

# Report of a Working Group on *Beta* and the World *Beta* Network

Third Joint Meeting, 8-11 March 2006, Puerto de la Cruz, Tenerife, Spain

L. Frese, L. Maggioni and E. Lipman, editors





Bioversity International is the operating name of the International Plant Genetic Resources Institute (IPGRI).

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# Report of a Working Group on *Beta* and the World *Beta* Network

Third Joint Meeting, 8-11 March 2006, Puerto de la Cruz, Tenerife, Spain L. Frese, L. Maggioni and E. Lipman, *editors*  **Bioversity International** is an independent international scientific organization that seeks to improve the well-being of present and future generations of people by enhancing conservation and the deployment of agricultural biodiversity on farms and in forests. It is one of 15 centres supported by the Consultative Group on International Agricultural Research (CGIAR), an association of public and private members who support efforts to mobilize cutting-edge science to reduce hunger and poverty, improve human nutrition and health, and protect the environment. Bioversity has its headquarters in Maccarese, near Rome, Italy, with offices in more than 20 other countries worldwide. The organization operates through four programmes: Diversity for Livelihoods, Understanding and Managing Biodiversity, Global Partnerships, and Commodities for Livelihoods.

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#### Citation

Frese L, Maggioni L, Lipman E, editors. 2009. Report of a Working Group on *Beta* and the World *Beta* Network. Third Joint Meeting, 8-11 March 2006, Puerto de la Cruz, Tenerife, Spain. Bioversity International, Rome, Italy.

#### Cover illustrations

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ISBN 978-92-9043-815-1

Acknowledgements to Dr L. Currah for English language editing and to F. Ferraiuolo for graphics and layout.

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## PART I. DISCUSSION AND RECOMMENDATIONS

#### Introduction

#### Opening of the meeting

Manuel Caballero, Scientific Director of the Instituto Canario de Investigaciones Agrarias (ICIA), opened the meeting on behalf of the President of ICIA and Director of the Jardín de Aclimatación de La Orotava, Puerto de la Cruz. He welcomed all the participants to the Canary Islands and explained the richness of botanical diversity and the rare plants growing in these islands, with 600 endemic species, including some representatives of the *Beta* section. He wished the Group a successful meeting.

On behalf of the International Plant Genetic Resources Institute (IPGRI, now Bioversity International), Lorenzo Maggioni, Coordinator of the European Cooperative Programme for Crop Genetic Resources Networks (ECP/GR)<sup>1</sup> welcomed the Group to the third joint meeting of the Working Group on *Beta* and the World *Beta* Network. He thanked the local hosts for their kindness, patience and efficiency during the preparation of the meeting. He was also pleased to note that for the first time a meeting was to be associated with a practical action, i.e. monitoring of the wild *Beta* populations occurring on the island of Tenerife, during the fourth day. Looking forward to the outcome of this meeting, largely focused on *in situ* conservation of wild relatives, he expressed the wish for a constructive meeting to a small, but dedicated group.

Lothar Frese, Chair of the ECP/GR Working Group on Beta and Secretary of the World Beta Network, reminded the Group of the history of the development of collaborative work on Beta genetic resources, dating back to the first ad hoc meeting held in 1987 in Wageningen, the Netherlands. This was a phase of germplasm collecting. After the establishment of the World Beta Network, in 1989, the Group entered the phase of establishment of the International Database for Beta (IDBB), which was followed by a phase dedicated to characterization and evaluation (1990s) and the creation of an alliance from 1998 onwards between the Working Group on Beta (with ECP/GR funding) and the World Beta Network. With the meeting of 2002 in Bologna, Italy, a phase of increased task sharing was encouraged, and concepts for sharing responsibilities for conservation were outlined; tasks were also shared by subgroup coordinators within the Network. This was also the phase of a shift towards themes related to *in situ* management, including through the collaboration with the EU-funded project PGR Forum. Currently the challenge consists in finding new and fascinating goals for the Group that can be useful to the user community. This meeting is opening with the focus dedicated to investigating the Why, How and Where of in situ management of Beta wild relatives.

After approval of the agenda, the participants briefly introduced themselves.

<sup>&</sup>lt;sup>1</sup> Following the decision of the 10th meeting of the ECPGR Steering Committee in September 2006, the name of the Programme was simplified to "European Cooperative Programme for Plant Genetic Resources" and the acronym was also modified to "ECPGR", removing the traditional slash of "ECP/GR".

### Section I. Scientific and technical aspects of in situ management

Available full papers of the presentations summarized below are included in Part III of the present report.

## **The PGR Forum project: some conclusions and recommendations** (*paper pp.* 27-30)

Brian Ford-Lloyd gave a description of the PGR Forum project and its results, essentially the Crop Wild Relatives (CWR) list, obtained from the Mansfeld's World Database and the Euro+Med PlantBase. The list is inclusive and it has reached a number of nearly 24 000 taxa. Criteria are needed to prioritize species in order to focus the limited resources for conservation.

The global Red Lists do not seem to provide suitable criteria, since only 163 species included in the CWR list have been globally red-listed and appear as threatened species, while many more taxa are listed in the national Red Lists.

An approach was chosen, based on the number of geographical units in which the taxon is recorded. Any taxon which occurs in more than 10 geographical units is considered not threatened and therefore not a priority for conservation. Geographical units are defined within the Euro+Med database and correspond to those used by the Flora Europea. Admittedly these do not all have the same weight, but the system can be used with some precautions and adjustments.

An application of the geographical units criteria to the wild *Beta* species results in a prioritization of all the species except a few: *B. macrocarpa* (occurring in 20 geographical units), *B. trigyna* (16), *B. vulgaris* subsp. *maritima* (62) and *Patellifolia patellaris* (or *B. patellaris*) (16). Further criteria for prioritization can be added in terms of use of the crop, with food, fodder/forage and industrial crops as the most important. A further criterion is based on current conservation status.

The selected criteria for prioritization justify from all points of view the conservation of most wild species of *Beta*.

Further activities that can be planned to better implement conservation consist in adding *Beta* priority species to the existing global Red Lists; undertaking population assessment and monitoring; and molecular population genetic assessment.

It may be questionable whether we are already in a position to designate, design and establish new reserves and whether appropriate monitoring is taking place in existing reserves.

One overarching problem for the funding of plant conservation is the occurrence of major threats such as bird flu and other stresses which are likely to require a large part of the available financial resources from potential donors.

During the discussion, it was noted that there may be more genetic diversity in species that are more widespread, such as *Beta vulgaris* subsp. *maritima*, and that there may be specific populations that are known to host useful traits and important richness in variation, such as is the case of the Po Valley populations. The prioritization criteria described are therefore a very useful starting point, but conclusions should also be drawn on the basis of specific expertise and the knowledge of interest groups.

## Reconstructing the evolutionary history of the Beta section with molecular data. A focus on the Canary Islands

(paper pp. 31-37)

Sarah Villain presented a study of the chloroplast diversity of *Beta* section *Beta*, which was analysed on a sample representative of the geographical distribution of three taxa, i.e. *B. vulgaris* subsp. *maritima*, *B. vulgaris* subsp. *adanensis* and *B. macrocarpa*. Chloroplast DNA fragments and nuclear regions were sequenced to analyse intraand interspecific diversity. Polymorphism analyses indicate that *B. vulgaris* subsp. *maritima* exhibits relatively greater levels of haplotype and nucleotide diversity. In general, it can be concluded that the *Beta* section has low levels of polymorphism. The structure of the haplotype network seems to indicate a recent differentiation of *B. vulgaris* subsp. *adanensis* from *B. vulgaris* subsp. *maritima* in the Aegean Sea. The diploid types of *B. macrocarpa* contain a major haplotype, distributed from the Canary island of Fuerteventura to Turkey. The two tetraploid Canarian accessions from Gran Canaria and Tenerife share a single haplotype with *B. vulgaris* subsp. *maritima*, confirming the hybrid origin of these tetraploid accessions as a result of a cross between *B. v. maritima* (as the male parent) and the diploid *B. macrocarpa*.

Phylogeographic studies of *B. vulgaris* subsp. *maritima* indicate that assumptions can be made on the glacial refugia from which postglacial recolonization started. An eastern European refugium is suggested, while the Iberian Peninsula or, most likely, the Moroccan area was the source for colonization of the Atlantic coasts.

Studies on the possible origin of the Canarian tetraploid *B. macrocarpa*, based on mini- and microsatellite markers, indicate that an hybridization of *B. vulgaris* subsp. *maritima* x *B. macrocarpa* (2x) might have occurred (perhaps in Morocco) before colonization of the Canary Islands. The diploid *B. macrocarpa* is hypothesized to deserve the status of a different and new species.

## Protection and distribution of Beta (s.l.) species in the Canary Islands: perspectives of conservation

Arnoldo Santos Guerra gave an account of the taxonomy, distribution, ecology and conservation of *Beta* in the Macaronesian region.<sup>2</sup> The actual accepted taxa include, within section *Beta, Beta macrocarpa* Guss., *Beta vulgaris* subsp. *maritima* L. and *Beta patula* Aiton; and within section *Procumbentes, Beta patellaris* Moq., *Beta procumbens* C. Sm. ex Hornem and *Beta webbiana* Moq. A number of taxonomic uncertainties were highlighted. These include: the doubtful attribution to *B. procumbens* of the species growing in the island of Salvajens; and the likelihood that *B. hastata* Link in Buch may be the legitimate name for *B. procumbens*. Also the distribution of the species still requires further investigation. A record of the presence of *B. patellaris* in the Socotra region was highlighted and recommended for verification. The only species that can be considered threatened is *B. patula*, growing in Madeira, Porto Santo and Desertas. *B. webbiana* also needs to be checked since its taxonomic status may be confused. The Canarian *Beta* are not considered threatened, since they largely grow within protected areas, covering nearly 35% of the Canarian territory. In particular, *Beta* 

<sup>&</sup>lt;sup>2</sup> Macaronesia is a biogeographical region which includes the Canaries, Madeira and the Azores.

*patellaris* grows within the National Parks of Timanfaya and Caldera of Taburiente, *Beta macrocarpa* within Timanfaya National Park, *Beta procumbens* in several protected Natural and Rural parks and *Beta webbiana* in the Isleta protected area. Regarding the management of the protected areas, it was made clear that the governments of the individual islands are responsible for them, except for the national parks, which are under the national authority.

## Biodiversity of Beta species in the Transcaucasus region (Armenia, Azerbaijan, Georgia, Iran)

(paper pp. 38-44)

Guram Aleksidze described in detail the distribution of *Beta* species in the Transcaucasus area. The following list of species was given for the respective countries:

#### Armenia

*B. vulgaris* L.; *B. vulgaris* subsp. *maritima* (L.) Arcang.; *B. corolliflora* Zosimovich; *B. macrorhiza* Steven; *B. lomatogona* Fischer et Meyer; and *B. trigyna* Wald. et Kit.

*B. corolliflora, B. macrorhiza* and *B. lomatogona* are represented in *ex situ* collections in Armenia, although these collections need support for long-term maintenance.

#### Azerbaijan

*B. vulgaris* subsp. *maritima* (L.) Arcang.; *B. lomatogona* Fischer et Meyer; *B. macrorhiza* Steven; *B. patula* (Soland) W. Aiton, Hortus Kewensis; *B. trigyna* Wald. et Kit.; and *B. vulgaris*.

#### Georgia

*B. vulgaris* L. subsp. *vulgaris* (cultivated leaf beet, garden beet, fodder beet and sugar beet); *B. corolliflora* Zosimovich; and *B. trigyna* Wald. et Kit.

Only five accessions of cultivated *B. vulgaris* are conserved in the genebank.

#### Iran

B. lomatogona and B. vulgaris subsp. maritima.

In the case of *B. lomatogona* in Iran, population size is apparently decreasing in a number of localities, suggesting the need for protection of this natural reservoir of potentially useful traits. Collecting missions were carried out and *in situ* conservation was established in the Ardabil Research Station.

There are strong political, biological and economic reasons why wild beet populations should be considered for *in situ* management in the Transcaucasus region. Detailed information is available on the distribution of the species and individual populations. Good taxonomic and biosystematics knowledge is available, as well as a central crop database for *Beta*, which could be used as an *in situ* management tool. There is a strong interest among scientists in investigating the specific requirements for *in situ* management of *Beta*.

There is also a serious need to organize collecting missions to collect *Beta* species and to increase *ex situ* collections with new accessions, as well as to increase on-farm conservation.

During the discussion, the validity of the presence of *B. patula* in Azerbaijan was questioned, considering that this species is considered endemic to Madeira Island.

#### Survey of Beta nana in Greece

#### (paper pp. 45-52)

Lee Panella described the joint exploration mission organized in 2005 by the United States Department of Agriculture (USDA), the Federal Centre for Breeding Research on Cultivated Plants (BAZ)<sup>3</sup> and the Greek Gene Bank to survey Beta nana populations in Greece. Beta nana, an inconspicuous diploid belonging to the monotypic section Nanae, only grows at high altitude (above 1800 m) and is endemic to Greece. Little is known about its genetic variation, while phenotypic variation is considered low within and between populations. The species is considered rare but not endangered. However, species in alpine regions are sensitive to global climate change. Reproduction and migration mechanisms of the species have not been investigated, demographic processes are not well understood, the impact of climate change is difficult to predict and gene flow and genetic variation within and among populations are little understood. The exploration was therefore meant to: 1) obtain fresh seed from a few, larger populations, enabling further research; 2) jointly develop an *in situ* management programme with the Greek counterparts; 3) develop a database tool suited to document monitoring data; 4) determine environmental conditions to design efficient *ex situ* regeneration procedures. This mission was also a follow-up to the previous recommendations of ECP/GR to monitor the status of this species. Twenty-six populations were found in mountainous areas throughout Greece, in grazed places, at medium risk of genetic erosion. Mount Olympus was identified as the most suitable site for *in situ* conservation, since it is already a national park.

There still remains a significant area to explore in Greece in order to confirm the full extent of the species distribution. It is also needed in order to understand how effectively *B. nana* populations are protected by national or provincial nature park statutes, when they are located in or near these areas. The extent of overlap between legally protected areas and growing sites needs to be explored by integrating geographic information system (GIS) and floristic studies to determine the scope of the relationship between conservation measures and plant species survival. It is also necessary to match the distribution of *B. nana* to existing protected areas and determine overlap, in order to suggest locations for the development of genetic conservation sites.

A more informed choice of populations and sites for conservation priority should be based on genetic distance measures and genetic variation detected within and among populations. Studies with microsatellite markers are proposed. How effectively agro-environmental measures subsidized by the EU Commission can be deployed for managing these sites also remains a question to be verified.

<sup>&</sup>lt;sup>3</sup> The German Federal Ministry of Food, Agriculture and Consumer Protection (BMELV) merged the former Federal Centre for Breeding Research on Cultivated Plants (BAZ), the Federal Biological Research Centre for Agriculture and Forestry (BBA), and parts of the Federal Agricultural Research Centre (FAL). Since January 2008 the new institution is called Federal Research Centre for Cultivated Plants – Julius Kühn-Institut.

In the discussion which followed it was remarked that it is easier to identify and conserve populations already included within protected areas. However, the question remains of how much diversity is present within or outside these areas.

It was also noted that research with microsatellites is likely to take place in the USA on the collected material, in the hope that the markers used for section *Beta* also work on section *Nanae*.

It was concluded that an important outcome of this mission had been to realize that *B. nana*, which had been believed possibly to be extinct, was still present and moreover, with several populations. The collected material will be very useful for further research and characterization and evaluation work.

### Survey of Beta vulgaris subsp. maritima populations in Ireland

(paper pp. 53-58)

Dermot Grogan reported on a survey of *Beta vulgaris* subsp. *maritima* populations that took place in Ireland in 2002-2003, when 56% (24) of the sites previously sampled in 1987 were checked, as well as three new sites. Geographical locations of the populations were mapped on the basis of a high global positioning system (GPS) accuracy (<3 m). Plants were always found in proximity (within 100 m) of the sea edge. Habitats included stone walls, concrete walls, coastal defences, cliffs, road verges, reclamation areas, park benches, caravan parks, yacht parks and (less often) sand dunes. The populations were generally quite well established, except for a few. The proximity to cultivated sugar beet was estimated to be within 1 km in 28% of the cases, between 1 and 5 km (30%), >5 km (44%). In no cases could the populations be considered to have been completely isolated throughout history. Diseased plants were found very rarely and in only one case was a probable virus disease noticed.

The ongoing threats to the habitats of maritime beet populations in Ireland are coastal erosion and human leisure and commercial activities.

Two sites are under threat of extinction and should be designated for conservation: 1) Ross's Point, Co. Sligo, where no plants are present any longer in this area and the actual habitat is not very suitable for *B. vulgaris* subsp. *maritima*; 2) Dunmore East, Co. Waterford, where less than 10 plants were found.

A number of sites are included in Special Areas for Conservation, although the species is not mentioned in the lists of "interesting" plants. Recommendations need to be made to the authorities to take these plants into consideration.

In the discussion, it was noted that in Italy too, *B. vulgaris* subsp. *maritima* only grows in close proximity to the sea. However, B. Ford-Lloyd mentioned that more and more occurrences of maritime beet growing inland were being recorded (e.g. in Minorca).

The observation was also made that in the South of England it seems that smaller amounts of seed are being produced, possibly as an effect of global warming, which could reduce the vernalization effect. This observation was also confirmed for the case of Sweden, where huge plants with no seed at all had been noticed. On the other hand, this tendency towards seed set reduction could not be confirmed for Ireland, but possibly the Irish populations might already have become adapted to a lower requirement for vernalization.

## The International Database for Beta (IDBB) and in situ management: potential role and functions

#### (paper pp. 59-74)

Christoph Germeier explained to the Group the arguments in favour of a cropspecific approach combining *in situ* and *ex situ* information:

- Managing *ex situ* and *in situ* data are complementary issues. Thus they need a common information source.
- *Ex situ* collecting sites can be used to find interesting *in situ* populations and vice versa.
- Evaluation and characterization of *in situ* populations will imply repeated sampling and keeping reference samples as genebank accessions.
- Users of genetic resources will use both sources and need a tool for integrating research for interesting traits within *in situ* and *ex situ* collections.
- Breeding-related interests (e.g. characterization and evaluation) are cropspecific. Multicrop information systems in these domains impair simplicity of use and performance.

Interacting information systems for *in situ* management were described, including the European Nature Information System (EUNIS): information on nature reserves with emphasis on nature protection, geographical, faunistic and floristic focus – multi-species inventories with special regard for biologists' favourites; the PGR Forum Crop Wild Relative Information System (CWRIS): lists of wild species of potential importance for breeding and agriculture and their habitats; and the Central Crop Databases (CCDBs), integrating data on occurrence *ex situ* and *in situ*, characterization and evaluation. Cooperation and integration among the above-mentioned databases is an option that needs to be pursued. This can be achieved by making use of shared open source software, such as CropForge. CropForge is a collaborative software development site, maintained by the biometric and bioinformatics unit of the International Rice Research Institute (IRRI), providing tools and a centralized workspace for developers to control and manage software development.

The representation of wild species in the IDBB (http://idbb.bafz.de) corresponds to 33% (3506 accessions) of the total number of accessions, and most of these are well documented for site information.

New features of the IDBB include pictures of some accessions and interactive maps of the collecting sites. Evaluation and characterization data can be geographically mapped as well. The IDBB records 16 067 observations related to the wild species, mainly for *B. vulgaris* subsp. *maritima*, *B. vulgaris* subsp. *adanensis* and *B. macrocarpa*. This represents 53% of the total number of observations in the IDBB.

A data model for *in situ* field observations was created. It is necessary to define the population. A certain geographic point (reference site) on the map can identify a population of a certain taxon. Population data can be linked to site data and habitat data, as well as to data for a survey of the population carried out at a defined time. Moreover, patch and demography data correspond to the observations made on a subpopulation or part of a population.

While there are good reasons to create and manage a database at the crop level, some elements of the database that are not crop-specific should be managed by

a central repository, at the multicrop level, for instance the site information. The currently available taxonomy backbone can derive from CWRIS and/or the Global Biodiversity Information Facility (GBIF). However, several taxonomic problems still remain to be solved.

## Screening techniques for root growth parameters under dry and compacted conditions in sugar beet germplasm

(paper pp. 75-78)

Eric Ober described ongoing work on improving tolerance to water stress. The rooting characteristic of the plant is an important phenotype that can be correlated to drought tolerance. Evaluation is made by looking for root variation in the field. Genotypic differences in rooting patterns and water use at the different soil layers were measured in an experiment under managed drought conditions in the field. It was also noted that greater water extraction corresponds to greater sugar yield. However, differences may not be large enough for breeders to use. A related character that needs to be considered is the differential ability to root under compacted conditions.

In the discussion, it was noted that research at Broom's Barn is still continuing, although funding cutbacks have been made. The most interesting recent development is the identification of a dozen proteins that could be candidate genes for drought tolerance.

E. Ober also announced the launching of an online newsletter on germplasm evaluation and pre-breeding, which should be published soon, following his request for contributions. The request was sent to 265 email addresses and 19 responses were obtained, including from public and private breeders. The Group considered the newsletter initiative very commendable and is looking forward to receiving the first issue.

#### Recent progress in Beta germplasm evaluation in the USA

Lee Panella reported that the US National Plant Germplasm System (NPGS) has 470 986 accessions. It is a cooperative effort by public (state and federal) and private organizations to preserve the genetic diversity of plants. Considering that scientists must have access to genetic diversity to help bring forth new varieties that can resist pests, diseases, and environmental stresses, the NPGS aids the scientists and supports the need for genetic diversity by acquiring, preserving, evaluating, documenting and distributing crop germplasm.

The system is loosely coordinated, in the sense that there is not one single head of the NPGS. The Sugarbeet Crop Germplasm Committee (CGC) advises the NPGS on the *Beta* collection. It includes federal, state (university), and industry members and coordinates a national *Beta* evaluation programme. It is funded by competitive grants from the United States Department of Agriculture-Agricultural Research Service (USDA-ARS) National Programme Staff.

An organization and a scientist (private or public) are responsible for the evaluation of each of the descriptors of interest. After evaluation, data are processed and entered into the Genetic Resources Information Network (GRIN) database. Resistant accessions are re-screened and often selected as parents for pre-breeding. ARS scientists begin pre-breeding at the different locations and the resulting germplasm is released to seed company breeders.

Currently, the following research is under development:

- Beet cyst nematode pre-breeding (ARS-Salinas and Fort Collins). Germplasm to be released in 2005-2006 (Salinas);
- Beet cyst nematode marker development (ARS-Salinas, Fort Collins and Fargo);
- Continued development of rhizomania resistant germplasm, especially against emerging races (Salinas);
- New populations for *Cercospora* resistance (Fort Collins). Release planned for 2007 or 2008;
- Molecular mapping of Aphanomyces resistance (ARS-East Lansing);
- Continued breeding for sugar beet root maggot resistance (ARS-Fargo). Germplasm is continuously released;
- Research into Fusarium race structure and species composition (ARS-Fort Collins).

What makes the evaluation programme so important in the USA is that, after evaluation, the useful traits are introgressed into sugar beet germplasm and released for cultivar development.

## Evaluation of beet germplasm and progress towards the development of sugar beet for disease resistance and root structure

(*paper pp. 79-85*)

Mohammad Nasser Arjmand reported that the most important research activities of the Sugar Beet Seed Institute (SBSI), Karaj, Iran, concerning utilization of *Beta* germplasm in sugar beet breeding programmes, are as follows:

- 1. Evaluation of germplasm for resistance to Polymyxa betae
- 2. Transfer of rhizomania resistance gene(s) from *B. vulgaris* subsp. *maritima* and cultivated sources to sugar beet
- 3. Transfer of beet cyst nematode resistance genes from resistant sources to sugar beet
- 4. Transfer of root roundness and smoothness from fodder beet and red beet to sugar beet.

*An additional paper on the "Genealogical structure of a collection of beet" was provided by V. Burenin (VIR) and is also included in Part III (pp. 86-89).* 

## Poster session

The following posters were displayed and discussed:

### Beta genetic resources in Morocco

Y. El Bahloul<sup>(1)</sup>, P. van Cutsem<sup>(2)</sup>, M. Sadiki<sup>(3)</sup> and C. Al Faiz<sup>(1)</sup>

<sup>(1)</sup> National Institute of Agronomy Research, Genetic Resources and Plant Breeding Unit, CRRA-Rabat, BP 415, Rabat RP, Morocco

<sup>(2)</sup> University of Namur, Plant Cell Biology Unit, Rue de Bruxelles 61, 5000 Namur, Belgium
<sup>(3)</sup> Agronomy and Veterinary Institute Hassan II, BP 6202 Rabat Instituts, Morocco

Wild *Beta* species are widespread in Morocco. *B. maritima* and *B. macrocarpa* are widespread all over the country, while *B. patellaris* can only be found on the southern coast. Pastures and urbanization are putting many sites at risk.

It was explained that no landraces of sugar beet or leaf beet are present in Morocco.

## Germination ability of sugar and fodder beet seeds after long-term storage in the Polish genebank

#### Kamilla Kuzdovicz

*Plant Breeding and Acclimatization Institute (IHAR), Bydgoszcz, Poland* The efficiency of the system used was confirmed, but differences were detected in accessions' response.

## Genetic diversity analysis in four elite diploid populations of sugar beet (*B. vulgaris* L.) using RAPD and ISSR

#### H.M. Srivastava, S. Srivastava, P.S. Gupta and V.X. Saxena

*Division of Crop Improvement, Indian Institute of Sugarcane Research, Lucknow, India* Four populations of sugar beet were screened and the ability of DNA-based markers to detect a high degree of polymorphism among these populations suggested the possibility of screening a higher number of anonymous loci in sugar beet to enable selection of the most suitable parents to obtain new genetic combinations.

## Section II. Technical meeting, including review of the workplan

## General briefing on ECP/GR

L. Maggioni described the current status of the ECP/GR programme. He explained that the ECP/GR had entered its VIIth Phase (2004–2008) with some modifications made to the structure and mode of operation by the Steering Committee at its last meeting in Izmir, Turkey, in October 2003.<sup>4</sup> With specific relevance for the Working Group on *Beta*, it should be noted that the former Industrial Crops Network changed its name to the "Sugar, Starch and Fibre Crops Network", which includes three Working Groups (*Beta*, Potato and the new Working Group on Fibre Crops (Flax and Hemp)).

The Steering Committee endorsed four priority areas for Phase VII: 1) Characterization and evaluation; 2) Task sharing; 3) *In situ* and on-farm conservation; and 4) Documentation.

The Steering Committee also requested a Network Coordinating Group (NCG) to define two priority groups within the Network and to make proposals, in consultation with the Working Groups, for actions on the basis of a budget of about 83 000  $\in$  allocated to the Network. As a result of this exercise, carried out during 2004, the Working Group on *Beta* was included among the priority Working Groups for Phase VII, together with Fibre Crops (Flax and Hemp). The following use of funds relevant for *Beta* was eventually approved:

- June 2005: Network database managers meeting (3600 €)
- March 2006: Third *Beta* WG meeting (11 500 €)
- March 2006: *Beta in situ* assessment day (3600 €)
- March 2006: Meeting of all Networks' Coordinating Groups (on a different budget line)
- 2006: Beta WG sub-coordinators ad hoc meeting (2000 €)
- Publication of meeting report (4000 €).

For further information on ECP/GR, the ECP/GR Web site can be consulted, where several reference documents are available, including the Networks' budget and the Terms of Reference for the ECP/GR operational bodies. A specific Web page is also dedicated to the Working Group on *Beta*, and this can be improved with the help of Group members and according to the needs of the Working Group.

