks of the PGR Secure informatics work package; namely, the development of a Trait Information Portal (TIP). The TIP envisaged will provide a unique entry point for the breeder community to access trait-specific information to help direct their research and allow them to obtain germplasm of CWR and LR for their breeding programmes.

The main concern in the development of the TIP has been to conceptualize a portal that can accommodate the varying needs of data providers and users, including the needs of traditional and the more specialized users such as pre-breeders, breeders and farmers. In developing this tool, the aim is to address the needs of a broad community of practitioners that are interested and want to be part of this new era of using and conserving plant genetic resources for present and future food security, without duplicating resources or efforts already made for similar tools. While assessing the need for such a tool, a major driver has stood out: the need to develop a concept that can accommodate the overall information and data for *in situ* and on-farm material (e.g., inventories, varieties names, location, species and many more) and hyperlink it to *ex situ* information, among other data sources.

“the TIP will include trait information on accession and population data generated within the PGR Secure project, in addition to existing data”

Examples from *ex situ* data gathering, sharing and publishing are available and are being used in the project scenario, but we want to go one step further by making use of current knowledge and challenging the way information and data can be made available in order to facilitate the use of crop wild relatives (CWR) and landraces (LR) for breeding and crop improvement, and at the same time provide a system to manage CWR and LR conservation data. The main ingredients in this challenge are the different data sources, type and nature of the data, and their uses. We are talking about using novel information technology methods and tools to develop a CWR and LR TIP that will include or link up with new sources of information such as phenomics, genomics and transcriptomics data, in addition to the national and European CWR and LR inventories.

Based on this concept the TIP will include trait information on accession and population data generated within the PGR Secure project, in addition to existing data. To achieve this, several activities were planned:

- Conceptualization of a CWR and LR TIP. This activity has been commissioned to develop a TIP infrastructure framework, including its ontology (controlled vocabulary) and technical specifications, and will involve all partners, including breeders;
- The TIP will be able to link to data generated in other PGR Secure work packages; namely the CWR and LR inventories, and characterization data (predictive, phenomic, genomic and transcriptomic);
- Definition of the key information sources that will permit the development of the TIP and its core infrastructure. In particular, the external sources such as the Crop Wild Relative Information System/Population Level Information System (CWRIS/PLIS) (<http://aegro.jki.bund.de/aegro/index.php?id=168>), EURISCO (<http://eurisco.ecpgr.org>), relevant crop-specific European Central Crop Data Bases (ECCDBs) ([www.ecpgr.cgiar.org/germplasm\\_databases.html](http://www.ecpgr.cgiar.org/germplasm_databases.html)) (i.e., *Avena*, *Beta*, *Brassica* and *Medicago* databases) and the European Molecular Biology Laboratory (EMBL) will constitute other sources of information;
- Development of a preliminary version of the TIP for testing and adjustment;
- Testing and validation of the preliminary TIP version with the stakeholder community and revising of the TIP to ensure that it meets the end users' (e.g., breeders') demands;
- Publishing and web-enabling the TIP.

During the project the conceptualization framework for the TIP and its first generic ideas were presented to the consortium at the project kick-off meeting on 15–16 March 2011, in Lyme Regis, United Kingdom, where active feedback and lines of vision were received and discussed. The second step consisted in the laying of the foundations for a wider understanding and up-take of the TIP concept and further identification of needs for the tool.

At the Joint PGR Secure/European Cooperative Programme for Plant Genetic Resources (ECPGR) workshop, 'Conservation strategies for European crop wild relative and landrace diversity', held on 7–9 September 2011, in Palanga, Lithuania (a second PGR Secure project event), the TIP concept was presented under the title 'Development of a European information system for CWR and LR conservation and use data and implementation of the Trait Information Portal—Pieces of the Puzzle' ([www.pgrsecure.bham.ac.uk/sites/default/files/meeting\\_s/palanga/P06\\_European\\_CWR\\_and\\_LR\\_Info\\_System\\_and\\_TIP\\_Dias.pdf](http://www.pgrsecure.bham.ac.uk/sites/default/files/meeting_s/palanga/P06_European_CWR_and_LR_Info_System_and_TIP_Dias.pdf)) where the main focus of the TIP concept on this occasion was to promote the use of ontologies for traits, CWR, LR and crop-specific data in this system. This was a major challenge since the development of ontologies is not yet well understood by many within this community. It therefore provided an opportunity to introduce them and identify the way

forward to enhance the existing CWR and trait ontology to include terms that the user community needs, and to develop the LR ontology and the crop-specific ontologies respectively. The way forward is therefore to use a ‘Triontology’—CWR, LR and Crop-Trait Ontologies. The goal is to: (i) develop an ontology that describes the crops, traits, anatomical and morphological structures, and growth and developmental stages, among other data types; (ii) establish a semantic framework to query across crops, inventories and traits (genotype and phenotype datasets); and (iii) describe crops, CWR and LR data structures and the relationships among them. The presentation given at the meeting demonstrated the effectiveness and value in using these tools as a starting point. The curation and the annotation tools developed by the Bioversity team within the Generation Challenge Programme (GCP) have proven to be well developed and mature tools to be considered for use in the development of the TIP as part of the PGR Secure project. Considering all these already existing resources (curation tool, annotation tool, CWR descriptors, LR descriptors, CWR ontology, Trait ontology, *ex situ* platform, etc.), PGR Secure ontology groups for *Avena*, *Brassica*, *Beta* and *Medicago* CWR and LR will be established and links made to the respective ontologies.

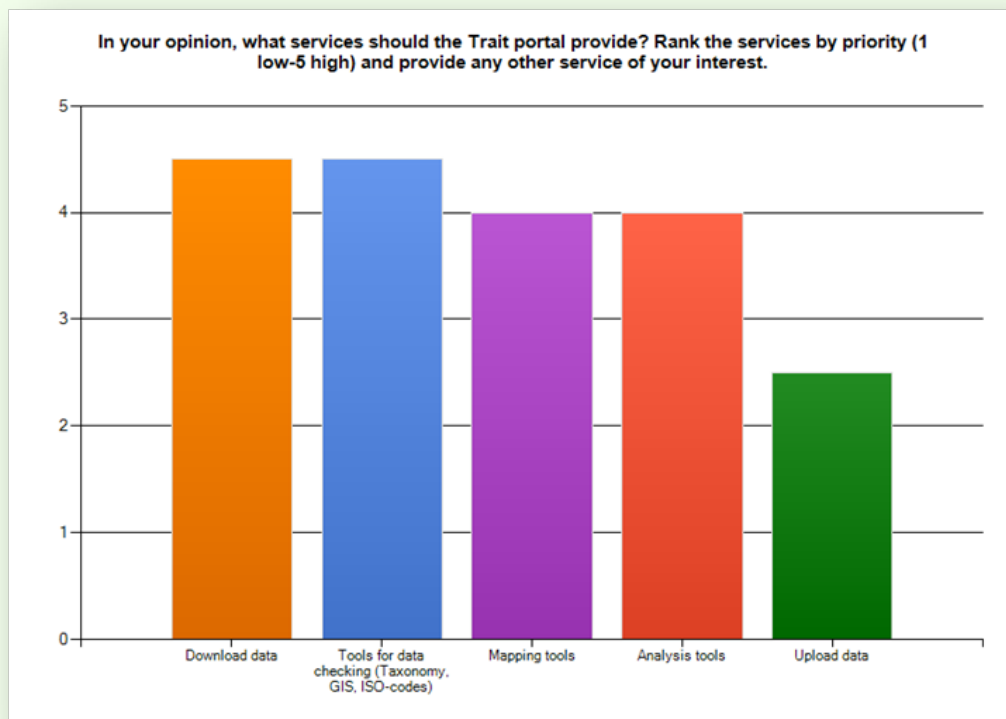
Key facilitators in introducing the TIP concept were the possibility to: (i) present the TIP at the project events; (ii) discuss and gather feedback and partners’ views; (iii) gather requirements and wish lists—the highlight of this process being primarily an external consultation with the breeders’ community. This consultation was conducted through a user requirements survey and the results were presented at the workshop in Palanga, Lithuania. The main survey questions were asked in order to: (i) understand what breeders were looking for; (ii) understand how they look for information; and (iii) provide an

opportunity to gather their expectations for this tool. Five priority services were identified through the breeders’ survey (Fig. 1), among which, the top priority was identified as the capacity to download data; secondly the availability of tools such as taxonomy, geo-references and any codes used for data checking, followed by the need for a mapping and analysis tool and, lastly the capacity to upload data (to send fresh data or update data in the information system).

