European Cooperative Programme for Crop Genetic Resources Networks

ECP/GR

Report of a Working Group on Forages

IPGRI

Eighth Meeting, 10–12 April 2003, Linz, Austria

B. Boller, E. Willner, L. Maggioni and E. Lipman, *compilers*

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The International Plant Genetic Resources Institute (IPGRI) 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 Future Harvest 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. IPGRI has its headquarters in Maccarese, near Rome, Italy, with offices in more than 20 other countries worldwide. The Institute operates through four programmes: Diversity for Livelihoods, Understanding and Managing Biodiversity, Global Partnerships, and Improving Livelihoods in Commodity-based Systems.

The international status of IPGRI is conferred under an Establishment Agreement which, by January 2005, had been signed by the Governments of Algeria, Australia, Belgium, Benin, Bolivia, Brazil, Burkina Faso, Cameroon, Chile, China, Congo, Costa Rica, Côte d'Ivoire, Cyprus, Czech Republic, Denmark, Ecuador, Egypt, Greece, Guinea, Hungary, India, Indonesia, Iran, Israel, Italy, Jordan, Kenya, Malaysia, Mauritania, Morocco, Norway, Pakistan, Panama, Peru, Poland, Portugal, Romania, Russia, Senegal, Slovakia, Sudan, Switzerland, Syria, Tunisia, Turkey, Uganda and Ukraine.

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The European Cooperative Programme for Crop Genetic Resources Networks (ECP/GR) is a collaborative programme among most European countries aimed at facilitating the long-term conservation and the increased utilization of plant genetic resources in Europe. The Programme, which is entirely financed by the member countries and is coordinated by IPGRI, is overseen by a Steering Committee composed of National Coordinators nominated by the participating countries and a number of relevant international bodies. The Programme operates through nine networks in which activities are carried out through a number of permanent working groups or through *ad hoc* actions. The ECP/GR networks deal with either groups of crops (cereals; forages; fruit; oil and protein crops; sugar, starch and fibre crops; vegetables, medicinal and aromatic plants) or general themes related to plant genetic resources (documentation and information; *in situ* and on-farm conservation; inter-regional cooperation). Members of the working groups and other scientists from participating countries carry out an agreed workplan with their own resources as inputs in kind to the Programme.

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CONTENTS

PART I. DISCUSSION AND RECOMMENDATIONS

Introduction

Opening of the meeting

On behalf of the Austrian Agency of Health and Foodsafety, Paul Freudenthaler welcomed all the participants and wished everybody a good stay in Austria and a successful meeting.

Welcome on behalf of the Austrian Minister of Agriculture

Mrs Hedwig Wögerbauer, National Coordinator, addressed the Group on behalf of the Austrian Minister of Agriculture:

Dear Participants of the Eighth Meeting of the Working Group on Forages,
Dear Guests,
Ladies and Gentlemen,

On behalf of the Federal Minister of Agriculture and Forestry, Environment and Water Management, I have the pleasure to welcome you to the city of Linz, situated in the heart of Upper Austria. Due to other very important and urgent commitments it was impossible for the Minister to be with you today in person, but he sends his warmest greetings to all participants and wishes great success for the forthcoming meeting.

Since the foundation of the European Cooperative Programme for Crop Genetic Resources Networks (ECP/GR) in 1980, Austria has been an active participant. The programme is based on the recommendations of the United Nations Development Programme (UNDP), on the Food and Agriculture Organization of the United Nations (FAO) and on the Gene Bank Committee of the European Association for Research on Plant Breeding (EUCARPIA), and is collaborative among most European countries, including Israel. It aims to ensure the long-term conservation of plant genetic resources in Europe and to facilitate their increased utilization. Generally the programme is based on the participating countries' contributions in kind, and in Austria it is financed by the budget of the Federal Ministry of Agriculture and Forestry, Environment and Water Management, for which my division is responsible.

There has been a close connection between long-term conservation and the increased utilization of plant genetic resources, which led to the foundation of the European Central Crop Databases. Data are stored there to facilitate use of genetic material for scientific and practical purposes. Austria is very proud to host the European Databases for *Phaseolus* and *Vigna*, situated in Linz at the Agency of Health and Foodsafety. These activities also play an active role within the World Information and Early Warning System (WIEWS) at FAO, a system that should help to prevent the loss of endangered cultivated plant species and to warn of their possible extinction.

I have studied very carefully the provisional agenda of the eighth meeting of the Working Group. It seems to me that one of the main items of this meeting is the sharing of responsibilities between all the members of the ECP/GR. Due to the restriction of financial means the sharing of responsibilities for the conservation of genetic resources becomes more and more important for the Federal Ministry of Agriculture and Forestry, Environment and Water Management. Strengthened cooperation and mutual commitment in determined fields, e.g. evaluation and renewal of material and storage, should help to reduce costs. Therefore minimum standards for regeneration, storage and conservation of the material should be elaborated under the auspices of IPGRI and, in order to manage the variability of materials, so-called core collections should be developed.

There is another very important point on the agenda for Austria concerning on-farm and in situ conservation, and this aspect is thoroughly explained in the paper presented today by Kaspar Holaus, the Austrian member in the Working Group on Forages. The reference to this short overview of the situation in Austria presented by K. Holaus brings me to the end of my introductory statement. I do believe the success of this meeting will help to contribute to better conservation of forage crop genetic resources in the future.

H. Wögerbauer was also happy to say that, although she was a lawyer, with this meeting she was returning to the agricultural roots of her family.

General briefing on ECP/GR

Lorenzo Maggioni, ECP/GR Coordinator, welcomed the participants to the eighth meeting of the Working Group on Forages and briefly summarized the objectives and structure of the programme, with two new member countries joining since the last meeting in 1999 (Albania and Armenia).

He then mentioned that during Phase VI of the programme (1999-2003) activities within the Forages Network consisted of two meetings of the Working Group on Forages (Elvas, Portugal, in 1999 and the current meeting in Austria). He briefly outlined the process leading to the selection by correspondence of the new Chair (Beat Boller) and Vice-Chair (Evelin Willner) after Petter Marum had resigned from the chairmanship in 2002.

He then said that the Web page recently prepared by the Secretariat for the Working Group was planned to become, in the near future, a more active vehicle for distribution of up-to-date information on the Group's activity. Comments and contributions to improve the use and effectiveness of this tool would be welcome.

Regarding the outcomes of the mid-term meeting of the ECP/GR Steering Committee held in St. Petersburg, Russian Federation, on 14-17 October 2001, a few relevant highlights for the Group were summarized. In particular, regarding sharing of responsibilities in Europe for the conservation of genetic resources, the Steering Committee recommended that the possible practical options be analyzed in more detail and that the definition of genebank quality standards should receive careful attention.

In order to develop a strategy for the next Phase (VII), a task force composed of a few Steering Committee members produced a draft document containing a set of recommendations. This document will also be circulated to all the Working Group Chairs for comments in advance of the End of Phase Steering Committee meeting to be held in Turkey in October 2003. According to this draft, the programme should increasingly focus its action on five specific areas: 1) documentation; 2) molecular markers and genomics; 3) task-sharing; 4) characterization and evaluation; 5) *in situ* and on-farm conservation. The Working Groups would remain the operational units, but only those with more urgent and clearly identified priorities and measurable targets would receive approval for funding of meetings and other actions.

Report of the Working Group Chair

Petter Marum

The last meeting of the Working Group on Forages was held in Elvas, Portugal, 18-20 November 1999. A report from the meeting of 227 pages was published by IPGRI in 2000.[1]

[1] Maggioni, L., P. Marum, N.R. Sackville Hamilton, M. Hulden and E. Lipman, compilers. 2000. Report of a Working Group on Forages, Seventh meeting, 18-20 November 1999, Elvas, Portugal. International Plant Genetic Resources Institute, Rome, Italy.

European Forage Databases

Since our last meeting several new databases have been made available on the Internet. They include the *Agropyron*, annual *Medicago*, *Arrhenatherum*, *Trisetum*, *Trifolium subterraneum* and the Minor Forage Legumes. Several databases have been updated.

In Elvas an alternative method of organizing and updating the forage databases was discussed. It was agreed to test an alternative method for the compilation process of the new Minor Forage Crops Database (European Minor Forage Grasses Database and Minor Forage Legumes Database).

In April 2000 the EPGRIS (European Plant Genetic Resources Information Infra-Structure) project was approved. As it is expected that the EPGRIS project, once it is operational, will efficiently advance the collection of all the passport data, the development of the databases was generally put on hold.

Sharing of responsibility

During our last two meetings the WG discussed and proposed a mechanism to establish a European decentralized collection with formal responsibilities. In Elvas precise instructions on how forage database managers could identify Most Original Samples (MOSs) were discussed. Several database managers performed this task of identifying MOSs in the Central European Forage Databases, but others encountered some problems. The greatest problem was that our databases were not good enough. Essential data were missing. In some cases, e.g. for the Nordic regional genebank with regional responsibility, the algorithm would not work. In that case no MOSs would be found.

I believe that the curators of the national collections in cooperation with breeders and people who established the collections are the best prepared to assign "Originality" to the accessions. I believe in a little personal touch to this task and not only to rely on the computer. In preparing for this meeting many of us have completed this task and hopefully this will give us momentum to complete the mechanism of "sharing of responsibility" and the creation of a decentralized European Forage Collection with formal responsibilities.

ICONFORS

In Elvas R. Sackville Hamilton reported on an EU project proposal with the title: "*Improving germplasm conservation methods for perennial European forage species*". The project proposal was approved and the project started on 1 January 2001. Members from the United Kingdom, Czech Republic, Denmark, the Netherlands, Portugal and Norway participate in the project. The main objective is to optimize our regeneration protocols for perennial forage species. The project is now halfway through. Later in the meeting we will learn more about this project.