## A European Genebank Integrated System (AEGIS)

A short account was given of the ECP/GR-funded project AEGIS (A European Genebank Integrated System), which is planning, initially through a feasibility study, to promote the creation of a rational European plant genetic resources genebank system of genetically unique and important accessions, in order to conserve them safely in the long term, at the same time ensuring their genetic integrity, viability and availability to users. According to a draft "Strategic Framework" document which was prepared during the feasibility study, the operational principles of an integrated system for the operation of genebanks in Europe would be the following:

<sup>&</sup>lt;sup>4</sup> See Report of the Ninth Steering Committee Meeting, also available on Internet at http:// www.ecpgr.cgiar.org/SteeringCommittee/SC9.htm

- Transparency of information
- Agreed quality standards of operation
- Joint planning for rational conservation
- Sharing resources (facilities and expertise)
- Joint financing (common fund)
- Ready and easy access to germplasm.

In the most likely scenario, germplasm would continue to be conserved in the same location where it is currently stored, but management of conservation of the accessions which are voluntarily designated by the member countries as being part of the system would be coordinated regionally at the crop level. ECP/GR Crop Working Groups would prepare and coordinate implementation of crop conservation action plans, possibly delegating (part of) these tasks to Coordinating European (Lead) Institutions.

Principal benefits of the operation of A European Genebank Integrated System (AEGIS) would be the following:

- Improved collaboration among European countries and a stronger unified Europe
- Cost-efficient conservation activities
- Reduced redundancy in European collections
- Improvement of quality standards across Europe
- More effective regeneration
- Facilitated access to germplasm
- Improved security of germplasm through safety-duplication
- Improved linkages to in situ conservation and users.

The AEGIS establishment process foresees the need for a formal approval by the ECP/GR Steering Committee in September 2006 of the "Strategic Framework" and of the establishment process itself, including the definition of a financial strategy. ECP/GR member countries would then be offered the opportunity to sign a Collective Memorandum of Understanding, defining countries' responsibilities.<sup>5</sup>

More information on AEGIS is available from http://aegis.cgiar.org/.

### **Country reports**

#### Critical assessment of the achievements at country level

The Chair asked attending members of the Working Group to report highlights of national genetic resources activities carried out since the last meeting of the Group, in particular on the following subjects:

- In situ management
- On-farm management and management strategies complementing the *ex situ* work

<sup>&</sup>lt;sup>5</sup> The Strategic Framework paper was finalized as a "Policy Guide" in January 2009 (available at http://aegis.cgiar.org/documents/constitutional\_documents.html): ECPGR. 2009. A Strategic Framework for the Implementation of A European Genebank Integrated System (AEGIS). A Policy Guide. European Cooperative Programme for Plant Genetic Resources (ECPGR). Bioversity International, Rome, Italy.

- Data exchange
- Regeneration guidelines
- Quality standards
- Task sharing
- Research and utilization
- Funding opportunities.

Available full papers of the presentations summarized below are included in Part III of the present report.

An additional paper on "Beta genetic resource activities in India (1990-2005) – a review" was provided by H.M Srivastava and is also included in Part III (pp. 94-103).

#### Caucasus/Transcaucasus

(See also above, p. 4 and full paper, pp. 38-44).

Guram Aleksidze described the situation for the entire Transcaucasus area, explaining that Azerbaijan is in good position to make progress with *in situ* management, since an experimental station has been established for this purpose; also that Armenia and Iran have already completed excellent preliminary surveys. On the other hand, in Georgia it is still necessary to first undertake surveys of the resources existing *in situ*.

Cooperation among Caucasus countries was said to be very good, especially for exchange of information. The need to identify funding opportunities is common to all the countries.

#### Germany

#### (paper pp. 90-93)

Lothar Frese introduced the main highlights for Germany:

- *In situ* management: only on the North Sea island of Helgoland and in a small area in the Baltic Sea region south of Denmark do a few wild *Beta* populations occur which have been surveyed. It seems that *B. vulgaris* subsp. *maritima* is expanding in the Baltic Sea area, a development which was not expected 10-15 years ago. Opportunities for *in situ* management are very limited in Germany.
- **Data exchange:** international cooperation is exemplified by the commitment to manage the International Database for *Beta* (IDBB), even though there are often insufficient personnel to properly administer the database.
- Regeneration guidelines: all curators have returned information related to regeneration practices and the document is online on the ECP/GR Web site. This document needs continual amendments. The Institute of Plant Genetics and Crop Plant Research (IPK) will need to make improvements in the course of its ongoing reorganization.
- **Quality standards:** the merger of the two German collections will result in the definition of a concept for quality standards, which will be developed by IPK (in a similar way to what was done by the Centre for Genetic Resources, Wageningen (CGN), where the ISO standards were adopted).
- **Research and utilization:** very few institutes are undertaking this task: some are, such as the Institute for Sugarbeet Research at Göttingen and the University of Kiel (looking for resistance genes and investigating the bolting

gene). The BAZ is a federal breeding research centre having no clear mandate for *Beta* breeding research.

Funding opportunities: EC Regulation 870/04 is one opportunity, but there are also several foundations which could be tapped for funds. For example, the Humboldt Foundation allows senior scientists to undertake sabbatical periods in Germany. The Humboldt Foundation also manages the Marie-Curie Foundation, which allows exchanges of young female scientists, on the basis of very competitive grants. One example of a project funded by the Marie-Curie Foundation is a botanic survey of the mountain species of Greece.

The Group was reminded that no further seed requests can be processed by BAZ Braunschweig, and that they should be sent to IPK.

#### Ireland

(paper p. 104)

Dermot Grogan explained that Ireland needs to develop a strategy for dealing with *in situ* populations, following the completion of the wild *Beta* survey. A proposal will be prepared with the aim of promoting the establishment of a national conservation policy.

Regeneration of the existing seed bank germplasm is needed, but this task cannot be proposed for any realistic plan at present, since funds for conservation are being focused exclusively on crops with national importance, i.e. potato and forages.

Data on *in situ* accession sites are available.

Very limited research and utilization is carried out.

Funding is available in Ireland through co-funding at 50% and this opportunity should be available every year for the next two years to undertake work on maritime beets.

#### Italy

Enrico Biancardi reported that locations of new sites of sea beets were found near Bari, Foggia and in other places. New localities were also found in Croatia, within the context of the EU-funded Sixth Framework Programme project for Sustainable Introduction of GMOs into European Agriculture (SIGMEA).

The overall objective of SIGMEA is to set up a science-based framework, strategies, methods and a practical toolbox for assessing ecological and economic impacts of genetically modified crops and for effective management of their development within European farming systems. The main objectives of the Project are: i) to bring together programmes studying crop-to-crop gene flow across Europe within a diversity of agricultural systems; ii) to collate and synthesize existing and developing experimental information on gene flow and environmental impacts; iii) to conduct landscape scale evaluations of gene flow and develop biogeographical models of outcrossing, seed dispersal and persistence, covering whole farms and regional scales.

The aim of the researches carried out at Rovigo is the detailed study of variation in natural populations of sea beet in some areas of Italy and Croatia. A further objective is the evaluation of the influence of the presence of cultivated beet on the biodiversity of the populations in these areas. Data on population genetic structure would allow an estimation of gene flow between crop and wild populations of beet in the central Mediterranean area. The information gained during this study would be of great value for the estimation of gene flow, the protection of the biodiversity within sea beet in relation to the influence from cultivated beet, and for the correct management of the sugar beet crop.

The Istituto Sperimentale per le Colture Industriali (ISCI) in Rovigo is continuing to collect in the Po Delta and the Venice lagoon. The collected seed samples are then grown in a greenhouse for evaluation. Correlation was established between stress situations (drought) and the development of the root system. This work is carried out in collaboration with Mitch McGrath from the USDA-ARS of East Lansing, Michigan.

#### Poland

#### (*paper pp.* 107-108)

Kamilla Kuzdowicz explained that the *Beta* collection in Poland is situated in the Bydgoszcz Research Division of the Plant Breeding and Acclimatization Institute, as part of the National Centre for Plant Genetic Resources (NCPGR) based in Radzików which coordinates, finances and provides storage facilities for crop genetic resources in Poland.

There are no species of *Beta* in the wild flora of Poland. The *Beta* collection in Poland consists of wild species from other countries, old cultivars and breeding materials of sugar and fodder beets. At present, it contains 343 accessions: 112 sugar beets, 199 fodder beets and 32 wild forms belonging to sections *Beta*, *Corollinae* and *Procumbentes*. This collection is conserved in the Long-Term Storage Laboratory in Radzików as seed samples kept in glass jars at -15°C and 5-8% moisture content. Accessions were obtained mainly from national breeding institutions and through exchange among beet collections and foreign research laboratories. Evaluation for morphological, cytological and biochemical traits, seed quality and seed germination tests are carried out in Bydgoszcz. Each year 20-25 accessions are evaluated. During the last ten years some of the accessions have been evaluated for two economically important beet diseases: *Aphanomyces cochlioides* Drechsler and *Cercospora beticola* Sacc. At present, accessions are being evaluated for tolerance to drought stress. The collected and evaluated germplasm is used in sugar and fodder beet breeding and in several research programmes. Information and seed samples are distributed freely.

#### United Kingdom

Brian Ford-Lloyd explained that a "Gene flow" project had been completed and the results were in press. This work includes a genetically modified organism (GMO) risk assessment study.

The University of Birmingham is engaged in studies on prioritization of wild beets for conservation and a lot of experience was accumulated by doing threat assessment red-listing. There is now an International Union for Conservation of Nature (IUCN) Species Survival Commission (SSC) Specialist Group for crop wild relatives. Threat assessment red-listing for *Beta* was initiated and the work will be completed in six months for submission to the Specialist Group for consideration. There may be a need to contact members of the *Beta* WG in the near future in order to complete this work. E. Ober reminded the Group that Broom's Barn participated in the *Beta* GENRES project. Accessions were evaluated in terms of drought stress and disease resistance and these descriptors' data are now publicly available in the IDBB. Standardization of abiotic stresses is still considered to be an issue where an agreed standardized test is required. Evaluation of phenotypic characteristics could be worth the establishment of a consortium project, on the model of the previous GENRES project. This project could be multicrop and include abiotic stresses.

A large project on *Polymyxa* resistance is ongoing, looking for new sources in genebank material. A likely source of resistance was found and a mapping exercise is currently going on.

Storage conditions for material at Broom's Barn are not ideal (glass jars kept at 5-7°C), with limited storage life. A database system is used to manage the institute's seed inventory, with a bar-coding system for each stored accession. The system can be shared with other institutes upon request.

Regarding the opportunity to standardize tests for abiotic stresses, C. Germeier commented that it is more important to describe exactly how the tests are done than to standardize the test precisely, since the methods used are always evolving due to the continual progress of modern technologies.

#### Statements and comments of other WBN countries

The Chair asked representatives of other WBN member countries to add comments, if they so wished, reporting the situation in their respective countries.

#### Iran

Mohammad Nasser Arjmand informed the Group that exploration will be continued to find new sites of *Beta* germplasm. Characterization and evaluation activities will also continue, as well as regeneration. Recently, 85 landraces of garden beet were characterized and evaluated for several traits.

The Sugar Beet Seed Institute (SBSI) *Beta* genebank contains beet germplasm plus varieties from Germany, Poland and USA, as well as local and foreign species, landraces and breeding material.

Information is openly exchanged with all colleagues.

Many projects are going on regarding utilization of material resistant to *Rhizoctonia* and other diseases. All activities are focused on sugar beet.

The Ministry of Agriculture is funding the national projects and funds from international organizations would be welcome

#### Morocco

#### (paper pp. 105-106)

Activity on wild *Beta* genetic resources is new for Morocco, but there is an active beet breeding programme, going on in parallel with the identification of suitable sites for sugar beet seed production, which are limited in Morocco. Collecting of wild species seed samples has started. Some plants are being evaluated for phenotypic traits.

Projects are taking place to evaluate biotic and abiotic (salinity and drought) resistance in wild populations and plans were made to continue exploration of the

distribution of wild species in Morocco. The opportunity to link with the ongoing work of S. Villain on the distribution of *B. macrocarpa* was an immediate result of interactions established during the current meeting.

Funding for *Beta* genetic resources are very limited, since this is not a priority for Morocco.

The opportunity for breeding local sugar beet varieties is under investigation. There are a few cultivated garden beets, but all are foreign varieties.

#### Spain

Arnoldo Santos explained that work on taxonomy is carried out in the Canary Islands. He would like to get engaged in projects on morphology, taxonomy, genetic diversity and typification of Macaronesian material. A project is in progress to collect seed for the botanic garden seed bank in Gran Canaria., while a project is being started to collect seed in Tenerife for a local seed bank.

The availability of help for interested scholars to collect wild *Beta* material in the Canary Islands was re-confirmed.

#### Sweden

Geert Janssen explained that the Syngenta breeding company does not have a genebank for wild species, but only a collection of hybrids and landraces. Discussions are in progress on whether this collection should be passed on to the Nordic Gene Bank.<sup>6</sup>

He thought that it would be desirable to ensure some commitment from breeding companies for genetic resources collaboration and for funding, especially for precompetitive research. Ideally, breeding companies could join the effort in order to offer a joint commitment for the conservation of genetic resources.

#### USA

The main activity is evaluation. There are 2513 accessions conserved, including lines, old varieties and 571 *Beta vulgaris* subsp. *maritima*. Much research is based on pathology, ranging from epidemiology to the detection of markers.

An ongoing project is the study of *B. vulgaris* subsp. *maritima* from the coasts of France. Plants are grown in a garden and quantitative measurements are being taken, with the aim of determining where is the largest genetic variation of the collection, and also in order to help identifying suitable candidate accessions for a core collection. Another aim is finding morphological differences and correlating these with genes. M. McGrath is trying to locate genes and doing mapping.

The only collecting mission carried out recently was focused on *Beta nana* in Greece and previously reported (see above, pp. 5-6).

Following an ECP/GR ad hoc meeting on the *Beta* core collection, the national core collection was coordinated with the GENRES *Beta* project core collection.

<sup>&</sup>lt;sup>6</sup> As of 1 January 2008, the Nordic Gene Bank (NGB) has joined with the Nordic Forestry Resource and Nordic Animal Genetic Resource institutes to form NordGen, the Nordic Genetic Resource Center of the five Nordic countries (Denmark, Finland, Iceland, Norway and Sweden).

#### Standardized reporting format and future workplan

L. Frese presented to the Group the draft standard report of the activities of the Working Group, which is expected to be submitted to the ECP/GR Steering Committee for consideration before its September 2006 meeting. He opened the discussion on the revision and finalization of this document.

Discussion took place on whether the previously planned sub-working group moderators' meeting should actually be held, and it was no longer considered necessary. It was preferred to utilize the available funds for a technical meeting aiming at the development of a descriptor list and data model for *in situ* monitoring of wild *Beta* species (see Appendix I, section III. Plans).

Regarding germplasm evaluation, it was decided to publish the newsletter on evaluation and pre-breeding being compiled by E. Ober, and then to consider the opportunity for the preparation of a project. Many workers know what the other groups are doing, but there could be developing countries' groups which are not aware of what is going on. It seems sensible to first verify who is doing what through the newsletter, then possibly to arrange a meeting and plan joint activities.

The need to include evaluation data into the IDBB was stressed. Currently it is possible to send evaluation and characterization data to the IDBB manager in Excel format (see report of the second meeting, 2002, Bologna).<sup>7</sup> A possibility for the future should be to enable everybody to input their own data online. This would require a major programming project.

It was commented that the update of the IDBB was being kept on hold until the AEGIS project can give an indication of the future of the ECCDBs. The hope is that database managers will be able to operate more rationally after this reorganization. Another reason not to proceed with updating in the traditional way is the recent possibility offered by the European Internet Search Catalogue (EURISCO) of directly downloading passport data.

Further work on the quality concept was also put on hold, pending the development of AEGIS. Development of a quality concept for *Beta* is a task for genebank managers.

The revised standardized reporting format, including results, contributions to priority activities, analysis and plans for the future of the Working Group, is attached as Appendix I (pp. 115-120).

#### Establishment of ad hoc working groups, if desired

A group composed of L. Frese, C. Germeier, B. Ford-Lloyd, A. Santos Guerra, D. Grogan and B. Harris was formed and met in the morning of 10 March to discuss the preparation of a project to be submitted to the second call of EC Regulation 870/04. The project could become a workpackage of the planned European Genetic Resources *In Situ* Inventory (EGRISI) project or a separate one. As opposed to the current EGRISI concept, the new workpackage would include field work in a limited number of potential genetic reserves. The group wishes to include non-EU countries (Caucasus/Transcaucasus and North Africa) in the project to cover the whole range of life forms of *Beta* and a broad range of sites with distinctive features.

<sup>&</sup>lt;sup>7</sup> Frese L, Germeier C, Lipman E, Maggioni L, compilers. 2004. Report of a Working Group on *Beta* and World *Beta* Network. Second joint Meeting, 23–26 October 2002, Bologna, Italy. International Plant Genetic Resources Institute, Rome, Italy.

Focusing on the climate change issue, B. Ford-Lloyd had compiled further suggestions that could be elaborated in a project proposal which could be for instance submitted under the Seventh Framework Programme of the EU.

#### **Recommendations and conclusions**

Many data on new collection sites have been generated by surveys and research projects that can be used to produce point precise distribution maps of Beta. However, these do not always relate to ex situ genebank samples. When new structures become available, it will be possible to document this kind of material in the IDBB. How to manage and publish them seems also to be a legal and political matter. It will be necessary to find a legal solution to make it possible for the CCDBs to operate. One possible solution will be to make data providers aware that uploading the data implies that these are held in the public domain.

The meeting was a joint one with botanists. It is recommended to encourage cooperation between botanists/conservation biologists and the PGR sector to allow for better exchanges of knowledge and information. Close cooperation between such experts is an essential requirement for the implementation of the in situ management concept in practice.

Changes in European agriculture impact on sugar beet production acreages, and there is growing interest in production areas outside Europe: production there may lead to new pest and disease problems. Though the production of sugar within the EU may decrease, the need for breeding improved varieties adapted to new growing areas and the need for genetic resources to be well maintained ex situ and in situ will remain unchanged.

*Climate change consequences for the maintenance for* **Beta** *in natural habitats were discussed in relation to drought, flowering patterns and changes in disease patterns. These are considered issues for future research targets.* 

Countries located in important parts of the distribution areas of Beta are encouraged to seriously consider nominating members to attend the Working Group meetings.

### Visit to the Institute

The botanic garden of La Orotava is the second oldest of Spain, founded in 1788. It is part of the Canarian Institute for Agricultural Research. There are two units in the gardens, i.e. the "botanical garden" and the "botanical unit", which does research. There are 40 000 stored herbarium sheets.

A. Santos guided the participants around the garden and also showed them a population of *B. procumbens* growing in the garden.

## Introduction to in situ management methodology with emphasis on threat assessment

The quantitative method for genetic erosion risk assessment developed by Guarino  $(1995)^8$  was introduced to the Group as well as some descriptors suited to describe

<sup>&</sup>lt;sup>8</sup> Guarino L. 1995. Assessing the threat of genetic erosion. In: Guarino L, Ramanatha Rao V, Reid R, editors. Collecting Plant Genetic Diversity, Technical Guidelines. CAB International, Wallingford, UK. pp. 67-74.

populations located in a genetic reserve. The applicability of Guarino's method and the descriptors were discussed in the field on Saturday 11 March (see Part II, pp. 21-24).

### Conclusion

#### Election of the Coordinating Committee of the ECP/GR Working Group on Beta / World Beta Network

The Group wished to thank Lothar Frese for effectively chairing the ECP/GR Working Group for several years and offered him the opportunity to continue in this role until the next meeting of the Group. He kindly accepted this offer and suggested, with the consent of the Group, nominating Guram Aleksidze as Vice-Chair: he kindly accepted.

Dermot Grogan was elected Chair of the Coordinating Committee of the World *Beta* Network.

Sub-working groups' moderators were also reconfirmed:

Eric Ober (Evaluation and breeding)

Brian Ford-Lloyd (Genetic diversity)

Ayfer Tan (Genetic resources management).

#### **Closing remarks**

The Group discussed the possible location for the next meeting. It was suggested that the meeting could be held in 2009 either in France or in the location where the meeting of the Study Group Breeding and Genetics of the International Institute for Beet Research (IIRB) will be held. It was also acknowledged that Dr H.M. Srivastava had kindly offered to hold the next meeting in India. However, considering that the largest part of the group is based in Europe and that ECP/GR only provides funds for meetings in Europe, it was not considered practical to accept the offer from Dr H.M. Srivastava, which was politely declined.

## PART II. VISIT TO KNOWN *BETA* COLLECTING SITES. TRAINING ON THREAT ASSESSMENT AND POPULATION MONITORING IN THE NATURAL HABITAT

An excursion to *Beta* collecting sites visited 25 years ago was organized on 11 March by the Instituto Canario de Investigaciones Agrarias on request of the Working Group. The sites visited were the following:

- 1. Near Garachico Village
- 2. Caleta de Interián-Sibora
- 3. Punta de Teno
- 4. Masca
- 5. Parque Nacional Cañadas del Teide.

This activity aimed at the discussion of a quantitative approach to estimate the threat of genetic erosion that a particular taxon faces in a defined area (Guarino 1995)<sup>9</sup> for two reasons. Firstly, the risk of extinction of a particular species is a function of the loss of genetic variability within individual populations of that species and in consequence its adaptability. Secondly, if a particular population is to be selected for protection in a genetic reserve, a threat assessment is required to determine the survival chances of this population. Factors affecting the genetic diversity and persistence of this population can then be reduced or eliminated. The discussion was held at the sites to check the descriptors elaborated in the office against the real data recording situation during a collecting or monitoring mission.

It turned out very soon that the descriptors can roughly be categorized into those that are related to the specific population and can only be recorded at the site, partly with the help of local administration and experts, and those that can only be found in information sources such as flora, habitat maps or weather stations. The descriptor list and notes referring to the discussions are provided in Table 1. Descriptors more related to the population for which the assessment is being made were qualified with the addition "at site". The model promoted by Guarino (1995) does not yet sufficiently take into account the species biology. Some species may even need accidental fires (descriptor 1.4) or some degree of disturbance resulting from grazing (descriptor 3.9) or irrigation schemes (descriptor 3.13) to form persistent populations. A ratio of present livestock density to estimated carrying capacity at a site of <0.5 (score 0) may even impair the species survival. It was recommended not to mix site and plant traits in a single descriptor. Instead, the threat should be calculated by weighting site traits with biological traits of the species. Any population threat assessment must always be species-specific and reasoned in the context of its ecology and ought to consider the specific conditions at the site.

<sup>&</sup>lt;sup>9</sup> Guarino L. 1995. Assessing the threat of genetic erosion. In: Guarino L, Ramanatha Rao V, Reid R, editors. Collecting Plant Genetic Diversity, Technical Guidelines. CAB International, Wallingford, UK. pp. 67-74.

### Table 1. Population threat assessment (based on Guarino 1995)

(the scores do not refer to the assessment of a specific population, but are given as an example of the types of scores that were obtained during assessment of *Beta nana* populations in Greece)

Factor	Score	Scored	Data source
1.1 Taxon distribution			
Rare	10		
Locally common	5	5	CWRIS, CCDB
Widespread or abundant	0		
1.2 Drought			
Occurred in 2 or more consecutive years	10		
On average one or more times every ten years, but not in consecutive years	5		Closest weather station
Less than once every ten years	0	0	
1.3 Flooding, at site			
Area known to be very flood prone	10		Topographic map, local
Area not known to be flood prone	0	0	administrations such as coast guards
1.4 Accidental fires, at site			
Area known to be very prone to fires	10		Local administrations such
Area not known to be prone to fires	0	0	as forestry departments
1.5 Potential risk from global warming			
Summit areas or low-lying coastal areas	10	10	Topographic map, closest weather station, botanical surveys
3.1 Extent of wild habitat of target species within stu	dy area		
Very restricted (<5%)	15	15	
Restricted (5-15%)	10		Geological & soil &
15-50%	5		topographic maps, in particular babitat maps
Extensive (>50%)	0		particular habitat hiapo
3.2 Conservation status of target species			
Species not known to occur in any protected area	10		
Species known to occur within a protected area, but protection status poor or unknown	5	5	EUNIS, CWRIS, national and state red lists
Species known to occur within a protected area, and protection status good	0		
3.3 Extent of use of wild habitat of target species, at a	site		
Industrial exploitation	15		
Exploitation by surrounding populations (e.g. fuelwood gathering from nearby towns)	10	10	Local administrations,
Hunting and gathering by small local communities	2		NGOS
Completely protected	0		
3.4 Extent of use of target species, at site			
Industrial exploitation	15		
Exploitation by surrounding populations	10		Local administrations,
Local exploitation	5		NGOs
Protected or not used	0	0	

# **Table 1. (cont.)** Population threat assessment (based on Guarino 1995) (the scores do not refer to the assessment of a specific population, but are given as an example of the types of scores that were obtained during assessment of *Beta nana* populations in Greece)

Factor	Score	Scored	Data source			
3.5 Agricultural pressure on wild habitat, at site						
Large-scale cultivation within habitat margins	15	15				
Subsistence cultivation areas within habitat margins	12		Local administrations			
Land suitable for cultivation, cultivated areas within 3 km of habitat margins	5		georeferenced images			
Land unsuitable for cultivation	0					
3.6 Human population growth rate per year, close to site						
>3%	10					
1-3%	5		Local administrations			
<1%	0	0				
3.7 Availability of agricultural land, at site						
> 70 ha / sqkm cultivated	10	10				
30-70 ha / sqkm cultivated	5		Local administrations,			
< 30 ha / sqkm cultivated	0		georereneed integes			
3.8 Species palatability						
High	10					
Medium	5	5	CWRIS			
Low	0					
3.9 Ratio of present livestock density to estimated car	rrying cap	oacity at s	ite			
>1	10					
0.5 – 1	5		Local administrations			
<0.5	0	0				
3.10 Average proximity to borehole or other all-year ro	ound wate	er supply	at site			
<10 km	10	10				
10-20 km	5		Survey, collecting			
>20 km	0		111331011			
3.11 Distance to major population centre, at site						
<20 km	10	10				
20-50 km	5		Survey, collecting			
>50 km	0		111331011			
3.12 Distance to major road, at site						
<10 km	10	10				
10-30 km	5		Survey, collecting			
>30 km	0		111331011			
3.13 Distance to development projects (irrigation sche	me, touris	m comple	ex, mining site), at site			
<20 km	10	10				
20-50 km	5		Survey, collecting			
>50 km	0		111551011			
MAXIMAL POINTS = 200						
SUM OF ACTUAL ASSESSMENT		115				

It was also noted that the demographic structure of a "population" cannot be determined readily in species having a prostrate growth habit and where plants deposit seeds in the immediate vicinity of the mother plant. These features impede the identification of single plants in *B. patellaris*, *B. procumbens* and *B. webbiana* which form closed canopies. Individuals of species of the sections *Beta*, *Nanae* and *Corollinae* can be identified more easily.

Before doing any threat assessment, some terms used in the descriptor list have to be defined, such as "human population growth rate", "major population centre", "major road" and geographic terms such as "region", "area", and "site" as well as terms like "habitat" and "patch" which compound biological aspects with geographical. The human population growth rate in a remote study area may be quite different from the national average growth rate and trends in population movement may also need to be considered. In Azerbaijan young people are moving back from the major cities into remote areas while in Greece the rural population is decreasing. Both demographic trends have their specific impacts on land use.

An interesting discussion began when the participants standing at the first site tried to define the limits of the *Beta* population growing there. Since this is impossible without any further knowledge of the species' geographic distribution pattern, spatial barriers between groups of plants and the extent of gene flow, it was suggested that observers should take records on defined patches of plants and keep these data separate. The limits of a "population" may be determined by genetic distance measures later and, in the case of clinal variation, adjacent groups of plants can perhaps be delineated by definition of threshold levels of genetic distance.<sup>10</sup>

In summary, the excursion was not only enjoyable but also instructive.

<sup>&</sup>lt;sup>10</sup> (Note by L. Frese on 16 March 2006): The task of defining a population is well known in forest genetics where the delineation of provenances is an important issue. See: Kleinschmit JRG, Kownatzki D, Gregorius H-R. 2004. Adaptational characteristics of autochthonous populations – consequences for provenance delineation. Forest Ecology and Management 197:213-224.