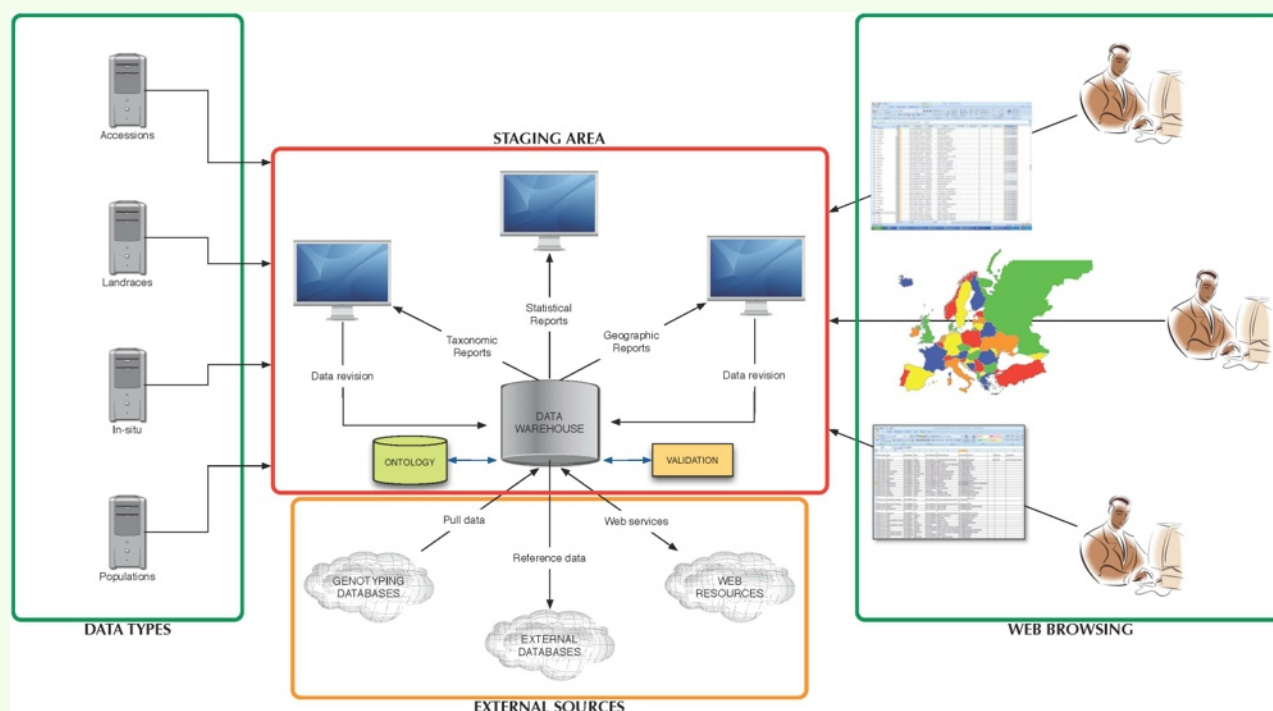
In this development process many questions were raised in order to plan the product for long-term sustainability beyond the end of the project phase. Some of the questions were: 1) Who will host the TIP? 2) How do you see the links—are they just re-directing the users to other sources or are they provid-

**“The way forward is to use a  
‘Triontology’—CWR, LR and  
Crop-Trait Ontologies”**

ing direct access to further information? 3) Is the proposed concept what you are looking for? 4) What features besides the ones indicated would be needed (taking into account the data types provided)? 5) Which data types will be downloadable and in which formats? 6) What data sources are missing? 7) What kind of access to analytical tools is needed? In addition to the survey, feedback from the PGR Secure project partners (with whom dependencies exist) was sought in order to have a better understanding of data types to be used, features envisaged, type of links, ontology groups established and feedback and guidance on the TIP to the development team (Bioversity International).



**Figure 1** Results of the breeders’ survey on what services the TIP should provide and priority services of interest to breeders



**Figure 2** TIP platform architecture

With all the information in hand, at the first annual PGR Secure consortium meeting held in Perugia, Italy in December 2011, the TIP framework concept was presented and confirmed by the consortium. The TIP framework is now in its second phase which consists of reproducing the TIP framework presented.

#### TIP concept framework description

The TIP will have a simple platform architecture accommodating input and output data types, as follows and as illustrated in Figure 2. The system will:

1. Use a document store database system;
2. Have an upload system with flexible template driven options for data being sent by providers;
3. Include and use the Generation Challenge Programme (GCP) data annotation and trait ontology curation tools developed by the Bioversity team;
4. Be searchable through ontology-driven views;
5. Include information on traits, locations, trial sites, georeferences, geographical information;
6. Use web scraping (gather related information/data) to include external data sources, molecular data, bibliography, characterization and evaluation data, images, etc.;
7. Link with external information sources;
8. Provide data analysis outputs.

Additionally, the TIP will include three different entry points (trait information, CWR and LR inventories), allowing users to choose their entry/access point to the information they require, while maintaining the capacity to link or tap into existing online sources of information such as GENESYS, EURISCO and ECCDBs.

This concept has been planned to create a system that primarily serves the data provider so that it can efficiently serve the users. To make the most of this idea the rationale for the TIP framework conceptualization was to use existing developments and resources, focusing the development team's efforts towards using and further enhancing existing and evolving resources being developed in other communities of practice. This is the spirit which has driven the development and evolution of the TIP during the first year of the PGR Secure project.

We expect to have the TIP mockup ready by 2013, after which the testing phase using project data will be initiated. More information is available on the PGR Secure project website ([www.pgrsecure.org](http://www.pgrsecure.org)).

The Trait Information Portal development team at Bioversity International is: Sónia Dias, Milko Skofic, Luca Matteis and Elizabeth Arnaud.

# Where and how? Genetic reserve site selection and development of common quality standards

J.M. Iriondo<sup>1</sup>, M. Parra-Quijano<sup>2</sup>, C. Lara-Romero<sup>1</sup>, F. Carreño<sup>1</sup>, N. Maxted<sup>3</sup>, S. Kell<sup>3</sup> and B.V. Ford-Lloyd<sup>3</sup>

<sup>1</sup> Area de Biodiversidad y Conservación, Universidad Rey Juan Carlos, Móstoles, Madrid, Spain. Email: [jose.iriondo@urjc.es](mailto:jose.iriondo@urjc.es)

<sup>2</sup> Depto. Biología Vegetal, Universidad Politécnica de Madrid, Madrid, Spain. Email: [mauricio.parra@agrobiodiversidad.org](mailto:mauricio.parra@agrobiodiversidad.org)

<sup>3</sup> School of Biosciences, University of Birmingham, Edgbaston, Birmingham, B15 2TT, UK. Email: [s.kell@bham.ac.uk](mailto:s.kell@bham.ac.uk)

Crop wild relatives (CWR) are a key component for food production and security and for the maintenance of agro-ecosystems. CWR are successfully being used in plant breeding, but at the same time, many of them are becoming increasingly threatened. The establishment of genetic reserves is one of the approaches for CWR conservation that has been assessed in Europe under the AEGRO project (AGRI GENRES 057 'An Integrated European In Situ Management Work Plan: Implementing Genetic Reserves and On Farm Concepts' <http://aegro.jki.bund.de/aegro/>) co-funded by the European Commission, DG AGRI within the framework of council regulation 870/2004.

Such reserves can be more effective if they are established in protected areas where conditions are more amenable for conserving CWR populations. Furthermore, genetic reserves should preferentially be located in sites that meet certain requirements such as: 1) the effective presence of the greatest number of populations of the focal threatened or, otherwise, priority CWR species, 2) maximum legal protection of the area, and 3) maximum coverage of populations of other non-focal CWR species.

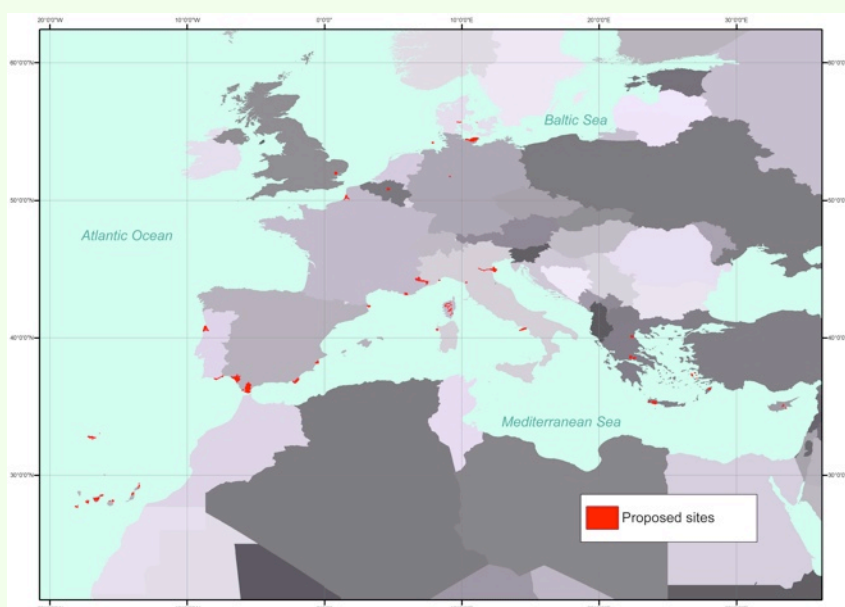
At a different scale, it is also advisable that the resulting network of genetic reserves allows for the conservation of the maximum genetic and ecogeographic diversity of the focal CWR species. Taking into account these considerations, and as part of the AEGRO project, a methodology was developed to select potential sites for the establishment of genetic

reserves using GIS tools, ecogeographical maps and species distribution models. Thus, a GIS platform was established for the European territory that included the following layers: a) geographic coordinates of CWR species of the four AEGRO model genera (*Avena*, *Brassica*, *Beta* and *Prunus*), b) polygons of the sites that comprise the Natura 2000 Network, c) ecogeographical characterization maps of the territory elaborated *ad hoc* (in the case of *Beta*) and d) species potential distribution models.