Operational structure and effectiveness of Working Group activity

In the report from the Working Group to the Steering Committee in September 2001, the WG expressed the opinion that the new operational structure, with a Network Coordinating Group and a Working Group, was not workable in our case. For a working group to function it has to meet more often. Commitment to operate WG activities is usually offered by WG members during the meeting and in the absence of these it will become increasingly difficult to agree on group workplans. During a 4 to 5-year time span, many WG members are likely to change. In the absence of meetings, the new members will find it difficult to be integrated in the Group. The continuity of the programme is bound to suffer. Experience also shows that WG activities are more intense around the meeting's event, which is essential to give momentum to the action of the Working Group. I also strongly believe that face-to-face discussions are important.

Aims of the meeting and welcome address from the new Chair

Beat Boller welcomed the participants from 28 countries and mentioned particularly three new countries: Croatia, Estonia, and Macedonia (FYR). Seven other countries were represented by new participants in the Group: Austria, Bulgaria, Greece, Israel, Portugal, Switzerland and Turkey. He thanked the organizers and complimented them for the nice facilities and the good organization so far.

The first objective of the meeting is to bring people together and to exchange information. In this respect, he stressed the importance of a common terminology and of an agreement about which descriptors of the accessions are considered the most important. Such agreements are an indispensable prerequisite for central databases to become a basis for further progress. At this meeting in particular, we hope to utilize these tools to move forward in the sharing of responsibilities.

While this is true for any crop, forages have some peculiarities that make work with their genetic resources especially challenging:

Forage crops are a comparatively new phenomenon. It is only during the last few centuries that they have been grown from seed, compared to millennia for cereals. Perhaps because of this, it is often a rather short step from genetic resources such as ecotypes to an advanced new cultivar. One or two generations of recurrent selection may be sufficient for a practical plant breeder. Moreover, Europe is the centre of origin for many important forage species. This facilitates collections of genetic resources but also means that we must assume the responsibility to maintain them for future generations.

While travelling to Austria, B. Boller realized how little importance political borders have for forage plant genetic resources. Natural conditions and cultivation methods were very similar across borders. He concluded that perhaps too much emphasis is given to national collections and too little to regional approaches. The example of the Nordic countries shows that such regional approaches can be effective and helpful to promote cooperation.

European Central Forage Databases

Progress in development of a European Internet Search Catalogue (EURISCO): implications for Forage Databases

L. Maggioni gave a brief account of the progress of the EU-funded project EPGRIS (http://www.ecpgr.cgiar.org/epgris/index.htm) for the establishment of a plant genetic resources infrastructure. The objective is to establish a European Internet Search Catalogue (EURISCO) with passport information of plant genetic resources maintained *ex situ* in Europe. Before the end of 2003, the first version of EURISCO is expected to be launched on-line and to contain a combination of data available from the existing national inventories and from the existing Central Crop Databases (CCDBs). The new logo of the catalogue was shown, and a brief demonstration of the search options of the catalogue was given, based on the EURISCO demo site currently under development.

EURISCO is expected to gradually develop and become the most complete and reliable source of passport data in Europe. The catalogue will carry an important minimum set of passport data, frequently and automatically updated from the national inventories. These data will be based on the revised version of the FAO/IPGRI Multicrop Passport Descriptor List (MCPDv2) finalized in December 2001 (available on the Internet at http://www.ipgri.cgiar.org/publications/pubfile.asp?ID_PUB=124).

National focal points, already designated in all European countries, will be responsible for data sources, data quality and accuracy, data availability and provision of data in the EURISCO/MCPD format. The central node receiving the data at IPGRI will be responsible for checking data compatibility with the catalogue, providing feedback to national partners, importing data into EURISCO and developing and maintaining the front end.

The launching of the first version of EURISCO is expected to take place at the occasion of the final meeting of the EPGRIS project, which is planned for September 2003 in Prague, Czech Republic, jointly with a meeting of the ECP/GR Documentation and Information Network. On this occasion, all European National Inventory focal persons and Central Crop Database managers will have the chance to discuss the future relationship between EURISCO and the CCDBs.[2] A document distributed in April 2002 to the ECCDB managers by the EPGRIS project suggested a way forward in this relationship, i.e.:

1. CCDBs to harmonize their structure with EURISCO (MCPDv2 + 6 EURISCO specific descriptors). CGN is already undertaking this harmonization.
2. CCDBs to continue gathering data until EURISCO becomes the preferred source of passport data.
3. Once EURISCO becomes operational, consider retrieving data from EURISCO.

Three possible scenarios are also expected to exist at any point in time, depending on the specific crop, i.e. that (1) EURISCO may contain less data than the CCDB; (2) EURISCO may contain more data than CCDB; and (3) EURISCO may contain different data than CCDB. The transition phase is considered likely to last 2-3 years before a good harmonization between EURISCO and the CCDBs. The role of the CCDBs and their managers will also be a point in the agenda of the EPGRIS meeting in Prague. It is foreseen that this role will increasingly focus on helping improve data quality, tracing duplicates, gaps, MOSs, gathering characterization/evaluation data, analyzing information (GIS, etc.), providing the users with data in various formats, helping define core collections, safety-duplication and collecting needs, etc.

Major advances in the development of individual forage crop databases

The following presentations were given *(see Part II, pp. 20-28):*

European *Trifolium subterraneum* and annual *Medicago* species Databases
Mónica Murillo

European *Arrhenatherum* and *Trisetum* Databases
Magdalena Ševčíková

European *Dactylis* and *Festuca* Databases
Włodzimierz Majtkovski

Discussion

Regarding the use of the algorithm for the identification of Most Original Samples (Appendix I of the report of the seventh WG meeting), it was acknowledged that its use needs to be complemented by the direct judgement of the curator/database manager, in order to reach meaningful decisions.

M. Ševčíková commented that the EURISCO catalogue and the CCDBs should be merged, in order to avoid unnecessary duplication of efforts.

W. Majtkowski pointed out the taxonomy problems faced by the database manager, Grzegorz Żurek, since the *Dactylis* and *Festuca* databases contain names which do not appear in the literature. The need for standardization of the taxonomy in the central databases was also expressed by other DB managers in the Group. It was accepted that the main reference is

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[2] As of September 2003, EURISCO is available on-line at http://eurisco.ecpgr.org

Flora Europaea, although the European databases often contain also non-European species for which it is worth consulting the USDA Germplasm Resources Information Network (GRIN).

L. Maggioni informed the Group that the issue of taxonomy in the central databases would be an item for discussion in the upcoming meeting of the ECP/GR Documentation and Information Network (Prague, Czech Republic, September 2003).

L. Horváth and L. van Soest pointed out that Hungarian and Dutch data for *Dactylis* were missing from the central database and they provided the data during the meeting. I. Thomas also pointed out that UK data were missing and were available from the IGER (Institute of Grassland and Environmental Research, Aberystwyth, UK) Web site.

A question was raised whether the European *Vicia* spp. Database was still managed by IDG (Istituto del Germoplasma, Bari, Italy), since it had recently not been possible to access data from or deliver data to the database. The latest news available to the Group on the database progress dates back to 1996. L. Maggioni informed the Group that the Italian National Coordinator wished to verify, in the coming weeks, whether it was possible for IDG to continue and strengthen this commitment, after recent changes had taken place at the institute. M. Ševčíková informed the Group that M. Hýbl (AGRITEC, Research, Breeding and Services, Ltd., Šumperk), was ready to offer maintenance of the European *Vicia* database on behalf of ECP/GR. This proposal was welcomed by the Group, in case IDG would not be able to ensure rapid improvement of the database and its accessibility from the Internet. It was also suggested that this database could be merged with the "European other Vicieae Database" currently maintained by the University of Reading, UK, which was not making any progress either in the view of the Group.[3]

National collections

A brief account of the national collections was presented by the representatives of Albania (K. Tahiraj), Austria (K. Holaus), Bulgaria (Y. Guteva), Croatia (S. Bolarić), Czech Republic (M. Ševčíková), Estonia (R. Aavola), France (B. Montegano), Germany (E. Willner), Greece (C. Iliadis), Israel (N. Yonash), Lithuania (N. Lemežienė), Macedonia FYR (Z. Dimov), the Netherlands (L. van Soest), Nordic countries (L. Bondo), Poland (W. Majtkovski), Portugal (J.P. Carneiro), Romania (T. Marusca), Slovakia (J. Drobná), Slovenia (V. Meglič) and Turkey (H. Özpinar) *(see Part II, pp. 29-100 – this section includes the contributions received later on from representatives of Hungary (L. Horváth) and Latvia (B. Jansone)).*

Discussion

The Group was pleased to see substantial improvements in the development of the forage crop collection and facilities in Albania and Bulgaria, and that the difficulties of the past seemed to have been overcome.

R. Aavola specified that *M. sativa* does not grow naturally in Estonia (except naturalized hybrids with *Medicago falcata*).

L. van Soest asked about the type of arrangements for transfer of material from VIR (N.I. Vavilov Research Institute of Plant Industry, St. Petersburg) to Estonia and it was clarified

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[3] In June 2003, the Italian National Coordinator confirmed to the ECP/GR Secretariat that the Italian Plant Genetic Institute (former Germplasm Institute) was still interested to continue managing the European *Vicia* spp. database and the responsible manager, Giambattista Polignano, had been designated and is still based in Bari.
In September 2003, University of Reading was contacted by the ECP/GR Secretariat with a proposal to consider a temporary transfer of the management of the "European other Vicieae Database" to a different institute.

that the Estonian accessions conserved at VIR are preferentially repatriated for multiplication in Estonia, since regeneration capacity is limited at VIR.