## PART III. PRESENTATIONS AND PAPERS

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## Prioritization of wild Beta species for conservation: the PGR Forum experience

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#### Introduction to PGR Forum

Coordinated by Nigel Maxted, Brian Ford-Lloyd and Shelagh Kell at the University of Birmingham, PGR Forum was a project funded under the European Union Fifth Framework Programme for Energy, Environment and Sustainable Development. The project brought together 23 partners from 21 countries across Europe, together with partners representing the International Union for Conservation of Nature (IUCN) and the International Plant Genetic Resources Institute (IPGRI, now Bioversity International). A broad cross-section of the professional European plant genetic resources (PGR) community was represented, including PGR conservationists, policy-makers and end-users. PGR Forum's objective was to provide a European forum for the assessment of taxonomic (species) and genetic (molecular) diversity of European crop wild relatives, and to develop appropriate methodologies that could be applied to conserve this diversity.

#### Crop Wild Relatives (CWRs)

In order to address the main aims and objectives of PGR Forum it was necessary to provide a definition of a crop wild relative. The definition adopted was "A crop wild relative (CWR) is a wild plant taxon that has an indirect use derived from its relatively close genetic relationship to a crop; this relationship is defined in terms of the CWR belonging to gene pools 1 or 2, or taxon groups 1 to 4 of the crop" (Maxted et al. 2006). In the context of PGR Forum "Crops" included food, fodder, industrial and forage crops, medicinal plants, condiments, ornamental and forestry species, minor crops and underutilized species.

#### The CWR Catalogue

The development of the "PGR Forum CWR Catalogue for Europe and the Mediterranean" was one of the major outputs of PGR Forum (Kell et al. 2008). The methodology used for creating the initial list of European CWRs was a process of data harmonization and cross-checking between a number of databases, PlantBase (http://www.emplantbase. primarily Euro+Med and org/home.html) Mansfeld's World Database of Agricultural and Horticultural Crops (Hanelt and IPK 2001; http://Mansfeld.ipk-gatersleben.de/ Mansfeld/). The initial list was therefore created by selecting the taxa contained in Euro+Med PlantBase within genera matching those contained in Mansfeld's World Database of Agricultural and Horticultural Crops. The same process was then repeated for forestry, ornamental, medicinal and aromatic plant species. A list of forestry genera was extracted from "Enumeration of cultivated forest plant species" (Schultze-Motel 1966); a list of ornamental genera was obtained from the Community Plant Variety Office (CPVO); and medicinal and aromatic plant genera were extracted from the database MAPROW (Medicinal and Aromatic Plant Resources of the World) (Shippmann 2004, personal communication).

It had been agreed early on in the process that it should be as inclusive as possible, and to include all categories of plants of socio-economic importance: food, fodder/forage, industrial, forestry, ornamental, herbs and spices and medicinal.

#### **Prioritization of taxa**

The final Catalogue contains in excess of 25 000 species and more than 273 000 records of taxon occurrences in 130 geographical units representing 58 nations across the Euro-Mediterranean region (see http://www.pgrforum.org/cwris/cwris.asp). More than 17 000 of these species occur in Europe alone. In order to effectively plan conservation it is necessary to prioritize this list of CWRs in order to determine which are of immediate concern, and which, in the European context, should receive the most attention because they are under threat in some way and/or are of significant potential economic value. Many of the taxa in the CWR list do not require any conservation action, while others are relatively unimportant as genetic resources in Europe.

By way of Euro+Med Plantbase (http://www.emplantbase.org/home.html) it has been possible to determine in how many geographical units every taxon within the CWR catalogue has been recorded. The argument was made for using this as a simply proxy for threat because the more geographical units in which a taxon occurs, the less likely it is to be under threat in all of the units, simultaneously needing urgent conservation action; generally speaking if a taxon has been recorded in 15 or 20 countries in Europe, it is unlikely to be in danger overall. Taxa which are endemic to a particular geographical unit or country may or may not actually be threatened, but should receive initial attention in terms of conservation to ensure that their future is secured as much as possible.

Summarizing why we need to prioritize:

- It is not possible to measure or monitor genetic erosion and pollution, or design *in situ* reserves for all 17 000 CWR species in Europe.
- We do not have threat assessments for all 17 000 species for all of Europe (but they may exist nationally).
- We need to be able to identify the taxa on which it is most important to spend time and money.
- Giving red-listed CWR taxa high priority for conservation would be an obvious way forward for us in dealing with the 17 000 species, but the 2004 IUCN Red List of Threatened Species, only listed 161 CWR species occurring in the Euro-Mediterranean region that have been globally red-listed, and nearly all of these are tree species.

Applying this simple proxy criterion to the CWR catalogue of over 25 000 species resulted in a substantial reduction in the number of species that could be recommended for further conservation assessment. There were around 2500 species occurring in fewer than 10 Euro+Med geographical units.

#### How are wild Beta species placed in terms of this prioritization?

Using occurrence in fewer than 10 geographical units as the cut-off (i.e. listing any taxon which occurs in fewer than 10 geographical units), what are the priorities for *Beta* CWR *in situ* conservation? Table 1 indicates that at least 11 out of the 14

wild species require conservation action, although the situation with regard to the *Patellifolia* species (former *Beta* section *Procumbentes*) requires further scrutiny because of taxonomic uncertainty. Notwithstanding this, two of the three *Patellifolia* species clearly do require consideration for conservation based upon this simple approach to prioritization, and are clear targets for *in situ* conservation by way of the establishment of *in situ* reserves.

Genus	Species/subspecies	No. of geographical units in which taxon occurs	Prioritize?
Beta	vulgaris subsp. adanensis	5	Yes
	corolliflora	3	Yes
	intermedia	1	Yes
	lomatogona	3	Yes
	macrocarpa	20	No
	macrorhiza	2	Yes
	nana	1	Yes
	patula	2	Yes
	trigyna	16	No
	<i>vulgaris</i> subsp. <i>maritima</i>	62	No
Patellifolia	patellaris	16	(*)
	procumbens	10	Yes
	webbiana	5	Yes

Table 1. Prioritization of 13 wild relatives of cultivated beet

(\*) The situation with regard to the *Patellifolia species* (former *Beta* section *Procumbentes*) requires further scrutiny because of taxonomic uncertainty.

#### Further prioritization and conservation measures for beet CWRs

PGR Forum recommended further prioritization based upon economic/use value (Ford-Lloyd et al. 2008). It is likely that there will be substantial agreement that wild beet taxa should be highly prioritized as they are relatives of food, fodder and industrial crops. If ease of use, meaning how easy it is to use the taxa for transferring genes into the crop, is to be considered an important criterion, then this would affect the prioritization. Those wild species occurring in the same section of the genus as cultivated beet (section *Beta*) would then change in priority, species belonging to the section *Beta* receiving higher prioritization than those in sections *Corollinae* and *Nanae* as well as in the genus *Patellifolia*.

The prioritization would further be altered if current conservation status taking into account *ex situ* holdings, were to be determined via a gap analysis (Scholten et al. 2008). Then it is likely that prioritization would move in favour of the taxa outside section *Beta*.

We hope that this account serves to illustrate that determining priorities for conservation is complex, and is likely to vary among different user communities and different nationalities. It could be argued that it illustrates that a simple approach (involving the proxy threat criterion as above) will allow a more rapid planning of conservation action. This should take the form of identifying the extent to which prioritized taxa already exist in protected areas, and ensuring that those protected area management plans take into account the existence of these important CWR genetic resources. For those taxa where such protection does not already exist, then the process of designing and establishing new reserves needs to be initiated (Scholten et al. 2008), ensuring that this is coordinated on a Europe-wide basis.

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# Reconstructing the evolutionary history of Beta section Beta with molecular data: a focus on the Canary Islands

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### Introduction

The genus *Beta* belongs to the subfamily Betoideae and the family Chenopodiaceae. Sections within the genus *Beta* were first proposed by Transhel (1927). Different authors have modified these sections over the years (Ulbrich 1934; Buttler 1977; Ford-Lloyd 1986; Letschert 1993; Lange et al. 1999; Hohmann et al. 2006; Kadereit et al. 2006). Currently, two sections are recognized in the *Beta* genus: *Beta* and *Corollinae*, the latter now includes the former section *Nanae* (Hohmann et al. 2006). The species and subspecies of section *Beta* are wind-pollinators mainly distributed along the European coasts. The *Beta* section is composed of three almost exclusively coastal species: *Beta vulgaris* (including the subspecies *maritima, adanensis* and *vulgaris* (cultivated forms)), *B. patula* and *B. macrocarpa* (Letschert 1993). Both diploid and tetraploid forms are found within *B. macrocarpa*, which is exclusively distributed on the Canary Islands. The tetraploid type is believed to be a natural amphidiploid hybrid between diploid *B. macrocarpa* and an unknown diploid of the *B. vulgaris* complex (Abe and Tsuda 1987).

## Chloroplastic diversity in the section Beta

The goal of this study is to characterize patterns of genetic diversity within each species of the section *Beta* through chloroplastic fragment sequencing (trnL-trnF, trnD-trnT, trnH-psbA and a large part of matK, with a total of 4kb). This analysis was conducted on a sample covering the geographical distribution of each species (one individual analysed by population), with 33 accessions of *B. vulgaris* subsp. *maritima*, 12 of *B. vulgaris* subsp. *adanensis* and 12 of *B. macrocarpa* (Fig. 1).

*Beta* seeds were obtained from the Federal Centre for Breeding Research on Cultivated Plants, Gene Bank, Braunschweig, from the University of Birmingham, and from the collection held by the University of Lille (Table 1).

Overall, in the section *Beta*, total haplotype diversity  $(0.915 \pm 0.018)$  is relatively high while nucleotide diversity  $(0.00097 \pm 0.00008)$  is quite low. Strong differences in the level of diversity between autogamous (*B. vulgaris* subsp. *adanensis* and *B. macrocarpa*) vs. allogamous (*B. vulgaris* subsp. *maritima*) taxa were observed. An haplotype network was built by statistical parsimony using TCS software (Clement et al. 2000). It is composed of a total of 24 haplotypes relatively close to each other (Fig. 2).



Fig. 1. Map of the samples (Beta section Beta) used for the phylogenetic study.



**Fig. 2.** Haplotype network of the section *Beta*. Haplotypes are indicated by circles. Each line between haplotypes represents a mutational step. The "name" of this line indicates the position of the mutation (ex: LF 124=mutation on the 124th nucleotide on the *trnL*-*trn*F fragment). The black points indicate intermediate haplotype states between observed haplotypes that, however, were absent in our sample.

Taxon / No.	IDBBNR (1)	O_COUNTRY (2)	DISTRICT/LOCATION	DONOR (3)
B. v. maritima				
1	5905	IRL	Sligo	BGRC
2	5915	GBR	North Yorkshire	BGRC
3		GBR	Ramsgate	GEPV
4		GBR	Land's End	GEPV
5		NLD	Zwin	GEPV
6		FRA	Roscoff	GEPV
7		FRA	Charente	GEPV
8		FRA	Erromardie	GEPV
9		ESP	Foz	GEPV
10		ESP	Punta Fouxeira	GEPV
11		ESP	Playa De La Lanzada	GEPV
12	7069	PRT	Obidos	BGRC
13	8550	MAR	Casablanca	BGRC
14	8560	MAR	Essaouira	BGRC
15	8556	MAR	Safi	BGRC
16	6069	PRT	Madeira	BGRC
17		PRT	Ponta Do Pargo	GEPV
18		ESP	Los Arenetes	GEPV
19		FRA	Bages:Leucates	GEPV
20	9452	ITA	Toscana	BGRC
21	9461	ITA	Lazio	BGRC
22	2205	ITA	Sicily	BGRC
23	8615	MLT		BGRC
24	3542	TUN	Sfax	BGRC
25	415	TUN	Bor. Djilidj	BGRC
26	9481	ITA	Veneto	BGRC
27	6952	YUG	Istria	BGRC
28	139	GRC	Levkas	BGRC
29	208	GRC	Khalkidhiki	BGRC
30		GRC	Crete	GEPV
31		GRC	Lesbos	GEPV
32	9742	EGY	Matruh	BGRC
33	8440	TUR	Hatay	BGRC

**Table 1.** Taxa analysed in this study, their sites of origin and their identification number in the International Database for *Beta* 

(1) IDBBNR: Unique identification number assigned to an accession by the International Database for Beta

(2) O\_COUNTRY: country of origin

(3) BGRC: Braunschweig Genetic Resources Collection (Germany)

GEPV: collection of the Laboratoire de Génétique et Evolution des Populations Végétales, University of Lille (France) Birmingham: University of Birmingham collection (UK)

Taxon / No.	IDBBNR <sup>(1)</sup>	O_COUNTRY <sup>(2)</sup>	DISTRICT/LOCATION	DONOR (3)
B. v. adanensis				
a1		GRC	Samos	GEPV
a2	3010	TUR	Canakkale	BGRC
a3		GRC	Lesbos	GEPV
a4	3016	TUR	Izmir	BGRC
а5		GRC	Chios	GEPV
a6		GRC	Kos	GEPV
а7		GRC	Kokinos	GEPV
a8	7119	CYP	Paphos	BGRC
a9	8462	TUR	Aydin	BGRC
a10	3798	ISR	Philistean Plain	BGRC
a11	8623	IRN	Khouzestan	BGRC
a12	8622	IRN	Hormozgan	BGRC
B. macrocarpa				
m1	1570	USA	California	Birmingham
m2	1631	ESP	Fuerteventura	Birmingham
m3	1571	ESP	Tenerife	Birmingham
m4	8569	ESP	Gran Canaria, N	BGRC
m5	8549	MAR	Driouch	BGRC
m6	1771	DZA	Mostaganem	Birmingham
m7		GRC	Chios	GEPV
m8	1188	TUR	Izmit	BGRC
m9	6371	GRC	Karpathos	BGRC
m10	7127	CYP	Limassol	GEPV
m11	4779	PRT	Alcochete	GEPV
m11	4779	PRT	Alcochete	GEPV

Table 1. (cont.) Taxa analysed in this study, their sites of origin and their identification number in the International Database for *Beta* 

(1) IDBBNR: Unique identification number assigned to an accession by the International Database for Beta

(2) O\_COUNTRY: country of origin

(3) BGRC: Braunschweig Genetic Resources Collection (Germany)

GEPV: collection of the Laboratoire de Génétique et Evolution des Populations Végétales, University of Lille (France) Birmingham: University of Birmingham collection (UK)

## • B. vulgaris subsp. maritima

*B. vulgaris* subsp. *maritima* shows the highest levels of diversity of the section *Beta*. This is probably associated with its long evolutionary history and its wide geographical distribution, which would have allowed the accumulation of genetic variation.

One ancestral haplotype of *B. vulgaris* subsp. *maritima*, with a broad distribution, was identified in the middle of the network, suggesting that it was present in ancestral

populations in glacial refuges and subsequently spread to new areas during postglacial recolonization (see also Fénart et al. 2006).

# • B. vulgaris subsp. adanensis

This subspecies showed low levels of polymorphism. That could be the consequence of a recent divergence from *B. vulgaris* subsp. *maritima* within a limited part of the species distribution area (Aegean Islands). The two subspecies, *B. vulgaris* subsp. *maritima* and *B. vulgaris* subsp. *adanensis* show very little taxonomic differentiation according to allozyme data (Letschert 1993). Our results confirm this close relationship. Moreover, the structure of the haplotype network and phylogenetic trees could be the signature of a recent divergence of *B. vulgaris* subsp. *adanensis* starting from *B. vulgaris* subsp. *maritima* in the Aegean Islands, and/or contemporary hybridization between both subspecies.

# • B. macrocarpa 2X

Despite its wide geographical distribution along the southern part of the Mediterranean Basin, diploid *B. macrocarpa* exhibited exceptionally low levels of polymorphism, which might be attributed to historical demographic events in the species. A past dramatic bottleneck followed by a rapid range expansion is suggested by our results and supported by the wide distribution of this species.

# • B. macrocarpa 4X

Since maternal transmission of cpDNA has been found in the majority of flowering plants, the parent that transmitted cpDNA to hybrids is very likely to be the maternal parent (Corriveau and Coleman 1988; Harris and Ingram 1991; Mogensen 1996). The two *B. macrocarpa* 4X accessions share the same cpDNA haplotype found within the *B. vulgaris* subsp. *maritima* cluster (Fig. 2).

Therefore, tetraploid forms of *B. macrocarpa* in the Canary Islands are probably the result of hybridization between *B. vulgaris* subsp. *maritima* as the maternal parent and diploid *B. macrocarpa* as the pollen donor.

## Phylogeography of B. vulgaris subsp. maritima

Chlorotype distribution allowed the identification of two major evolutionary lineages from an ancestral haplotype of *B. vulgaris* subsp. *maritima* which occurs in every biogeographical area.

A first lineage, with the mutation "LF 118", has an Atlantic distribution. The second lineage, with the mutation "LF 124", is distributed east of the Italian peninsula (Fig. 2).

The pattern of haplotype distribution is not consistent with a rapid range expansion where a "star-like" phylogeny of haplotypes would be expected. The lack of population expansion was further supported by the multimodal mismatch distribution and the non significant Tajima's D and Fu & Li's D\* values. Results of mismatch distribution and raggedness index (Slatkin and Hudson 1991; Rogers and Harpending 1992), suggest that population growth in the west Mediterranean sector is more recent than in other domains. Moreover, the shape of the chloroplastic haplotype network and particularly the presence of Mediterranean haplotypes found at the edges of the network (nos. 18, 19 and 23) derived from western and eastern haplotypes could be the signature of post-glacial Mediterranean recolonization from glacial refuges. Nested clade analysis complemented this hypothesis and identified past fragmentation as having had significant effects. The same haplotype (the more common one) might have been present in distinct eastern and western refugia. This genetic structuration is probably a vestige of the glaciation effects but the glacial period(s) involved could not be identified with certainty because of the lack of molecular clock calibration in this coastal plant species.

Nuclear and cytoplasmic data suggest that the present-day *B. vulgaris* subsp. *maritima* populations were derived from two main and different sources: a southwestern European/North African source and an eastern one. The Atlantic and northern Mediterranean coasts were probably essentially colonized from the southwestern refuge whereas the eastern Mediterranean populations are probably derived from eastern refugia.

## Origin and diversity of B. macrocarpa in the Canary Islands

Both diploid and tetraploid *B. macrocarpa* populations were sampled on the five most easterly of the Canary Islands, with eight sympatric locations. Tetraploid populations were found on every island, while diploid populations were mostly found in the eastern ones. Phenotypic and phenological analyses in the greenhouse (under artificial light) show highly significant differences between diploid and tetraploid *B. macrocarpa* (e.g., 4X plants flower earlier, are smaller, and have many more and smaller glomerules than 2X) (data not shown). In natural conditions, the differences between 2X and 4X *B. macrocarpa* are less obvious but glomerules of 4X plants are still smaller and smoother than those of 2X *B. macrocarpa* plants.

A genetic study shows that diploid, autogamous *B. macrocarpa* are monomorphic for all analysed microsatellite markers (10 loci). For the tetraploid form, a maximum of two different alleles per individual is found, suggesting the fixation of alleles through an autogamous mating system in the hybrids. In addition, microsatellite markers provide further evidence for their genetic origin: 1) one allele, i.e. the common allele of all diploid *B. macrocarpa*, is always present in each hybrid, and 2) the second allele is polymorphic among plants and similar to continental *B. vulgaris* subsp. *maritima* alleles.

The close proximity of the Canary Islands to the African continent, and the finding of significant differences in the distribution of alleles (western islands versus eastern ones) support the hypothesis that tetraploid *B. macrocarpa* are the result of at least two independent colonizations/hybridizations in the region. Western 4X *B. macrocarpa* populations have close genetic affinities with Atlantic populations of *B. vulgaris* subsp. *maritima*, whereas eastern Canarian populations are closer to Moroccan *B. vulgaris* subsp. *maritima*. We propose that the allotetraploid *B. macrocarpa* be considered as another species, and therefore, await description. Further investigations are in progress to better describe this potentially new species and understand its origin.

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# Biodiversity of Beta species in the Caucasus Region (Armenia, Azerbaijan, Georgia, Iran)

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# Introduction

The Caucasus hotspot for biodiversity, historically interpreted as the isthmus between the Black and Caspian Seas, covers a total area of 580 000 km<sup>2</sup>. It includes the nations of Armenia, Azerbaijan and Georgia, the North Caucasus portion of the Russian Federation, northeastern Turkey and part of northwestern Iran (Fig. 1).

It is one of the 25 richest and most threatened reservoirs of plant and animal life on Earth. These areas, called biodiversity hotspots, covering only 1.4% of the planet, yet contain 60% of all terrestrial species diversity.



Fig 1. The Caucasus hotspot.

# Importance of crop wild relatives

Crop wild relatives are essential components of natural and semi-natural habitats, as well as agricultural systems, and are critical for maintaining ecosystem health. Their conservation and sustainable use is vital for improving agricultural production, increasing food security, and maintaining the environment.

N.I. Vavilov realized the importance of crop wild relatives in the early years of the 20th century, particularly for their ability to exchange genes with the crops

themselves. Crop wild relative germplasm has been utilized by humankind for thousands of years to improve the quality and yield of crops. Natural crosses between crops and their wild relatives have occurred since the beginnings of agriculture. Farmers have used traditional breeding methods for millennia, and more recently, plant breeders have utilized crop wild relatives' genes to improve a wide range of crops, including wheat (*Triticum aestivum*), maize (*Zea mays*), rice (*Oryza sativa*), barley (*Hordeum vulgare*), potato (*Solanum tuberosum*), cassava (*Manihot esculenta*), and grain legumes such as *Phaseolus*, *Vicia*, *Vigna*, *Lens*, *Lathyrus* and *Cicer*. Improvements include resistance to pests and diseases, and to abiotic stresses such as drought and salinity. Other useful characters include increased protein and vitamin content. The improvement of medicinal plants and pharmaceuticals is another interesting aspect.

Europe is an important centre for crop wild relative diversity. Major crops such as oats (*Avena sativa*), sugar beet (*Beta vulgaris*), apple (*Malus domestica*), annual meadow grass (*Festuca pratensis*) and white clover (*Trifolium repens*) have wild relatives in Europe. Many minor crops have also been developed and domesticated in the region, such as arnica (*Arnica montana*), asparagus (*Asparagus officinalis*), lettuce (*Lactuca sativa*) and sage (*Salvia officinalis*).

# Distribution of Beta species in the Caucasus and Iranian region

#### Taxonomy, genepools and geographic distribution

The wild species of the genus *Beta* are native to Europe and the adjacent areas.

The section *Beta* (primary genepool) occurs along the shores of the Mediterranean basin and along the Atlantic coasts from the Canary Islands in the most southerly part of its distribution, up to the South of Sweden.

The three base species and two hybrid species of section *Corollinae* (secondary genepool) are mainly found in Turkey, the centre of diversity of this section, and in the adjacent Caucasus and Transcaucasus region (Table 1).

Genepool	Taxon	Distribution in the Causasus/ Transcaucasus regions
Primary genepool	Section Beta syn. Vulgares Ulbrich B. vulgaris L. subsp. vulgaris (cultivated beets) Leaf Beet Group Garden Beet Group	Azerbaijan, Armenia, Georgia, Iran
Secondary	Fodder Beet Group Sugar Beet Group subsp. <i>maritima</i> (L.) Arcang. Section Corollinae Ulbrich	Armenia, Azerbaijan
genepool	Base species <i>B. corolliflora</i> Zosimovich <i>B. macrorhiza</i> Steven <i>B. lomatogona</i> Fischer & Meyer	Armenia, Azerbaijan, Georgia, Iran Armenia, Azerbaijan Armenia, Azerbaijan, Iran
	Hybrid species <i>B. intermedia</i> Bunge <i>B. trigyna</i> Wald. & Kit.	Armenia, Georgia Armenia, Georgia

 
 Table 1. Taxonomy of the genus Beta and geographic distribution in the Caucasus/ Transcaucasus region

## Beta species in Armenia

### Section Beta

The wild species and subspecies of this section are adapted to very different edaphic and climatic conditions.

*B. vulgaris* subsp. *maritima* is mainly distributed along the sea shores where plants are most prevalent on beaches in a narrow band between the high tide zone and the start of the dense coastal vegetation. In Armenia wild and weedy forms are reported. *Beta vulgaris* subsp. *maritima* plants, with a characteristic thick root, grow on tertiary red clays or on stony slopes in the Idjevan, Yerevan and Darelegis floristic regions, at altitudes ranging from 800 to 1800 m, an area which has climatic features rather different from the main distribution area of the sea beet in the Mediterranean area or in Western Europe. Especially at the higher altitudes the winters can be fairly cold in Armenia. It is not yet understood how the sea beet survives under these conditions in our country. The plants also occur in the Ararat valley, where they sometimes occupy slightly saline ground. The relationship to *B. perennis* (L.) Halaczy, a taxon not recognized by Letschert (1993), needs to be investigated.

## Section Corollinae

- *B. lomatogona* is specifically adapted to arid conditions. The competitiveness of the species ceases quickly with increasing humidity of the climate and soil (Buttler 1977).

The diploid (2n=2x=18) *B. lomatogona* Fischer & Meyer is native to the Transcaucasus, Asia Minor and North-West Iran.

In Armenia it occurs in Shirak and Yerevan floristic regions, growing on dry rocky slopes in mountain steppes, at altitudes ranging from 1500 to 1800 m.

- *B. corolliflora* is a frequent weed in farm fields and also grows along field margins and roads. Only 10% of the detected sites were part of the natural vegetation (watercourse margins, hill meadows).

The tetraploid (2n=4x=36) *B. corolliflora* Zosimovich ex Buttler is native to Transcaucasus, East Turkey and North-West Iran.

In Armenia it is recorded in Lori, Idjevan, Sevan, Aparan and Darelegis floristic regions, occurring in the middle and the upper mountain zones (from 1500 to 2100 m), where it grows on slopes, meadows, edges of forests and ruderal sites.

- *B. macrorhiza* is a typical ruderal species colonizing landfall areas, i.e. fresh gravel and soil at the foot of hillsides or steep cliffs. The sites are humid, as slope water is available to the plants even in prolonged dry periods.

The diploid (2n=2x=18) *B. macrorhiza* Steven is native to Dagestan, the Southern and Eastern Caucasus, Eastern Turkey, and North West Iran.

In Armenia it occurs in the Sevan and Darelegis floristic regions. It occupies wet places, banks of lakes and fallow fields at altitudes ranging from 1400 to 2100 m.

### Beta species in Azerbaijan

It is established that five species of wild Beta are found in Azerbaijan:

- *B. perennis* (L.) Freyn. = *B. vulgaris* subsp. *maritima* (L.) Arcang.
- B. lomatogona Fischer & Meyer
- B. macrorhiza Steven
- B. patula (Soland) W. Aiton, Hortus Kewensis
- B. trigyna Waldstein et Kitaibel.

#### Section Beta

- Perennial beet - *Beta perennis* (L.) Freyn., now referred to as the sea beet = *B. vulgaris* subsp. *maritima* (L.) Arcang. Comparative studies are required to confirm the status of *Beta perennis* and its relationship with the sea beet.

It is widespread in the Araksinskoi lowlands, Kurinskoj and Steppe plateau and is also found in lowlands and in foothills, and on the coasts of the Caspian sea from the railway station of Gaza Mamaldi, and in Alyt, Salyan, and Neftchala. The area of the species covers the territory of certain areas of Azerbaijan: the given area is located between 39-40°N latitude and between 44°20E and 49°80E in longitude. The basic landscapes which favour the species are desert and semi-desert vegetation.

During the international expeditions organized by the Genetic Resources Institute of Azerbaijan in 2003 and 2004, the ecological features of the species were studied. Seeds are preserved in the Institute's Gene Bank.

#### Section Corollinae

- *B. lomatogona* Fischer & Meyer in Azerbaijan is widespread in the mountainous part of Lenkorani, on stony and dry slopes. Its roots contain up to 25-30% of sugar and the species is considered drought tolerant.