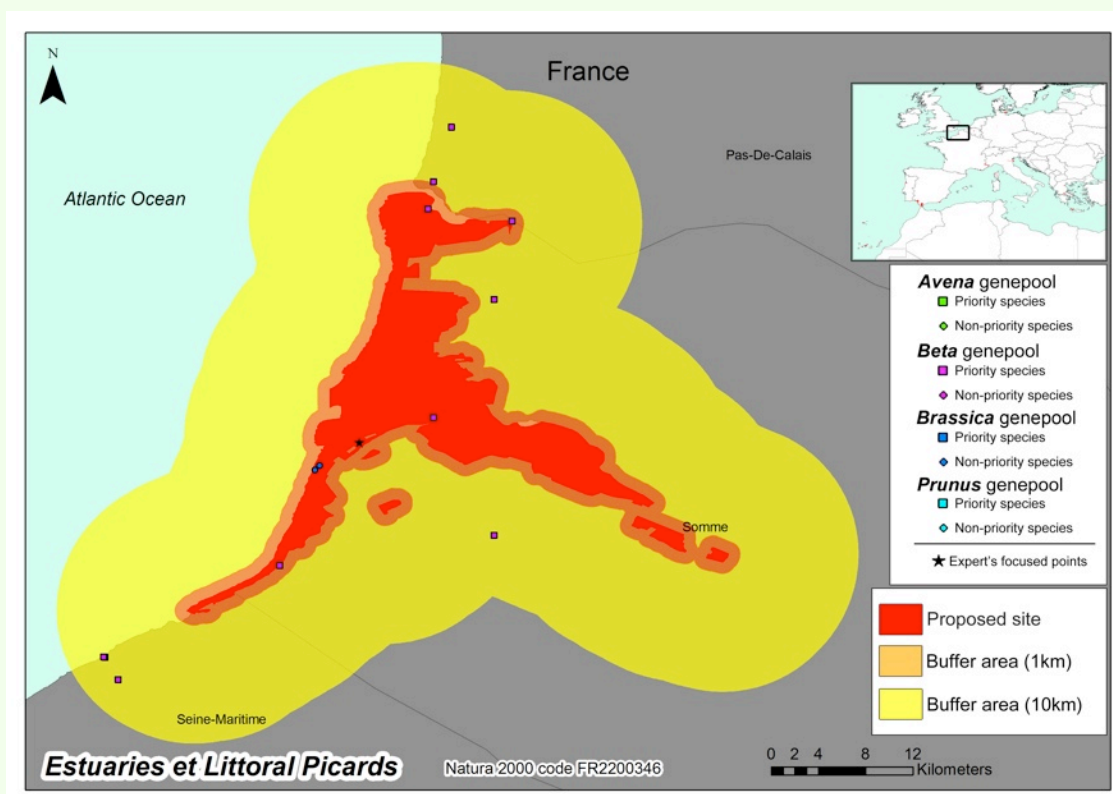
From all this information and the creation of additional maps of occurrence, density and species richness, a stepwise strategy for the selection of genetic reserves was formulated. Areas were selected that had a high presence of focal prioritized CWR species, a high probability of occurrence of other prioritized and non-prioritized CWR that are included in sites of community interest (SCI), and that were most representative of the overall ecogeographical range of the focal CWR species. This procedure sets the stage for a subsequent evaluation of the ability of selected genetic reserves to conserve populations of focal species and to provide an umbrella for the indirect conservation of a significant amount of populations of other non-target CWR species.

This stepwise strategy to select potential genetic reserves for CWR was described in detail by Parra-Quijano *et al.* (2012). In a final step, scientists and experts for each model crop gene pool determined whether the list of protected areas selected as potential genetic reserves was the most appropriate or whether it needed the addition or deletion of some areas. The final selection of protected areas as potential genetic reserves for the four model crop gene pools considered in AEGRO project is shown in Figure 1 and published on the web as the Genetic Reserves Information System (GenResIS) ([www.agrobiodiversidad.org/aegro/](http://www.agrobiodiversidad.org/aegro/)). On this website, the plant genetic resources community can find not only the list of the potential genetic reserves, but also detailed information about ecogeographical characterization of each protected area considered as a potential genetic reserve, the umbrella effect on other non-focal species and interactive and downloadable maps (Fig. 2).

A second deliverable of the AEGRO project was the formulation of quality standards for genetic reserve conservation of CWR. The generation of this set of quality standards is meant to ensure that conservation efforts are carried out following



**Figure 1** Sites selected as potential locations for the establishment of genetic reserves as part of the AEGRO project



**Figure 2** Estuaires et Littoral Picards, a Natura 2000 site chosen for the establishment of a genetic reserve of *Beta vulgaris* subsp. *maritima* showing the boundaries of the Natura 2000 site and a 1 km and a 10 km buffer. It also presents information on populations of other species of the four priority AEGRO crop gene pools that lie within the area.

the most logical and efficient procedures and that positively contribute to achieving the objectives. The adoption of standards of good practices relies on the assumption that the projects or programmes that are executed this way will be more rigorous in the process of decision-making and more efficient in both the use of resources and in pursuing the objective of conservation. The main objectives that are pursued with the development of a set of quality standards are therefore to increase the efficiency and quality of genetic reserve conservation of CWR. Furthermore, they provide a useful tool for conservation managers to plan, execute and evaluate conservation actions for CWR according to protocols that are considered as 'good practice'.

In the formulation of standards we set two levels; namely, 'minimum' and 'optimal' quality standards. 'Minimum' quality standards assemble the baseline traits that should be required of any genetic reserve established in a protected area to operate and fulfill its conservation objectives. In contrast, 'optimal' quality standards gather a more stringent set of traits that should be aimed at for genetic reserves in the longer term. The quality standards include a set of specifications and requirements that are meant to be applied to the protected areas selected for the establishment of genetic reserves and to the genetic reserves themselves. These specifications and requirements concern location, spatial structure, target species, populations and management.

The elaboration of the final set of standards followed a step-wise process to involve the participation of interested researchers and practitioners in the CWR conservation community. Thus, a draft text for the set of quality standards was pro-

duced after analysing the need for implementing effective *in situ* conservation procedures in CWR and reviewing existing literature on genetic reserves and natural protected areas. There were several rounds of reviews and improvements of the draft involving contributions from AEGRO project partners. The result of this first consultation process was disseminated to the CWR conservation community through the presentation of an oral communication at the international Symposium 'Towards the establishment of genetic reserves for crop wild relatives and landraces in Europe' held in Funchal, Portugal, in September 2010 ([www3.uma.pt/cem/aegro/ecpgr/symp/index.html](http://www3.uma.pt/cem/aegro/ecpgr/symp/index.html)). In parallel to this process a website was designed and established at: <https://sites.google.com/site/qualitystandardsforcwrs/home>. The consultation process on the quality standards was complemented with the distribution of the draft to specialized mailing lists of the plant genetic resources and protected areas communities and the Europarc Federation. Furthermore, the managers of the protected areas selected as potential sites for the establishment of genetic reserves for priority species of *Avena*, *Beta*, *Brassica* and *Prunus* formulated in the AEGRO project were contacted concerning this issue to gather additional feedback. Relevant feedback was used to produce a final version of the quality standards (Iriondo *et al.*, 2012). This final version is also available at: <https://sites.google.com/site/qualitystandardsforcwrs/home>.

This set of quality standards developed by the AEGRO project provides a useful tool to increase the efficiency of genetic reserve conservation of CWR. The quality standards may be used by protected area managers interested in conserving priority CWR in their protected areas (bottom-up approach), but also they may be employed in the last stage of the process



of selecting the locations for genetic reserves when multiple alternatives exist according to main selection criteria (top-down approach). A necessary next step is to test the applicability of this set of quality standards on existing initiatives for the establishment of genetic reserves. In this sense, it would be a helpful exercise to test their applicability on the candidate locations

“a set of quality standards developed by the AEGRO project provides a useful tool to increase the efficiency of genetic reserve conservation of CWR”

identified for the establishment of genetic reserves for priority species of *Avena*, *Beta*, *Brassica* and *Prunus* formulated in the AEGRO project.

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## Development of a report and monitoring system for the *in situ* conservation of genetic resources of crop wild relatives in Brandenburg, Germany

R. Vögel

Brandenburg State Office for Environment, Health and Consumers Protection (LUGV), Trammer Chaussee 2, Eberswalde Brandenburg 16225, Germany. Email: [Rudolf.Voegel@LUGV.Brandenburg.de](mailto:Rudolf.Voegel@LUGV.Brandenburg.de)

#### Aims of the project

The aim of the project was to support the conservation and sustainable use of *in situ* plant genetic resources (PGR) in Brandenburg, Germany, by transparent documentation and information, particularly with regard to crop wild relatives (CWR), as well as by listing historic and recent cultivars of crops. This aim was to be achieved by exemplary development of a report and monitoring system for *in situ* PGR.

#### Project partners

Project partners are the Brandenburg State Office for Environment, Health and Consumers Protection (LUGV Brandenburg), the Eberswalde Forestry Research Institute of the State of Brandenburg (LFE) and the University of Applied Sciences Eberswalde (HNE).

#### Objectives achieved

- Creation of a list of relevant CWR in Brandenburg; selection according to taxonomy, use, and threat status; classification by habitat types; identification of need for action.
- Establishing a concept for an exemplary database for *in situ* documentation in Brandenburg.
- Development of an IT-supported procedure for the consolidation of all data from Brandenburg CWR and on-farm data sources to promote conservation and sustainable use of existing resources.
- Exemplary realisation of statistical analysis and data aggregation for reporting at national and regional levels.

#### Contact:

Project coordinator: Rudolf Vögel, LUGV [[Rudolf.Voegel@LUGV.Brandenburg.de](mailto:Rudolf.Voegel@LUGV.Brandenburg.de)]

The project was sponsored by the Federal Agency for Agriculture and Food (BLE) and was of three years duration, 2007–2010.