The innovative approach made by the French networks of creating new variability through pooling of accessions for natural pollination was appreciated by the Group.

The Group was favourably impressed by the large number of samples regenerated each year in Germany.

L. van Soest mentioned the difficulties of cooperation with breeding companies for regeneration, since they need isolation space for their own regeneration and cannot devote their resources to genebank material. E. Willner explained that, in Germany, breeding companies were happy to collaborate, especially those belonging to the association GFP (Gemeinschaft zur Förderung der privaten deutschen Pflanzenzüchtung e.V.). However, the regeneration protocols suggested by the Working Group in recent meetings were considered too demanding for the private companies to comply with. Therefore, the German genebanks primarily collaborate with private companies in evaluation projects.

E. Willner also asked the Group for help, wherever possible, for the regeneration of legumes (red clover and alfalfa).

L. Bondo confirmed the trend shown by E. Willner, indicating that requests for seeds from genebanks had declined in the past few years. She also said that the requests to the Nordic Gene Bank (NGB) came from all the continents, i.e. also from southern latitudes with high altitudes.

J. Drobná specified that various species of *Bombus* were used in Slovakia for pollination of accessions under regeneration, and that some species were more effective than others. They were provided by the Czech University, where bumble bees are bred.

L. van Soest asked what had been the aim behind collecting missions in Slovenia. V. Meglič replied that samples were collected of interesting species for breeding. Moreover, collecting in the Alpine region was aimed at finding cold resistance and persistence, with collecting sites visited at a distance of at least 5-6 km one from another.

International cooperation for collecting missions

(see Part II, pp. 101-111)

Evelin Willner presented the results of two international collecting missions in Ireland and Bulgaria, with collaboration between Germany and the target countries.

L. Bondo presented the results of a VIR/NGB international collecting mission in Karelia.

I. Thomas presented the results of an international collecting expedition in the Pyrenees mountains.

Discussion

E. Willner confirmed that all the information collected in these missions, including collecting data and characterization/evaluation data, is planned to become available on the Internet in the next three years. I. Thomas asked whether any reference had been made to a previous collecting mission in Ireland and suggested comparing the results of that mission with those of the current one. L. van Soest also confirmed that a previous expedition to Ireland involved CGN (Centre for Genetic Resources, Wageningen, the Netherlands), IGER (Institute of Grassland and Environmental Research, Aberystwyth, UK) and others but he was unsure whether any samples were still conserved anywhere, since they were not present at CGN. I. Thomas thought that they were conserved at Aberystwyth.

Workplan

E. Willner will check with the Welsh and Irish genebanks on the availability of seed from previous collecting missions and will plan to include such samples in the future characterization or evaluation trials.

Implications of the International Treaty on Plant Genetic Resources for Food and Agriculture for international collecting missions and exchange of material

L. Maggioni gave a brief account of the main international agreements that are currently regulating access to genetic resources and the sharing of the benefits derived by their use. Article 15 of the Convention on Biodiversity (CBD) (1993) recognizes the sovereign rights of States over their natural resources and that the authority to determine access to genetic resources rests with national governments and is subject to national legislation. This framework sets the scene for the need to establish mutual agreements between countries in order to guarantee facilitated access and equitable sharing of the benefits. The Bonn guidelines on access to genetic resources, adopted in April 2002 by the Conference of the Parties to the CBD, are voluntary guidelines prepared to assist governments in the structuring of legislation to ensure fair access to genetic resources and sharing of benefits. These recommend legal certainty and clarity and suggest the definition of national focal points to inform applicants on procedures for acquiring prior informed consent and mutually agreed terms. Examples of monetary and non-monetary benefits that could be derived from the exchange of material are also listed in the guidelines.

Recently an International Treaty to facilitate access to genetic resources for food and agriculture and to ensure the sharing of the benefits deriving from their use has been adopted.

This Treaty, which is in harmony with the Convention on Biodiversity, was endorsed in November 2001 in Rome after 7 years of negotiations, with 116 countries voting in favour and two abstaining (Japan and the USA). The International Treaty goes far beyond the Bonn guidelines, since it is a legally binding document. The Treaty establishes a Multilateral System (MLS) for facilitated exchange, which is an alternative approach to the system of bilateral agreements and will enter into force 90 days after ratification by the 40th country (15 countries have ratified the document so far)[4]. The Multilateral System will guarantee facilitated access to material that is "under the management and control of the Contracting Parties and in the public domain" and is restricted to a list (Annex I) including 35 genera of food crops and 80 forage species. The list had to be defined by consensus and the veto raised by individual countries excluded from the list a few important crops, such as soybean and tomato. Material Transfer Agreements will have to be signed whenever germplasm is exchanged. Some difficulties can already be envisaged for the near future, since the definition and implementation of a text for Material Transfer Agreement still needs to be negotiated and additions to the list of crops have to be made by consensus. Moreover, the legal interpretation of parts of the document will not always be straightforward.

The adoption of the "International Treaty on Plant Genetic Resources for Food and Agriculture (PGRFA)" is seen as a very positive development, although several questions remain unanswered. In order to address issues raised by this and other international agreements, an ECP/GR Task Force established by the Steering Committee will discuss in the near future how genebank operations will be affected and the best way to implement international obligations.

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[4] Having reached the required number of instruments in order for the International Treaty to enter into force (40), the date of entry into force was 29 June 2004.

The question was raised whether the Treaty also applies to PGRFA obtained before 1994 and it was confirmed that this is the case for the crops listed in Annex I of the International Treaty.

R. Schachl commented that, however useful and necessary the International Treaty is in principle, it also suffers from being limited to those species listed in Annex I, which do not include several crop species essential for food security, as well as wild relatives of important crop species. Thus it would be advisable to think about a European solution ensuring free exchange of all genetic resources for food and agriculture within the European region.

H. Wögerbauer informed the Group that the representatives of 10 new countries joining the European Union in 2004 ("access countries") would participate in Brussels in the Council Working Group starting in May 2003 and she thought that they should raise the issue of the International Treaty and try to influence the Council Working Group on Genetic Resources in order to shed more light on specific unclear areas of the Treaty and define how the Community can interpret them.

Sharing of responsibilities

Definition and identification of Most Original Samples (MOSs)

During the 1999 meeting at Elvas, the Group decided on a mechanism to define responsibility for the maintenance of the Most Original Samples (MOSs) in the forage collection. An algorithm was to be applied by the central crop database managers to provisionally classify the samples, and this classification should have been checked by the collection curators. However, little progress was made in the proposed mechanism, mainly due to difficulties in applying the algorithm in situations where important information was missing in the databases.

To speed up the process, collection curators were asked prior to the meeting to assign an "originality" descriptor to the accessions in their collection, either by using the algorithm or by going through their collections manually.

L. van Soest and P. Marum confirmed that the algorithm may yield odd results in some cases that need correction. For example, in the case of bred varieties the algorithm would almost never result in the identification of a MOS. It was agreed that when breeders donate samples of their own varieties to their national genebank with the aim of ensuring their long-term conservation, these samples should be assigned a "1" (i.e. MOS).

B. Boller showed the example of how MOSs were classified. A number of samples with no donor would be considered "original", but this information was checked against original data of the collecting missions, making it possible to confirm the originality of the samples. The samples put in category 3 by the algorithm needed to be considered on a case-by-case basis. Detailed knowledge of these samples enabled the curators to decide whether to confirm the category or not.

P. Marum confirmed that in the case of the Nordic countries, the algorithm would not work, since the Nordic Gene Bank only coordinates collecting missions and most of the samples were collected and donated by other Nordic institutions that do not conserve the samples themselves. Therefore most samples were assigned to category 1 even though the donor field was not empty.

E. Willner realized that samples collected in collaboration with the Czech Republic were assigned to category 1 by both Germany and Czech Republic and that a consultation is necessary to decide which genebank should be given the primary holding responsibility.

Recommendation

The Group agreed that in all collaborative collecting missions, a consultation among the collecting partners should help in defining which genebank would be assigned the primary responsibility for any given accession.

W. Majtkowski told the Group that very many MOSs are held by Polish genebanks and a second step of concentration would be necessary to ensure regeneration and continuing maintenance, possibly by establishing a national core collection. Similarly, I. Thomas said that the number of MOSs at IGER that are in need of regeneration is too large to be regenerated with their ordinary budgets, but additional funding had been secured from DEFRA (Department for Environment, Food and Rural Affairs, UK) to regenerate some priority accessions and to create safety-duplicates.

In spite of these difficulties, the Group agreed that the identification of MOSs was a first and important step in obtaining benefits from sharing of responsibilities among genebanks. It must result in the assumption of the responsibility of maintaining an accession as its primary holder. The latter requires the involvement of CCDB managers to identify cases where more than one institute holds an accession with the highest level of originality.