During expeditions seeds were collected from the district of Meidan, in the territory of Zuvanda. New sites on Digah mountain were detected, at 2000 m above sea level (asl): it is mountainous with meadows and a soil pH of 7.5. The area covers Lenkoran–Talish, Diabar, located between 40-50°N latitude and 40-48°E and 40-50°E longitude.

- *B. macrorhiza* Steven is widespread in Azerbaijan, in eastern and western parts of the big Caucasus chain, especially in the Kuban area and in the vicinities of villages, including the village Khinalug, from which this species was described for the first time. It grows on the summits and at medium elevations of mountain belts, on stony slopes. The area of distribution of this species covers the territory of Kuban, Khinalug, Gonagkend and the Shirvan mountain. This region is located between 40-42°N latitude and 46-50°20′E longitude. The roots contain 12% of sugar on average.

#### Beta species in Georgia

The following species are known to occur in Georgia:

#### Section Beta syn. Vulgares Ulbrich

 B. vulgaris L. subsp. vulgaris (cultivated beets) Leaf Beet Group Garden Beet Group Fodder Beet Group Sugar Beet Group

# Section Corollinae Ulbrich

- Base species: B. corolliflora Zosimovich is found at the following sites:
  - Bakuriani (1752 m altitude), situated in the Borjomi district of Georgia. It is located on the northern slope of the Trialeti Range, at an elevation of 1700 m asl. The region around Bakuriani is covered by coniferous forests (mainly spruce). The resort lies 30 km from Borjomi and is located within the so-called Bakuriani depression/caldera. The present-day area of the town was built up by the lava flows from the Mukheri volcano. The climate of Bakuriani is transitional from humid maritime to relatively humid continental. The winters are cold and experience significant snowfall while the summers are long and warm. Average annual temperature of the town is 4.3°C. The average temperature in January is -7.3°C while the average August temperature is 15°C. The annual precipitation is 734 mm. The depth of snow from December to March is 64 cm.
  - Village Baraleti (1681 m altitude)
  - Akhalkalaki (1707 m altitude) is a small city in Georgia's southern region of Samtskhe-Javakheti. It lies on the edge of the Javakheti Volcanic Plateau, about 30 km from the border with Turkey. Javakheti is one of the geologically oldest regions of Georgia, distinguished by its originality and uniqueness. Unusual and diverse is its landscape with its alternating grandiose mountain slopes covered all over with boulders, deep winding canyons and the wide flat Akhalkalaki plateau. The greater part of Javakheti is a mountainous plateau surrounded by ridges, peaks of which reach up to 2300-3000 m asl. Three geographic areas are distinguished in Javakheti: northern (Chobareti-Tabatskuri), central (Akhalkalaki Valley) and southern (Kartsakhi–Paravani and Niala). Akhalkalaki plateau slopes along the East-West axis and heights above sea level range between 1200 and 1800 m. Average annual temperature in Akhalkalaki is 4.9°C. Vegetation period is 138 days, the total precipitation 122 mm.
- Hybrid species: B. trigyna Wald. & Kit. is found at the following sites:
  - Bakuriani, 1752 m altitude
  - Khertvisi, near Artvin, 1197 m altitude.

# • Beta species in Iran

# Section Corollinae

In Iran the wild beet species *Beta lomatogona* Fischer & Meyer (*Beta* section *Corollinae*) is an element of the Iranian Flora. Hohenacker (1838) detected the species in the Talysch Mountains at Tatuni. Buttler (1977) considered *B. lomatogona* as a model plant for the Irano-Turanian flora because the limits of distribution of this wild beet species are almost congruent with the oriental Turanian geobotanical area. This species has its main distribution area in Turkey. Its abundance decreases from eastern Turkey to northwestern Iran and Azerbaijan.

*B. lomatogona* used to grow among various field crops, in pastures and alongside streams and orchards in Ardabil (1332 m altitude). Due to severe drought, land management changes and overgrazing, populations of this species have suffered

badly, so that no plants of this species were found during a collecting mission in 1999. In fact, the population size of *B. lomatogona*, surveyed several times in the past few years, is apparently decreasing in a number of localities, suggesting the need for protection of this natural reservoir of potentially useful traits. The collecting team drew attention to this and strongly suggested action to establish *in situ* conservation to rescue this wild beet (Frese et al. 2001). The Sugar Beet Seed Institute (SBSI) agreed to establish *in situ* conservation in Ardabil Research Station. The pericarp caps of fruit balls of *B. lomatogona*, collected in Gardeh, were removed manually and sown in April 2000 in the greenhouse in Karadj in one-litre pots filled with sterile soil. Some seeds did not germinate due to damage during cap removal.

Seedlings were maintained in the greenhouse until they reached a well-developed stage. In 2001 150 plants were transported to the research station of Ardabil and transplanted to the prepared plot. These plants survived the winter conditions of Ardabil. Seed stalks appeared in late May and seeds were harvested in bulk in August 2002. The plants are kept in the research station of Ardabil.

### Ex situ Beta collections

#### Armenia

Three wild species out of four are present in *ex situ Beta* collections in Armenia: *B. corolliflora* (20 accessions), *B. macrorhiza* (5) and *B. lomatogona* (4).

These collections urgently need support for seed storage.

The collections also include 12 accessions of cultivated *B. vulgaris*.

#### Azerbaijan

The Institute of Plant Genetic Resources holds the following species: *Beta crassa, B. lomatagona, B. macrorhiza, B. maritima, B. matola, B. perennis, B. sikla* subsp. occidental europae, *B. vulgaris, B. vulgaris* cv. esculentum and *B. vulgaris* cv. saccharifera.

#### Georgia

The collection is held at the N.Y. Lomouri Institute of Farming, PGR genebank. Only five accessions of cultivated *B. vulgaris* are present.

In Georgia beet pests and diseases are widespread. The most important are leaf miner moths, aphids, flea beetles, bugs, cercospora leaf spot, oidium, phomopsis and rust.

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# Survey of Beta nana (Boiss. & Heldr.) in Greece

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#### Introduction

The genus Beta is made up of the sections Beta, Corollinae, Nanae, and Procumbentes. Beta nana Boiss. & Heldr. constitutes a monotypic section within the genus, and is a relict species endemic to Greece that has a very limited distribution area (Buttler 1977; Letschert 1993). It occurs in the mountains at high altitudes, in limestone substrates and on short open turf at the edges of meadows with late snow. It is the only alpine species of the genus distributed in Greece (Strid 1995) but not the only alpine Beta species (Buttler 1977). Species of section Beta exist at lower elevations in Greece. The general habitat for B. nana is in closed or open depressions with relatively moist soil above 1800 m elevation. The climate at such an altitude is cool and moist because clouds often build up around the mountains. The plant populations are mainly found on ranges facing east or northeast, where temperatures are lower during the summer afternoons. Plants also grow in crevices between rocks and in disturbed areas, such as rough tracks or severely grazed open plant communities. The prostrate growth habit protects the head of the storage root from being damaged by grazing animals (Dale 1980, 1981). It is possible that a certain degree of grazing, however, may keep the associated flora short, thereby promoting the survival of the species. Strid (1995) described the species in detail. It is an inconspicuous, diploid (Franzen and Gustavsson 1983) plant species, with a small rosette of leaves approximately 10-20 cm in diameter, depending on the fertility of the soil. The plant is said to be self-fertile, producing few seed stalks with 10-25 flowers per spike between June and August. The monogerm seedballs dehisce to the ground in the vicinity of the seed plant while still green. Dale (1980) noted that germination of the seed has proved difficult, and assumed that the extremes of temperatures in the natural habitat, leaching of inhibitors, as well as enzymes in the gut of animals may all play a part in successful germination. He further argued that the species might not be easy to cultivate outside its very specific natural habitat. In contrast, Strid (1995) reported that the species is easy to cultivate from seed, and that plants have survived in the Copenhagen Botanic Garden for almost 15 years, where they have regenerated spontaneously.

<sup>&</sup>lt;sup>11</sup> The German Federal Ministry of Food, Agriculture and Consumer Protection (BMELV) merged the former Federal Centre for Breeding Research on Cultivated Plants (BAZ), the Federal Biological Research Centre for Agriculture and Forestry (BBA), and parts of the Federal Agricultural Research Centre (FAL). Since January 2008 the new institution is called Federal Research Centre for Cultivated Plants – Julius Kühn-Institut.

Little is known about the patterns of genetic variation in *B. nana*. Phenotypic variation observed by Dale (1980) in the natural habitat was low within and between populations. Allozyme patterns of *B. nana* have been analysed by Nagamine and Ford-Lloyd (1989) who found five unique and invariant alleles compared to a range of other species investigated in the same study, indicating a unique phylogenetic position of *B. nana* within the genus. Shen and co-workers (1998a, 1998b) and earlier studies (Jung et al. 1993) showed that *B. nana* is more closely related to the section *Corollinae* than to any other section, although it was a clearly separated group when using random amplified polymorphic DNA (RAPD) banding patterns. Like *B. nana*, *Corollinae* species occur at high altitudes and are part of the secondary genepool in relation to cultivated beet (Buttler 1977). *Beta nana* is of interest to the beet breeding community because of its novel genetic variation, potential cold tolerance and monogermicity.

B. nana is considered a rare but not threatened species requiring no specific conservation measures. Some of the populations are growing in protected areas (Strid 1995). However, species living in alpine regions are particularly prone to extinction risk from climate change (Grabherr et al. 1994; McCarty 2001; Parmesan and Yohe 2003). B. nana is a highly specialized species of the alpine regions of the Greek mountains. Reproduction and migration mechanisms of the species have not been investigated, nor are demographic processes understood; thus the impact of climate change on the ecological niche of the species and its long-term population viability cannot be predicted. Moreover, it is not understood how the genetic variation is distributed within a population, among populations within a region, or among regions; and to what extent gene flow between populations within a region and among regions occurs. Short distance gene flow by seed dispersal (a few hundred metres) is facilitated, perhaps, by melt water flows, grazing animals, and birds. Because a few plants were found during the exploration in 2005 growing in cracks of steep cliffs, it is hypothesized that birds may play a role in long-distance dispersal by depositing undigested seeds.

In 1980 the International Board for Plant Genetic Resources (IBPGR, now Bioversity International) charged M.F.G. Dale with a survey of *B. nana*. The major finding of his missions (summarized in Table 1) indicated genetic erosion within the species caused by overgrazing and road construction. The destruction of several populations of a species does not necessarily endanger the species as a whole. However, because plant breeders are especially interested in the maintenance of the within-species variation, the ECPGR Working Group on *Beta* recommended repeating the survey and assessing the need for *in situ* conservation actions (Maggioni et al. 2000).

Therefore, there are good reasons to monitor the demographic development of this wild crop relative and to acquire a better understanding of the biology, ecology and landscape genetics of the species. The *B. nana* exploration in 2005 was organized to provide a baseline to support future research to address these questions. Specific objectives of the joint plant exploration were:

- collection of fresh seeds from a few, larger populations for use in research;
- joint development of an in situ management programme with Greek collaborators;
- development of a database tool suited to document monitoring data; and
- determination of the environmental characteristics (soil, microclimate, associated vegetation) of the natural growing sites to design more efficient *ex situ* seed multiplication procedures.

Mountain surveyed	
Taigetos (1980, 1981)	1980: One population, extremely limited distribution, site intensively grazed by sheep and goats. 1981: Two quite large and other smaller populations were found.
Chelmos (1980, 1981)	No sign of <i>B. nana</i> . Vegetation heavily grazed by sheep and goats.
Parnassos (1980, 1981)	1980. <i>B. nana</i> widely distributed in this part of the mountain range. Intensively grazed by sheep and goats. Insect damage observed. Road construction may have caused drying out of previously suitable habitats. 1981: Disturbance caused by Athens Ski Club. Approximately 30-40% of the original suitable habitats/populations had been destroyed in 1981.
Giona Oros (1980, 1981)	1980: A number of populations observed. Man's activities are certain to have an effect on the populations of <i>B. nana.</i> 1981: Limited impact of bauxite mining on some populations.
Timfristos (1980, 1981)	No sign of <i>B. nana</i> . Natural habitat may not be suited for this species.
Lakmos (1980, 1981)	No sign of <i>B. nana</i> though potentially suitable sites were observed.
Smolikas (1980)	No sign of B. nana. Local shepherds recognized samples of the species.
Olympos (1980, 1981)	1980: Small populations were found throughout this area. The region was grazed by sheep and goats. 1981: Many populations were showing signs of drought stress.
Kato Vermiou (1981)	No sign of <i>B. nana</i> . Few suitable habitats.
Vitsi (1981)	No suitable habitats.
Tymphe (1981)	No sign of <i>B. nana</i> . Few areas retaining moisture.
Kaliakouda (1981)	No sign of <i>B. nana</i> . Little retention of surface water.
lti (1981)	No sign of <i>B. nana</i> . Suitable habitats were observed.
Vardousia (1981)	Populations were found in open depressions. Grazing by sheep and goats evident.

 Table 1. Results of a B. nana survey in Greece conducted by Dale (1980, 1981)

## Materials and methods

The itinerary of the recent survey was based on Dale's reports and an extensive literature search. Unfortunately, Dale did not describe the exact collection sites nor did he estimate the geographical coordinates based on topographic maps. Populations were found using Dale's general site descriptions, current topographic maps, and local experts. Once found, the location of a population was determined using a Garmin 12XL global positioning system (GPS) (Garmin International, Inc., Olathe, Kansas, USA), and further site and plant characters were determined as shown in Table 2. Leaf tissue and plant sap samples were also collected from 12 to 24 plants from selected populations for future DNA extraction. The sap samples were collected using Clone Saver<sup>™</sup> cards (Whatman International, Middlesex, UK).

A data model for recording monitoring data was tested during the trip and is described by Frese and Germeier (see paper on "The International Database for *Beta* and *in situ* management – potential, role and functions", this volume, pp. 59-74). Using a model proposed by Guarino (1995), the threat of genetic erosion was assessed for each site by using proxy data. Proxy data are recorded to study a phenomenon for which a direct measurement in a given situation is not possible. The model uses scores for factors such as the extent of wild habitat of the target species within the study area, the ratio of present livestock density to estimated carrying capacity of a site, or the distance of the site to development projects such as tourism complexes or mining sites.

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Date	Accession	No. of	No. of	No. of	Site data								
collected	number	seed collected	plants collected	plants found	Site size (m²)		GPS Latitude		GPS Longitude	Elevation (m asl)	Aspect	Physical description	Locality
08/23/05	GR05-009	14	ى ا	20	30	z	36.95351	ш	22.35788	1873	East	Shallow gully on upper slope of a large ridge.	Mt. Taygetos
08/23/05	GR05-012	78 (*)	S	38	30	z	36.95905	ш	22.35826	1870	East	Medium sized rocky drainage on upper slope of major ridge.	Mt. Taygetos
08/24/05	GR05-023	109 (*)	6	>70	100	z	37.97605	ш	22.19042	2077	West	Mid-slope on large ridge.	Mt. Chelmos
08/24/05	NO SEED Collected					z	37.97138	ш	22.19068	2174	West		Mt. Chelmos
08/26/05	GR05-024	80 (*)				z	38.68164	ш	22.13107	1938	West	Bench on mid-slope on large ridge. Site has large to small scattered rocks.	Mt. Vardousia
08/26/05	GR05-025	395 (*)	65	>300	1500	z	38.68099	ш	22.13164	1927	West	Bench on mid-slope on large ridge. Site has large to small scattered rocks.	Mt. Vardousia
08/26/05	NO SEED Collected		0	-	10	z	38.68272	ш	22.12892	1920	South	Eroded road cut.	Mt. Vardousia
08/26/05	GR05-026	51 (*)				z	38.68276	ш	22.13358	2080			Mt. Vardousia
08/26/05	NO SEED Collected					z	38.68432	ш	22.11334	2081			Mt. Vardousia
08/26/05	NO SEED Collected					z	38.68419	ш	22.13177	2000			Mt. Vardousia
08/26/05	NO SEED Collected					z	38.68486	ш	22.13065	1980			Mt. Vardousia
08/27/05	GR05-037	216 (*)	20	>300	400	z	38.64036	ш	22.28575	1917	North- northeast	Shallow vale (3000m <sup>2</sup> ) surrounded by rocky, limestone ridges. At or close to to to of range.	Mt. Giona
08/27/05	GR05-039	21	12	>200	100	z	38.63937	ш	22.28613	1912	West	Small shettered area surrounded by high rock walls.	Mt. Giona
08/28/05	GR05-040	276 (*)	25	>100	500	z	38.58549	ш	22.24103	1881	Northeast	Rocks over red sandy loam. Small (50x100m) bowl at top of high ridge.	Mt. Giona
(*) = Seed	solit between I	IS Western	Regional Plar	nt Introduct	tion Station	<b>WRF</b>	PIS) and Gr	) yee	Gene Bank				

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Date	Accession	No. of	No. of	No. of	Site data								
collected	number	seed collected	plants collected	plants found	Site size (m²)		GPS Latitude		GPS Longitude	Elevation (m asl)	Aspect	Physical description	Locality
08/28/05	GR05-041	28	4	75	1200	z	38.59762	ш	22.23894	1937	Northwest	Rocks over red sandy loam. Small (50x100m) bowl at top of high ridge.	Mt. Giona
08/28/05	GR05-042	126 (*)	18	06<	200	z	38.59763	ш	22.23896	1950	West	Upper slope of high ridge.	Mt. Giona
08/28/05	GR05-043	338 (*)	25	>300	3000	z	38.61639	ш	22.22629	2095	South	Very rocky over red sandy loam. Not much soil. Large open bow//valley on top of ridge.	Mt. Giona
08/29/05	GR05-044	355 (*)	25	75	10000	z	38.54584	ш	22.59277	1928	North	Large bowl mid-slope on large ridge.	Mt. Parnassos
08/29/05	GR05-045	-	-	34	6000	z	38.53913	ш	22.61411	2052	AII	Flat area surrounded by steep rocky slopes	Mt. Parnassos
08/29/05	GR05-046	127 (*)	14	32	200	z	38.54222	ш	22.60944	2058	Southeast	Small gully. Very rocky.	Mt. Parnassos
08/29/05	GR05-047	290 (*)	69	143	3000	z	38.53795	ш	22.61266	2061	AII	Small (3000 $m^2)$ flat area in a rocky bowl. Like a sink hole.	Mt. Parnassos
08/31/05	GR05-053	385 (*)	approx. 50	250	400	z	40.07534	ш	22.3592	2492	AII	Large, deep, open bowl on upper- slopes of mountain.	Mt. Olympus
08/31/05	GR05-054	222 (*)	>100	>1000	1200	z	40.07799	ш	22.35632	2639	AII	Small open gully off main ridge.	Mt. Olympus
08/31/05	GR05-055	103 (*)	100	>1000	75	z	40.07762	ш	22.35483	2675	East- northeast		Mt. Olympus
08/31/05	NO SEED Collected					z	40.07222	ш	22.35216	2626	East- northeast	Valley/saddle between two peaks. Plants found in lowest areas.	Mt. Olympus
08/31/05	GR05-056	82 (*)	100	006<	1600	z	40.07004	ш	22.35731	2558	East	Wide swale between rocky hill tops.	Mt. Olympus
(*) = Seed	split between	US Westem F	Regional Plar	nt Introduct	ion Station	(WR	PIS) and Gr	eek	Gene Bank.				

# Results

Populations of *B. nana* were found in these mountains: Taygetos, Chelmos, Vardoussia, Giona, Parnassos and Olympos (Fig. 1). In contrast to Dale's reports (1980, 1981), the extent of the occurrence of the species was confirmed to be as reported in 1908 by Halacsy (cited by Akeroyd, 1986). Except for a very sheltered site at Mount Parnassos, all sites proved to be grazed to various degrees, primarily by goats. Estimates for the risk of genetic erosion were fluctuating around 100 on a scale of 0 (=no risk) to 200 (=very high risk). Especially on Mount Olympos, the species had reproduced well at several sites within the surveyed area. The overall population sizes ranged from more than 1000 individuals on Mount Olympos to a few individuals on Mount Taygetos. All sites had full exposure to the sun, and the slopes ranged from 0 to 25 degrees.



Fig. 1. 2005 Beta nana survey sites and population locations.

Maxted et al. (1997) suggested a new conservation technique called "genetic reserve conservation" to complement *ex situ* conservation efforts. Conservation of a wild species in its natural habitat requires the designation of an exact location for a genetic reserve, the development and implementation of a site management plan, the

engagement and funding of a reserve manager, and the monitoring of the location to assess the impact of the site management on the target species. The distribution area on Mount Olympos would be most suited for the establishment of a genetic reserve because the sites are already located within or close to the Nature Park. Two further sites, one on Mount Parnassos and a second on Mount Giona, would also be possible choices. Fig. 2 shows the Vathia Lakka site located on Mount Giona. In the background of the left part of the photo, rocks formed a U-like structure, which could easily be fenced by a shepherd and grazed by sheep or goats in a controlled manner favouring reproduction of the species.



Fig. 2. Vathia Lakka, Mount Giona. The site on the left part of the photo (arrow) would be suited to establish a genetic reserve.

## Conclusions

Although populations of *B. nana* can be found on the mountains identified in this study, there still remain significant areas in Greece to survey to confirm the full extent of the range for this species. It is noted that sites which have extant populations of *B. nana* on Mount Olympos, Mount Parnassos and other mountains are located within or nearby protected areas. This is significant in that it may be that preservation of habitat is beneficial, but it is not known how effectively *B. nana* populations are protected by National or Provincial Nature Park statutes. This relationship needs further study. The extent of overlap between legally protected areas and natural sites of *B. nana* needs to be explored by integrating geographic information systems (GIS) and floristic studies to determine the scope of the relationship between conservation measures and plant species survival. One approach would be to match the distribution of the *B. nana* populations to existing protected areas, and then systematically determine the obvious overlaps. Researchers could then suggest locations for the development of genetic reserve conservation sites.

The choice to be made, of which populations and sites should be protected in genetic reserve conservation sites, needs to be based on genetic distance measures and/or the amount of genetic variation detected within and among populations. For this work, which is still to be done, it is proposed to use microsatellite markers. The question of how effectively agro-environmental measures subsidized by the EU Commission can be deployed for the controlled management of selected sites also needs to be investigated.

#### Acknowledgements

The plant exploration was permitted by Mr A. Chatziathanssiou, Ministry of Agriculture Development and Food, General Directorate of Agricultural Extension and Research, Directorate of Natural Planning and Environmental Protection, Athens, Greece. This exploration was funded by the USDA-ARS National Plant Germplasm System and supported by the German Ministry of Consumer Protection, Food and Agriculture (BMVEL).

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# Survey of Beta vulgaris subsp. maritima populations in Ireland

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# Introduction

The survey was carried out during 2002 and 2003 to establish the current extent and level of potential threat to *in situ* populations of maritime or sea beet (*Beta vulgaris* L. subsp. *maritima* L.) that had been sampled by the international collaborative collection carried out during 1987 in Ireland (Fig. 1).



**Fig. 1.** *Beta maritima* distribution in 1987 (Doney et al. 1990).

Doney et al. noted in their 1990 report that:

"The distribution of sea beet was similar to earlier observations (in 1962). However, many small populations were in danger of elimination, or had disappeared...Factors threatening or causing extinction of local populations included livestock grazing (particularly sheep), slippage of mud cliffs, industrialization, sea ports, and recreational activities."

# Extent of changes since 1987 to native *B. vulgaris* subsp. *maritima* populations represented in *ex situ* germplasm collections

To date a total of 24 (56%) of the sites visited in 1987 have been re-visited and surveyed, while 3 new sites have been also recorded.

Full global positioning system (GPS) details of ten selected plants were recorded at each location. Estimates were taken of habitat population numbers and the proximity of maritime populations to sugar beet crops. Indications of potential threats to habitat were also recorded. Digital photographs were taken at all sites from as many angles as possible to give an indication of the current status of the habitat. Future comparisons can be used to gauge the extent of any changes to the habitat.



An example of data recorded is shown in Fig. 2.

Fig. 2. Site Ballyheigue, Co. Kerry

(Accession identifiers for the International Database for Beta (IDBBNR) = 3873, 5896).

Ireland represents one extreme of the maritime beet habitat. At the northwestern edge of Europe, the generally cool and wet climate, with mild winters, means that maritime beet can survive as a perennial under our conditions. Individual plants thrive in favourable locations giving rise over time to "colonies" of stems supported by a sturdy taproot. They usually occupy a relatively narrow strip of supralittoral sediment, which occurs between the high water mark and more normal non salt-tolerant vegetation. This gravel or shingle area usually accumulates drift material rich in nitrogenous organic matter. Populations in a number of locations have progressed inland, spread by operations to prevent coastal erosion, or car-park construction.

All stages of growth have been observed:

- early emergence,
- first true leaf stages,
- juvenile growth stages,
- flowering, and
- seed ripening.

Plants were found growing in:

- stone walls,
- concrete walls,
- coastal defences,
- cliffs,
- road verges,
- reclamation works,
- under park benches,
- caravan parks,
- yacht parks, and
- less often in sand dunes.

Populations were summarized into five broad categories:

- 1. No plants or colonies found
- 2. Scarce (1-10)
- 3. Sufficient (10-50)
- 4. Plentiful (50-100)
- 5. Abundant (>100).

The distribution of these categories is shown in Fig. 3.



**Fig. 3.** Populations of maritime beets in Ireland, 2002 and 2003 (% of sites surveyed).

Of the sites surveyed, 28% were present within 1 km of commercial sugar beet crops, 30% had a crop between 1 and 5 km away, while the remaining 44% were farther than 5 km from commercial crops.

There was little evidence of any foliage diseases at the sites surveyed, with just one plant found at each of three sites (Woodstown Strand, Skerries and Malahide) showing leaf disease symptoms. One plant in Dungarvan showed symptoms of viral infection.

## Seed collected

Seed was collected from ten selected plants, representative of the location, from each of four sites (Shanagarry, Ballyheigue, Fenit and Ardmore Bay). The location of each individual plant sampled was recorded on the GPS receiver. This should allow future researchers to locate the exact source of seed collected.

All seed samples were dried, cleaned, sub-sampled and are now stored at the Seed Department in Mallow. Germination tests were carried out using the sub-samples. The average dried weight of seed collected is 25 g, ranging from less than 1 g per sample to 75 g. Germination levels were disappointing, with an average of 7% and ranging from 0% to 72% (Fig. 4). The low level of germination indicated that timing of seed collection is critical to ensure viable seed is collected for *ex situ* storage.



**Fig 4.** Percentages of germination of maritime beet seed samples collected in 2002 and 2003 in Ireland.

# Threats to habitats

The ongoing threats to the habitats of maritime beet populations are:

- Coastal erosion, and
- Human leisure and commercial activities.

Some specific examples of erosion damage are visible at:

- Ardmore,
- Dunmore East,
- Wexford,
- Garrettstown,
- Skerries, and
- Rough Point.

Where populations are relatively large, or growing vigorously in suitable conditions, the plants appear capable of colonizing disturbed conditions such as:

- New picnic sites at Ballyheigue,
- Building rubble at Rossglass,
- A car-park at Tramore, and
- Coastal protection measures at Ardmore Bay.

Both commercial and residential developments can be seen to have impacted on habitat in:

- Wexford,
- Duncannon,
- Ardglass,
- Tramore, and
- Dingle.

Evidence of the impact of erosion and human activity on habitat was evident at almost all sites, but such is the tenacity and seed production potential that most populations appear to be more numerous than those reported in 1987, with newer colonies of younger plants evident at most sites.

The exceptions were:

- Ross's Point, Co. Sligo, and
- Dunmore East, Co. Waterford.

In these two cases, the number of plants found was lower than in 1987. Of the two, Ross's Point is at the point of extinction – only four juvenile plants were found in vulnerable places. No other populations were found at a number of other locations visited on the North Sligo coast. In Dunmore East, erosion has reduced easily accessible plants to less than ten, while some more plants could be observed on inaccessible cliffs.