#### For further information:

[www.mugv.brandenburg.de/cms/detail.php/bb1.c.215088.de](http://www.mugv.brandenburg.de/cms/detail.php/bb1.c.215088.de)

[www.isip.de/coremedia/generator/isip/Start,nodeId=15413.html](http://www.isip.de/coremedia/generator/isip/Start,nodeId=15413.html)

[www.hnee.de/Wald-und-Umwelt/Mitarbeiter/Professoren/-innen/Pierre-Ibisch/Research-projects/Current-research-projects/Projekt-Erhalt-pflanzengenetischer-Ressourcen-CWR/Projekt-zur-Erhaltung-pflanzengenetischer-Ressourcen-E4144.htm](http://www.hnee.de/Wald-und-Umwelt/Mitarbeiter/Professoren/-innen/Pierre-Ibisch/Research-projects/Current-research-projects/Projekt-Erhalt-pflanzengenetischer-Ressourcen-CWR/Projekt-zur-Erhaltung-pflanzengenetischer-Ressourcen-E4144.htm)

# Collecting genetic resources of crop wild relatives in Crimea, Ukraine, in 2009

A. Diederichsen<sup>1</sup>, R.V. Rozhkov<sup>2</sup>, V.V. Korzhenevsky<sup>3</sup> and R.L. Boguslavsky<sup>2</sup>

<sup>1</sup> Plant Gene Resources of Canada, Agriculture and Agri-Food Canada, Saskatoon, Saskatchewan, Canada. Email:

[Axel.Diederichsen@AGR.GC.CA](mailto:Axel.Diederichsen@AGR.GC.CA)

<sup>2</sup> National Centre of Plant Genetic Resources of Ukraine, Ukrainian Academy of Agricultural Sciences, Kharkiv, Ukraine. Email:

[ncpgru@gmail.com](mailto:ncpgru@gmail.com)

<sup>3</sup> Nikita Botanical Garden, National Scientific Centre, Yalta, Crimea, Ukraine.

*Very diverse ecogeographic zones and floral elements typical for the Mediterranean, Near East, European and Caucasian regions occur in Crimea. Due to rapid infrastructure developments during recent years, several natural habitats are endangered. Some endemic Crimean wild relatives of crop plants are rare species. This includes the three wild species of the genus *Lens* Mill., which were the focus of a joint Ukrainian-Canadian collecting mission in 2009. During this expedition, 127 seed samples of 55 genera were collected. Of these, 43 samples are wild relatives of crop plants and 54 samples have potential as forage grasses or legumes. The remaining seed samples included potential medicinal and ornamental species. The collected germplasm will be preserved by the national genebanks of Ukraine (National Centre of Plant Genetic Resources of Ukraine, NCPGRU, Kharkiv) and Canada (Plant Gene Resources of Canada, PGRC, Saskatoon), and made available to researchers and breeders. The lentil material will be assessed for its resistance to the diseases anthracnose and ascochyta blight.*

Crimea has attracted the attention of botanists and geographers for many years because of the Mediterranean climate on the Crimean south coast, which is facilitated by the climatic barrier of the Crimean Mountains that stretch from the southwest to northeast (Regel, 1943). The northern part of Crimea is lowland that has a continental climate comparable to many parts of Eastern Europe and the Ukrainian mainland. The Crimean Mountains consist of three ranges that run parallel to the southeastern coast of Crimea, protecting the southeastern coastline facing the Black Sea from continental cold and allowing for a Mediterranean to nearly subtropical climate on a strip of land along the coast. The tallest mountain range close to the coast is of Jurassic origin and sometimes referred to as the Yayla. It reaches an altitude of 1545 m at peak Roman Kosh and has several other peaks higher than 1200 m. The Yayla drops steeply towards the coast of the Black Sea.

Extremely different eco-climatic zones with diverse floral elements typical for the Mediterranean, Near East, European and Caucasian regions can be found in close proximity. Walter (1943) distinguished the following three major landscape types of Crimea: (1) the steppe, ranging from semi-desert to grassland north of the Crimean Mountains and including most of the Kerch Peninsula; (2) the Crimean Mountains, with oak forests, beech forests and nearly subalpine meadows resembling temperate to oceanic European landscapes; and (3) the southeastern coast with Mediterranean climate and flora.

The steppe zone is used as grassland or for agricultural production with emphasis on cereal production. Grazing of the Crimean Mountain meadows by sheep and cattle has declined during recent years. On the southern slopes of the Yayla, vineyards that have been recently re-established can be seen, while other agricultural or horticultural activities are not of sig-

nificance and seem restricted to occasional home gardens. The most important industry in southern Crimea is tourism and a lot of recent development of related infrastructure can be seen. Housing construction is very active. Many buildings of the bungalow style, consisting of crude walls with simple tin roofs and not ready to be inhabited have been erected on former meadows or other open spaces during recent years, obviously bypassing any planning regulations of state authorities. This rapid development has put natural habitats at risk and reduced the agricultural land base.

As early as 1812, the Nikita Botanical Garden was established in close proximity to the city of Yalta. The flora of Crimea is well-documented in the herbarium of the Nikita Botanical Garden as well as in the scientific literature. The Crimean botanist E.V. Vul'f (1885–1941) made outstanding contributions that scientifically connected botany to the sphere of plant genetic resources. He worked at the Nikita Botanical Garden and systematically compiled the flora of Crimea which was published in three volumes between 1927 and 1969. In 1926, upon invitation by N.I. Vavilov, Vul'f moved from Crimea to Leningrad (now St. Petersburg) to lead the volatile oil plants section and later on the herbarium and geography section at the All-Union Research Institute of Plant Industry, which is now the Russian national genebank named after N.I. Vavilov (Agajev, 1994).

A recent inventory of the Crimean flora by Golubev (1996) lists 2775 species, of which 2560 are endemic. Of these, 904 species were classified as being rare, very rare, disappearing or critically threatened, indicating the need for close monitoring to prevent loss of diversity. This inventory also provides information about biological features and potential economic uses of each species, highlighting the richness of endemic species that have potential as forages, as sources of volatile oil and as medicinal plants.



**Figure 1** Map of Crimea indicating the collecting sites. Sites in close proximity are only represented by a single dot (Underlying map from Microsoft Encarta 2007)

### Rationale for collecting

The motivation for this collection mission was the interest in genetically diverse material from wild relatives of cultivated lentil (*Lens culinaris* L.). Three wild lentil species (*L. ervoides* [Brign.] Grande, *L. nigricans* [Bieb.] Webb et Berth., and *L. orientalis* [Boiss.] Schmalh.) are very rare endemics of Crimea (Barulina, 1930; Golubev, 1996). Due to their close relationship to the cultivated lentil, they are sometimes considered as subspecies of *L. culinaris* (Cubero, 1984). *Lens ervoides* has been reported by pathologists to be a source of resistance to anthracnose disease in Canada (Tullu *et al.*, 2006). Access to this material may contribute to the sustainability of lentil production in Canada and other countries where production is affected by this disease.

Information from specimen labels of the State Nikita Botanical Gardens Herbarium (YALT) was used to determine locations of wild lentil species. Germplasm of other crop wild relatives (CWR) or wild plants that may have potential for cultivation and utilization was also collected. Recent collecting missions conducted in Crimea as collaborations among Ukrainian institutions and the United States Department of Agriculture (USDA) focused on CWR and potential forage legumes with emphasis on the family Poaceae Barnhart (Bockelman, 1999) and the genus *Medicago* L., respectively (Greene, 2008). These missions resulted in 500 accessions deposited in the NCPGRU as well as the USDA National Plant Germplasm System, and include two accessions of wild lentil species.

The present collecting mission was conducted from June 26 to July 7, 2009, as a joint project between the national genebanks of Ukraine (NCPGRU) and Canada (PGRC). Local expertise for Crimea was provided by botanists from the State Nikita Botanical Garden, Yalta. A driver with local experience from the Nikita Botanical Garden was part of the collection team. Germplasm was collected from 57 locations mostly along the mountainous southern coastline of Crimea, including the Mys Martjan nature reserve close to the village of Nikita, and the Aju-Dag archaeological park (Fig. 1). On the Kerch Peninsula, collections were made in the Opuk and Kazantip nature reserves on the coasts of the Black Sea and Sea of Azov, respectively.

### Collected plant material

A total of 127 accessions covering 16 families, 55 genera and at least 66 species were collected (Table 1). In 35 instances, mostly with grasses and forage legumes, the mature plants could only be identified to the genus level; species identification will be conducted during the first genebank regeneration. Poaceae (41%), Fabaceae (36%) and Brassicaceae (9%) dominated the collected material, as the remaining 13 families were only represented by one or two species each.

About 34% of the collected accessions are wild relatives of field crops. The populations of wild lentils occurred on dry rubble in sparse stands of native oak (*Quercus pubescens* Willd.) and juniper (*Juniperus excelsa* Bieb.). Spring 2009 was dry and by the end of June most wild lentil plants had shattered their seeds so they had to be picked from the ground (Fig. 2). Eight accessions of *L. ervoides* and *L. nigricans* were found at different locations. These accessions will be further investigated regarding their resistance to lentil anthracnose disease.