The Group aims at defining the primary holder of the accessions of the most important species by the end of 2003 and decides on the following:

Workplan

1. *Collection curators/WG members to add a descriptor "Originality"* (ORIGINALITY) *to the data they prepare for transmission to the CCDBs and assign it values of 1 to 5 for: 1 = "MOS"; 2 = "with MOS"; 3 = "one away"; 4 = "more away"; 5 = "unknown" according to Appendix I of the report of the seventh WG Meeting.*
2. *Collection curators/WG members to send information about originality to CCDB managers **by 1 July 2003.***
3. *CCDB managers to analyze originality information and make a proposal for "Holder of primary collection" (*PRIMCOLL*) for each accession, i.e. holder of the sample with the highest level of originality, indicating specifically possible items that need clarification.*
4. *CCDB managers to send compiled data back to curators/WG members **by 1 October 2003**.*
5. *Curators/WG members to check compiled data, seek solutions for items that need clarification and approve proposal of primary collection holder **by 31 December 2003**.*

*The Group aims at completing the action above **by end of 2003** for the following genera:*

Dactylis	Lolium	Phleum	Trifolium
Festuca	Medicago	Poa	

Recommendation

The Group is aware that in many situations genebanks will need to rationalize their collections that might have reached unsustainable dimensions.

As a further step to help genebanks to maintain valuable collections, the Group recommends that the support of international initiatives (such as the Global Crop Diversity Trust[5]) be ensured.

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[5] The Global Crop Diversity Trust is building an endowment to provide a permanent source of funding for the *ex situ* conservation of the world's most important plant diversity collections. For more information, see http://www.startwithaseed.org.

Safety-duplication

The table on safety-duplication capacities (Appendix V of the sixth meeting's report) was reviewed and its updated version is given below (Table 1).

Table 1. Survey on safety-duplication capacities

Country	Institute / storage conditions / comments
Belgium	DvP, -10°C and -20°C
Bulgaria	Space is available to host safety-duplicates but the buildings need technical reconstruction
Croatia	Capacity to host safety-duplicates at -18°C; modalities to be determined
Czech Republic	-18°C, limited capacity
Estonia	-20°C, limited capacity
France	GEVES, -18°C, limited capacity
Germany	IPK, Gatersleben -15°C / IPKM, Malchow -20°C
Greece	no space available at present, but possibly after new storage facilities are built, according to a new national programme pending final approval
Israel	Israeli Gene Bank, Volcani Centre will have space at -20°C within a year
Lithuania	-18°C, limited capacity
Poland	IHAR, -18°C
Portugal	BPGV, -20°C (contact Ana Maria Barata at bpgv@draedm.min-agricultura.pt)
Slovakia	only -18°C, in limited capacity
Spain	-18°C at Centro de Recursos Fitogenéticos, Madrid (contact Celia de la Cuadra at cuadra@inia.es)
Switzerland	RAC, Changins –20°C, limited capacity (contact Geert Kleijer at geert.kleijer@rac.admin.ch)
Turkey	AARI, -18°C, limited capacity
United Kingdom	IGER, only –25°C - depends on the volume of seed

Minimum standards for regeneration

(see Part II, pp. 112-119)

Status of current EU-funded research project ICONFORS: Improving germplasm conservation methods for perennial European forage species

Ian Thomas

Discussion

The Group is looking with interest at the large amount of data that are being generated by this project and acknowledges that it is very difficult to reach practical conclusions based on current scientific knowledge.

Experiences with the new regeneration strategy at the Nordic Gene Bank

L. Bondo explained that a balanced bulk of single plant harvests from at least 49 individual plants had been adopted for the rejuvenation of the base collection, while the active collection and newly collected material is regenerated by a bulk harvest of at least 30 plants. It was not considered possible to apply the single plant harvest method for regeneration of the active collection due to its high cost.

The active collection is regenerated from the active collection only, the base collection from the base collection seed.

A few members questioned the system whereby the base collection is not used for the regeneration of the active collection and the fact that, out of the 20 000 seeds stored in the base collection, only around 50 would be used to regenerate it.

E. Willner, L. van Soest and M. Ševčíková stated that their genebanks adopted the bulk harvest approach, since the single plant harvest would be too expensive (also the seed threshing and cleaning by single plants would significantly increase the cost).

Recommendation
The Group is looking forward to the indications that the ICONFORS project will be able to give regarding the extent of the genetic effect of the choice of the bulk versus single plant harvest. Also the economic analysis that ICONFORS will undertake will hopefully give more clear indications to the Group.

The Group agreed that the single plant harvest would be recommendable, whenever it is considered absolutely essential to maintain the genetic integrity of the accessions, while bulk harvesting would remain acceptable, considering current limitations, as the most practical choice for economic reasons.

The Group appreciated learning about the methodology used by the NGB and noted that it is following acceptable standards and aiming at higher standards for regeneration of the base collection. It was agreed that it was important to publish the methodologies used by all genebanks in order to increase the transparency of operations.

Workplan
All Working Group members will send a description of the regeneration standards used for forage species in their respective country's genebank. This text will be sent to the Working Group Chair and the ECP/GR Coordinator ***by 1 August 2003****. A table will be prepared by E. Willner and B. Boller and provided to the WG. This will be published on the Forages Network Web site and eventually included in the final report (Appendix I).*

Core collections

(see Part II, pp. 120-140)

The *Medicago* "core collections" subgroup established at Elvas in 1999
Valeria Negri

Discussion
E. Willner commented that the *Medicago* catalogue for Europe could be used. V. Negri replied that her group attempted to define the best methodology on the basis of the existing literature. The *Medicago* group wishes to receive some input from the entire Group regarding the need to establish a core collection before proceeding with the project.

Progress in analysis of the evaluation of the European *Lolium* core collection: genetic diversity between and within ryegrass populations of the ECP/GR collection by means of AFLP markers
An Ghesquiere

Discussion
The conclusions are complicated by the fact that the geographic structure of the populations can be distinguished by the molecular markers, but still the majority of variance is within populations. Since AFLPs tend to represent non-coding parts of the genome, grouping of accessions should be confirmed by analysis of heritable plant characters.

I. Thomas informed the Group that IGER is prepared to continue the statistical analysis of the *Lolium* project and will pay the cost of about £13 000 for a preliminary analysis to be carried out by HRI Wellesbourne. This preliminary analysis will be completed by the end of

2003 and a report will be distributed to the Group members on request (see Appendix II for preliminary analaysis).

The Group was pleased to see that conclusions and analysis are still being drawn from the results of the *Lolium* project and recommended IGER to continue with the analysis and appreciated the financial contribution that IGER is prepared to make. I. Thomas urged those who have additional evaluation data to send them (again) to IGER; they could then also be included in the data analysis. This applies to digestibility data obtained in Switzerland, Belgium and Norway, as well as to disease resistance data from Germany and possibly more.

A. Ghesquiere informed the Group that all the data from the AFLP analysis carried out at DvP (Department of Plant Genetics and Breeding, Melle, Belgium) are available upon request.

T. Marusca proposed that future analysis should include the consideration of the altitude of origin of the accessions, since these can be related to physiological traits.

Lolium perenne core collection - results and conclusions from Poland

Włodzimierz Majtkovski

Discussion

L. Horváth suggested that a sub-regional grouping of accessions from the Danube valley could be more meaningful than separating accessions on the basis of the country of origin. This comment applies to other regions as well.

The question was raised as to how the Group should proceed with the concept of core collections.

L. van Soest expressed concern that core collections might not be used by breeders, although they had been most in favour of creating these collections originally. His experience with other crops is that breeders are mainly interested in very specific core collections, depending on specific problems. He quoted the example of the barley core collection that disappointed many plant breeders, since traits of interest were not found. He also noted that core collections sometimes could not be used since there were not sufficient seeds available. In the case of the *Lolium* core collection, the Group agreed that it would be important to ensure that the seed is available. I. Thomas confirmed that with very few exceptions, seed could be retrieved from IGER. However, he does not know of specific requests to IGER made for samples of the *Lolium* core collection. The Group hopes that new interest may arise for the *Lolium* core collection once complete evaluation data are made publicly available.

V. Meglič thought that the core collection, if evaluated, might offer access to the traits of interest. P. Marum considered that there are situations where building a core could be useful, such as in the case of *Festuca*, since this could be a useful step to help genebanks to prioritize the maintenance of a manageable number of accessions. He also thought that definitions of cores, based on simple criteria, such as geographic source, could be a useful basis for submitting and undertaking research projects, even without a commitment to carry out the evaluation work initially. L. Horváth commented that, from the point of view of the genebank manager, the preparation of core collections would facilitate prioritization of work, mainly in the case of forage legumes. However, it does not seem realistic to undertake an evaluation in order to define the core, but rather to define the core in order to have a limited number of accessions to evaluate.

V. Negri commented on behalf of the *Medicago* core collection group that it is firstly important to define a clear purpose to prepare a core collection. If the need exists, then the Group should look for funds. B. Montegano announced that a meeting of the French network would take place in France and the question would be raised there.

Recommendations

The Group agreed that there does not seem to be an immediate need to engage in building new core collections, except perhaps in the case of Medicago. *Further indications on the importance of building a core collection of* Medicago *will be sought by V. Meglič at the EUCARPIA forage meeting planned in September and from the French network by B. Montegano.*

On-farm / *in situ* conservation

(see Part II, pp. 141-148)

On-farm conservation in Finland

Louise Bondo

Discussion

L. Bondo and P. Marum explained that the farmers would be able to get a small subsidy if they grew the registered landraces. Farmers would also be allowed to sell the seed of the local varieties, provided there was proof of distinctness, obtained through a specific certification process.

On-farm conservation/improvement of forage crops in Norway

Petter Marum

Forage crop landraces in central Italy

Valeria Negri presented a survey of the cultivation of landraces of *Medicago sativa* and *Onobrychis viciaefolia* in Umbria. These are cultivated by relatively old farmers on large farms (>70 ha) for local use and due to their better performance and persistence.

She explained that the continued existence of landraces in cultivation had been mainly due to the possibility of selling the seed, which is now no longer legally possible. The Finnish approach would be a good model to follow, since so far EC Directive 98/95 has not been put in practice in Italy – there is also a fierce opposition from the seed companies toward its implementation.