A number of the sites visited (Ballyheigue, Bunmahon, Dunmore East, Duncannon, Ross's Point and Killard) are already located in designated Special Areas of Conservation.

However information displays at access points and car-parks do not identify maritime beet as a useful or interesting plant (despite its presence around signs and under park benches at Ballyheigue and Bunmahon).

It was recommended that the site at Ardmore (IDBBNR 3863) should be designated as an area of special scientific interest, as a selection from that site has been screened and found to have good resistance to *Cercospora beticola* (leaf spot) disease (Panella and Frese 2000). In conclusion it appears that from the locations visited to date, the majority of *in situ* populations are not under any immediate threat of destruction.

#### Acknowledgements

This project was co-funded by grant aid from the Irish Government's Department of Agriculture and Food as part of their Programme to Conserve Genetic Resources for Food and Agriculture, and by Irish Sugar Ltd.

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# The International Database for Beta and in situ management – potential, role and functions

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#### Introduction

The second International Biological Programme stressed the importance of plant genetic resources for food and agriculture (Frankel and Hawkes 1975) and the need to control genetic erosion in crops and their related wild species. The inevitable consequences arising out of the loss of genetic variability were recognized in the 1970s and since then strenuous efforts have been made to mitigate the loss by preserving germplasm samples in *ex situ* collections. According to FAO (1996) approximately 6.1 million accessions are stored in holdings worldwide; 10 485 accessions of wild and cultivated *Beta* are listed in the International Database for *Beta* (IDBB). The management of these large collections is facing some unresolved difficulties, in particular in frequently outcrossing species such as *Beta*. Results published by Chebotar et al. (2003) indicate that the genetic integrity of populations can be affected when *ex situ* accessions of outcrossing species such as *Secale cereale* are repeatedly subject to the selection pressure of the "genebank environment". Anyone who has ever tried to regenerate seed samples of wild species knows, without any sophisticated experiments, that the loss of genotypes not adapted to genebank germplasm management procedures can already occur during the first seed multiplication.

Jain (1975) and his generation of conservationists had already understood that manmade environmental changes will increasingly interfere with the genetic variability of species. Without sufficient genetic variability for adaptive traits, species will not be able to evolve at a speed to match the environmental changes taking place today. Hence, loss of genetic variability would not only occur in genebanks but also in the natural genetic reservoirs which the agriculture sector may expect to exist forever. Jain (1975) stressed the urgent need for programmes aiming at the protection of plant species in their natural habitats, which obviously needs to include the protection of the habitats themselves. About 20 years after Jain's publication, *in situ* conservation of species was fostered politically by the Convention on Biological Diversity (CBD 1993). In 2002 the Sixth Conference of the Parties (COP6) finally adopted the Global Strategy for Plant Conservation (GSPC) striving for a significant reduction of the species loss rate by the year 2010. At the time of writing there are only four years left before "Target 2010" to prove that at least some progress has been achieved through improved cooperation in Europe.

In the field of crop wild relatives (CWRs), both the conservation biology and the germplasm user communities could effectively collaborate to implement Target 2010 in practice.

<sup>&</sup>lt;sup>12</sup> The German Federal Ministry of Food, Agriculture and Consumer Protection (BMELV) merged the former Federal Centre for Breeding Research on Cultivated Plants (BAZ), the Federal Biological Research Centre for Agriculture and Forestry (BBA), and parts of the Federal Agricultural Research Centre (FAL). Since January 2008 the new institution is called Federal Research Centre for Cultivated Plants – Julius Kühn-Institut.

The *Beta* germplasm user community is represented by the ECPGR Working Group on *Beta* and the World *Beta* Network (WBN). Conservation biology is represented by the World Conservation Union (IUCN) and its Crop Wild Relative Specialist Group (CWRSG) as well as the European Environment Agency (EEA) and affiliated institutions at the national level. These groups should improve their cooperation to safeguard *Beta* germplasm.

*Ex situ* and *in situ* management have often been looked at as complementary, since both of them have specific weaknesses and strengths. Considerable work has been done to develop databases for information management in the *ex situ* collections. Beginning in the 1980s the ECPGR initiated the development of the current 57 Central Crop Databases (CCDBs), combining crop-wise data from the European *ex situ* germplasm holdings. The International Database for *Beta* (IDBB) was established in 1989 to serve as a central crop database within a worldwide network of decentralized *ex situ* collections. CCDBs were considered as instruments facilitating task-sharing between European holdings and consequently the rational maintenance of germplasm. For this purpose passport data of *Beta* collections existing within the network were compiled in a central system. A second role, for users of germplasm holdings rather more relevant, consists in the gathering of characterization and evaluation data in a CCDB, which allows the users to search via a crop portal in the network of decentralized holdings for germplasm to meet his/her needs.

## Rationale for collaboration with the conservation biology discipline

Why should we in addition to well-managed *Beta ex situ* holdings care about the *in situ* management of the genetic diversity in the genus *Beta*? It is generally accepted that global climate change has effects on all levels of ecological systems, causing phenomena such as life history changes and shifts in the geographical distribution range of a species (McCarty 2001). *Beta* species occur in habitats which are particularly prone to changes caused by global warming, such as the narrow band between high tide and 10 to 20 m inland along the European coasts. They can also be encountered in mountainous areas of Turkey and the Caucasus/Transcaucasus. In the case of *B. nana*, populations exist solely at high altitudes close to the summits of mountains in Greece. How a rise in sea level or the upwards expansion of alpine species more competitive than for example *B. nana* will affect these species is not yet known, but should be observed at regular time intervals to ascertain the survival and future availability of these important resources for beet breeding.

While it is beyond the capabilities and facilities of the ECPGR Working Group on *Beta* and the WBN to implement monitoring in the wide distribution area of the genus, it is well within our power to provide, to the national and local authorities responsible for monitoring, the data that we gather and document in our public central crop-specific database.

Conservation of biodiversity relies on general approaches such as the protection of habitats, and if these harbour *Beta* species, nature conservation meets the specific interest of the ECPGR Working Group on *Beta* and the WBN. In well-justified cases our interest group may wish to protect selected populations more actively. How can these wishes be organized and work concepts realized? One essential step needs to be undertaken by the ECPGR Working Group on *Beta* and the WBN: raising awareness on the significance of wild beet species within the community of conservation biologists in the whole distribution area. In spite of this growing concern in the *Beta* experts' circles, organizations responsible for nature conservation projects in countries located in the distribution area may not be fully aware of the actual and potential contributions of their institutions to the task of safeguarding genetic diversity of the genus *Beta*. There is therefore a need for the botanists and crop experts to close these gaps through improved communication. Communication can be supported by data exchange.

# The Crop Wild Relatives Information System (CWRIS) links the genetic resources community with the conservation biology discipline

In the PGR Forum project a first attempt was made to combine a floristic database (Euro+Med PlantBase) with the Mansfeld database focusing on genetic resources. Based on these main information sources the Crop Wild Relatives Information System (CWRIS) was developed by the University of Birmingham (PGR Forum CD-ROM version, October 2005 and http://www.pgrforum.org/cwris.htm). It was launched to emphasize the importance of crop wild relatives (CWRs) and to provide an online searchable database containing a comprehensive catalogue of all CWR species (23 818 CWR taxa) in Europe, and may provide the missing link between conservation biologists working in their respective countries and *Beta* expert groups.

Currently, the most important features of the system are "Taxonomy" and "Search". The taxonomy module not only provides a list of species but also links them with other data sources such as the European Union Nature Information System (EUNIS). The "Search" module allows the tracing of information in case studies, to which the user community should add many more to improve the usefulness and attractiveness of the system. The search module has three levels: (i) the taxon information with the layers biological data, conservation action, ecogeographical data, nomenclature, population information, references, threat, utilization and other information; (ii) the site and population information has further layers such as site location, population size, structure, dynamics, management, biotic interaction, ethnography, characterization and evaluation, local threat to specific population, conservation measures applied to specific populations, and other population information; (iii) the geographical information level of CWRIS is still under construction.

One of the significant messages of the PGR Forum team is that communication between crop experts interested in both the conservation of genetic resources and conservation biology can indeed be improved for the mutual benefit of both groups. ECPGR Working Groups such as ours should be proactive and seek cooperation with nature conservation programmes, agencies and local authorities to help them in setting priorities and to assist in the organization of conservation actions. The organization of collaboration requires ready access to data and information which institutions are keeping in their databases such as EUNIS and the new CWRIS.

# Why develop a crop-specific approach combining in situ and ex situ information?

This reflects the primary crop-specific interests of users of genetic resources in breeding and crop science. Multicrop information systems in these domains impair their simplicity of use and their performance as well as the resolution and quality of data they contain.

As the management of germplasm *ex situ* and *in situ* is complementary, has common (documentation and management) issues and often relies on identical data sources referring to taxonomy, the distribution of taxa, geographical localization of collecting and maintenance sites, it is evident that crop-specific germplasm managers and users should

preferably work with a common information source. *Ex situ* collecting sites can be used to find interesting *in situ* populations and vice versa. Evaluation and characterization of *in situ* populations will imply repeated sampling of genetically delineated and well-defined populations which function, from the user's point of view, in a similar way to an *ex situ* accession. Plant material identified as a "valuable donor" during expensive characterization and evaluation work will preferably be conserved *ex situ*. Information resulting from characterization and evaluation work on *ex situ* accessions may be indicative also for *in situ* populations from which these accessions may originate. The geographical distribution patterns of traits will gain interest, particularly if traits are confined to specific areas or habitats. If this is the case, these areas can be highlighted as evolutionarily important areas and populations within these areas as "evolutionary significant units (ESU)" (Luck et al. 2003), at least from the breeders' point of view.

When *ex situ* as well as *in situ* collections of genetic resources are accessible for use, users of genetic resources will use both sources and will need a tool for integrative research into both collection types for traits in the focus of their interest.

# Representation of wild species in the IDBB (*ex situ* passport, characterization and evaluation data)

The representation of *ex situ* accessions of crop wild relative species in the current IDBB is shown in Table 1. One third of the whole database refers to crop wild relatives belonging to 13 taxa. Considerable information on collecting sites is available. Site descriptions were extensively used to extract geographical information from maps during 1986-1990. Thus geographical coordinates are available with the collecting site information.

For these accessions 16 039 observations are available on 56 evaluation and characterization traits, which comprises 44% of the total characterization and evaluation information. These include resistance observations to virus diseases (beet mild yellow virus 323 observations, beet yellows virus 366, rhizomania 335) and fungus diseases (*Aphanomyces cochlioides* 379, *Cercospora beticola* 344, *Erysiphe betae* 368, *Polymyxa betae* 15, *Pythium ultimum* 346, *Rhizoctonia solani* 257).

Taxon	Accession	าร	% of these with g	eographic inform	ation available
	Number	% of total number of accessions	Site description	Coordinates	Elevation
B. vulgaris subsp. maritima	1985	18.9	86	86	77
B. vulgaris subsp. adanensis	78	0.7	99	99	96
B. intermedia	338	3.2	93	93	92
B. lomatogona	281	2.7	69	69	62
B. patellaris	163	1.6	37	37	32
B. corolliflora	133	1.3	79	79	77
B. trigyna	124	1.2	17	17	10
B. macrocarpa	106	1.0	74	74	59
B. macrorhiza	95	0.9	61	61	54
B. procumbens	81	0.8	49	49	48
B. nana	59	0.6	88	88	88
B. webbiana	41	0.4	37	37	37
B. patula	22	0.2	14	14	5
Total	3506	33.4			

Table 1. Representation of wild relatives in the IDBB

# How to retrieve data for crop wild relatives from the IDBB

Generally there is no difference between using the IDBB for extracting information from cultivated or wild species. Yet the focus of interest is slightly different in each case. Pictures are helpful to learn species identification and the range of variation in a specific species. Geographical information is of great importance in the exploration of wild species. The importance of using geographical information when collecting for certain traits interesting for breeding is stressed in approaches like the Focused Identification of Germplasm Strategy (FIGS) (http://figstraitmine.org/index. php?dpage=11, copyright 2005-2007).

During the past few years we have concentrated on the development of such modules for the IDBB. The search engine has been described in some detail by Germeier and Frese (2004). More information is available in the online help (http://idbb.bafz.de/CCDB\_PHP/idbb/Help.html). The new modules can be accessed for the selected accessions from display results (Fig. 1).

	IDBB	
MT .	International Database for Beta	
TAR CARDON SAL	Display results	aggregated
	Cross table for selected characterisation and evaluation descriptors	V
Selection of	Observations for selected characterisation and evaluation descriptors	
	Standard observations listed for selected characterisation and evaluation descriptors	
	Observation methodology used for the selected characterisation and evaluation descriptors	
	Experimental details for the selected characterisation and evaluation observations	
		include duplicates
	Passport data. for the selected accessions as available	
	Genebanks holding the selected accessions	
Salanel Y	Pictures for the selected accessions as available	
	Map collecting sites found for the selected traits and accessions	

Fig. 1. Results, which can be displayed for selected accessions.

## Pictures

A module has been implemented to list and display pictures (Fig. 2, a and b). Particularly for learning the identification of species *in situ* and the range of variation naturally occurring in one taxon, it would be useful to be able to see online the available pictures of unambiguously determined specimens of different origins. Pictures from multiple accessions of the various species would therefore be necessary. Currently 130 pictures of 48 accessions from 12 taxa are available. The number of pictures currently available for wild species is shown in Table 2.

Taxon	Number of pictures	Accessions represented
B. corolliflora	1	1
B. intermedia	1	1
B. macrocarpa	4	2
B. macrorhiza	11	9
B. patula	7	1
B. trigyna	1	1
B. vulgaris adanensis	2	1
B. vulgaris maritima	34	16

Table 2. Pictures of wild relatives	in	in the	DBB
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Fig. 2. Displaying pictures in the IDBB.

a (top). List of thumbnails with passport information.

b (bottom). Picture with accession, origin and selected characterization and evaluation data.

### Interactive geographical mapping

The IDBB application uses the Google Maps API (http://www.google.com/apis/maps/) to visualize collecting sites of *Beta* accessions (Fig. 3). Google Maps offers map display, satellite images and a hybrid map of both.

Characterization and evaluation data can be visualized in the selected set of accessions by the use of different colours of symbols. A search engine for specific traits is displayed with the map. It allows for filtering specific value expressions (e.g. disease resistance) in the map display. By clicking to a marker, passport, characterization and evaluation information or pictures can be retrieved for the respective accession (Fig. 4).



**Fig. 3.** Collecting sites of accessions with sample status wild and weedy collected in Portugal and Spain displayed on a Google Maps satellite image.



Fig. 4. Visualizing rhizomania reaction in wild and weedy accessions from Portugal and Spain. Resistant accessions can be filtered in the display, by clicking on a marker, the passport, characterization and evaluation data or a picture as selected in the radio button can be displayed for the respective accession.

## Modelling a new module for in situ monitoring

Concepts have been presented and discussed for modelling new modules for *in situ* management information on the population level in the CCDBs. First results are presented below.

# Ecogeographic basis: site, area and habitat

Fig. 5 displays a data model showing the most important entities describing the environment of an *in situ* population. The site is a concept referred to as "collecting site" in the FAO/IPGRI *Multi-crop Passport Descriptors* (MCPDs). As the term is used in *ex situ* plant genetic resources databases it is geographically not very well defined: mostly rough verbal descriptions like (in the best case) "Cabo de Gata, 2 km E of" are found. If geographical coordinates are available, they are point coordinates and understood as a reference point. Indications of extension (shape and size of the population site) are not given and not foreseen in the MCPDs. Therefore it is suggested that a "site" should be defined as a locality geographically described or referenced, but without an explicitly determined extension.

When extension is explicitly referenced, it is suggested that the term "area" (cf. protected area) should be used instead of site. Collecting sites as reference points may be located in defined (protected) areas. It is assumed that management as a protected area implies that the managed area is geographically exactly defined (see Fig. 5).



Fig. 5. Data model and potential data sources for the ecogeographic basis of an *in situ*/on-farm information system.
Sites and areas relate to ecogeographic concepts which describe the environmental conditions encountered there: they have certain climates and soils which form habitats bearing certain flora (and fauna). Similar environmental conditions may be found at many sites or areas - one to many relations, which will normally be represented by layers in a geographical information system.

Data will be available from several sources: meteorological services, which are proposed to be made available as Web services by UNIDART (http://www.dwd. de); European Soil Database (http://eusoils.jrc.it/ESDB\_Archive/ESDB/index. htm); site and habitat information from EUNIS (http://eunis.eea.europa.eu/sites. jsp; http://eunis.eea.europa.eu/habitats.jsp) or the World Database on Protected Areas (http://www.unep-wcmc.org/wdpa/).

#### Levels of aggregation •

Monitoring of wild plants can be done at different levels of aggregation (Fig. 6), which will have to be considered in an *in situ*/on-farm information system. Natural levels of aggregation are individuals and patches which can be directly observed in the field. An operational definition for monitoring will consider an individual plant found at a certain time at a certain place. Individuals can be revisited only if marked. A patch is an agglomeration of individuals of the same species. On revisiting, if a recorded patch area overlaps with a previous recorded one, they are considered to be the same patch. After fusion of two patches one of them is considered the successor of both. Individuals and patches may belong to a population, which is generally the entity of interest. The delineation of a population requires further analysis (genetics, dynamics of (sexual) reproduction) and is therefore a matter of dispute.

Monitoring methodologies may involve experimental aggregation into monitoring plots and transects. These again may contain individuals and patches.

#### A. Natural **B. Experimental** MONITORINGPLOT INDIVIDUAL PATCH POPULATION Агеа TRANSECT D PopulationCode Survey Survey ID SITE PlotNumber FirstRecord FirstRecorded Longitude PlotNumber SiteCode TreatmentCode RecordedBy Transect Latitude RecordedBy Block CountryCode GeodeticSystem TaxonName ReferenceSite Туре Lane n State Length Uncertainty System TaxonName District Plot Width FirstRecord Population System SiteCode Location Permanence RecordedBy Successor Evidence Status StartLatitude TaxonName PlotLength StartLongitude PlotWidth System Azimuth PlotSize Sex Area Patch l n Permanence Population n ReferenceLatitude Marked ReferenceLongitude ReferenceAzimut GeoObject GeodeticSystem Uncertainty

Fig. 6. Levels of aggregation in monitoring plant genetic resources in situ.

### • Monitoring

Monitoring can be done on all levels observable in the field. Again the target of interest is normally population monitoring. Monitoring data recorded at the observable levels are aggregated to population level. Fig. 7 shows details of monitoring entities for the population level indicating lower levels as layers behind with only the title indicated.

Monitoring data on different aggregation levels are not directly comparable. Thus they should be kept separate in similarly modelled layers. This also facilitates their automatic aggregation to higher levels, if the necessary data are available.



Fig. 7. Monitoring of plant genetic resources in situ.

A monitoring event is defined within a certain survey, targeted to a certain object (individual, patch, transect, plot, population) at a certain date. In the higher aggregation levels (above the individual) it covers additional summary information on the total number of individuals and their reproductive effectiveness (total and effective size), the range (minimum, maximum) and average development stage of the individuals, the area covered and the extent of coverage, and trend estimates (increasing, declining), if available from a single point observation.

Most important for management monitoring is demography, which is covered in more detail in the demography tables. These differentiate the population into different cohorts (individuals of similar development stage like seedling, flowering, mature, aged), indicating their number and percentage. Further observations may be taken to characterize and evaluate the populations, e.g. morphology or traits interesting for users (pests, diseases, chemical traits). These are comparable to characterization and evaluation data on *ex situ* accessions. The relationships to descriptors and methodology and further details can be found in Germeier and Frese (2001).

#### Information systems for in situ management and their possible interactions

Biodiversity is generally categorized at three levels: ecosystem diversity; species diversity; and genetic diversity within species. EUNIS deals with ecosystem and species diversity, CWRIS will focus on plant species diversity, while the ECPGR Central Crop Databases already operate with data at the level of genetic variation within species.

#### EUNIS and CWRIS: use for Beta

Independently from genetic resources information systems, the nature conservationists developed electronic information systems for keeping records of species occurrence *in situ*. The importance of this information for the localization of collecting and *in situ* management sites is evident. A prominent European example is the European Nature Information System EUNIS (http://eunis.eea.eu.int). It is operated by the European Environment Agency with the purpose of assisting the Natura2000 process and is required for environmental reporting and informed political decisions.

It provides access to data on species, habitats and sites compiled in the framework of Natura2000, information on nature reserves with an emphasis on nature protection and a geographical, faunistic and floristic focus. It is a multi-species inventory, covering all groups of organisms ranging from "A" = algae to "R" = reptiles. The database contains information on 19 222 taxa of flowering plants. EUNIS has a number of useful search facilities. A combined search for "Beta macrocarpa" and "Portugal" provides the user with taxonomic information, the general geographical distribution, the references and a list of related sites with additional, detailed information. The site description of the "Parque Nacional de Donana" in Spain, last updated on 22 December 2005, not only informs the user on the legal status of the site but also contains a species list: among them B. macrocarpa and B. vulgaris subsp. maritima and other crop wild relatives such as Avena byzantina, A. barbata and A. longiglumis are to be found. The system would allow for descriptions of the population size and the species status but these data have mainly been recorded for birds. The various habitats within the site are also catalogued. The mapping tool is another useful feature of EUNIS as exemplified for the B. macrocarpa site "Salinas del Cabo de Gata" (Fig. 8), where a small population of *Beta patellaris* was sampled by E. de Meijer (Frese et al. 1990).

The mapping tool is also suited to determine the coincidence of collecting sites with protected areas. The *ex situ* accession FL/89 005, *Beta vulgaris* subsp. *maritima* was sampled in Portugal, Cascais, Boca de Inferno, an area known for genetic variation in the cytoplasmic component of male sterility. Using the geographical coordinates recorded by the collector, a search in EUNIS produces the protected site Cabo da Roca, Corine code C21200071, which is not far away from Boca de Inferno.

EUNIS pursues a broad approach to nature conservation and is suited to monitoring biodiversity at the ecosystem and species level. The broad approach has shortcomings once the details begin to get important. The EUNIS species list lacks records of two donors of the important *Heterodera schachtii* resistance, *B. procumbens* and *B. webbiana*. Furthermore, no species of section *Corollinae* except for *B. trigyna* are mentioned though the countries where the species occur are listed. These shortcomings could be overcome by a closer interaction with more specialized information systems like the CCDBs.



**Fig. 8.** Occurrence in EUNIS and collecting sites listed in the IDBB for the Parque Nacional de Donana/Salinas del Cabo de Gata. Clicking to the points in the IDBB gives the respective passport, characterization and evaluation data.

Fig. 9 shows suggested relationships between different information systems. All of them would require common tools such as a taxonomic backbone, mapping facilities etc. The Crop Wild Relative Information System (http://www.pgrforum.org/CWRIS. htm) used Euro+Med PlantBase (http://www.emplantbase.org) as its taxonomic backbone. A regular automatic update of CWRIS according to the newest taxonomic results in Euro+Med PlantBase would be feasible. CWRIS was designed to allow for the monitoring of CWR species in Europe including a threat assessment mainly at the taxon and country level. This central system will enable decision-makers to prioritize conservation needs and to organize conservation measures more effectively. One conservation method complementing *ex situ* holdings has been discussed by the PGR Forum group intensively: the genetic reserve concept. The location, management and monitoring of genetic diversity in natural wild populations within defined areas for active, long-term conservation is called genetic reserve conservation (Maxted et al. 1997).

Although a genetic reserve is established for a specific target taxon or population, a reserve should ideally also benefit associated CWR taxa and become a multi-species conservation site. CWRIS will be the information platform where all individual conservation projects can be catalogued, and where experts responsible for different crops can exchange information on the ongoing conservation projects and attune multi-species genetic reserves to their crop-specific needs.



Fig. 9. Information systems in the domain of *in situ* management of genetic resources and their possible cooperation.

Overlay of occurrence data, given as collecting sites in *ex situ* or herbarium collections with existing protected areas will be a major task for the identification of genetic reserves. Geographical information on protected areas is available from the European Nature Information System (http://eunis.eea.europa.eu/index.jsp) and from the World Database on Protected Areas (http://www.unep-wcmc.org/wdpa/). Historical and recent occurrence data are available in the Global Biodiversity Information Facility (http://www.gbif.org/), from *ex situ* collections as collecting sites in the European Internet Search Catalogue EURISCO (http://eurisco.ecpgr.org/), the International Database for *Beta*, and genebank information systems like GRIN (http://www.ars-grin.gov/). Population level information comprising the occurrence and extent of natural populations together with information on characterization and evaluation data for these natural populations should be included in the CCDB as explained before.

The integrity and, from the users point of view, quality of an information system increase the more the user community gets involved in its development and information uploading. Having said this, it seems only logical that the basic information on genetic reserves should be collected by crop experts, uploaded into the crop-specific information system and provided, perhaps in a more aggregated form, to CWRIS. The exchange of data between the fine-scaled crop-specific systems and the CWRIS operating on the next scale level needs to be organized. Fig. 10 shows possible technical ways of interaction and collaboration between the information systems. Cooperation can be by automatic data exchange via CGI, XML and Web services, but also by cooperative building of source code libraries and sharing them in a central repository like CropForge.



Fig. 10. Suggested interoperation of information systems.

# Role and function of the IDBB

The ECPGR Central Crop Databases are currently focused on information from *ex situ* collections. This also comprises population, geographical and habitat information from the collecting sites, sometimes even including some characterization at the collecting site. Monitoring (*in situ*) will integrate occurrence, characterization and evaluation information.

Since genetic reserves for Beta do not yet exist, a scenario is needed. Let us imagine that the ECPGR Working Group on Beta and the WBN come to the conclusion that populations of *B. vulgaris* subsp. *maritima* distributed along the sea shores of the Baltic Sea in Denmark and Germany should be managed actively. Imagine further that genetic investigations have shown the BNYVV gene occurring in Normandy is different from the Danish source and that local authorities responsible for landscape/nature reserve management have accepted this genetic finding as a sufficient justification for the development of a genetic reserve project at the Baltic Sea coast. BNYVV causes the rhizomania disease of sugar beet. It is further known that this subspecies is living in a very dynamic habitat. The number of individuals of local populations changes over time, at some sites the local settlements get lost while other sites are newly colonized, and some populations prove to be more stable at very suitable, sheltered sites over time. Hence, the subspecies forms a meta-population in this region and the management task consists of maintaining the BNYVV gene (and the possible further evolution of this gene) in that meta-population (see Fig. 11). Which data and IT tools do we need for the *in situ* management of this germplasm? How can we monitor and perhaps even predict the population development in the region both in space and time? Concepts for recording data in the field described in preceding chapters actually matured after a test trip to the Baltic Sea coast in the summer of 2005. A close interaction between the developer of information systems and the user community is crucial for its functionality and acceptance. This is certainly not a new finding but it is one which is sometimes forgotten.



**Fig. 11.** How a *B. vulgaris* subsp. *maritima* genetic reserve aiming at the maintenance of variation for BNYVV resistance could look. The size would depend on the distribution pattern of the BNYVV gene(s) and the size of a meta-population required to safeguard the gene(s) within this area.

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# Evaluation of differential rooting and water use characteristics of sugar beet genotypes under field drought conditions

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# Introduction

Water availability limits sugar beet production in the UK and in many other regions worldwide where rainfall is insufficient and irrigation is not possible or is a limited resource. We are engaged in working with breeders to develop varieties with greater drought tolerance and water use efficiency. An important characteristic of such varieties is the ability of root systems to access stored soil water. Currently, there is no information on genotypic diversity for depth of rooting and water uptake capacity in sugar beet. Therefore, we have studied a range of diverse sugar beet hybrids under managed drought conditions in the field to discover whether certain genotypes can express greater ability to mine soil water than others. Identification of superior genotypes will enable breeders in the future to incorporate favourable alleles into current breeding material.