Wild relatives of the major cereals wheat, barley, oat and rye were found. With the exception of wild einkorn, *Triticum baеoticum* Boiss., the cereal wild relatives found during this mission were not classified as being rare in Crimea (Golubev, 1996). The first report of *T. baеoticum* is by M. Bieberstein from 1809. After 1967 it was missing according to Dorofeev *et al.* (1979); however, Bockelman (1999) reported collecting this species in the region. The reduction of agricultural intensity after 1990 in southern Crimea may have allowed for cryptic populations of *T. baеoticum* to become larger in recent years. Among the seven accessions of wild einkorn, forms with white, red and black awns occurred together in mixed populations of considerable size (Fig. 3). Currently, there are very few locations with occurrence of this species reported for Crimea. The typical habitats were abandoned fields or field margins. Cultivation of the closely related cultivated einkorn, *T. monococcum* L., does not presently occur in Crimea but was part of agriculture practised by the native Tartars that were expelled from Crimea in 1944 (Dorofeev *et al.*, 1979; Bagrov and Rudenko, 2004). A search for *Aegilops tauschii* Coss. on the coast of the Sea of Azov on the Kerch Peninsula was not successful, although its occurrence at the location was documented in 1982 by a herbarium specimen collected at that time. A wild relative of rye, *Secale sylvestre* Host, was found on dry grassland close to the Sea of



**Figure 2** Mature *Lens ervoides* (Brign.) Grande in native habitat (left) with seeds mostly shattered (right)

Table 1 Germplasm collected in Crimea (Ukraine) in 2009

Usage group	Genus	Species	Authority	Family	Conservation status <sup>1</sup>	No. Accessions	
Crop	<i>Coriandrum</i>	<i>sativum</i>	L.	Apiaceae	n.a.	1	
	<i>Cuminum</i>	<i>cyminum</i>	L.	Apiaceae	n.a.	1	
	<i>Sinapis</i>	<i>alba</i>	L.	Brassicaceae	n.a.	1	
	<i>Cicer</i>	<i>arietinum</i>	L.	Fabaceae	n.a.	1	
	<i>Phaseolus</i>	<i>vulgaris</i>	L.	Fabaceae	n.a.	1	
	<i>Hordeum</i>	<i>vulgare</i>	L.	Poaceae	n.a.	1	
	<i>Triticum</i>	<i>aestivum</i>	L.	Poaceae	n.a.	1	
	Crop wild relative - Brassicaceae	<i>Brassica</i>	<i>incana</i>	Ten.	Brassicaceae	1	1
		<i>Camelina</i>	<i>microcarpa</i>	Andrz.	Brassicaceae	11	1
		<i>Crambe</i>	<i>koktebelica</i>	(Junge) N. Busch	Brassicaceae	9	1
<i>Crambe</i>		<i>pinnatifida</i>	R. Br.	Brassicaceae	11	1	
<i>Crambe</i>		<i>pontica</i>	Stev. ex Rupr.	Brassicaceae	11	1	
Crop wild relative - cereal - barley	<i>Hordeum</i>	<i>bulbosum</i>	L.	Poaceae	13	3	
	<i>Hordeum</i>	<i>murinum</i>	L.	Poaceae	13	1	
Crop wild relative - cereal - oat	<i>Avena</i>	<i>fatua</i>	L.	Poaceae	12	1	
	<i>Avena</i>	<i>ludoviciana</i>	Durieu	Poaceae	13	8	
Crop wild relative - cereal - rye	<i>Secale</i>	<i>sylvestre</i>	Host	Poaceae	12	1	
Crop wild relative - cereal - wheat	<i>Aegilops</i>	<i>biuncialis</i>	Vis.	Poaceae	14	1	
	<i>Aegilops</i>	<i>cyllindrica</i>	Host	Poaceae	14	2	
Crop wild relative - flax	<i>Aegilops</i>	<i>triuncialis</i>	L.	Poaceae	14	1	
	<i>Dasyphyrum</i>	<i>villosum</i>	(L.) Borb.	Poaceae	13	3	
	<i>Triticum</i>	<i>baeoticum</i>	Boiss.	Poaceae	3	7	
	<i>Linum</i>	<i>austriacum</i>	L.	Linaceae	13	2	
	Crop wild relative - lentil	<i>Lens</i>	<i>ervoides</i>	(Brign.) Grande	Fabaceae	11	2
		<i>Lens</i>	<i>nigricans</i>	(Bieb.) Webb et Berth.	Fabaceae	11	3
	Grass	<i>Lens</i>	sp.	Mill.	Fabaceae	n.a.	3
		<i>Agropyron</i>	<i>dasyanthum</i>	Ledeb.	Poaceae	11	2
		<i>Agropyron</i>	<i>desertorum</i>	(Fisch. ex Link) Schult.	Poaceae	6	1
		<i>Agropyron</i>	<i>pectinatum</i>	(Bieb.) Beauv.	Poaceae	14	1
<i>Agropyron</i>		sp.	Gaertn.	Poaceae	n.a.	2	
<i>Arenatherum</i>		<i>elatius</i>	(L.) J. et C. Presl	Poaceae	12	1	
<i>Bromus</i>		<i>secalinus</i>	L.	Poaceae	9	1	
<i>Bromus</i>		<i>squarrosus</i>	L.	Poaceae	14	1	
<i>Bromus</i>		sp.	L.	Poaceae	n.a.	3	
<i>Cynosurus</i>		<i>echinatus</i>	L.	Poaceae	12	1	
<i>Dactylis</i>		<i>glomerata</i>	L.	Poaceae	14	2	
<i>Festuca</i>		sp.	L.	Poaceae	n.a.	1	
<i>Koeleria</i>		<i>cristata</i>	(L.) Pers.	Poaceae	14	1	
<i>Leymus</i>		<i>racemosus</i>	(Bieb.) Tzvel.	Poaceae	13	1	
<i>Lolium</i>		<i>austriacum</i>	L.	Poaceae	14	1	
<i>Melica</i>		sp.	L.	Poaceae	n.a.	1	
<i>Puccinellia</i>		<i>distans</i>	(Jacq.) Parl.	Poaceae	12	1	
<i>Stipa</i>		sp.	L.	Poaceae	n.a.	1	
<i>Taeniatherum</i>		<i>asperum</i>	(Simonk.) Nevski	Poaceae	14	1	
Forage legume		<i>Astragalus</i>	sp.	L.	Fabaceae	n.a.	2
	<i>Coronilla</i>	<i>scorpioides</i>	(L.) Koch	Fabaceae	13	1	
	<i>Coronilla</i>	<i>varia</i>	L.	Fabaceae	14	1	
	<i>Lathyrus</i>	<i>aphaca</i>	L.	Fabaceae	14	1	
	<i>Lathyrus</i>	<i>pratensis</i>	L.	Fabaceae	13	1	
	<i>Lathyrus</i>	sp.	L.	Fabaceae	n.a.	3	
	<i>Medicago</i>	<i>arabica</i>	(L.) Huds.	Fabaceae	13	1	
	<i>Medicago</i>	<i>denticulata</i>	Willd.	Fabaceae	12	1	
	<i>Medicago</i>	<i>lupulina</i>	L.	Fabaceae	14	1	
	<i>Medicago</i>	<i>minima</i>	(L.) Bartalini	Fabaceae	14	1	
	<i>Medicago</i>	<i>orbicularis</i>	(L.) Bartalini	Fabaceae	14	1	
	<i>Trifolium</i>	<i>pratense</i>	L.	Fabaceae	14	1	
	<i>Trifolium</i>	sp.	L.	Fabaceae	n.a.	5	
	<i>Trigonella</i>	<i>monspeliaca</i>	L.	Fabaceae	14	1	
	<i>Vicia</i>	<i>dalmatica</i>	A. Kerner	Fabaceae	12	2	
	<i>Vicia</i>	<i>ervilia</i>	(L.) Willd.	Fabaceae	1	1	
	<i>Vicia</i>	sp.	L.	Fabaceae	n.a.	7	
	Medicinal	<i>Echballium</i>	<i>elaterium</i>	(L.) A. Rich.	Cucurbitaceae	11	1
<i>Lepidium</i>		<i>perfoliatum</i>	L.	Brassicaceae	13	1	
<i>Nigella</i>		<i>damascena</i>	L.	Ranunculaceae	13	1	
<i>Plantago</i>		<i>lanceolata</i>	L.	Plantaginaceae	14	1	
<i>Rumex</i>		<i>euxinus</i>	Klok.	Polygonaceae	13	2	
<i>Securigera</i>		<i>securidaca</i>	(L.) Degen et Doerfl.	Fabaceae	13	1	
<i>Verbascum</i>		sp.	L.	Scrophulariaceae	n.a.	1	
<i>Ziziphora</i>		<i>tenuior</i>	L.	Lamiaceae	12	1	
Ornamental		<i>Allium</i>	<i>rotundum</i>	L.	Alliaceae	14	1
		<i>Bellevalvia</i>	<i>sarmatica</i>	(Pall. ex Georgi) Woronow	Hyacinthaceae	6	1
	<i>Erodium</i>	sp.	L'Hér	Geraniaceae	n.a.	1	
	<i>Hedysarum</i>	<i>tauricum</i>	Pall. ex Willd.	Fabaceae	13	1	
	<i>Hedysarum</i>	<i>candidum</i>	Bieb.	Fabaceae	13	1	
	<i>Iris</i>	<i>pumila</i>	L.	Iridaceae	13	1	
	<i>Matthiola</i>	<i>odoratissima</i>	(Bieb.) R. Br.	Brassicaceae	9	1	
	<i>Salvia</i>	<i>pratensis</i>	L.	Lamiaceae	13	1	
	<i>Tulipa</i>	<i>schrenkii</i>	Regel	Liliaceae	11	1	
	<i>Tulipa</i>	sp.	L.	Liliaceae	n.a.	1	
Wild	<i>Lepidium</i>	<i>crassifolium</i>	Waldst. et Kit.	Brassicaceae	11	1	
	Unknown			Brassicaceae	n.a.	3	

<sup>1</sup> Conservation status according to Golubev (1996): 1=occurs only at one location; 3=occurs at 6-10 locations; 6=very rare; 9=rare; 11=quite rare; 12=scarce; 13=quite common; 14=common.

Azov coast, and wild relative species of barley, *Hordeum* L., were frequent.

The hexaploid wild oat species *Avena sterilis* L. was common in dry, open and slightly disturbed locations. *A. fatua* L. was found as a weed in a bread wheat (*Triticum aestivum* L.) field. The tetraploid *A. barbata* Pott ex Link and the diploid *A. eriantha* Durieu, both documented in the Nikita Botanical Garden Herbarium, were not found at the locations expected.