Discussion

P. Marum questioned the idea that cultivation of landraces should become a task to be assigned preferentially to organic farmers. V. Negri agreed that what is important is to maintain the dynamic development of the landraces, independently of the type of cultivation. She also explained that the better performance of landraces over bred varieties was due until now to the fact that breeding companies were not interested in breeding adapted varieties that could compete with ecotypes, since farmers had the opportunity to sell the seed of ecotypes.

The presentations were followed by a general discussion on on-farm conservation initiatives in Europe.

Discussion

P. Marum mentioned a project that was recently started in Norway to identify sites for *in situ* conservation. L. van Soest also mentioned a Dutch project to identify the forage diversity within protected areas. P. Freudenthaler reported that regulations subsidizing special cultivation for conservation exist in Austria.

I. Thomas mentioned the risk that *in situ* conservation, if run by environmental agencies, would aim at improving the general biodiversity and this might lead to disappearance of productive species from the fields.

V. Negri suggested that *in situ* conservation should be given priority at all levels, since it helps to maintain a dynamic conservation.

Experiment on grassland – Mondsee

Rudolf Schachl presented an experiment that was started in 1968 and that was originally designed to study the influence of different fertilizer and manure applications on an absolutely untouched and natural grassland, and their effect on fodder quality and its effect on cattle fertility. This study was completed and discontinued, and interest in it was lost.

However, this experiment, running for almost 35 years in its original form, started to show a remarkable influence on the composition of the grassland, as well as—and this is the interesting thing—on the biotypes themselves. Thus it was decided to continue this experiment under the responsibility of the genebank in Linz.

Change in the composition of grassland is a well-known effect, while the change of the biotype is not so much understood. In this context, it should be mentioned that *Lolium* types with completely changed botanical characters and showing the typical feature of a landrace were developed when fertilizer was applied in excessive quantities.

Therefore the question was raised whether the formation of a landrace is a unique process or a repeatable one. The application of fertilizer and agrochemicals can be considered an evolutionary factor, and R. Schachl stated his opposition to excluding their application as a matter of principle during the maintenance of genetic resources, since it might be an important factor for *in situ* conservation.

This experiment, however, will be able to show the possible evolutionary steps that can be expected when collecting genetic resources from agriculturally managed grassland, and will answer the question whether collection has to be repeated, and if so, at what intervals.

He invited researchers who are interested in this evolutionary question to seek close contact with the Linz genebank in order to discuss and study this matter. He finally launched a strong appeal to continue this experiment, which is probably unique in Europe, and is just starting to produce highly valuable results.

The Group appreciated the offer from R. Schachl for collaboration and will seek to disseminate this information to interested researchers.

Research activities

(see Part II, pp. 149-181)

Cyprus: collection and evaluation of local germplasm for forage production
Demetrios Droushiotis

Screening of tall oatgrass and yellow oatgrass germplasm held in the European Central Crop Databases
Magdalena Ševčíková

Evaluation of important traits as the basis for an assessment of *Poa* genetic resources for breeding purposes
Evelin Willner

Study of quantitative features in *Phalaris arundinacea* accessions in two different environments in Romania
T. Marusca

Safeguarding a unique collection of former Swiss red clover landraces
Beat Boller

Evaluation of orchard grass *(Dactylis glomerata* L.) populations collected in Turkey
Huseyin Özpinar

Evaluation of the agronomic value of perennial fodder legumes in Georgia
Avtandil Korakhashvili

The use of phenological data for the estimation of global warming "fingerprints" on forage grasses
Grzegorz Żurek

Update on the Framework 6 Programme and the EC 1467 Regulation of the European Union

L. Maggioni gave a brief account of the prospects for the opening of calls for projects to be funded by the European Commission on genetic resources. Regarding the Multiannual Sixth Framework Programme for Research (2002–2006) (http://fp6.cordis.lu/fp6/home.cfm), he reported that the terms of reference for past and upcoming calls for proposals did not provide much opportunity for proposals for genetic resources research. A more positive expectation could however be derived from the expected launching of the new proposal for a Council Regulation for a Community programme on the conservation, characterization, collection and utilization of genetic resources in agriculture and repealing Regulation (EC) 1467/94. The first call for proposals was expected at the end of 2003 or early 2004 and a leaflet including a summary of the information currently available was distributed to the Group.

Discussion
The Group thought that it would be worthwhile to submit proposals to the new EC programme in order to have a better description and evaluation of forage crop collections for better utilization. It was considered that there were good prospects for approval of proposals from this Group since the EC Regulation in its previous version funded no work on forage crops. Moreover, forage crops seemed to satisfy the objectives of the programme, i.e. diversification of production, improved product quality, sustainable management and use and better care for the environment. Suggestions were also made to include NGOs and to focus the subject on *in situ* conservation and perhaps on organic farming in order to improve the chances of success. However, the Group wondered whether *in situ* approaches could fit into the stepwise approach of the previous regulation that seems to have been re-adopted in the new version.

Recommendation
The Group encouraged its members to seek for volunteers that could become coordinators for the preparation of an EU project. It was recorded that M. Humphreys (IGER, Aberystwyth, UK), J. Baert (DvP, Melle, Belgium) and D. Reheul (University of Gent, Belgium) or the Institute for Organic Farming (FibL, Berlin, Germany) could be suitable candidates.

The opportunity to wait for the second call in 2005 and therefore to be able to extend the project to the new countries joining the EU was also recommended. In this case, V. Meglič (Ljubljana, Slovenia) was suggested as a potential coordinator.

Workplan
The Group members agreed to communicate ideas for a project title and project coordinator to the Chair ***within the next 4 months***.[6]

Possibilities of funding forage crop genetic resources projects as part of national "Plans of Action" to implement the "Global Plan of Action", following the Rio Convention on Biodiversity

The members were asked to report on the possibilities of receiving funds for genetic resources projects as a consequence of the establishment of national plans of action for plant genetic resources for food and agriculture. It was confirmed that both in Norway and Switzerland it was possible to obtain funds for forage crop genetic resources activities from the national government. Also in Denmark this opportunity was said to exist, although no projects on forage crops had been funded so far.

No other countries offering similar opportunities were known to the Group.

Conclusion

The section *Discussion and Recommendations* of the report was presented to the participants and was approved with minor modifications.

Beat Boller and Evelin Willner were reconfirmed by the Group as respectively Chair and Vice-Chair.

Closing remarks

The Group thanked the local organizers for the smooth and effective organization and the warm hospitality.

The Group wished to give special thanks to Rudolf Schachl, who recently retired from the Austrian Agency of Health and Foodsafety, after having devoted his career to plant genetic resources.

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[6] During the EUCARPIA Fodder Crops Section Meeting at Brno (Czech Republic) in September 2003, several Working Group members gathered to collect further ideas on the topics that could be submitted in a proposal to the follow-up regulation of EC 1467/94 and welcomed the offer of Joost Baert to coordinate the project.

PART II. PRESENTATIONS AND PAPERS

Note: This section includes all full papers submitted for publication; when no full papers were submitted, the abstracts provided before the meeting were included.

European Central Forage Databases

European Trifolium subterraneum *and annual* Medicago *species Databases*

Francisco González López and Mónica Murillo Vilanova

Servicio de Investigación y Desarrollo Tecnológico (SIDT), Finca La Orden, Guadajira, Badajoz, Spain

Updating of the databases

After the last meeting of the Forages Working Group in Portugal (November 1999) new data were added to both databases. The **European *Trifolium subterraneum* Database** now contains a total of 4776 accessions (Tables 1 and 2) and the **European annual *Medicago* Database,** 2321 accessions (Tables 3 and 4), belonging altogether to 12 holding institutes. The increase in the number of accessions since 1999 is due to:

- incorporation of two new databases, one from ISCF (Istituto Sperimentale per le colture foraggere, Lodi, Italy – ITA009) and the other from RIPP (Research Institute of Plant Production, Piesťany, Slovakia – SVK001);
- updating of the data from SIDT (Servicio de Investigación y Desarrollo Tecnológico) with the material obtained in three recent collecting expeditions, one in 1998 (southwestern Spain and Portugal) and two in 2000 (southern Spain and Extremadura).

In the updated *T. subterraneum* database, 54% of the accessions, mainly collected in Spain and Portugal, are held at the SIDT genebank (ESP001) (Table 1). In the *Medicago* database only 29% of the accessions are held at SIDT (Table 3) and originate from this area.

Both collections held at the SIDT genebank (*Trifolium subterraneum* and annual *Medicago)* have been multiplied and safety-duplicates have been sent to the Centro de Recursos Fitogenéticos (CRF) in Madrid.

Passport descriptors have been changed to FAO/IPGRI Multi-crop passport descriptors (version December 2001) and include 56 descriptors, according to the list of Forage Passport Descriptors (1997) and additional descriptors such as soil characteristics, rainfall, population, abundance, etc., which are considered relevant for the species.

The databases are searchable on-line on the Internet server of Junta de Extremadura:

- *Medicago:* http:www.juntaex.es/consejerias/aym/dgpifa/sia/r_genetico_medicago.htm
- *Trifolium:* http:www.juntaex.es/consejerias/aym/dgpifa/sia/r_genetico_Trifolium.htm

Identification of the Most Original Samples (MOSs)

The MOSs of both databases have been updated (Tables 5 and 6). In the *Trifolium subterraneum* database there are still 1717 accessions provisionally declared “unknown”, until the ISCF database is updated.