# Materials and methods

Experiments were conducted in the field at Broom's Barn from 2003 to 2005, with 9, 6 and 14 diverse hybrids in each trial, respectively. A randomized complete block design was used with four replicates. At approximately 40 days after sowing, plots were covered with polythene using large tunnels (Ober et al. 2004) to impose drought until two weeks before harvest in early October. Adjacent to the polytunnels, a duplicate trial was conducted under fully irrigated conditions to assess unstressed yield potential. Plots were harvested by hand and root and top mass were measured. Roots were processed and sugar content was determined as described previously (Ober et al. 2004).

Root activity in the soil was estimated by measuring changes in soil water content through the soil profile throughout the season, beginning when the plots were covered and drought was initiated. Soil moisture content was measured with a capacitance-type soil moisture sensor (Diviner 2000, Sentek Ltd., Australia) which was lowered into PVC tubes installed in plots to a depth of 1.2 m. Weekly or fortnightly changes in water content indicated extraction of water by roots, and therefore indicated the extent of rooting at each depth of measurement from the soil surface (Ober et al. 2005a). In a preliminary test of the capacitance probe, similar data were obtained in adjacent plots using a neutron probe (Ober et al. 2005b). This indicated that the capacitance probe, which is easier to use, was a valid tool for the study.

# **Results and discussion**

Genotypes showed significant differences in patterns of soil water extraction at different times during the drought period (Ober et al. 2005a). In particular, there were significant genotypic differences in rates of water extraction deep in the soil profile (Fig. 1). The amount of water removed from 110 cm from the surface was positively correlated with sugar yield under droughted conditions. There was also a positive correlation with yield under irrigated conditions (Fig. 2).



**Fig. 1.** Patterns of water extraction from a 10 cm thick layer of soil centred at 110 cm from the soil surface. Measurements were made fortnightly over the course of the season under droughted conditions in 2004.





**Fig. 2.** Relationship between water use and sugar yield under droughted and irrigated conditions. Water use was measured on droughted plots from a 10 cm layer measured at 110 cm from the surface, summed over the season. Results are shown from experiments in 2003 and 2004. Symbols are the means of four replicates. The coefficient of determination is significant at P < 0.05 (\*) or P < 0.01 (\*\*) where indicated.

Experiments in the other years with different sets of hybrids showed similar results (data not shown). This indicates that deeper rooting was a function of overall growth vigour. Therefore, the deeper root systems of genotypes with larger droughted yields may not have been a response to drought per se. However, the data indicate that it is possible to detect relatively small genotypic differences in rooting patterns in the soil under field conditions, and that these differences could be exploited by breeders. However, current methods are not appropriate for large-scale screening of breeding programmes. Therefore, marker-assisted selection would probably be required. These results show that parental lines for developing mapping populations could be selected using the techniques described here.

Despite differences in patterns of water extraction deep in the soil profile, absolute differences in the quantities of water were small in comparison with the season-long water use summed over the entire soil profile. The genotypic differences in total seasonal water use were small and statistically insignificant (Ober et al. 2005c). However, there were large differences in sugar and total dry matter yield; hence, there were significant genotypic differences in water use efficiency. This means that some genotypes that showed smaller yields "wasted" water compared with higher yielding genotypes. These results show that genotypes with poor water use efficiency can be eliminated from breeding programmes, which is particularly important for semi-arid areas such as Iran.

#### Conclusions

There is significant genotypic diversity within sugar beet germplasm for rooting traits, soil water extraction from deep soil layers, and water use efficiency. Screening techniques under managed drought conditions in the field have been established, and could be used to screen a larger, more diverse array of sugar beet germplasm in order to identify sources of much greater differences in rooting patterns than those observed in the studies reported here.

#### Acknowledgements

This research was funded by the British Beet Research Organisation, with further support from KWS SAAT AG, Germany. Thanks to Abouzar Rajabi from the Sugar Beet Seed Institute of Iran who made some of the measurements as part of his PhD Thesis work, and to the Crop Productivity Group team and field staff at Broom's Barn.

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# Evaluation of beet germplasm and progress towards the development of sugar beet for resistance to diseases and for root structure

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#### Introduction

The importance of resistance/tolerance genes controlling biotic stresses and disease infections has led to extensive evaluation of sugar beet germplasm. In recent years, root diseases have become more prevalent throughout the sugar beet growing areas of Iran. Yield in crop plants has been reported as a function of morphological and physiological characteristics. Superior varieties in sugar beet consist of plants producing desirable root yield and uniform structure.

The Sugar Beet Seed Institute (SBSI) has carried out research activities aimed at the development of materials resistant to rhizomania, nematodes etc. The most important topics are as follows.

#### Evaluation of germplasm for resistance to *Polymyxa betae* Keskin.

Polymyxa betae is the carrier of the beet necrotic yellow vein virus (BNYVV). This soil-borne fungus transfers the virus to sugar beet. Resistance to this agent has been reported in some species of the genus Beta (Paul et al. 1993; Barr et al. 1995). Adding the fungus resistance gene(s) into sugar beet varieties along with resistance to the virus can guarantee strongly rhizomania-resistant varieties. Sixteen accessions from four different sources of beet germplasm (B. vulgaris, B. vulgaris subsp. maritima, B. macrocarpa and B. procumbens) were evaluated for resistance to Polymyxa betae under greenhouse conditions (Etemadifar et al. 2004). Seeds of each accession were sown in sterile sand and watered with nutrient solution regularly. Seedlings were planted in an infested soil at the 2- to 4-leaf stages. After 70 days, roots were examined microscopically for fungus spores. The results showed that spores were not observed in the roots of the species B. procumbens and B. macrocarpa originating from the Canary Islands and Germany, respectively (Table 1). A few plants of the two other accessions, B. vulgaris and B. vulgaris subsp. maritima, were not infected by fungus. It seems that these plants had escaped from disease infection. Therefore more studies are being conducted to identify the most interesting accessions and to incorporate resistance genes into breeding programmes of sugar beet.

Accession	Origin	No. of	plants	
		Total	No. of susceptible plants	No. of resistant plants
B. vulgaris	Greece	41	41	-
B. vulgaris subsp. maritima	Ireland	34	34	-
B. macrocarpa	Germany	24	-	24
B. vulgaris subsp. maritima	Turkey	15	13	2
B. vulgaris subsp. maritima	Greece	20	20	-
B. vulgaris	China	37	37	-
B. vulgaris subsp. maritima	Iran	18	18	-
B. vulgaris subsp. maritima	Iran	22	22	-
B. vulgaris	Germany	15	14	1
B. procumbens	Canary Islands	33	-	33
B. vulgaris	Spain	38	38	-
B. vulgaris subsp. maritima	France	36	36	-
B. vulgaris subsp. maritima	Ireland	28	28	-
B. vulgaris subsp. maritima	Holland	32	32	-
B. vulgaris subsp. maritima	Tunisia	24	24	-
Susceptible check		41	41	-

Table 1. Evaluation of Beta germplasm for resistance to fungus Polymyxa betae

# Transfer of rhizomania resistance gene(s) from *B. vulgaris* subsp. *maritima* and cultivated sources into sugar beet

Rhizomania is one of the most important viral diseases influencing sugar beet production in the world (Tamada and Baba 1973; Rush and Heidel 1995). A general solution to overcome this obstacle is to develop resistant varieties. Two resistant sources named WB42 (B. vulgaris subsp. maritima) and Holly (a cultivated source) were used to transfer desirable genes into the existing monogerm lines (Mesbah et al. 2005a). Plants of both sources were crossed to four male sterile monogerms (428, 419, 261 and 231) and F1 generations were evaluated for resistance in the field as well as by using enzyme-linked immunosorbent assay (ELISA) tests. The resistant plants were backcrossed to the recurrent parent. At the same time some of the F1 plants were crossed to an unrelated recurrent parent. BC1-F1 generations were tested under infested soil conditions and the selected resistant plants intercrossed in the isolated plots to produce BC1-F2 generations. Progenies from the cross between sugar beet and wild WB42 required more backcrosses to eliminate undesirable habits from the wild donor parent. Results of BC1-F2 generations in the field under severe infection by rhizomania are presented in Table 2. The performance of some backcrosses was as high as that of the resistant check varieties. The genotype No. 3 yielded 30.55 t/ha roots, higher than the resistant check at 22.35 t/ha (entry No. 12). Indeed, this genotype demonstrated the highest white sugar yield (3.78 t/ha) and its optical density (OD) appeared to be as low as 0.047. Entry No. 5 was also another promising BC1-F2 which performed higher than the check variety with an OD value 0.028 as compared to the other crosses. Some crosses performed as well as the resistant check with respect to the root yield, sugar content and other characteristics (entries Nos. 6 and 8). These resistant BC1-F2s are promising breeding materials that can be considered for use in breeding programmes to improve resistant pollinators and monogerm inbred lines.

No.	Genotype <sup>(1)</sup>	Root yield (t/ ha)	Sugar content (%)	White sugar yield (t/ha)	<b>OD</b> <sup>(2)</sup>
1	428 x (428 x 1013)	19.75	10.94	1.29	0.362
2	31 x (231 x W-114)	16.53	12.37	1.44	0.172
3	231 x (231 x 1013)	30.55	15.40	3.78	0.047
4	261 x (261 x 1013)	19.45	15.10	2.36	0.111
5	261 x (261 x W-114)	27.25	15.33	3.33	0.028
6	231 x (231 x 1012)	22.22	14.10	2.28	0.218
7	419 x (419 x W-114)	9.92	13.88	1.02	0.632
8	231 x (428 x 1013)	21.56	14.18	2.35	0.101
9	231 x (261 x W-114)	19.05	12.73	1.67	0.380
10	231 x (419 x W-114)	15.87	15.63	1.95	0.287
11	261 x (419 x W-114)	7.54	13.07	0.69	0.379
12	Resistant check	22.35	15.30	2.85	0.204

 Table 2.
 Evaluation of BC1-F2 generations of beet in a severely infested field of rhizomania in Shiraz, 2004

<sup>(1)</sup> 1012, 1013 and W-114 = cultivated resistant sources

<sup>(2)</sup> OD = Absorbance at 405 nm

# Transfer of beet cyst nematode resistance genes from resistant sources to sugar beet

Sugar beet cyst nematode (*Heterodera schachtii* Schmit.) is considered as one of the limiting factors in sugar beet cultivation. For the development of resistant varieties, many efforts have been made to transfer resistant genes from wild species to sugar beet. One accession of the wild subspecies *B. vulgaris* subsp. *maritima* showing partial resistance to sugar beet nematode was used in a crossing programme. In addition, one translocation line containing the HS1Pro-1 resistance gene originating from *B. procumbens* was also crossed with sugar beet monogerm lines.

Resistant plants of F1 generations which had been selected from infested soil in greenhouse conditions were backcrossed to nuclear male sterile plants of a monogerm O-type line. Selected resistant F1s were crossed to sugar beet to produce the BC1-F1 generation. Different BC1 crosses were tested under normal and also severely infested field conditions (Table 3). Some selected plants of the later generations showed high yields under infested conditions as compared to the resistant check (entries Nos. 1, 9 and 10). Individual selections of resistant plants can be taken for developing either pollinator or cytoplasmic male sterile (CMS) lines.

No.	Genotype <sup>(1)</sup>	Normal con	nditions Infested co			nditions	
		Root yield (t/ha)	Sugar content (%)	White sugar yield (t/ha)	Root yield (t/ha)	Pf/Pi <sup>(2)</sup>	
1	231 x (MSNB1 x W-1009)-3	64.27	14.66	7.33	46.42	2.95	
2	231 x (MSNB1 x W-1009)-2	65.11	14.13	6.93	35.17	0.71	
3	231 x (MSR x W-1009)-3	63.65	13.50	6.43	39.83	0.66	
4	231 x (MSC2 x W-1009)-1	56.17	14.19	5.91	35.75	1.30	
5	231 x (MSC2 x W-1009)-2	59.58	13.29	5.76	38.08	1.27	
6	231 x (MSNB1 x W-1009)-1	67.50	13.95	6.94	39.67	0.98	
7	231 x (MSR x W-1009)-2	62.08	12.50	5.18	37.17	0.52	
8	231 x (MSC2 x W-1009)-3	60.52	13.61	5.98	28.50	0.66	
9	231 x (MSG x W-1009)	58.85	13.76	5.92	48.33	-	
10	231 x (MSR x W-1010)	49.51	13.63	4.91	45.11	-	
11	231 x (MSC2 x W-1010)	41.46	13.65	4.17	36.44	-	
12	231 x (MSNB1 x W-1010)	52.92	14.34	5.85	38.97	-	
13	231 x (MS261 x W-1010)	56.12	16.00	7.41	34.89	-	
14	W-1009	65.21	12.85	5.84	37.83	0.58	
15	Resistant check	68.85	12.14	5.35	48.08	0.72	
16	Resistant check	55.16	13.59	5.44	43.26	0.88	
17	Susceptible check	74.17	13.26	6.88	32.56	-	

 
 Table 3. Evaluation of nematode resistant BC1 beet lines under normal and nematodeinfested soil conditions in Chenaran, northeast Iran, in 2004

<sup>(1)</sup> W-1009 = translocation line; W-1010 = wild resistant source

<sup>(2)</sup> Pi = Nematode population in 100 g soil before planting; Pf = Nematode population in 100 g soil after harvesting

# Transfer of root roundness and smoothness from fodder beet and red beet to sugar beet

A primary evaluation of fodder and red beet landraces was made in Karaj during 1996-1998. Important characters like root shape, root colour, chemical components, grooved roots, petiole colour and agronomic characteristics including root weight and sugar content were also compared among populations (Arjmand et al. 2004) (Fig. 1).

The development of smooth-root sugar beet without grooved roots started in 1998 by crossing between sugar beet x fodder beet and sugar beet x table beet (Mesbah et al. 2005b). Crosses were made between the selected landraces and nuclear male sterile of O-types which resulted in segregating populations of sugar beet x fodder beet and sugar beet x table beet. Selected roots were then backcrossed to sugar beet for two generations and the subsequent F2 populations were examined for root shape, root smoothness, sugar yield, root yield, sugar content and non-sugar components (Table 4). The results showed that sugar content has been raised up to 14.87% with sugar extractability of 82%. In these populations, the frequency of smooth-root architecture with globe-shaped and conical-shaped roots has considerably increased in the improved generations (Fig. 2).



Fig. 1. Structure of fodder beets (right) compared with sugar beet (left).



Fig. 2. Root structure of selected F2 generations from crosses between sugar beet and fodder beet.

Selected materials in BC1-F2 segregating populations from crosses between fodder beet x sugar beet and sugar beet x red beet resulted in smooth uniform roots (entries Nos. 4, 7, 8, 9, 14 and 15, Table 4). Sugar beet root structure shows great variations according to genetics and environments. Sucrose content continues to be the greatest challenge in breeding smooth-rooted varieties under semi-arid conditions, since the amount of sodium has been consistently higher in smooth-rooted genotypes than in current commercial varieties.

These materials are desirable for the identification of O-types as they have been backcrossed to nuclear male sterile (NMS) O-types for two generations. More research should be conducted on the selection of pollinators for high sugar and low sodium and potassium contents on the basis of individual roots and family lines.

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So.	Genotype	Root	Sugar	White	Root shaj	pe (%)				
		yield (t/ha)	content (%)	sugar yield (t/ha)	Circular	Broad elliptic	Smooth conical	Narrow- grooved broad elliptic	Deep- grooved broad elliptic	Narrow obtriangular
	231 x (MSNB1 x W-1002)	52.12	14.87	6.25	5.71	5.71	57.15	17.14	8.57	5.71
2	231 × (MS261 × (MSNB1 × 7221))	63.19	13.20	6.04	11.90	7.14	47.62	4.76		28.57
с	231 × (MSNB1 × (MS261 × W-1002-I))	68.07	12.33	6.50	20.00	32.50	20.00	5.00	7.50	15.00
4	231 × (MSNB1 × (MS261 × 7105))	61.39	13.72	6.63	16.67	19.44	44.45	·	5.56	13.89
5	261 × (MS261 × W-1002-I)	48.32	12.42	3.93	13.64	18.18	31.82	60.6	9.09	18.18
9	261 × (MS261 × (MS261 × W-1003)	47.50	11.43	3.30	8.57	8.57	62.86	5.71	2.86	11.43
7	261 × NO-6-I	66.57	11.58	4.71	7.50	5.00	72.50			15.00
ω	261 × (MSNB1 × N0.6-I)	57.22	11.72	4.13	6.67	11.11	71.11			11.11
6	261 × (MS261 × N0.6-I)	54.38	11.88	4.02	2.63	5.26	47.37	7.89	7.89	28.95
10	261 × (MS261 × (MS261 × W-1002-I))	45.00	12.68	3.86	7.14	3.57	46.43	28.57	3.57	10.71
£	261 × (MSNB1 × (MSNB1 × W-1002-II))	58.33	11.50	4.26	8.33	12.50	41.67	20.83	16.67	
5	261 × (MS261 × (MSNB1 × W-1003))	53.19	13.70	5.30	16.67	16.67	25.00	37.50		4.17
13	261 × (MS261 × (MS261 × W-1003))	43.61	10.92	2.69	3.33	3.33	30.00	43.33		20.00
4	261 × (MS261 × (MSNB1 × 7221-I))	55.53	11.15	4.06	9.38	12.50	59.38	12.50		6.25
15	231 × (MS261 × (MS261 × W1003))	68.08	12.25	5.87	6.25	6.25	65.63	3.13	3.13	15.63
16	231 × NO.6-I	70.41	10.00	4.04	25.00	6.25	37.50	3.13	3.13	25.00
17	W-1005 check	68.95	12.75	6.06	93.33	ı	6.67			
18	W-1006 check	59.21	12.58	5.32	76.00	,	24.00			
19	W-1014 check	64.58	14.53	7.41	89.74	·	10.26			

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### Genealogical structure of a collection of beet

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#### Chronology of receiving beet samples in the collection

The first beet accessions in the collection of the N.I. Vavilov Research Institute of Plant Industry (VIR) were brought by N.I. Vavilov from Afghanistan (1924), the countries of the Mediterranean (1926-1927) and Indochina (1929). Subsequently the collection has constantly been replenished by collecting missions on the territory of Russia and abroad, and by means of seed exchange with genebanks and breeding companies.

It is possible to demarcate several periods in the updating of the VIR's collection. The first is the pre-war period (until 1941), when a total of 238 accessions were collected: 87 of table beet, 88 of fodder beet, 18 of spinach beet (chard), 55 of wild beet and only 10 accessions of sugar beet. During the post-war period, starting in 1950, the collection was replenished basically with diploid populations. In the same period the first tetraploid forms were obtained. During the third (modern) period, starting in 1980, hybrid accessions of beet prevailed.

The accessions may be divided into three groups: (1) primitive forms, (2) old varieties, and (3) breeding varieties.

Accessions of the first group include 'Long smooth', 'Palkovidnaya' and 'Chernokrasnaya'. Characterized by long and frequently very branchy roots, they are promising for breeding cultivars with such traits as intense flesh colour, non-bolting and long-term root storage ability.

Accessions of the second group are represented by old varieties such as 'Bassano', 'Granat', 'Cameroon', 'Trevize' and 'Hediv', characterized by high root density, with root shapes varying from flattened spherical to conical and cylindrical; some of them possess disease resistance. Varieties of the 'Granat' type, with an oval-cylindrical root shape, were the source of modern breeding varieties of the 'Cylindra' type.

The large third group (about 50%) includes advanced varieties such as 'Egyptian Flat', 'Crosby' and 'Detroit'. This group also includes early-ripening varieties ('Early Wonder'), cold-resistant ones ('Eclipse', 'Granat'), and those suitable for long conservation ('Ohio'). A special group consists of table beet varieties for ornamental purposes ('Black Queen', 'Dracenolistnaya' and 'Crapoden') and for green leaves ('Green Top' and 'Long Season'), possessing drought and heat resistance.

The degrees of relationship and origins of different beet species and forms may be ascertained by interspecific crossings. For example, the species of the section *Patellares* are practically impossible to cross with cultivated beet and species from other sections. Hence, the species of this group are the most remote from the others, and their genomes must be significantly divergent. Crosses between species of the section *Corollinae* are difficult to obtain, and their hybrids have reciprocal differences. For example, *B. vulgaris* x *B. lomatogona* hybrids are sterile, while *B. lomatogona* x *B. vulgaris* hybrids are fertile. *B. vulgaris* and *B. trigyna* are crossable, but their hybrids are sterile. Crosses between *B. lomatogona* and *B. trigyna* yield hybrids of two types: triploid and hexaploid, with only the hexaploids being fertile. Hence, the species of this section have significant variation in their genomic structure. In the section *Beta (Vulgares)* crossing goes easily,

hybrids possess high fertility, and their progeny is prolific, which testifies that the species of this section are closely interrelated and possess a common genome.

Most researchers regard the species *Beta vulgaris* subsp. *maritima* (L.) Arcang. as the ancestor of cultivated beet. Owing to its plasticity and high polymorphism, *B.v. maritima* spread over a vast area: from India via the Mediterranean coasts to England and Norway. This resulted in its significant differentiation and the development of biennial forms (in VIR's collection accessions of this type are called 'Northern wild beet'), distinguishable from annual forms, such as 'Algerian'. Selection of *B. v. maritima* biotypes (leafy forms with incrassate roots and a biennial development cycle) led to the breeding of modern root beet (Krasochkin 1971). VIR's collection contains many primitive and transitional semi-root forms ('Balykessirskaya', 'Alashehirskaya', 'Badahshanskaya') from Central and Western Asia to confirm this assumption. Further distribution and improvement of semi-root and primitive root forms enables breeders to develop modern, highly productive varieties of table and fodder beets.

There are different opinions concerning the origin of sugar beet. Some authors argue that sugar beet evolved as a result of natural hybridization between fodder beet and chard. Their arguments include the high density of flesh, large number of conducting bundles and high sugar content in chard and sugar beet roots (Zosimovich 1940, 1968; Krasochkin 1971). At the same time, V.T. Krasochkin (1971) noted that Northern wild beet, the richest in sugars among all wild forms (up to 14-20%), may have taken part in the genesis of sugar beet together with chard.

In our experience, Northern wild beet (K-1384) from Sweden was characterized by incrassate roots which after twice- or thrice-repeated selection became closer to those of sugar beet. According to the data of the Biochemistry Laboratory of VIR, the content of dry matter in Northern wild beet roots reached 22-23%, and sugars up to 14-15%. Notably, the sugar content in dry matter amounted to 62% (in sugar beet 71%, fodder beet 58.5%, and table beets 59.5%), i.e. it was the same as in chard. When sugar beet was crossed with Northern wild forms, the F1 hybrids and subsequent generations had less branchy roots than its hybrids with chard, which is further evidence of their relationship.

Additional data were obtained after the immunochemical structure of seed fibres had been analysed (Burenin and Gavrilyk 1982). The component specific to seed fibres of biennial *B. v. maritima* (from Sweden) was found in the fibres of cultivated beet but was absent in chard and annual *B. v. maritima* occurring on the Mediterranean coasts. The analysis of hybrid seeds showed that the similarity with *B. v. maritima* proteins was observed only in the combination *B. v. maritima* x sugar beet, whereas its hybrids with table beet and fodder beet had a different protein structure, which testified to their divergent origins.

#### Genealogy of table beet varieties

Multifactor studies of the collection have shown that the greatest diversity of original ancient forms of beet came from Asia Minor and the Transcaucasus (Krasochkin 1959, 1971; Burenin 1983; Burenin and Pivovarov 1998). Anthocyanin-pigmented biotypes, close to modern table beet cultivars, were identified among them. Various primitive forms with lengthened conical or shortened conical, less frequently oval-shaped roots were found in the populations collected in Central Asia and the Mediterranean region (Cyprus and Crete). The analysis showed that these forms had originated in Western Asia (Krasochkin 1971).

According to experimental data, 'Balykesirskaya' beet was the first form to develop from wild beet through chard to the root form (see Fig. 1); its populations were initially collected in Balykesir area in Asia Minor, then in Transcaucasus and Afghanistan. Its root is stalky, branchy and white-coloured, with white or pinkish flesh. Traits inherited from wild beet are slow seed germination and aptness to bolt.

'Alashehirskaya' beet (accessions collected in Alashehir, Turkey) is close to 'Balykesirskaya' beet by its biological properties; the plant and especially its root are distinguished for various hues of red colour (Krasochkin 1960).

'Adanskaya' (bright orange roots of rounded shape) and 'Af'onkarahissarskaya' (white oval-conical roots) beets are considered transitional to cultivated forms, but 'Abkhazskaya Krasnaya' is the closest to modern varieties (roots which are intensively red-coloured, rounded or flat occur in populations). It is valuable for breeding for heat resistance. 'Shotlandka' accession, represented by populations from Abkhazia, Armenia and Turkey, has a root similar to the Egyptian type, with a yellowish shade of flesh and rough skin; it is characterized by earliness and non-lodging of the seed-bearing plant. Together with 'Polosatochereshkovaya', 'Turkestanskaya' and 'Chardjuiskaya' beets, they represent those primitive table beet varieties that were the initial source of the modern ones (Fig. 1).

'Bassano', an old Italian variety of apparently Western Asian origin, may also be classed as an old landrace. It has pink-violet or red-violet roots of round-flat shape, with pink-red or violet-red flesh, and shows resistance to salinity and a number of diseases. This group also includes: 'Grushevidnaya Chernaya', with its piriform-ovoid root and red leaves; 'Covent Garden' with red leaves, dark-red conical root and darkred flesh with violet tinge; 'Crapoden', which has an elongated root of almost blackred colour, speckled (reticulate) skin and black-red flesh; 'Green Top' with a cinnabarred oval-conical root, bright red flesh with an orange tinge, and green leaf blades.

Among advanced varieties nine convarieties have been described, six of which are most widespread (see Fig. 1). They are characterized by dark-red skin colour and dark-red (without light rings) root flesh. Morphological differences are mainly expressed in the root shape, varying from flat and flat-rounded to roundish, oval-round or elongated (cylindrical). They considerably differ in their biological properties (drought and heat resistance, cold resistance, non-bolting, earliness/ lateness and resistance to diseases).

Such varieties as 'Egyptian Flat' are the most widespread all over the world; they are characterized by their flat root shape, shallow submersion in soil, dark-red skin and flesh, earliness and resistance to bolting. Convar. 'Egyptian Round' ('Crosby') is close to 'Egyptian Flat', but differs in its round-flat root shape.

Varieties of the 'Detroit' type from Canada penetrated into the USA and became widely distributed all over the world. They are characterized by round-oval and ovalcubic root shape, dark-red flesh, marketability, transportability and good keeping qualities. 'Bordeaux 237' was bred at Gribovskaya Experimental Station (nowadays All-Russian Institute of Vegetable Breeding and Seed Production, VNIISSOK) by cross-pollination between several table beet varieties and accessions of the 'Detroit' type; it is characterized by a round-oval root, dark-red flesh and earliness. This variety is cultivated practically everywhere in the Russian Federation.

Convar. 'Eclipse' is characterized by round or oval-round roots with skin and flesh colour similar to 'Egyptian Round'. Varieties are cold-resistant and early-ripening;

their flesh quite often has pink-white rings because of rapid accretion of the root. In recent years varieties such as 'Cylindra' have become widespread in the Russian Federation and abroad. They originated from very old landraces of the 'Globe' type with an oval-conical root.

The presence of diverse and well-investigated source material, as well as knowledge of the pedigree of the collection accessions, will undoubtedly promote their effective use in breeding practice.



Fig. 1. Genealogical tree of table beet varieties.

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# Beta genetic resources activities in Germany (2003–2005)

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The 01 February 2002 marked the start of the establishment of the federal central genebank at the Institute for Plant Genetics and Crop Plant Research (IPK). Between 2003 and 2005 the germplasm collection held by the Federal Centre for Breeding Research on Cultivated Plants (BAZ) at Braunschweig was merged into the IPK holding. The reorganization of genebank work was financially supported by the Ministry of Science and Technology (BMBF) as well as the Ministry of Food, Agricultural and Consumer Protection (BMELV).