The perennial wild cabbage *Brassica incana* Ten. (synonyms: *B. sylvestris* [L.] Mill. subsp. *taurica* Tsvet. and *B. oleracea* L. subsp. *incana* (Ten.) Gladis et Hammer) was collected from the Black Sea coast at Cape Mys Aju-Dag, which is difficult to access from land (Fig. 4). A healthy population of this rare species exists there and represents the most eastern occurrence of the disjunctive distribution range of the wild species of the genus *Brassica* in the Mediterranean–Atlantic area of Europe and the Near East (Lizgunova, 1984; Snogerup *et al.*, 1990). This species belongs to the primary gene pool of cultivated cabbage (*B. oleracea* L.) and to the secondary gene pool of oilseed rape (*B. napus* L.) (Gladis and Hammer, 2001; Diederichsen and McVetty, 2011). Interestingly, the mature and dry siliques of *B. incana* were indehiscent (i.e., they did not shatter seeds at all). Non-shattering of seeds is part of the so-called 'domestication syndrome' (Hammer, 1984) and *B. incana* could be a useful source of genetic diversity for improving seed retention in oilseed crops. Possibly, this population escaped from historic cultivation of *Brassica oleracea* at this location and adapted to survival in the wild. Christensen *et al.* (2011) reported such adaptation of an escape from cultivation for a leafy kale type (*B. oleracea* convar. *acephala* (DC.) Alef.) in Denmark. Cape Mys Aju-Dag may become more influenced by tourists due to its easy accessibility by boat from the seaside; such excursions seem quite popular and the location is close to the tourist centres Yalta and Alushta. At the same location, plants of *Pisum elatius* Bieb. were found, but the seeds were all shattered and could not be collected. It has been suggested that *P. elatius* Bieb. may be an escape from medieval gardens of a monastery that existed on Mys Aju-Dag and naturalized in the wild (Korzhenevsky, personal communication).

A remarkable richness of the genus *Linum* L. was noticed during the field collections and Golubev (1996) reports 16 wild species of this genus for Crimea. However, *L. bienne* Mill., the



**Figure 3** Black, red and white glumes and awns of *Triticum baeticum* from a population at one location close to Feodosia



**Figure 4** Fruiting perennial *Brassica incana* Ten. at Cape Mys Aju-Dag; note the indehiscent siliques

wild progenitor of cultivated flax, is not reported for Crimea. Seed material of *Linum* species, belonging to the tertiary gene pool of flax was only collected from *Linum austriacum* L., while the other species found were in full flower when encountered (Fig. 5). The *Linum* species are cultivated as ornamentals but not crops in the agronomic sense. The wild habitats of the Crimean Mountains, the southern coast and the Kerch Peninsula are particularly rich in grassland plants and many leguminous species (Fabaceae) that have economic potential as forages. Features such as cold and drought tolerance may be found in this germplasm. Nine accessions collected have potential as medicinal plants (Table 1).

Seven seed samples of crop germplasm were either bought at local markets (coriander, cumin, chickpea, garden bean) or collected from ruderal sites (barley, wheat, yellow mustard). Compared with other regions of Ukraine, home gardening and small-scale agriculture were rare in the area covered by this expedition and seed-saving of local material may also be rare. An exception may be a local type of red skinned and flattened *Allium cepa* L., which was offered frequently by street vendors in the region around Yalta. Based on this observation, it seems most likely that landraces of field or garden crops have vanished from Crimea, although the Tartars who have been returning to the Crimea since the 1990s may bring some landrace material back from western Siberia or Central Asia.

The Kerch Peninsula has a continental climate and very low precipitation compared with the southern coast of Crimea. The nature reserves Kazantip and Opuk on the Kerch Peninsula showed a wide diversity of plants that deserve attention as ornamental plants for harsh continental climates in western Canada, where the Canadian national genebank is located. Wild relatives of oilseed crops of the genera *Camelina* Crantz and *Crambe* L. were frequent at the locations visited on the Kerch Peninsula. Both the Kazantip and Opuk nature reserves have been assigned the highest priority regarding the preservation of Crimean biological diversity (Korzhenevsky and Sa-



**Figure 5** Left to right: *Linum tenuifolium* L., *L. corymbulosum* Reichenb. and *L. austriacum* L. from a dry meadow close to the village of Orionoe, southern Crimea

dogursky, 2006). Material collected from the Kerch Peninsula may be of value when looking for winter hardiness and for drought and salt tolerance in the related cultivated species. Material collected from the mud volcanos close to the village of Bondarenkovo (e.g., *Lepidium crassifolium* Waldst. et Kit.), is adapted to grow on very basic soils (pH 10.4) with high concentrations of toxic salts (Korzhenevsky and Klyukin, 1991), and is of interest when looking for such extreme adaptations.

### Conclusions

CWR from Crimea deserve attention as plant genetic resources for food and agriculture. Maxted *et al.* (2008) have drawn attention to the recently increasing threats to CWR in the Mediterranean flora of other parts of Europe. Such pressure on the native flora is increasing in Crimea. The change to a market economy after 1990, the active development of tourism and recreational housing in recent years, and the return of the native Tartars claiming land for housing influence the native habitats. The dynamics in the populations of CWR over time were obvious, as some species were more frequent than expected (e.g., *Triticum baecoticum*) while others documented in the recent past could not be found (e.g., *Aegilops tauschii*). The State Nikita Botanical Garden at Yalta will have an important role in protecting the plant diversity of Crimea for future generations. Depositing such germplasm in genebanks for *ex situ* conservation will be part of a strategy, but protection of the natural habitats, including monitoring of the population dynamics for efficient *in situ* conservation, is the highest priority.

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# Papua New Guinea: a much neglected hotspot of crop wild relative diversity

R. Kambuou<sup>1</sup>, T. Okpul<sup>2</sup> and D. Hunter<sup>3</sup>

<sup>1</sup>National Agricultural Research Institute (NARI), Papua New Guinea.

<sup>2</sup>University of Technology, Papua New Guinea.

<sup>3</sup>Bioversity International, Italy. Email: [d.hunter@cgiar.org](mailto:d.hunter@cgiar.org)

*The crop wild relatives (CWR) of Papua New Guinea (PNG) constitute an enormous and unique resource of genetic diversity which may be vital for future crop improvement and food security, particularly because they may harbour many valuable traits for agricultural adaptation to changing climate. Yet we know so little about the CWR in a country so biodiverse as PNG. While PNG has attracted much interest from international agencies and NGOs working in the area of biodiversity and conservation, most, if not all, have overlooked the role and importance of CWR. Sadly, we know so little about their numbers, their distribution or those species or populations most at risk. All this while habitats in PNG continue to be lost or degraded.*

PNG occupies the eastern half of the big island of New Guinea, just north of Australia. Covering a land area around 460,000 km<sup>2</sup>, it contains some of the most unique species, landscapes and ecosystems in the world (Anonymous, 2010). The country is justifiably considered among the most bioculturally diverse in the world. Its unique environment has supported hundreds of social groups and tribes for millennia and given rise to over 800 distinct indigenous languages. Today, PNG is ranked among the most mega-biodiverse countries in the world. In terms of flora, PNG is estimated to have around 20,000 identified species of plants, of which at least 30 % are considered endemic; yet less than 300 are believed to have been assessed according to IUCN Red List Criteria (Anonymous, 2010). The plants of PNG are also unique because they arise from two distinct biogeographic sources—the Gondwana flora of the south and the flora of Asian origin from the west (Bourke and Harwood, 2009). Sadly, recent years have witnessed the expansion of forest clearing, logging, oil palm exploitation and mining activities which have all impacted on PNG's environment. These threats will be increasingly added to by climate change. What is most alarming is that so little of PNG's unique and diverse wilderness is afforded formal protection of any sort, with estimates of less than 4 % of the terrestrial area currently

covered by around 50 protected areas, few of which have inventories of the plant species they contain or functional plans to manage them (Anonymous, 2010).

**“The country is justifiably considered among the most bioculturally diverse in the world”**



**Figure 1** A wild taro found in a stream in East New Britain Province, PNG (Photo: Peter Matthews)

PNG has a long history of agriculture, largely dependent on vegetatively propagated species, and it remains the major livelihood for the majority of the population with subsistence agriculture providing about 80 % and 75 % of food energy and protein respectively (Bourke and Harwood, 2009). The authors also claim that early agriculture and domestication of certain cultivated species was probably happening in PNG around the same time as similar events elsewhere in the world and that a large number of starchy staples, vegetables, fruits and edible nuts are likely to have been domesticated in PNG, including banana, taro, sago, greater yam, sugarcane, breadfruit, pandanus, and other important tree species. Edible nuts of *Pandanus antaresensis* and *P. brosimos* have been collected and eaten for thousands of years (possibly as far back as 30,000 years for the former) and it appears that the edible nut pandanus commonly found in the highlands, *P. julianettii*, was domesticated from *P. brosimos* about two millennia ago (Bourke and Harwood, 2009). There is also evidence that taro (*Colocasia esculenta*), believed to be one of the world's oldest food crops, was cultivated at Kuk archaeological site in a

swamp in the Western Highlands Province of PNG from around 9,000 years ago, and starch remains on stone tools indicating a vastly older history of use (Matthews, 2010). Yet we know so little about the ecology and diversity of wild taro (Fig. 1) or their interactions with cultivated taro. PNG is also known as a secondary centre of genetic diversity for many other crops, including sweet potato (*Ipomoea batatas*), cassava (*Manihot esculenta*) and aibika (*Abelmoschus manihot*).

Despite the existence of a significant number of CWR and corresponding reservoir of genetic variation and growing awareness of the impact of threats, PNG has not yet undertaken national inventories or ecogeographic surveys of important CWR in the country. Yet wild stands of many fruit and nut tree species like *Pometia pinnata*, *Canarium indicum* and okari nuts (*Terminalia* spp.) are declining in areas of the country due to logging activities (Anonymous, 2009). While some plant diversity rich areas in the country have been declared as nature reserves and national parks, there are no corresponding detailed inventories of the plant species in these areas, nor appropriate management plans due to a lack of training, awareness, manpower and other resources.