Table 1. European *Trifolium subterraneum* Database - accessions classified by contributing institute

Institute code	Advanced cultivars	Breeders' lines	Weedy	Wild	Unknown	Total
BGR001	3	-	-	4	5	12
DEU001	1	-	-	401	-	402
ESP010	39	118	1	2334	75	2567
GBR016		-	-	1	6	7
GRC006	1	-	-	1	-	2
ITA004	-	-	-	10	-	10
ITA009				1686		1686
ITA015	1	-	-	52	10	63
RUS001					15	15
TUR001	-	-	-	12	-	12
Total	**45**	**118**	**1**	**4501**	**111**	**4776**

Table 2. European *Trifolium subterraneum* Database - accessions classified by country of origin

Country of origin	Advanced cultivars	Breeders' lines	Weedy	Wild	Unknown	Total
Not registered	4	4	-	321	76	405
AUS	32	12	-	1	12	57
BGR	-	-	-	4	7	11
CYP	-	-	-	18	-	18
DZA	-	-	-	10	1	11
ESP	9	82	1	1549	3	1644
FRA	-	-	-	39	1	40
GRC	-	6	-	66	-	72
ISR	-	1	-	1	-	2
ITA	-	1	-	1797	4	1802
MAR	-	2	-	425	3	430
MLT	-	1	-	-	-	1
NZL	-	-	-	-	1	1
PRT	-	6	-	214	2	222
TUN	-	2	-	47	-	49
TUR	-	-	-	6	-	6
USA	-	1	-	2	-	3
YEM	-	-	-	-	1	1
YUG	-	-	-	1	-	1
Total	**45**	**118**	**1**	**4501**	**111**	**4776**

Table 3. European annual *Medicago* species Database - accessions classified by contributing Institute

Institute code	Advanced cultivars	Breeders' lines	Landraces	Weedy	Wild	Unknown	Total
BGR001		-	-	-	9	118	127
DEU001	1	-	-	-	370	-	371
ESP010	14	2	-	1	647	14	678
GBR004	-	-	-	-	81	-	81
GBR016		-	-	-	23	29	52
ITA015	1	198	2	-	197	243	641
RUS001	-	43	-	-	314	-	357
SVK001	1	-	-	-	13	-	14
Total	**17**	**243**	**2**	**1**	**1654**	**404**	**2321**

Table 4. European annual *Medicago* species Database - accessions classified by country of origin

Country of origin	Advanced cultivars	Breeders' lines	Landraces	Weedy	Wild	Unknown	Total
Not registered	6	179	-	-	26	194	405
AFG	-	-	-	-	2	2	4
ARM	-	-	-	-	6	-	6
AUS	6	29	-	-	16	44	95
AZE	-	-	-	-	3	-	3
BEL	-	-	-	-	1	-	1
BGR	-	-	-	-	12	10	22
BLR	-	-	-	-	1	-	1
CAN	-	-	-	-	4	2	6
CHE	-	1	-	-	2	-	3
CHL	-	-	-	-	1	1	2
CHN	-	-	-	-	3	-	3
CYP	2	-	-	-	171	4	177
CZE	1	-	-	-	22	2	25
DEU	-	-	-	-	3	1	4
DNK	1	1	-	-	10	-	12
DZA	-	-	-	-	-	2	2
ESP	-	-	-	1	290	6	297
ETH	-	-	-	-	-	7	7
FRA	-	-	-	-	28	4	32
GBR	-	-	-	-	5	1	6
GEO	-	-	-	-	20	-	20
GRC	-	2	-	-	93	3	98
HUN	-	-	-	-	6	2	8
IND	-	1	-	-	-	-	1
IRL	-	-	-	-	1	-	1
IRN	-	-	-	-	-	6	6
IRQ	-	2	-	-	-	2	4
ISR	1	-	-	-	7	13	21
ITA	-	20	2	-	252	28	302
JOR	-	-	-	-	-	1	1
JPN	-	-	-	-	1	-	1
KAZ	-	-	-	-	1	-	1
KGZ	-	-	-	-	1	-	1
LBN	-	-	-	-	-	4	4
LBY	-	2	-	-	-	2	4
LTU	-	1	-	-	1	-	2
LVA	-	-	-	-	1	-	1
MAR	-	-	-	-	377	13	390
MLT	-	-	-	-	-	1	1
NLD	-	-	-	-	1	-	1
NOR	-	-	-	-	3	-	3
PER	-	-	-	-	4	-	4
POL	-	1	-	-	3	2	6
PRT	-	3	-	-	98	8	109
RUS	-	-	-	-	53	4	57
SAU	-	-	-	-	1	-	1
SVK	-	-	-	-	8	-	8
SWE	-	-	-	-	4	-	4
SWZ	-	-	-	-	1	-	1
SYR	-	-	-	-	64	7	71
TJK	-	-	-	-	3	-	3
TUN	-	-	-	-	1	3	4
TUR	-	-	-	-	12	14	26
UKR	-	1	-	-	15	-	16
USA	-	-	-	-	9	10	19
YEM	-	-	-	-	1	-	1
YUG	-	-	-	-	6	1	7
Total	**17**	**243**	**2**	**1**	**1654**	**404**	**2321**

Table 5. European *Trifolium subterraneum* Database. Identification of the most original samples (MOS)

ACCENUMB			**ORIGINALITY**					
GENUS	**SPECIES**	**ORIGIN**	**MOS**	**with MOS**	**One away**	**More away**	**Unknown**	**Total**
Trifolium	*subterraneum*	Bred	7	2	36		1	46
		Collected	1912	54	263	23	18	2270
		Unknown			35	708	1717	2460
Total			**1919**	**56**	**334**	**731**	**1736**	**4776**

Table 6. European annual *Medicago* species Database. Identification of the most original samples (MOS)

ACCENUMB			**ORIGINALITY**					
GENUS	**SPECIES**	**ORIGIN**	**MOS**	**with MOS**	**One away**	**More away**	**Unknown**	**Total**
Medicago	annual	Bred	65	41	22	2	4	134
		Collected	1036	86		3	12	1137
		Unknown			12	416	622	1050
Total			**1101**	**127**	**34**	**421**	**638**	**2321**

European Arrhenatherum *and* Trisetum *Databases*

Magdalena Ševčíková[1] and Ivan Hon[2]

[1] *OSEVA PRO Ltd., Grassland Research Station (GRS), Zubří, Czech Republic*

[2] *Research Institute of Crop Production (RICP), Prague, Czech Republic*

The European Central *Arrhenatherum* and *Trisetum* Databases are maintained by OSEVA PRO Ltd., Grassland Research Station at Zubří (Czech Republic) and managed under FoxPro software. Both databases have been made available on-line on the Internet server of the Research Institute of Crop Production (RICP, Prague) since February 2002 and are also searchable on-line:

- *Arrhenatherum*: http://www.ecpgr.cgiar.org/databases/Crops/arrhenat.htm
- *Trisetum:* http://www.ecpgr.cgiar.org/databases/Crops/Trisetum.htm

The **European *Arrhenatherum* Database** contains passport data for 254 accessions of 3 taxa stored in 15 European genebanks. The collections contain accessions from 25 countries of origin and include advanced cultivars (59), landraces (7), breeding material (2), wild material (107), ornamental type (1) and material of unknown status (78).

The **European *Trisetum* Database** includes passport data on 86 accessions of 4 taxa held by 11 European genebanks. The collections contain accessions from 9 countries of origin and include advanced cultivars (17), landraces (2), breeding material (3), wild material (53) and material of unknown status (11).

Seed requests were sent to the holding institutions and the quality of the seed obtained was monitored. The viable accessions were characterized and evaluated within the research project No. 521/99/163 supported by the Grant Agency of the Czech Republic.

European Dactylis *and* Festuca *Databases*

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Since the last meeting of the Forages Working Group in 1999, only small changes were made in both databases managed by the Botanical Garden of the Plant Breeding and Acclimatization Institute. The average increase was 8.1%, with 1184 records added for *Festuca* and 257 records for *Dactylis*. The databases currently contain 8779 records for *Festuca* and 9050 for *Dactylis* (Table 1).

Table 1. Changes in the European *Dactylis* and *Festuca D*atabases since 1999

Database	No. of records		
	last update	current status	increase (%)
Festuca	7595	8779	1184 (13.4)
Dactylis	8793	9050	257 (2.8)
Total	**16388**	**17829**	**1441 (8.1)**

The **European *Festuca* Database** covers nearly 100 species (Table 2); however only six species (F. *pratensis, F. arundinacea, F. rubra, F. ovina, F. gigantea* and *F. duriuscula*) account for more than 97% of all records.

The **European *Dactylis* Database** consists of only three species (*D. glomerata, D. marina* and *D. polygama*) with *D. glomerata* accounting for 99.7%.

Tables 3 and 4 provide the current status of both databases.

There is still a great need to standardize taxonomy, especially in the genus *Festuca*. For example in the case of *Festuca rubra* one accession could have different taxonomic names and all of them are correct. For example: *Festuca rubra* L. = *F. rubra* L. subsp. *fallax* (Thuill.) Hayek = *F. rubra* L. var. *commutata* Gaudin. = ***F. nigrescens*** Lam., ***F. trachyphylla*** = *F. duriuscula*, ***F. capillata*** = *F. tenuifolia.* Names in ***bold italics*** letters are correct according to the *Flora Europaea* (Tutin *et al*. 1980). It is suggested to use this source for the standardization of the taxonomic descriptions.

Reference

Tutin, T.G., V.H. Heywood, N.A. Burges, D.M. Moore, D.H. Valentine, S.M. Walters and D.A. Webb. 1980. Flora Europaea. Vol. 5. Cambridge Univ. Press.