### Information management

Within the framework of this project a new Genebank Information System (GBIS) is being developed and implemented in an Oracle environment. It consists of two parts: (1) GBIS/M, the internal genebank management software, offers various functions for the day-to-day genebank activities. Data are being migrated from the existing systems into GBIS. GBIS/M will be fully implemented during 2006; (2) GBIS/I, the Internet portal will include an online seed ordering component. Its availability is planned for summer 2006. In addition a Java application called GBIS/B supporting the use of Personal Digital Assistants (PDAs) for recording field observations was developed.

# Beta holding

The national holding is described in Table 1. Thirty-two percent of the total holding requires regeneration.

<sup>&</sup>lt;sup>13</sup> Now the Leibniz Institute of Plant Genetics and Crop Plant Research.

<sup>&</sup>lt;sup>14</sup> The German Federal Ministry of Food, Agriculture and Consumer Protection (BMELV) merged the former Federal Centre for Breeding Research on Cultivated Plants (BAZ), the Federal Biological Research Centre for Agriculture and Forestry (BBA), and parts of the Federal Agricultural Research Centre (FAL). Since January 2008 the new institution is called Federal Research Centre for Cultivated Plants – Julius Kühn-Institut.

Botanical name	No. of accessi	ons		
	Available	Needing	Total	
		regeneration		
Beta corolliflora	35	49	84	
Beta intermedia	95	120	215	
Beta lomatogona	30	68	98	
Beta macrocarpa	25	18	43	
Beta macrorhiza	14	18	32	
Beta nana	0	14	14	
Beta patellaris	33	5	38	
Beta patula	1	4	5	
Beta procumbens	5	0	5	
Beta trigyna	12	14	26	
Beta vulgaris	98	52	150	
Beta vulgaris - Fodder Beet	185	41	226	
Beta vulgaris - Garden Beet	245	32	277	
Beta vulgaris - Leaf Beet	183	36	219	
Beta vulgaris - Sugar Beet	222	35	257	
Beta vulgaris subsp. adanensis	32	5	37	
Beta vulgaris subsp. maritima	366	178	544	
Beta webbiana	3	0	3	
Beta sp.	0	46	46	
Total	1584	735	2319	

Table 1. Numb	per of acces	ions in the	German Ber	a collection	by species
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# **Quality standards**

In November 2005 IPK started to prepare the certification of the genebank core processes with the aim of reaching the standard required for the certificate DIN EN ISO 9001:2000 in April 2007. Genebank management, evaluation and documentation will be included. The procedures for *Beta* are based on or were derived from manuals existing at the IPK and from information available at http://www.ecpgr.cgiar.org/Workgroups/beta/beta. htm#management. It is intended to update the German data.

# Procedure (process instructions) for multiplication of wild and cultivated beets (*Beta* spp.) used at IPK

Introduction	Multiplications have to fulfil quality requirements to ensure
	maintenance of genetic identity and integrity and a high quality
	of the seed material.
Isolation	Wild and cultivated beets are almost all outbreeders. Material
	from these plants is therefore multiplied in small greenhouses or
	in the field under isolated conditions.
Population size	The multiplication is done on an average of 50-100 plants (at least
-	25, 150 at most). If material germinates poorly or very slowly, these
	findings are recorded in the multiplication protocol. The number
	of plants used in multiplication is recorded for every accession.
Cultivation	Beets are annual, biennial or perennial. Depending on the
	life cycle the assortment manager decides on the method of

	multiplication. During the cultivation a control of the material
	will be done. Plants are not selected. However, if an accession is
	more heterogeneous than was expected on the basis of passport
	data or if it is a mixture of species or types the assortment
	manager together with the person in charge of the group "genetic
	resources and reproduction" decides if and how plants may be
	selected. This decision is recorded in the multiplication protocol.
Pollination	Beets are almost all wind-pollinators. Therefore, a special method
	for pollination is not necessary.
Harvest	All inflorescences with ripe seeds are harvested (if necessary at
	several intervals).
Identity	During sowing, cultivation and harvest, accessions must be clearly
	marked with a label giving the accession and the batch number.
Seed quality	The bags with the harvested plant parts are dried in a drying
1 2	chamber. The seeds are cleaned and a final seed check is done by
	the assortment manager. After an effective germination test the
	seeds are transferred to the cold storage chamber (at $-15^{\circ}$ C).
Deviation	Any deviation during the cultivation is recorded and copied into
	the multiplication protocol.

# **Descriptors used at IPK**

Reference numbers listed in the "Descriptors for *Beta*" (IBPGR 1991; IPGRI 1996) are shown between brackets. These plant traits are used at the location Gatersleben for primary characterization.

	Passport data
	Sowing date/emerging date
	Hibernation
	Planting
	Bolting
	Flowering time
	Harvest date / yield / 1000 seed weight
	Remarks
	Plant height (cm)
(4.3.2)	Growth habit
	Leaf shape
(4.1.11)	Leaf colour
(4.1.12)	Leaf pigmentation
	Colour of petiole and leaf ribs
	Leaf surface
	Leaf hairiness
	Beet
	Beet shape in longitudinal section
	Beet shape in transverse section
(4.2.6)	Beet position in soil
(4.2.11)	Beet colour
(4.2.12)	Flesh colour
	Zonation
(4.2.15)	Colour of zonation

### Distribution of germplasm (2003–2005)

The number of accessions distributed in Germany and abroad is shown in Table 2.

Year	No. of accessions distribution	ted	
	within Germany	abroad	
2003	53	26	
2004	16	155	
2005	27	51	

Table 2. Distribution of germplasm (2003–2005)

### **Evaluation and research**

Evaluation of the national *Beta* genetic resources holding has been stopped pending further notice. In the public sector, the University of Kiel, Institute of Plant Breeding, is engaged in breeding research on *Beta*. The projects focus on the bolting gene *B*, genes responsible for storage root development and on the functional analysis of the *Hs1* gene. *Hs1* is the gene from *B. procumbens* conferring resistance to the beet cyst nematode. Breeding companies co-fund research projects dealing with *Rhizoctonia* resistance, the occurrence, spread and pathogenicity of various types of the rhizomania virus (beet necrotic yellow vein virus, BNYVV) at the Institute of Sugarbeet Research (Göttingen), as well as a bioinformatics project at the University of Hohenheim, all of which are to some extent based on or related to genetic diversity of *Beta*.

# Contributions to the ECPGR programme

*B. vulgaris* subsp. *maritima* is the only wild species with a very limited, although increasing distribution area in Germany. The BAZ has assumed tasks in the field of *in situ* and on-farm management which, in the case of *Beta*, can only be seen in a broader European context (see Frese and Germeier, this volume, pp. 59-74; Frese et al., this volume, pp. 45-52). The BAZ at Braunschweig operates the International Database for *Beta* (IDBB). Recent improvements to this information system are described in the above-mentioned paper by Frese and Germeier.

# Beta genetic resource activities in India (1990-2005) – a review

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#### Introduction

Sugar cane (Saccharum species complex) and sugar beet (Beta vulgaris L. subsp. vulgaris, Sugar Beet Group) are the two main sugar-producing crops in the world. Commercial cultivation of sugar beet as a supplementary sugar crop was started in India in 1971 to augment sugar production from sugarcane in the hot summer months of April and May when sugar recoveries from sugar cane show a steep declining trend. A cane cum beet diffusion plant was set up at Sriganganagar in 1971, which processed sugar cane from November to March, followed by processing of sugar beet from April to May (about 60 days). The highest beet sugar recovery from this plant was obtained during the crop seasons 1978-79 and 1979-80 when it was above 11.3% (Anonymous 1981; Srivastava and Bajpai 1985). However, for unknown reasons this beet processing plant was closed in the 1990s. Recently, large-scale commercial trials of sugar beet have again been started in the state of Maharashtra in 2001 through the efforts of Vasantdada Sugar Institute (VSI), Pune and the Cooperative Sugar Factories Federation of the state of Maharashtra. The Indian Council of Agricultural Research (ICAR), Government of India, at its special meeting in Pune in July 2004, sanctioned a new Sugar Beet Network Research Project with three centres located at the Indian Institute of Sugarcane Research (IISR), Lucknow (Uttar Pradesh), VSI, Pune and the Agricultural Regional Research Station, University of Bikaner (Rajasthan). IISR, Lucknow has its Sugar Beet Breeding Station at Mukteswar, Kumaon hills, for germplasm maintenance and breeding work.

Different cultivar groups of beets, namely Sugar Beet, Fodder Beet, Garden Beet, Leaf Beet and the closely related wild form *B. vulgaris* subsp. *maritima* have originated in North-East Europe. A wide distribution of the genus *Beta* is found along European coasts and the Mediterranean coasts of Africa and the Middle East. The distribution of cultivated and wild forms extends towards some Asian countries, namely Iran, Iraq, Afghanistan, Pakistan, India, China, Nepal, and Japan (Srivastava et al. 1992; Srivastava 1995b).

#### Organization of Beta research in India

The organizational arrangements for *Beta* genetic resources research in India are under the control of the Indian Council of Agricultural Research (ICAR) and some private and non-governmental organizations. Fundamental research in sugar beet breeding, genetic resources and breeders' seed production is now being done mainly at IISR, Lucknow, and some basic work on germplasm evaluation is done by the Centre for Research and Development for Sugar Crops for Sub-optimal and Stress

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Environments (CSCSE) at Lucknow. Sugar beet varietal trials are conducted at VSI, Pune, and some trials in farmers' fields are conducted by the Cooperative Sugar Factories Federation of Maharashtra State. Research projects on garden and leaf beets are mainly done at the Indian Institute of Vegetable Research, Varanasi, and the Indian Institute of Horticultural Research (IIHR), Bangalore (both ICAR institutes), and also in many Agricultural Universities and State Research Stations in India.

# Germplasm conservation and holdings

*Beta* germplasm collections of sugar beet and wild beets are conserved mainly at two places:

- Indian Institute of Sugarcane Research (IISR), Lucknow, where short-term storage of seed at 4°C is done. Working collections are maintained at Lucknow and Mukteswar. Seed is multiplied by IISR for research purposes at Mukteswar.
- National Bureau of Plant Genetic Resources (NBPGR), New Delhi, is the National Gene Bank for all the crop plants, and has facilities for medium-and long-term seed storage.

# Current status of Beta germplasm

The *Beta* germplasm holding in India comprised 131 genotypes in 1990, while it was 241 genotypes in 2002 (Table 1). Out of this holding, the percentage of sugar beet genotypes was 61.41%. The wild genotypes are 21.16%, followed by fodder beets at 12.46%. The rest are garden and leaf beets (Fig. 1). Details of the Indian *Beta* collection by sub-taxon are given in Table 2.

Type of accessions	No. of ac	accessions         2002         8         20         70         98         15         15         15         15         15         15         15         10         2         50         148         30         8         4
	1991	2002
Sugar beets		
a. Hybrid/commercial varieties		
(i) Diploid hybrids	8	8
(ii) Triploid hybrids	10	20
(iii) Anisoploid hybrids	60	70
Total hybrid/commercial varieties	78	98
b. Breeding lines		
(iv) Diploid genotypes (open pollinated)	10	15
(v) Diploid inbreds (indigenous)	10	15
(vi) Diploid inbreds (exotic)	8	8
(vii) CMS and O-types	5	10
(viii)Tetraploid lines (indigenous)	1	2
Total breeding lines	34	50
Total sugar beets	112	148
Fodder beets	2	30
Garden beets	2	8
Leaf beets	4	4
Wild beets	11	51
Grand total	131	241

# Table 1. Structure of Beta collections at IISR (1990-2002)



**Fig. 1.** Structure of *Beta* collections at IISR, Lucknow (2002).

Cultivar	Species	Subtaxon	No. of accessions		
Group			1991	2002	
Sugar Beet	B. vulgaris L.	subsp. vulgaris var. altissima	112	148	
Fodder Beet	B. vulgaris L.	subsp. <i>vulgaris</i> var. <i>vulgaris</i>	2	30	
Garden Beet	B. vulgaris L.	subsp. <i>vulgaris</i> var. <i>vulgaris</i>	1	8	
Leaf Beet	<i>B. vulgaris</i> L.	subsp. <i>vulgaris</i> var. cicla	2	3	
	<i>B. vulgaris</i> L.	subsp. <i>palonga</i>	1	1	
Wild Beet	B. vulgaris L.	subsp. maritima var. Arcang.	1	40	
	<i>B. vulgari</i> s L.	subsp. macrocarpa	2	2	
	B. patellaris Moq.	-	1	-	
	<i>B. trigyna</i> Wald. et Kit.	-	2	3	
	<i>B. webbiana</i> Moq.	-	1	2	
	B. lomatogona Fisch et Meyer	-	-	2	
	B. macrorhiza Steven	-	1	-	
Total			126	239	

<b>Table 2.</b> Indian <i>Beta</i> collections by taxon at IISR, Lucknow (1991-200	Table	<ol><li>Indian</li></ol>	Beta collections b	v taxon at IISR.	Lucknow	(1991-2002
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#### Germplasm evaluation and characterization

At IISR, Lucknow, the "Descriptors for *Beta*" (IPGRI 1996) are used for characterization of *Beta* germplasm for botanical attributes. Observations on a limited number of traits are recorded for the root crop, namely root shape, colour, root weight, top weight; while root length, crown size and numbers of rings are recorded along with leaf characteristics. Photographs of most of the cultivated forms are taken and kept as prints or slides. Many of them are now kept as computerized files. Flowering behaviour, multigermicity, days to flowering, seed maturity, seed shattering, seed yield/plant, stem colour, anther colour, growth habit, flowering habit and annuality are recorded in the hill growing areas for sugar beet genotypes while some of these characters are noted for wild beets in the plains. Data on agronomic and quality attributes are recorded in Table 3. About 150 genotypes were evaluated in multi-location trials conducted between 1976

and 2002. At IISR, Lucknow, evaluation of sugar beet genotypes for productivity attributes from 1980 to 2002 showed some very good genotypes for high yield and gross sugar content (Table 4).

Table 3. Evaluation of sugar beet germplasm	for botanical,	agronomic and	quality	attributes
at IISR, Lucknow, India (1990–2002)		-		

Attributes	Characters
1. Botanical	Quantitative characters as per descriptor list Flowering characters in seed crop Bolting behaviour in root crop
2. Agronomic	Root yield, top yield Gross sugar (t/ha)
3. Quality	Sucrose (%) Impurity index (Na, K, alpha-amino nitrogen) Recoverable sugar (%)

Table 4. Evaluation of sugar beet genotypes for yield and sucrose at IISR, Lucknow (1990-2002)

Attributes	Achievements	Genotypes identified
Evaluation for productivity attributes	Identification of high yielding genotypes	Maribo Magnapoly, Kawe Gigapoly, Kawe Megapoly, Virtus, Solid, Maribo Monova, Maribo Monozet, IISR Comp-1, LKC-2, LS-6, Ramonkaya-06
	Identification of high sugar types	LS-6, LKC-4, IISR-2, Pant S-10, CLR pb-II/79, AJ-Poly-2, Maribo Magnapoly, Polyrave-E, KWS-E, Desprez-Poly N, K-Sacchapoly, IISR-2

On the basis of multi-location trials (data of the All India Co-ordinated Research Project (AICRP) and the Sugar Beet Network project), the following varieties were identified by AICRP workshops held from time to time in India and recommended for commercial cultivation of these varieties and their release in India:

- Indian varieties: IISR Comp-1, LS-6, Pant S-10
- Exotic varieties: Maribo Magnapoly, Maribo Resistapoly, Maribo Monova, Maribo Monozet (Denmark), Virtus, Solid (Sweden), Ramonskaya-06 (Russia)
- Other varieties identified as highly promising: Kawe Megapoly, Kawe Gigapoly (Germany), Maribo Maropoly, Maribo Unica (Denmark), Desprez-Poly N, Desprez-Poly E (France).

# Evaluation of accessions for abiotic and biotic stresses

Sugar beet genotypes comprising commercial exotic genotypes and diploid genotypes (both exotic and indigenous) have been screened for abiotic and biotic stresses as listed in Table 5.

Attributes	Characters
Abiotic stresses	High temperature tolerance
	Drought tolerance
	Losses in quality / recoverable sugar at late harvest
Biotic stresses	Disease resistance / tolerance
	Root rot diseases (Sclerotium and Rhizoctonia)
	Leaf spot diseases (Cercospora and Alternaria)
	Viruses
	Pests

**Table 5.** Screening of sugar beet and wild beet germplasm for abiotic and biotic stresses in

 India (1990-2002)

Genotypes identified as tolerant for three main abiotic stresses are given in Table 6 (Srivastava et al. 1992; Srivastava 1995a). A team of pathologists and entomologists evaluated sugar beet germplasm for biotic stresses. *Sclerotium* root rot along with *Rhizoctonia* root rot is prevalent in most of India. Accessions were screened for *Sclerotium* root rot under natural field conditions as well as under inoculated conditions in root crops at Lucknow. *Cercospora* leaf spot incidence was studied under natural conditions at Lucknow and in seed plots at Mukteswar. Genotypes identified as tolerant are given in Table 7 (Srivastava et al. 1991, 1993; Srivastava 1995a).

Table 6. Evaluation of Beta germplasm for abiotic stresses at IISR, Lucknow (1990-2002)

Attributes	Genotype identified as tolerant
Drought tolerance	IISR Comp-1, LS-6, AJ-3, Ramonskaya-06, LS-7, Maribo Marocpoly
High temperature tolerance	IISR Comp-1, IISR-2, LS-6, AJ-3, AJ-4, OPH, Maribo Magnapoly, Maribo Marocpoly
Salinity tolerance	IISR Comp-1, Pant Comp-3, CLR pb II, Maribo Resistapoly

Table	7.	Scree	nina	of	sugar	beet	aenotype	es for	<ul> <li>biotic</li> </ul>	stresses	at	IISR.	Lucknow	(1990)	-2002)
							J					- ,		· · · ·	/

Attributes – Diseases	Genotypes identified as tolerant
(i) Sclerotium Root Rot	IISR Comp-1, LS-6, Ramonskaya-06, Maribo Magnapoly, Kawe Megapoly
(ii) Cercospora Leaf Spot	LS-6, IISR-2, Kawe Cercopoly, Maribo Resistapoly, Kawe Megapoly

# • Evaluation of wild Beta germplasm

Wild germplasm consisting of 37 genotypes of *Beta vulgaris* L. subsp. *maritima* obtained from the Federal Centre for Breeding Research on Cultivated Plants (BAZ, Germany) was studied for genetic diversity during the 1997-98 crop season. Data

on nine quantitative characters were recorded; based on Sparks Non-Hierarchical Cluster analysis these 37 genotypes were grouped in six clusters. On the basis of principal component analysis, the first two principal components accounted for 63.5% of the variability (Srivastava et al. 2000).

#### Cytogenetic and karyotypic studies

The karyotype and its components were used to decipher karyo-evolutionary trends within the species of genus *Beta*. Chromosomal details of four species of the genus *Beta*: *B. vulgaris* L., *B. vulgaris* L. subsp. *maritima*, *B. vulgaris* L. subsp. *orientalis* (Roth.) Aellen and *B. lomatogona* Fisch. et Meyer with chromosome numbers 2n=18 were studied to find karyotype relationships *vis-à-vis* meiotic features to ascertain the feasibility of using these species for development of interspecific hybrids and polyploids in sugar beet breeding programmes in the subtropical climate of India (Srivastava and Srivastava 2000).

Their karyotypes were basically asymmetric. Total haploid chromatin length ranged from 17.92 to 24.17 mm, whereas individual chromosome size ranged from 1.47 to 3.15 mm. The Stebbins' class of asymmetry among these species ranged from 2A to 4A, thereby confirming the evolutionary trend among the karyotypes. The karyotype of *B. vulgaris* L. breeding line LS-6, developed at IISR, Lucknow, was the most advanced and it fell in class 4A. The karyotypes of more than one species e.g. *B. vulgaris* subsp. *maritima* and subsp. *orientalis* were in the same class i.e. 3A. To quantify further gradations in the same class of asymmetry, chromosome Dispersion Index (DI) was calculated and the DI value of 0.479 for *B. vulgaris* L. breeding line LS-6 confirmed its high karyotypic specialization.

Meiotically, all the species exhibited predominantly open bivalent formation with a few ring bivalents (Srivastava and Srivastava 1999). At metaphase, chiasmata were apparently distally localized and interstitial chiasmata were occasionally formed. The low frequency of univalents that occurred in all *Beta* species during meiosis did not affect fertility, as distribution at anaphase was regular. Chiasmata association frequency of long arm was higher than the short arm. Chiasma formation per bivalent decreased with the increase in the length of pairing blocks. This suggested the possibility of a species-specific variable gradient of chromosome condensation because sugar beet karyotypes are relatively constant at somatic and meiotic phases.

# • Biochemical and molecular analysis

At IISR, Lucknow the Biotechnological Laboratory in the Division of Crop Improvement was set up in 1997-98. Isozyme analysis in four elite diploid genotypes was done. Seventy-one bands consisting of 28 isomorphs of 6 isozyme systems, namely superoxide dismutase, guaiacol peroxidase, malate dehydrogenase, amylase, esterase and aspartate aminotransferase were resolved: 18 bands in R-06, 21 in LKC-11, 16 in IISR Comp-1, and 16 in LS-6. Although these populations could be distinguished on the basis of polymorphic isozyme spectra (Fig. 2) showing both homology and diversity in their banding pattern, the high isozyme-genetic similarity indices (mean value 0.73) among these genotypes reflected the need to survey greater numbers of enzymes and to use DNA-based markers to explore polymorphism.



**Fig. 2.** Isozyme phenotypes of some isozymes and their distribution in four elite sugar beet genotypes under subtropical Indian conditions.

Srivastava and Srivastava jointly initiated molecular marker analysis in sugar beet genotypes. Genetic diversity of four populations of sugar beet (B. vulgaris L.) consisting of genotypes IISR Comp-1, LS-6, LKC-11 and R-06, was explored using the two DNA-based molecular markers RAPD and Inter Simple Sequence Repeats (ISSR). Highly polymorphic RAPD and ISSR band profiles were obtained with an average of 11.67 and 9.75 bands per primer in sugar beet populations. The genetic similarity (GS) matrix based on Dice coefficient analysis for each marker system ranged from 0.10 to 0.74 for RAPD and from 0.12 to 0.86 for ISSR. RAPD-GS coefficients and ISSR-GS coefficients were used to cluster the genotypes based on the Unweighted Pair Group Method with Arithmetic Mean (UPGMA) method and dendrograms were constructed. The X-axis contained IISR Comp-1 and R-06, and the Y-axis contained LS-6 and LKC-11. The ability of DNA-based markers to detect a high degree of polymorphism among these populations suggested the possibility of screening a higher number of anonymous loci in sugar beet to enable the selection of the best parents in order to obtain new genetic combinations (Srivastava et al. 2007).

#### Documentation of Beta germplasm in India

At IISR a passport database for *Beta* germplasm is available. These data have been sent to the IDBB. Data on different morphological and agronomic characters have been meticulously recorded. These data are gradually being computerized and will be sent to the International Database for *Beta*.

### Future plan of work on genetic resources activities in India

Three research organizations of Lucknow, namely the Indian Institute of Sugarcane Research (IISR), Lucknow, the Centre for Research and Development for Sugar Crops for Sub-optimal and Stress Environments, Lucknow, and Lucknow University will continue working on *Beta* during the next three years, on the following topics:

- Evaluation of *Beta* germplasm consisting of sugar beet and wild beets for abiotic tolerance, i.e. high temperature tolerance, and drought tolerance under Indian conditions;
- Cytological and biotechnological studies of some diploid sugar beet types and wild species of *Beta*, according to the availability of funds and other facilities.

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Irish Sugar Ltd, Mallow, Co. Cork, Ireland

#### Ex situ collection

Irish Sugar Ltd. are the holders of the only *ex situ* collection of sugar beet genetic resources in Ireland. The collection, known as the Historical Collection, is held in the Seed Department, Irish Sugar Ltd., Mallow, Co. Cork. This collection is mainly made up of breeding lines used by Irish Sugar in their own research programmes and in programmes carried out in conjunction with Hilleshog (Syngenta) from the mid-1980s. The collection is in good condition and is mostly made up of raw processed multigerm seed of the following selections:

- Mother seed lots
- Selected breeding lines
- Winter beet selections
- Selected pollinator seed
- Cytoplasmic male sterile selections
- Polycross selections
- Other bulk seed lots.

#### In situ collections

National funding has been given to identify and evaluate the current extent and diversity, including the identification of potential threats, of native populations of maritime or sea beet (*Beta vulgaris* L. subsp. *maritima* L.). A total of 27 sites on the south and west coasts of Ireland were surveyed and identified using the Global Positioning System. National funding to continue with this survey work is expected in 2006. A separate paper summarizes the outcomes of this survey to date (Grogan, pp. 53-58, this volume).

#### Survey of wild Beta genetic resources in Morocco

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#### Introduction

Genetic diversity is necessary for the breeding process that aims at improving useful traits of plants and animal. It contributes to supplying basic germplasm and satisfying the increasing requirements of human populations, in terms of production and quality of crops.

Access to genetic resources is therefore a *sine qua non* of plant improvement. It is particularly true for sugar beet, which entirely results from selection: all ancestors of cultivated sugar beet were wild species. Despite the variability present in cultivated beet germplasm, interest in wild genetic resources is continuously increasing. Many collecting missions have been conducted worldwide, particularly in Europe and in the Near and Far East for identifying wild species related to cultivated beet, their distribution and characterization for genes of interest for breeding programmes.

In Morocco, knowledge about existing species and their distribution is very limited. An important document published by Jahandiez and Maire (1932) mentioned the existence of three species in Morocco and described the areas where they were found.

In our recently initiated study, a systematic field survey has been conducted to identify existing wild beet species and their geographic distribution throughout the country. The main objectives are the identification of wild *Beta* species and the determination of their geographical distribution and abundance.

#### Material and methods

A survey was done to identify naturally occurring wild beet species. Populations were sampled along the Atlantic coast and within the country near the Atlas and Rif mountains. This itinerary was based on the map illustrated by Jahandiez and Maire (1932), to localize target sites for collecting.

Collected seeds were divided into three sets:

- 1. Seeds were dried and stored at -20°C for long-term conservation
- 2. Seeds were stored at 4°C for short- and medium-term use
- Seeds were sown and plants are being evaluated phenotypically and used for molecular genotyping (simple sequence repeats, SSR).

#### Results

Wild species were found in all the surveyed regions. Generally, the distribution of the species was as follows:

- Beta maritima: along the Atlantic coast
- Beta macrocarpa: from 30 km away from the sea and up to 1000 m above sea level
- Beta patellaris: only in the southwestern part of Morocco.

Collected accessions of wild beet species are presented in Table 1.

Species	No. of samples	No. of accessions	
Beta maritima	140	25	
Beta macrocarpa	130	20	
Beta patellaris	90	10	
Total	360	55	

Table 1. Beta species and accessions collected in Morocco in 2005

#### Conclusions

Wild *Beta* species were present in many regions of Morocco. *Beta maritima* and *B. macrocarpa* were widely spread all over collected sites while *B. patellaris* was found only along the southern coasts.

The abundance of species varied notably among different regions. All species were strongly affected by animal grazing since their leaves remain green later than most other fodder species. Urbanization is also rapidly developing at the expense of *in situ* biodiversity.

#### Reference

Jahandiez E, Maire R. 1932. Catalogue des Plantes du Maroc [Catalogue of Moroccan plants]. (Vol. 2). Imprimerie Minerva, Alger, Morocco. (in French).

#### The Beta collection in Poland

#### Kamilla Kuzdowicz

Plant Breeding and Acclimatization Institute, Bydgoszcz, Poland

The *Beta* collection in Poland is located at the Bydgoszcz Research Division of the Plant Breeding and Acclimatization Institute. This collection is a unit of the National Centre for Plant Genetic Resources (NCPGR), based in Radzików, which coordinates, finances and provides storage facilities for crop genetic resources in Poland.