The authors are not aware of any active *in situ* conservation of wild relatives in the country, yet PNG remains a high priority in terms of globally important food crops and their wild relatives. Macted and Kell (2009), in an extensive global background

study on the wild relatives of those crops identified as crucially important for food security and included in Annex 1 of the ITPGRFA, identified a number of priority wild relative taxa and sites for *in situ* conservation in PNG (Fig. 2). Using data collated from a variety of sources, they concluded that PNG was a high priority country for the *in situ* conservation of wild relatives of rice; namely, *Oryza longiglumis*, *O. minuta* and *O. schlechteri*.

The authors identified as potential sites for the *in situ* conservation of rice wild relatives in PNG: Tonda Wildlife Management Area (IUCN category VI and Ramsar site) for *O. minuta* and *O. longiglumis*; Neiru Wildlife Management Area (IUCN category VI) and Kikori Marine Park/Reserve (proposed IUCN site), and Bismarck-Ramu National Park (proposed IUCN site) for *O. schlechteri*. The authors also concluded that *Musa schizocarpa*, a wild relative of banana, was a high priority taxa for *in situ* conservation in PNG but its occurrence and distribution are not known.

The National Agricultural Research Institute (NARI), the premier agricultural research organization in PNG, has taken the initiative in collecting some species of CWR—especially fruits, nuts and traditional vegetables—and a small *ex situ* collection of this germplasm is maintained at NARI Research Centre, Keravat (Anonymous, 2009). In 2005, in collaboration with the National Institute of Agro-Biological Sciences (NIAS), Japan,

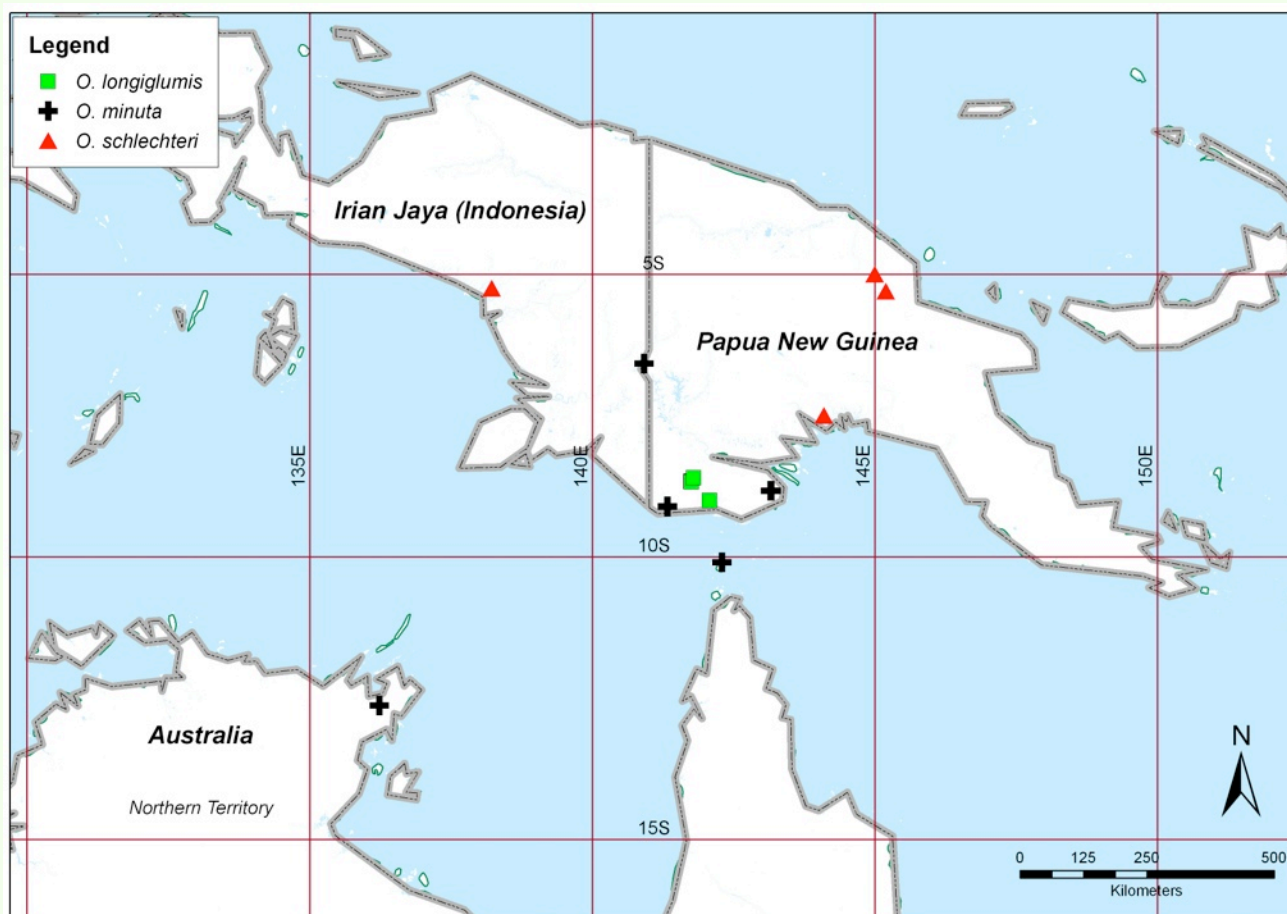


Figure 2 Distribution of high priority wild rice relatives in PNG (Source: Macted and Kell, 2009)





**Figure 3** *Oryza ridleyi* (Photo: Tom Okpul)

NARI undertook missions to collect wild rice and wild cowpea in Western, Gulf and Central Provinces in the southern part of PNG and East Sepik and Madang Provinces in the north. Three wild rice species were collected: *O. ridleyi* (Fig. 3), *O. rufipogon* and *O. schlechteri*. *O. ridleyi* was found and collected at four locations along the Sepik River and its tributaries and also found in the lower Ramu River area. *O. rufipogon* was collected in the Blackwater area of East Sepik and *O. schlechteri* was found in two locations along the Jamu River, a tributary of the Minajim River in Madang Province. *Vigna reflexo-pilosa* was found along the roadside in the forested hills between Wewak and Maprik in East Sepik. On the Sepik plains between Maprik and Pagwi an unidentified *Vigna* species, possibly a member of the *V. minima* complex, was found in two locations. Along the Sepik River and abundantly in the Chambri Lakes a species similar to *V. radiata* var. *sublobata* but with some distinctive characteristics was found. In Madang Province, *V. reflexo-pilosa* and *V. radiata* var. *sublobata* were collected in several locations (Vaughan *et al.*, 2005). Collected accessions are deposited in field collections at NARI Laloki and Bubia. A duplicate collection of wild rice is maintained by the University of Technology (UNITECH), Department of Agri-

“The authors are not aware of any active *in situ* conservation of wild relatives in the country, yet PNG remains a high priority in terms of globally important food crops and their wild relatives”

culture (DOA). However, little is known about the genetic diversity or ecology of wild rice and wild cowpea found in PNG.

To date, this represents the extent of collection and *ex situ* conservation of wild relatives in PNG—clearly much more is needed.

#### Future needs

There is an urgent need for attention and action on CWR conservation in PNG. Because of the cross-disciplinary and multi-stakeholder nature of CWR conservation, this will require a coherent and coordinated approach and effective working partnerships between groups and individuals from within the biodiversity and agriculture sectors, who traditionally do not collaborate. This is always a challenge, especially in a country the size of PNG where capacity and research on wild relatives is so limited. Action for CWR in PNG cannot afford to be piecemeal and demands a structured and logical framework. Many of the gaps and needs identified above could be addressed by the development of a national CWR conservation strategy (Hunter and Heywood, 2010).

There are many stakeholders in PNG with responsibility or mandates for biodiversity who need to work together for effective CWR conservation. The Department of Environment and Conservation (DEC) is the main government body responsible for biodiversity and conservation including the formulation of policy and legal issues as well as the maintenance of protected areas. The PNG National Forest Authority and their research and development organization, the PNG Forest Research Institute (FRI), look after forest resources. NARI has been mandated by the government to look after the rich genetic diversity of agricultural biodiversity including food crop species of the country. The national universities, including the University of Papua New Guinea (UPNG) and UNITECH, also contribute to research and capacity building on biodiversity and conservation, as do a range of local and international NGOs, including Conservation International, the Wildlife Conservation Society, World Wildlife Fund and The Nature Conservancy. All these organizations and others, including community-based and farmer groups, must be involved and an effective first step in trying to address this issue in PNG would be to develop a national CWR conservation strategy or action plan which would:

- Ensure coordination of planning and implementation of CWR conservation so that collaboration occurs and activities are harmonized between the relevant stakeholders and actors involved in PNG;
- Institutionalize and mainstream the practice of CWR conservation in PNG by embedding it in national planning mechanisms supported by relevant policy, legislative and financial measures;
- Promote awareness and understanding among policy-makers, researchers, development and conservation practitioners and the general public of the importance and value of CWR and their conservation to PNG;
- Provide a mechanism for reporting on progress towards PNG's targets and plans agreed under other agreements such as the CBD and its Global Strategy for Plant Conservation.