Table 2. Species currently recorded in the European *Festuca* Database

No.	Species	No. of accessions	No.	Species	No. of accessions
1	*F. airoides*	2	51	*F. koritnicensis*	1
2	*F. alpina*	2	52	*F. laevigata*	1
3	*F. altissima*	2	53	*F. lambinonii*	1
4	*F. amethystina*	3	54	*F. lemanii*	1
5	*F. ampla*	1	55	*F. liviensis*	1
6	*F. armoricana*	1	56	*F. longifolia*	5
7	*F. arundinacea*	1539	57	*F. magellanica*	1
8	*F. arvernensis*	3	58	*F. mairei*	1
9	*F. borderi*	1	59	*F. mathewsii*	2
10	*F. bosniaca*	1	60	*F. nigrescens*	2
11	*F. brigantina*	1	61	*F. niphobia*	1
12	*F. burgundiana*	1	62	*F. ovina*	102
13	*F. caesia*	1	63	*F. pallens*	4
14	*F. capillata*	6	64	*F. paniculata*	2
15	*F. carpatica*	2	65	*F. petraea*	2
16	*F. christiani-bernardii*	1	66	*F. polesica*	2
17	*F. cinerea*	6	67	*F. pratensis*	6060
18	*F. circummediterranea*	1	68	*F. pseudoeskia*	1
19	*F. costei*	2	69	*F. pseudotrichophylla*	1
20	*F. cretacea*	1	70	*F. pseudovina*	4
21	*F. curvula*	2	71	*F. pulchella*	1
22	*F. dimorpha*	1	72	*F. pumila*	1
23	*F. dolichophylla*	1	73	*F. punctoria*	2
24	*F. drymeja*	2	74	*F. rubra*	1540
25	*F. durandii*	1	75	*F. rupicarpina*	1
26	*F. duriotagana*	1	76	*F. rupicola*	3
27	*F. durissima*	2	77	*F. scariosa*	2
28	*F. duriuscula*	30	78	*F. scirpifolia*	1
29	*F. duvalii*	1	79	*F. semilusitanica*	1
30	*F. elatior*	9	80	*F. sibirica*	2
31	*F. elegans*	1	81	*F. skvortsovii*	1
32	*F. eskia*	1	82	*F. stricta*	2
33	*F. extremadura*	1	83	*F. tatrae*	2
34	*F. extremorientalis*	1	84	*F. tenuifolia (= F. capillata)*	4
35	*F. filiformis*	1	85	*F. trachyphylla (= F. duriuscula)*	5
36	*F. gaetula*	1	86	*F. tuberculosa (= Castellia tuberculosa)*	1
37	*F. gamisansii*	1	87	*F. vaginata*	2
38	*F. gautieri*	4	88	*F. valesiaca*	8
39	*F. gigantea*	18	89	*F. varia*	2
40	*F. glauca*	5	90	*F. vasconcensis*	1
41	*F. gracilior*	1	91	*F. violacea*	1
42	*F. halleri*	1	92	*F. yvesii*	2
43	*F. henriquesi*	1	93	species not defined	15
44	*F. hervierii*	2			
45	*F. heterophylla*	8		Species name defined but not present	
46	*F. huonii*	1		in accessible floras or checklists:	
47	*F. indigesta*	3	94	*F. nipicola*	1
48	*F. jeanpertiii*	1	95	*F. patrae*	1
49	*F. juncifolia*	1	96	*F. rusca*	1
50	*F. kirilovii*	1	97	*F. rubi*	1
Total no. of accessions = 8779					

Table 3. Current status of the European *Dactylis* Database (number of accessions)

Genebank (INSTCODE)	Advanced cultivars	Breeders' lines	Primitive cultivars, landraces	Semi-natural ecotypes	Wild ecotypes	Status unrecorded		Total for genebank	%
						named	unnamed		
BGR001	23	10	119	8			7	167	1.8
CHE001					11			11	0.1
CZE082	116	12	2					130	1.4
DEU001	88	24			669			781	8.6
ESP009					333	4		337	3.7
ESP119	1				22			23	0.3
FRA051	57				190			247	2.7
GBR004					58		7	65	0.7
LTU001	34			11		20		65	0.7
LVA0007	8	20						28	0.3
POL003	28				1			29	0.3
POL022	117				5919		136	6172	68.2
PRT084	6	1			136		1	144	1.6
ROM003	27	5			17			49	0.5
ROM007		3	25		19			47	0.5
SVK012	100	2		37	197			336	3.7
SVN019	1				27			28	0.3
SWE002	31	3			177			211	2.3
TUR001					180			180	2.0
Total	**637**	**80**	**146**	**56**	**7956**	**24**	**151**	**9050**	**100**
%	**7.0**	**0.9**	**1.6**	**0.6**	**87.9**	**0.3**	**1.7**	**100**	

Table 4. Current status of the European *Festuca* Database (number of accessions)

Genebank (INSTCODE)	Advanced cultivars	Breeders's lines	Primitive cultivars, landraces	Semi-natural ecotypes	Wild ecotypes	Status unrecorded		Total for genebank	%
						named	unnamed		
BEL087							81	81	0.9
[BELHBBRMA]					28	1		29	0.3
BGR001	12	1					14	27	0.3
CHE001					37			37	0.4
CZE079	3							3	0.0
CZE082	308	8	1		2			319	3.6
DEU001	123	9			529			661	7.5
ESP119					22			22	0.3
FRA051	59				116			175	2.0
GBR004					75		3	78	0.9
LTU001	20			**5**		65		90	1.0
LVA0007	15	308						323	3.7
POL003	134				106			240	2.7
POL022	141				4322		191	4654	53.0
ROM003	87	6			320		1	414	4.7
ROM007		2	38		35		13	88	1.0
SVK012	157	4		**100**	918	14		1193	13.6
SVN019	2	1						3	0.0
SWE002	67	5			242			314	3.6
TUR001					28			28	0.3
Total	**1128**	**344**	**39**	**105**	**6780**	**80**	**301**	**8779**	**100.0**
%	**12.8**	**3.9**	**0.4**	**1.2**	**77.2**	**0.9**	**3.4**	**100**	

National collections and collecting activities

The Albanian forage germplasm collection

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Introduction

Since the last meeting of the Working Group on Forages in 1999, the number of forage crop accessions has been increased following the establishment of a working group and with the support of international donor institutes. The working group, led by Karaman Tahiraj and Ismet Boka, organized several expeditions all over Albania as part of a project financed by the Ministry of Science and Education.

The Albanian collection of forage crops comprises more than 500 accessions including the genera *Lolium*, *Trifolium*, *Poa*, *Dactylis*, *Medicago*, *Festuca*, *Phleum*, *Pisum*, *Avena*, etc. Seeds are currently available from the collection of the Research Institute of Field Crops (RIFC) in Fushë-Krujë (Table 1).

Table 1. Current status of the RIFC forage collection (number of accessions)

Genus	Type of sample					Total
	Varieties	Local material	Wild material	Breeding material	Unknown	
Avena	6	2	2	3	6	19
Dactylis	3	2	2	0	3	10
Festuca	4	3	38	0	2	47
Hordeum	18	0	4	0	0	22
Medicago	25	3	26	1	13	68
Melilotus	6	0	3	0	1	10
Lathyrus	11	2	4	0	0	17
Lolium	4	6	4	2	0	16
Lotus	3	1	7	0	1	12
Phalaris	2	1	3	0	2	8
Phleum	4	2	10	0	0	16
Poa	0	1	3	0	0	4
Pisum	24	1	2	1	5	33
Soya	25	0	0	3	9	37
Sorghum	10	0	0	4	0	14
Trifolium	15	4	70	1	9	99
Vicia	40	4	20	0	8	72
Total	**200**	**32**	**198**	**15**	**59**	**504**

Conservation and safety-duplication

Considering the needs of researchers and farmers in Albania and the need to conserve and use the genetic diversity of forage crops, the concept of "collection and selection of landraces of forage crops" is presented as an integrated approach to:

- Evaluate populations at the local level and improve them more quickly than by traditional selection;
- Promote the development of adapted and competitive local populations of forage crops; and
- Promote the sustainable use of landraces of forage crops derived from local genepools and consequently their genetic diversity. There are good reasons to believe that diversity is important for sustainable agriculture (Swift and Anderson 1992).

Our current activities include *in situ* and *ex situ* conservation. New accessions which enter the collection (Table 2) have to be multiplied and characterized (primary evaluation).

To preserve this genetic diversity, forage crops have been collected and sent for safety-duplication to the genebank at NCSS (National Center for Seeds and Seedlings) in Tirana. Seed samples are dried to a moisture content of 4% and packed in foil bags. The storage temperature varies between -3°C and -4°C. There are currently 215 forage accessions stored as safety-duplicates at the genebank in Tirana.