The main aim of the collecting of beet materials is to save the genepool which exists in old multigerm cultivars, because the modern use of hybridization methods based on cytoplasmic male sterile (CMS) lines has led to the narrowing of the genetic background in new cultivars.

Wild species of the genus *Beta* are important as a source of resistance to diseases, pests and abiotic factors. There are no species of *Beta* in the wild flora of Poland. Malesterile ecotypes of subsp. *maritima* are kept and regenerated in *in vitro* cultures. In Bydgoszcz we have also our own perennial wild beet collection of section *Corollinae* (*B. macrorhiza, B. lomatogona, B. trigyna* and *B. corolliflora*) growing in the field. It is used for study of the *Beta* genome and for molecular biology research.

The *Beta* collection in Poland consists of wild species, old cultivars and breeding materials of sugar and fodder beets. At present, it contains 343 accessions: 112 sugar beets, 199 fodder beets and 32 wild forms belonging to sections *Beta*, *Corollinae* and *Procumbentes*. This collection is conserved in the Long-Term Storage Laboratory in Radzików as seed samples kept in glass jars at -15°C and 5-8% moisture content. Some of the new seed samples are stored in Bydgoszcz in medium-term storage (0-4°C) as a working collection. Accessions have mainly been obtained from national breeding institutions and by way of exchange among beet collections and foreign research laboratories. The material received from international expeditions (local populations) is also of great interest. The collection is annually increased by 10–30 accessions.

The evaluation of the collection is conducted according to the "Descriptors for *Beta*" (IPGRI 1996). Evaluation for the main agronomic traits is carried out at the Experimental Station in Konczewice, on 10-m<sup>2</sup> plots using two replications with standard check cultivars, over a 2-year cycle. Evaluation for morphological, cytological and biochemical traits, seed quality and seed germination tests are carried out in Bydgoszcz.

Seed is multiplied when the seed amount available from expeditions and other sources is insufficient or seed germination ability decreases to 40-50%. Multiplication of *Beta* accessions is carried out in field or greenhouse conditions under strict isolation.

Each year about 20-25 accessions are evaluated and statistical data analysed and documented. Some of them are photographed. Parts of the data are sent to the NCPGR in Radzików and then to the International Database for *Beta*.

Utilization of the collected materials is determined by the main trends in breeding and research. For the past ten years some of the accessions have been evaluated for two economically important beet diseases: *Aphanomyces cochlioides* Drechsler and *Cercospora beticola* Sacc. (Szymczak-Nowak et al. 2000, 2003; Dalke et al. 2002; Kuzdowicz and Wasacz 2005).

At present, we are trying to select some accessions tolerant to drought stress. The collected and evaluated germplasm is used in sugar and fodder beet breeding and in several research programmes.

Information and seed samples are distributed freely. A quarantine certificate is necessary to send samples abroad.

#### References

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#### Beta genetic resources: North American activities

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#### Introduction and status of the collection

The U.S. National Plant Germplasm System's (NPGS) *Beta* collection is housed at the U.S. Department of Agriculture (USDA)-Agriculture Research Service (ARS) Western Regional Plant Introduction Station (WRPIS) in Pullman, Washington, USA. This collection has a total of 2521 accessions of both cultivated and wild species of beet. Table 1 is a summary of the current holdings and status of the collection. Over the last four years we have incorporated 141 new accessions. The majority of these accessions are *Beta vulgaris* subsp. *vulgaris* cultivars but we also received 21 accessions of *B. v.* subsp. *maritima*, 20 accessions of wild *Beta* from other institutions, and 20 accessions of wild collected *Beta nana* from the 2005 NPGS-sponsored mission to Greece (see Frese et al., this volume, pp. 45-52). From 2003 to 2006, we distributed a total of 1644 seed packets. Table 2 summarizes our distribution activities.

Taxon	No. of accessions			
	Total	Available	Backed-up	
Beta corolliflora	4	1	3	
<i>Beta</i> hybrid	2	1	1	
Beta lomatogona	29	2	4	
Beta macrocarpa	16	12	13	
Beta macrorhiza	20	2	2	
Beta nana	21	0	0	
Beta patellaris	29	17	12	
Beta patula	3	2	3	
Beta procumbens	15	6	5	
<i>Beta</i> sp.	16	6	5	
Beta trigyna	48	6	5	
Beta vulgaris	21	9	16	
<i>Beta vulgaris</i> subsp. <i>maritima</i>	571	445	391	
<i>Beta vulgaris</i> subsp. <i>vulgaris</i>	1710	1276	1437	
Beta webbiana	8	0	1	
Beta x intermedia	8	1	1	

#### Table 1. Status of the U.S. Beta collection

Year No. of orders		No. of seed packets distributed			
		U.S.	Foreign	Total	
2003	32	371	109	480	
2004	35	340	38	378	
2005	49	357	96	453	
2006	38	311	22	333	

Table 2. Summary of distributions

Currently, 70.8% of the US *Beta* collection is available for distribution and 75.3% of the accessions have duplicate samples (back-up) at the USDA-ARS National Center for Genetic Resources Preservation (NCGRP) in Fort Collins, Colorado. Of the 1710 accessions of *B. v.* subsp. *vulgaris*, 74.6% are available and 84% have back-up samples deposited at NCGRP. The *B. v.* subsp. *maritima* collection is similarly available but with fewer accessions backed-up: 77.9% available, 68.5% backed-up. The majority of the accessions that are unavailable are the more difficult to germinate and regenerate wild *Beta* species.

To address the unavailability of the *Beta* species accessions we are currently focusing our regeneration efforts on this material along with *B. v.* subsp. *maritima*. At present, all our increases are done in the greenhouse. We use all available, suitable spaces in the WRPIS and Washington State University greenhouse systems, a total of 13-19 rooms.

Because there is still a considerable backlog of *Beta vulgaris* accessions we are again addressing field increase of this material. As reported previously (Panella et al. 2002) we have experimented with field increase and had some success in Pullman. Due to personnel changes in 2004 further experimentation and modifications to the field programme were temporarily stopped. We are once again looking at developing a field increase protocol. In autumn 2006, we planted four accessions in the field: three accessions of *B. v.* subsp. *vulgaris* and one accession of *B. v.* subsp. *maritima*. We are looking at cage size, testing two cage sizes to address heat and pest accumulation in the cages in the summer. We also are looking at fall versus spring planting in both *B. v.* subsp. *vulgaris* and *B. v.* subsp. *maritima* to address problems we have had in overwintering beet germplasm.

#### Evaluation

Evaluation of the U.S. *Beta* collection is coordinated by the U.S. Sugarbeet Crop Germplasm Committee (see report in Part I, this volume, pp. 8-9). Since 1994, 20 to 30 accessions per year have been evaluated for resistance to the following diseases: rhizoctonia, cercospora, root maggot, curly top, and rhizomania. In addition to disease resistance evaluations, descriptor data are collected on accessions being regenerated. Data from all evaluations are entered into the Germplasm Resources Information Network (GRIN) and available on the Web at http://www.ars-grin.gov/npgs/index.html. These data are being used by public breeders in the United States to begin the long-term process of introgression of novel resistance genes into agriculturally acceptable sugar beet germplasm (Panella and Lewellen 2007).

#### References

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## **A**PPENDICES

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# Appendix I. Progress report of the ECP/GR Working Group on *Beta* for the period (1999) 2002 to March 2006 and workplan for the future

#### I. RESULTS

Workplan (milestones)	What results have been obtained?	Which aims/goals have not been (fully) reached?	Completeness ratio (%)
Hold the third joint meeting of the WG on <i>Beta</i> and the World <i>Beta</i> Network in Spain.	The meeting was jointly organized with the Instituto Canario de Investigaciones Agrarias, Gobierno de Canarias, Puerto de la Cruz, Spain.	-	100
Hold an ad hoc meeting of the sub-working groups' moderators, to discuss topics related to germplasm evaluation, utilization, molecular markers, data documentation.	An email newsletter was designed by the sub- working group moderator for germplasm evaluation and distributed within the ECP/GR <i>Beta</i> Working Group and WBN. Colleagues were encouraged to publish short communications in the newsletter. The response to the sub-group moderator's initiative was very positive.	During the third meeting at Puerto de la Cruz the need for a sub-working group moderators' meeting was discussed. The plenum suggested using the funds for the development of a descriptor list for <i>in situ</i> management data and related data modelling.	5
Provide the IDBB manager with passport accession data in the EURISCO/MCPDv2 format.	On hold until the end of AEGIS project. It is hoped that the results of the project will strengthen the role of CCDB managers.	-	2
update, send to the curators a Windows application displaying duplicate groups	The CCDB manager received <i>Beta</i> passport data from Ukraine.		
and enabling them to fill in their agreements regarding responsibility, restrictions and resulting storage status for their accessions.	It is planned to exchange data with non-European countries. With respect to European countries the CCDB manager will wait for		
Make the result of the first round of decisions on sharing of responsibilities available online via the IDBB.	the completion of EURISCO and then download data for an update.		

a. Comparison of workplan (r	nilestones) versus results obt	ained (cont.)	
Workplan (milestones)	What results have been obtained?	Which aims/goals have not been (fully) reached?	Completeness ratio (%)
Following the scheme agreed during the meeting, develop	None. On hold. Part of the AEGIS procedure.	-	0
a draft document on quality concept for <i>Beta</i> genetic resources conservation and circulate it.	The development of a quality concept is considered a task of the curators of <i>Beta</i> holdings.		
Send comments and revisions to the proposed document.			
Circulate a revised document for final endorsement.			
Provide the IDBB manager with additional characterization and evaluation data, following the suggested guidelines.	Wild beet photos were added to the database.	-	1
Provide the <i>Beta</i> curators and/or WG members with a base document on seed production procedures.	Done.	-	100
Complete the document in accordance with specific local conditions and return it to the BAZ Gene Bank.			
Compile individual reports and publish them on the ECP/GR WG on <i>Beta</i> Web pages.			
Provide the IDBB with GIS software (Milestone from 1999).	An online mapping tool has been developed on the basis of the open source map server available from the University of Minnesota.	The methodology for recording and processing of geo- referenced data needs to be developed further.	80
Encourage inclusion of wild relatives of <i>Beta</i> , and especially <i>B. macrocarpa</i> Guss. and <i>B. nana</i> Boiss. et Heldr. and possibly species from section <i>Procumbentes</i> , in <i>in situ</i> conservation projects by the respective countries and in the monitoring of populations under potential danger (Milestone from 1999).	A Beta nana exploration was organized and successfully completed in 2005. A survey and risk assessment for <i>B. vulgaris</i> subsp. maritima was implemented in Ireland. The need for specific actions for <i>B. macrocarpa</i> , <i>B. patula</i> , <i>B. procumbens</i> and <i>B. webbiana</i> was discussed during the third ECP/GR - WBN meeting. Concepts for <i>in situ</i>		100
	management for <i>B. macrocarpa, B. patula,</i> and <i>Procumbentes</i> species have been developed and are in the process of being implemented.		

Workplan (milestones)	What results have been obtained?	Which aims/goals have not been (fully) reached?	Completeness ratio (%)
Prepare a funding proposal to hire additional staff in charge of entering GRIN, GENRES and additional evaluation data into the database (Milestone from 1999).	The database expert visited GRIN in the year 2000 and discussed opportunities for such joint work. The visit was partly funded by the German Ministry of Agriculture. The GRIN data model was made available to the IDBB managers. At an appropriate time a proposal aiming at the establishment of the International Database for <i>Avena</i> will be submitted to FAO. Since the <i>Beta</i> and <i>Avena</i> databases are based on the same data model the <i>Beta</i> will profit from such a project equally.	A funding proposal was not prepared. GRIN accession number refers to IDBB numbers. GRIN adopted the taxonomic system suggested by the WBN and used by the IDBB. The whole task is a very complicated matter since the data models for passport data are different. The development of a data warehouse was discussed. The continued support of a data warehouse would however require the permanent engagement of an administrator.	50

#### a. Comparison of workplan (milestones) versus results obtained (cont.)

#### b. Contribution to the four ECP/GR priorities for Phase VII

1. Characterization/evaluation (including modern technologies)

During the 2002 meeting of the *Beta* Working Group participants from Belarus, Bulgaria, Czech Republic, Germany, Hungary, Iran, Lithuania, the Nordic countries, Poland, Romania, Russia, Slovenia, Turkey, Ukraine, United Kingdom, and the USA reported on characterization, evaluation and research activities.

#### 2. Task sharing

On hold until the end of AEGIS.

Management of an international core collection for *Beta* has been discussed by an ad hoc group in the year 2000. The developed concept never completely materialized (see **I.d.** 2), except for GRIN.

#### 3. In situ/on-farm conservation and development

The Working Group Chair attended PGR Forum meetings and provided information on *Beta* to this project group. Reports presented by country representatives (Romania, Ukraine, Bulgaria, Italy, the Nordic Countries, Azerbaijan, Morocco and Spain) during the 2002 and 2006 meetings provided new information or confirmed data on the geographical distribution of the genus *Beta*. The Cape Verdean Islands were reported as the distribution area of the *Patellifolia* genus (syn. *Beta* section *Procumbentes*) in the old literature. This report was confirmed enabling the *Beta* network to close another geographic gap in *ex situ* holdings.

The Working Group Chair joined the above mentioned *B. nana* exploration largely financed by the USDA-ARS, Pullman, Washington. The Greek Gene Bank guided the group. A base line for monitoring of *B. nana* was established.

Sugar beet breeders are contributing to the "on-farm" management of beet genetic resources through buffer populations.

#### b. Contribution to the four ECP/GR priorities for Phase VII (cont.)

#### 4. Documentation and information

See report of the *Avena* Working Group. The elements developed for *Avena* are/can also be applied for *Beta*, since the information systems for both crops are based on the same object and data model. A tool for geographic mapping of characterization and evaluation data was developed that may be of interest to other CCDBs.

#### c. Relevance (regional / international)

Did your work and/or outputs have inter-regional dimension? (if it did, give precise details)

The ECP/GR Working Group on *Beta* emerged from the World *Beta* Network. There has therefore been a continuing cooperation with the USA (Fort Collins, Colorado; Pullman, Washington), the Asian (in particular Iran) and the North African regions since 1989 and this cooperation has been further evidenced by continuing participation of representatives from those regions. The *Beta* working group shares common interests with the working group Genetics and Breeding of the IIRB (International Institute for Beet Research, Brussels). The book "Genetics and Breeding of Sugar Beet" published in 2005 includes a chapter on genetic resources and it can be considered a product of international cooperation.

#### d. Lessons learnt (recommendations)

Which lessons learnt are also relevant for other Working Groups?

1. Working groups are composed of persons with different scientific backgrounds, interests and tasks. Knowledge in the fields of conservation biology, plant breeding and genetics, and information science is developing at high speed. While most of the Working Group members can communicate on croprelated agronomic matters as well as plant breeding and genetics, it is more difficult to generate the same common in-depth knowledge on information science, geo-informatics and conservation biology. Due to the size of the Working Group, the number of participants having this specific knowledge often is below a critical mass. The tasks to be solved by information scientists, geoscientists or conservation biologists are probably similar for many crops. Just because of the need to solve tasks, each Working Group starts developing individual solutions where a general one would be more appropriate and effective. There is therefore a need for more interaction between the crop-specific working groups and the thematic networks on cross-cutting activities such as informatics and *in situ* and on-farm management. This can perhaps be achieved by sending ECP/GR employed consultants to working meetings (on request of the Working Group) that on the one hand mediate knowledge between Working Groups as well as advise groups, and on the other hand collect ideas emerging during crop specific Working Group meetings.

2. The progress of such heterogeneous Working Groups largely depends on the ability of the Chair to keep contact with each individual member, to stimulate the cooperation and new projects. This ability clearly is a function of capabilities and facilities. The work the Chair and the database managers of Working Groups are investing is input in kind by their institutions, which is a weak basis for running a European Plant Genetic Resources Conservation Programme. The ECP/GR provides a framework for cooperation; the member states should also accept their role and tasks by assigning mandates to institutions that play a leading role for a specific crop. The mandate needs to be underpinned by a legal status and an earmarked budget / grant. The negotiation of mandates could be a task for the ECP/GR Steering Committee.

3. Cooperation with partners outside Europe was always considered by the ECPGR Working Group on *Beta* as a basic aim for two reasons. Wild species and especially landraces of leaf and garden beet are distributed in Asian and African countries. Significant evaluation work is implemented in the USA which in turn benefits all, since information and even improved material is freely available. The Global Crop Diversity Trust may be interested in funding a global conservation strategy for *Beta* which will allow partners in Asian and African countries to develop improved cooperation with ECP/GR partners.

4. The group discusses frequently how to acquire additional funds needed to fulfil the workplan. There is a large diversity of funding agencies and programmes. Some guidance by the ECP/GR in fund-raising would be helpful.

#### **II. ANALYSIS**

#### a. Bottlenecks

What were the bottlenecks experienced?	How do you plan to solve the bottlenecks?
1. Too little communication within the Group between the Working Group's meetings.	See <b>I.d. 2.</b> The publication of a newsletter
2. Only little progress in entering characterization and evaluation data.	See I.d. 2.
3. Knowledge in the field of geoscience needs to be improved.	Project application and I.d. 1
4. There should be more formalized contact with the main players in the field of nature conservation and the PGR sector. The potential of agro-environmental measures for <i>in situ</i> conservation projects must be better understood.	Working relation with IUCN exists and will be intensified by interaction with the IUCN/SSC CWR Specialist Group (CWRSG).

#### b. Internal support needed (Secretariat, Steering Committee, other Working Groups, etc.)

Secretariat: Support in the preparation and organization of Working Group meetings including report writing and editing is always very efficient and is really appreciated by the Working Group.

Steering Committee: Flexibility in the use of the NCG budget would allow us to overcome some of the work capacity bottlenecks. The Sugar, Starch and Fibre crops' Network Coordinating Group has launched a test case, the Flax Database proposal.

Other working groups: There is potential for cooperation in the field of *in situ* management concept development and implementation. The respective thematic network could play a coordinating role.

#### c. External resources needed (collaboration, external funding)

For reasons explained under **I.d. 2**., financial support is no longer requested from companies or other bodies to facilitate the participation of experts from Asian, Arabic and North African countries. The international working group for *Beta*, the WBN, has therefore become a smaller one more focused on Europe. The establishment of similar working groups for *Beta* in the Asian, Arabic and North African regions by IPGRI or any other funding mechanism to encourage the invitation of key persons from these regions would be helpful.

The Council Regulation 1467/94 allowed the *Beta* Network to achieve major progress in the field of evaluation. ECP/GR should strive for the establishment of an alternative funding mechanism.

#### III. PLANS

a. Planned activities, last part of Phase VII	b. Expected results
1. Identification of target populations for <i>in situ</i> conservation. Partners in Greece, Spain, Morocco and the Caucasus countries should explore the feasibility of creating genetic reserves as soon as possible.	Geographical location of populations identified.
2. Develop a data model and module for <i>In Situ</i> Management (ISM). Develop and agree on descriptor list for ISM.	12 taxa, each 1 within and 1 outside protected areas.
3. Organize a technical meeting aiming at the development of a descriptor list and a data model.	Meeting with BAZ, Birmingham, IPGRI, IBV
4. Solve some taxonomic problems.	Help to determine " <i>B. patula</i> " observed in Azerbaijan. Complete description of the geographical distribution of <i>B. patellaris</i> .
5. Inclusion of additional C&E data into the IDBB. IDBB manager will not request data. It is the task of the institutions producing data to submit them to the IDBB.	Depending on data received.
6. Establishment of a baseline of genetic diversity data for monitoring.	Some populations analysed by Syngenta as input in kind to the network.
7. Providing USDA-ARS, Pullman with the duplicate samples (Greece, Germany, UK) of F. Dale 1980-1981 <i>Beta nana</i> collecting mission.	Seed exchange organized and populations established for further research.

# Phase VIII - Suggested activities of the ECP/GR Working Group on Beta

- Meeting of the Working Group jointly with the WBN;
- Ad hoc Working Group meetings focusing on specific themes such as the creation of an online descriptor list for *Beta*, meeting of collection curators to decide on/organize responsibility and task sharing, etc;
- Continued characterization and evaluation work;
- Monitoring of genetic reserves once established and the initiation of characterization and evaluation projects for these populations;
- Maintenance of cooperation with the WBN in the field of characterization and evaluation is considered essential as there are limited capacities for evaluation work in Europe. Much of the pre-bred germplasm used in European breeding programmes is provided by the USDA-ARS system.

# Appendix II. Acronyms and abbreviations

AEGIS	A European Genebank Integrated System
BAZ	Federal Centre for Breeding Research on Cultivated Plants, Braunschweig, Germany
BGRC	Braunschweig Genetic Resources Collection, Germany
BNYVV	Beet Necrotic Yellow Vein Virus
CBD	Convention on Biological Diversity
CCDB	Central Crop Database
CGN	Centre for Genetic Resources, Wageningen, The Netherlands
CMS	Cytoplasmic Male Sterility
CWR	Crop Wild Relatives
CWRIS	Crop Wild Relatives Information System (PGR Forum)
ECP/GR	European Cooperative Programme for Crop Genetic Resources Networks ( <i>now European Cooperative Programme for Plant Genetic Resources, ECPGR</i> )
EGRISI	European Genetic Resources In Situ Inventory Project
EU	European Union
EUNIS	European Nature Information System
EURISCO	European Internet Search Catalogue
FAO	Food and Agriculture Organization of the United Nations, Italy
GBIF	Global Biodiversity Information Facility
GIS	Geographic Information System
GMO	Genetically Modified Organism
GRIN	Genetic Resources Information Network, USA
IBPGR	International Board for Plant Genetic Resources, Rome ( <i>now Bioversity International</i> )
IBV	Information and Coordination Centre for Biological Diversity, Bonn, Germany
ICIA	Instituto Canario de Investigaciones Agrarias, Tenerife
IDBB	International Database for <i>Beta</i>
IHAR	Plant Breeding and Acclimatization Institute, Poland
IIRB	International Institute for Beet Research, Brussels, Belgium
IPK	Institut für Pflanzengenetik und Kulturpflanzenforschung (Institute of Plant Genetics and Crop Plant Research), Gatersleben Germany ( <i>now the Leibniz Institute of Plant Genetics and Crop Plant</i> <i>Research</i> )
IRRI	International Rice Research Institute (CGIAR)
ISCI	Istituto Sperimentale per le Colture Industriale, Italy
ISM	In Situ Management

ISSR	Inter simple sequence repeat
IUCN	International Union for Conservation of Nature
NCG	Network Coordinating Group (ECP/GR)
NCPGR	National Centre for Plant Genetic Resources, Radzików, Poland
NGB	Nordic Gene Bank, Alnarp, Sweden
NPGS	National Plant Germplasm System, USA
RAPD	Random Amplified Polymorphic DNA
SBSI	Sugar Beet Seed Institute, Karadj, Iran
USDA-ARS	United States Department of Agriculture-Agricultural Research Service, USA
VIR	N.I. Vavilov Research Institute of Plant Industry, Russian Federation
WBN	World Beta Network
WRPIS	Western Regional Plant Introduction Station (of USDA-ARS)

### Appendix III. Agenda

#### Third Meeting of the ECP/GR Working Group on Beta and the World Beta Network 8-11 March 2006, Tenerife, Spain

Venue: Jardin de Aclimatacion de la Orotava, Puerto de la Cruz, Tenerife

#### Tuesday 7 March

Arrival of participants

# Wednesday 8 March - Joint scientific meeting of the World *Beta* Network representatives and the ECP/GR Working Group on *Beta*

#### Introduction

08:45 - 09:00	Opening of the meeting Welcome by the local host, L. Maggioni and L. Frese
Section I. Scientif	ic and technical aspects of <i>in situ</i> management
09:00 - 09:30	• The PGR Forum project: some conclusions and recommendations ( <i>B.V. Ford-Lloyd</i> )
09:30 - 10:00	• Reconstructing the evolutionary history of the <i>Beta</i> section with molecular data. A focus on the Canary Islands ( <i>S. Villain</i> )
10:00 - 10:30	• Protection and distribution of <i>Beta</i> (s.l.) species in the Canary Islands: perspectives of conservation ( <i>A. Santos Guerra and J.A. Reyes-Betancort</i> )
10:30 - 11:00	Coffee break
11:00 – 11:20	• Biodiversity of <i>Beta</i> species in the Transcaucasus region (Armenia, Azerbaijan, Georgia, Iran) ( <i>G. Aleksidze et al.</i> )
11:20 – 11:40	• Survey of <i>Beta nana</i> in Greece ( <i>R. Hannan et al., presented by L. Panella</i> )
11:40 – 12:00	• Survey of <i>Beta vulgaris</i> subsp. <i>maritima</i> populations in Ireland ( <i>D. Grogan</i> )
12:00 - 14:00	Lunch
14:00 – 14:20	• GIS application for the <i>in situ</i> conservation and management programme in Turkey ( <i>A. Tan</i> ) – <i>unable to attend</i>
14:20 – 14:40	• The International Database for <i>Beta</i> and <i>in situ</i> management: potential role and functions ( <i>C. Germeier and L. Frese</i> )
14:40 – 15:00	• Screening techniques for root growth parameters under dry and compacted conditions in sugar beet germplasm ( <i>E. Ober</i> )
15:00 - 15:30	Coffee break
15:30 - 15:50	• Recent progress in <i>Beta</i> germplasm evaluation in the USA ( <i>L. Panella</i> )

15:50 - 16:10	• Evaluation of beet germplasm and progress towards the development of sugar beet for diseases resistance and root structure ( <i>M. Mesbah et al., presented by M.N. Arjmand</i> )
16:10 - 16:30	• <i>Beta</i> genetic resources activities in India (1990-2005) - a review ( <i>H.M. Srivastava and S. Srivastava</i> ) - <i>unable to attend</i>
16:30 – 17:00	Break for individual discussions
Section II. Techni	cal meeting, review of the workplan
17:00 – 17:30	<ul> <li>General briefing on ECP/GR (<i>L. Maggioni</i>, 15 min)</li> <li>Report of the Working Group Chair (<i>L. Frese</i>, 15 min)</li> </ul>
17:30 - 18:00	• A European Genebank Integrated System (AEGIS) (L. Maggioni)
Thursday 9 Marc	h - Meeting of the ECP/GR Working Group on Beta
08:30 – 10:00	<ul> <li>Review of the workplan</li> <li>Critical assessment of the achievements and discussion of the next workplan by ECP/GR countries (attending members: Armenia, Georgia, Germany, Ireland, Italy, Poland, Turkey, UK)</li> <li>Statements and comments from other WBN member countries (France, Iran, India, Morocco, Spain, USA)</li> </ul>
10:00 - 10:30	Coffee break
10:30 - 12.30	Establishment of ad hoc working groups, if desired Recommendations and conclusions
12:30 - 14:00	Lunch
14:00 - 17:00	Visit to the Institute Introduction into the <i>in situ</i> management methodology with emphasis on threat assessment
Friday 10 March	
09:00 - 12:00	Drafting of the report by task force Ad hoc working group meetings to discuss project proposals to be submitted under EU Council Regulation 870/2004
13:00 - 14:30	Lunch
14:30 - 16:00	Plenary meeting and approval of the report
16:00 - 16:30	Coffee break
16:30 – 17:00	<ul><li>Election of new Chair and Vice-Chair for the Working Group</li><li>Closing remarks</li></ul>
20:00	Social dinner

#### Saturday 11 March

Visit to known *Beta* collection sites. Training on threat assessment and population monitoring in the natural habitat

#### Sunday 12 March

Departure of participants

## Appendix IV. List of participants

#### Third Meeting of the ECP/GR Working Group on Beta and the World Beta Network 8-11 March 2006, Tenerife, Spain

N.B. Contact details of participants updated at time of publication. However, the composition of the Working Group is subject to changes. The full list, constantly updated, is available on the Beta Working Group's Web page (http://www2.bioversityinternational.org/networks/ ecpgr/Contacts/ecpgr\_wgbe.asp).

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ISBN: 978-92-9043-815-1