Such a national strategy or action plan could ensure that the following measures, as outlined in Hunter and Heywood (2010) could be undertaken in PNG to strengthen the conservation of CWR:

### “There is an urgent need for attention and action on CWR conservation in PNG”

- The compilation of a first national inventory of CWR along the lines of those developed by other countries with a rich heritage of wild relatives.
- The selection of a list of priority species of CWR for which conservation action is proposed, either *in situ* or *ex situ* or both, and which is realistic in terms of manpower and resources.
- The development of baseline assessments for priority species—their ecogeographic status, including threat assessment.
- Gap analysis to establish where gaps exist in conservation measures.
- Proposals for *in situ* conservation action (including threat management) and identification of important areas for CWR conservation inside and outside protected areas.
- Collaborations and partnerships with local communities who directly utilize wild relatives in times of general food scarcity and upon whose land priority species occur.
- For priority species for which *ex situ* conservation is required, proposals for their sampling and storage in gene banks, botanic gardens or other long-term facilities.
- Proposals for relevant research on priority species, such as gene flow mechanisms between wild and cultivated taro, genetic diversity measurement between wild and cultivated populations, identification of important genetic traits in wild relatives for pre-breeding and crop improvement.
- The development of a national information system which covers PNG's wild relatives. In 2004, a National Steering Committee known as the PNG Biodiversity Network (PNGBioNeT) with the mandate of developing a 'national hub' for gathering, collating, storing and disseminating information and data on biodiversity of the country was set up (Anonymous, 2010). Ideally, CWR data and information should be integrated into such a national information system.
- Measures for ensuring national awareness of the importance of conserving and using CWR. PNG has two botanic gardens, the Lae Botanical Garden located in Morobe Province and the Port Moresby Botanical Garden on the same campus as UPNG—both could be utilized to showcase the country's important cultivated species and their wild relatives.
- A national capacity development plan that addresses CWR. Scientific and technical capacity is inadequate in PNG. PGR are included in the curricula at both UNITECH and Vudal University but the content is basic and contains little detail on CWR. Few, if any national scientists have been trained to postgraduate level in CWR conservation, management and sustainable utilization, yet the country is endowed with such a wealth of genetic resources. Training on CWR conserva-

tion is also required for staff involved in the management and administration of PNG's protected areas and should be included in training and teaching courses in biodiversity and conservation.

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## Recent publications

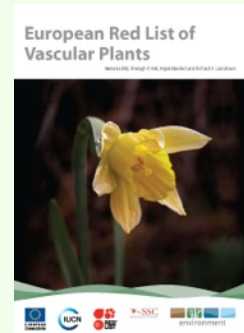
**Bilz, M., Kell, S.P., Maxted, N. and Lansdown, R.V. (2011) *European Red List of Vascular Plants*. Luxembourg: Publications Office of the European Union. ISBN 978-92-79-20199-8.**

Threat assessment is a routine tool used to help prioritize species most in need of active conservation; however, there are few examples of the tool being used for CWR species and to date, relatively few CWR Red List assessments have been published in the IUCN Red List of Threatened Species ([www.iucnredlist.org/](http://www.iucnredlist.org/)). As part of a recent EC-funded project to produce a European Red List (<http://ec.europa.eu/environment/nature/conservation/species/redlist/>), the Crop Wild Relative Specialist Group of the IUCN Species Survival Commission undertook regional IUCN Red List assessments of a sample of priority European CWR. Species were selected on the basis that they are in the same gene pool as crops which: a) are particularly important for food security in Europe, b) make a major contribution to the European economy, c) are listed in Annex I of the International Treaty on Plant Genetic Resources for Food and Agriculture (ITPGRFA), and d) have wild relatives native to Europe. The selected list of 591 CWR species, 188 of which are endemic to Europe, encompass 25 crop gene pools/crop groups. Nineteen species were assessed as Not Applicable, either due to their marginal distribution in Europe or because they were introduced to the region after AD 1500. The status of the remaining species was assessed at two regional levels: geographical Europe (572 species) and the area encompassed by the 27 countries of the EU (521 species). At the European level, 11.5 % (66) of the species are considered as threatened, with 3.3 % (19) of them being Critically Endangered, 4.4 % (22) Endangered and 3.8 % (25) Vulnerable—a further 4.5 % (26) of the species are classified as Near Threatened. A clear European priority is to focus conservation attention on the 92 threatened and Near Threatened species. However, although many of the species assessed as Least Concern (54.7 %) occur in several countries in Europe, a significant percentage are threatened at national level; therefore, these species should not be overlooked in conservation planning. It is also important to note that the assessments are at species level and the IUCN Red List Categories and Criteria (IUCN, 2001) do not take into account threat to genetic diversity within species. Furthermore, 29 % of the species assessed at European level are classified as Data Deficient which highlights the need for greater attention to be paid to CWR in national biodiversity monitoring programmes and for reassessment of these species when the necessary data become available. This publication will prove a useful resource for Europe, but the lack of population data remains a limitation to Red Listing in Europe and most likely in other regions too.

A pdf of the text can be downloaded from the European Red List website at: [http://ec.europa.eu/environment/nature/conservation/species/redlist/downloads/European\\_vascular\\_plants.pdf](http://ec.europa.eu/environment/nature/conservation/species/redlist/downloads/European_vascular_plants.pdf).

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**Maxted, N., Dulloo, M.E., Ford-Lloyd, B.V., Frese, L., Iriondo, J.M. and Pinheiro de Carvalho, M.A.A. (eds.) (2012) *Agrobiodiversity Conservation: Securing the Diversity of Crop Wild Relatives and Landraces*. CABI Publishing, Wallingford. ISBN 978-1-84593-851-2.**

Although there have been significant recent advances in agrobiodiversity conservation, there is still a lack of good illustrative examples of *in situ* CWR and LR conservation. This book, based on the 2010 symposium 'Towards the establishment of genetic reserves for crop wild relatives and landraces in Europe' held in Funchal, Madeira in September 2010, chronicles some of the latest discussion on agrobiodiversity conservation. The 43 chapters draw exemplar case studies (primarily from Europe) on CWR and LR conservation, priority setting and threat assessment, data management, the policy context and a set of invited papers on specific issues such as regional or global CWR/LR conservation and finishing with a report of a horizon scanning exercise for medium and longer term CWR/LR conservation priorities in Europe. By considering the benefits of understanding and preserving CWR and LR, it encompasses issues as wide-ranging and topical as habitat protection, ecosystem health and food security. The focus is on Europe, but the case studies presented have global relevance, so *Agrobiodiversity Conservation* will prove a useful resource for postgraduate students of conservation and environmental studies, conservation professionals, policy makers and researchers.

## Book review

**Offord, C.A. and Meagher, P.F. (eds.) (2009) *Plant Germplasm Conservation in Australia: Strategies and Guidelines for Development, Managing and Utilizing Ex Situ Collections*. Australian Network for Plant Conservation Inc., Canberra, Australia. ISBN 978-0-9752191-1-9.**

Given the fact that much of Australian agriculture is based on species introduced from the Mediterranean region, it is good to know that Australia is now turning toward conserving its own significant wealth of native CWR diversity which includes major diversity in rice (*Oryza* spp.), soybean (*Glycine* spp.) and eucalypts (*Eucalyptus* spp.), as well as many medicinal and ornamental species. The objective of this publication is to provide guidelines on the collection, curation and use of this diversity. The eight edited chapters cover various aspects of Australian plant genetic resources, introductions to prioritization, seed and plant material collection, seed banking, seed storage, tissue culture, cryopreservation and management of living collections. Despite one of the book's stated objectives, there is a minimal focus on germplasm use and some of the chapters do not cover all of the contemporary techniques available to develop and manage *ex situ* collections; but despite these quibbles, the text is a useful introduction to the subject for the Australian market.

## *Lathyrus belinensis*: a CWR discovered and almost lost

N. Maxted

School of Biosciences, University of Birmingham, Edgbaston, Birmingham B15 2TT, UK. Email: [nigel.maxted@dial.pipex.com](mailto:nigel.maxted@dial.pipex.com)

In 1987 while searching near Cavus, Antalya province, Turkey for food, fodder and forage legume species with Ayse Kitiki (Aegean Agricultural Research Institute, Menemen, Turkey) and Bob Allkin (Royal Botanic Gardens, Kew, UK), we discovered a new species of the legume genus *Lathyrus*. The species was subsequently published as *Lathyrus belinensis* by Maxted and Goyder (1988). The single population was growing alongside a new road that was just then being cut through fields between Kumluca and Tekirova (Fig. 1). The population appeared to have its greatest concentration in and around an ungrazed village graveyard in the village of Belin.

The new species was a member of *Lathyrus* section *Lathyrus* and most closely related to *L. odoratus* (sweet pea), being just as scented as sweet pea but with more hairy vegetative parts. The most striking and economically interesting distinguishing feature of *L. belinensis* is the flower colour which is yellow with conspicuous red veins (Fig. 2). This is in contrast with *L. odoratus* flowers which can be purple, blue, pink or cream, but never yellow. Thus the discovery of *L. belinensis* was an opportunity for horticulturalists to breed a yellow sweet pea—a goal of many contemporary sweet pea breeders.



**Figure 2** *Lathyrus belinensis* flower (Photo: Nigel Maxted)

The type population was found over an area of only 2 km<sup>2</sup> and although the species was published in 1988, no further populations have been reported. The only known population is found adjacent to a new main road that carries holiday traffic along the coast in an area ripe for tourism development and is in an area that was being planted with conifers at the time of original collection. On returning to the site in 2010 it was found that the original type location had been completely destroyed by earthworks associated with the building of a new police station (Fig. 3). Although some plants were still found in the area and seed is held *ex situ*, the richest area



**Figure 1** *Lathyrus belinensis* growing on a roadside bank (Photo: Nigel Maxted)

within the site had been lost. In part, to draw attention to the need for active *in situ* conservation of *L. belinensis* it has recently been assessed using the IUCN Red List Criteria and found to be Critically Endangered—the most highly threatened category. The species has real economic potential as a CWR gene donor, yet it is near extinct in the wild. Only time will show if action can be taken before we lose the opportunity to fully exploit this natural resource!

### Reference

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**Figure 3** Excavated area where *Lathyrus belinensis* formerly existed (Photo: Nigel Maxted)