Table 2. Increase of the RIFC forage collection, 1995-2003 (number of accessions)

Species	1995	1997	1999	2003
Avena sativa	9	4	18	19
Dactylis glomerata	5	2	1	10
Festuca arundinacea	23	10	2	47
Hordeum vulgaris	11	5	14	22
Lolium multiflorum	4	1	2	6
Lolium perenne	4	1	0	10
Lathyrus sativa	8	3	11	17
Lotus corniculatus	6	2	2	12
Medicago sativa	20	10	17	38
Medicago polymorpha	2	2	2	5
Medicago sativa x *varia*	2	1	1	5
Medicago hybrida	3	1	1	5
Medicago lupulina	4	1	1	7
Medicago falcata	2	1	1	4
Medicago nigra	1	0	1	3
Medicago arabica	0	0	0	1
Melilotus officinalis	5	1	4	7
Melilotus alba	0	0	0	3
Phalaris arundinacea	4	0	0	8
Phleum pratense	8	0	2	16
Pisum sativa	10	7	22	28
Pisum arvense	5	0	1	5
Poa pratense	2	0	0	4
Sorghum halepense	7	4	0	14
Soya hispida	18	8	18	37
Trifolium pratense	5	2	4	33
Trifolium repens	6	1	3	25
Trifolium alexandrinum	15	10	2	29
Trifolium squarrosum	3	1	1	6
Trifolium incarnatum	2	1	1	3
Trifolium subterraneum	2	0	2	3
Vicia sativa	12	7	13	44
Vicia faba	10	3	17	25
Vicia villosa	1	0	0	1
Vicia ervilia	0	0	2	2
Total	**218**	**89**	**166**	**504**

Regeneration

The concept "collection and selection of landraces and introduced varieties of forage crops" is based on the evaluation of these varieties/populations and seed lots in the institute's field plots. The experimental layout of the field trials and evaluation methods vary according to local conditions but should allow testing of landraces and selection of new strains, varieties or populations. The populations of many threatened species are doomed for demographic reasons, even though they may not disappear for many decades (Primack 1998).

Our genetic material is sown in the field. Each accession is planted in three rows with one replication and a control variety is planted after every 10 accessions (Allard 1960).

Since 2000 the activities of the Research Institute of Field Crops include:

- Performance testing of forage crop varieties to evaluate the new selected material for important agronomic traits such as yield, maturity, stand ability, quality, etc.
- Cultivation trials: evaluation of the genetic quality of different varieties by controlled planting. We cannot undertake genetic studies owing to the lack of modern laboratory equipment.
- Demonstration fields: presentation of current varieties (landraces and introduced varieties of forage crops) compared with the most important competing varieties.

Utilization

Our collection has been utilized in both research and plant breeding activities, especially in pre-breeding activities on landraces of the genera *Trifolium*, *Medicago* and *Pisum*. Moreover, even within traditional cultivars there is usually a high level of genetic diversity. The combination of distinctiveness and variability which characterized many landraces was well described by Harlan (1975). Landraces have a certain genetic integrity, are recognizable morphologically, and most important, are genetically diverse.

As mentioned at the last meeting, among the collected accessions, the 'Shishtavec' clover landrace (genus *Trifolium*) is found in the northeastern part of the country at about 1500 m asl. This annual plant is 120-150 cm tall. It produced 40-50 t/ha green mass in experimental conditions. Its hay is of a very high quality and seed yield is 200-300 kg/ha. Other interesting landraces include 'Tomin' for alfalfa and 'Voskopoja' for forage pea. Conservation of the populations is one of the main tasks of our institute in order to minimize genetic erosion. The value of biodiversity in all ecosystems and especially in agroecosystems has been more widely and dramatically realized through its disappearance rather than through its presence (Collins 2001). The species diversity of natural ecosystems is extremely variable (Edwards and Hilbeck 2001).

Future activities

All accession data have been recorded according to the IPGRI descriptor list. Collaboration with analogous institutions is improving, especially with the IPK Genebank (Institut für Pflanzengenetik und Kulturpflanzenforschung, Gatersleben, Germany), to which some forage samples were sent.

The main research topics in the future are:

- Increase of accessions from natural flora and from other countries, identification of most original samples including the following codes: GBK (institute holding the accession), DON (donor's institute), COL (collector's institute) and BRE (breeder's institute).
- Conservation and increase of seeds of accessions with high feeding value: landraces 'Tomin' (alfalfa), 'Shishtavec' (clover) and 'Voskopoja' (pea).
- Establishment of a genebank at our institute for the conservation of these accessions for long-term storage. No cold store is available for the moment. Two store rooms are needed, one at 0°C and one at -15°C.
- Computer data processing and collaboration with international genetic resources networks. The development of a computerized management system will make the routine work in the seed store more effective.

We plan to develop a database for forage crops but unfortunately do not have on-line access to genetic resources databases.

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Forage genetic resources in Austria

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With an area of 84 000 km^2, Austria is a relatively small country, but nevertheless very diverse. For example, in the east the altitudes are below 200 m asl while they reach nearly 3800 m in the Central Alps in the west. Correspondingly, the climatic conditions vary from a continental climate with roughly 500 mm rainfall in the east to the climate influenced by the Atlantic in the west, approaching almost the glacial zone. Especially here on the northern slopes of the Alps the rainfall increases to more than 2000 mm per year. The so-called "Tiroler Oberland" in Tyrol, the area embedded between the northern and southern ranges of the Alps, forms a remarkable exception with 500 to 600 mm rainfall only. These geographic and climatic conditions however cause a wide variation in the flora, which is also influenced by various farming methods.

In the east, field cropping and vineyards dominate. Up to 20 years ago, field cropping even expanded. Cultivation of fodder plants, quite important in the past, lost its importance and gave place to fallowing, which was encouraged and financially supported. With respect to genetic resources, only small areas could survive more or less untouched, and we must assume that a reasonable number of local genetic resources were lost.

To some extent this might be true also for the area north of the Danube and the northern Sub-Alpine region, though the loss seems to be less dramatic. In this context also larger areas in Styria and Carinthia need to be mentioned. The closer the mountains, the more important meadows and pastures become. But also this grassland has changed its natural features and plant composition drastically by intensification of farming. A main factor in this respect is and was re-sowing with commercial varieties, with or without ploughing first. Consequently in advanced areas with intensive agriculture, untouched natural habitats became quite rare, and where still present they are rather limited in size. Another point is that of mountain slopes: this form of grassland was also subject to intensification, but intensity decreased again, giving the grassland a chance to return to its natural state. The higher mountainous areas especially are characterized by this process of increasingly extensive management, even accompanied by reforestation. To sum up we can say that these areas are larger and still contain their original plants: there is less loss of genetic variability.

Grasslands however dominate in the central Alps in the west, although in smaller dryer locations field cropping can be found as well. There also, in advanced areas genetic variability must be assumed lost, or at least endangered by the intensification of the agriculture. On the other hand, a large number of small farms there, having additional income, retain their traditional extensive farming methods. Though in principle extensive farming would favour the survival of the original genetic variability, these areas are increasingly endangered by reforestation as mentioned above, especially when grassland is abandoned and no longer managed. Further, to some extent meadows are converted to pastures, thus changing the composition of the grassland which is inevitably accompanied by genetic erosion. The existence of pastures in cleared forests and the use of Alpine grassland for grazing at high altitudes should be mentioned as a peculiarity of this western part of the Alps. These sites, which cover a quite significant area, retain the original variability of the flora, even though this is to some extent also a product of the traditional farming, and these areas must be considered an important ecological niche for a large number of endangered species.

Let us reflect now on the endeavour to maintain this genetic variability of forage plants in Austria. *Ex situ* conservation of forage species does not play an important role at all. Only a very few local landraces collected in the Tyrol were preserved by the genebank in Linz. Collections were established for the purpose of plant breeding. No significant and systematic collecting activities were undertaken. Unfortunately, one of the largest collection held by the Plant Breeding Station Reichersberg in Upper Austria, comprising older commercial varieties as well as several local strains, was lost when the breeding programme for forages was terminated. The breeding activities and seed production of local races of forages in the Plant Breeding and Seed Testing Institute in Rinn, Tyrol, were also stopped recently when the institute was formally closed. Thus at present, breeding efforts are only carried out at the Federal Institute for Alpine Agriculture in Gumpenstein, Styria. This breeding programme focuses on the selection of certain local types suitable for re-sowing of grassland in higher regions, and is not concerned with real genebank activities or the maintenance of genetic resources. For the time being, no official genebank is properly equipped and staffed to take care of forage genetic resources; therefore it also makes little sense to start collecting as long as the maintenance of the material collected is not ensured.

Thus the most reasonable solution to prevent larger genetic erosion appears to be strengthened *in situ* conservation, and in this respect some initiatives have been started. For instance, in the recent past hay has been increasingly applied as organic matter when grassland is renewed; another recommended measure provides grass-cutting at a late stage, allowing the ripe seeds to be harvested. Seed harvested from such grassland, turned back to its original state, is available on the market in limited amounts and might be expected to gain increasing interest and importance in future. To be realistic, no large areas can be sown by this practice, but it is definitely a significant contribution to the maintenance of the genetic variability of our grasslands and will contribute to *in situ* conservation. In this context we should also mention the practice of saving the soil covered by natural grassland when construction work is needed and restoring it again when the work is finished.

The most significant contribution to preserve genetic variability is achieved by national parks, protected areas and legal instructions for maintenance of habitats. In Tyrol, for instance, damp or wet biotopes are protected and their maintenance has been financially supported for years; recently this has been extended to dryer biotopes as well, identified as protected areas. Guidelines are issued and financial support is provided for the investment necessary to maintain sustainable traditional farming. The attempt to move back to traditional management of Alpine grassland is also a step in this direction and includes mowing, besides the common use as pasture. All these activities are carried out by the Austrian government under the ÖPUL programme[7] which aims to reduce fertilizer application and the excessive number of cattle in order to protect natural environment.

In future absolute priority must be given to the documentation and inventory of species on grassland still showing its original genetic variability: the next step should emphasize the description of the various existing biotypes. Considering the increasing cuts of financial resources we may not expect significant support for scientific studies; furthermore, the material already collected may be threatened.

[7] ÖPUL = Osterreichisches Programm für umweltgerechte Landwirtschaft (Austrian Programme for Environmentally Compatible Agriculture)

The Bulgarian forage collection

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Status of the collection

Forage species in the Bulgarian collection are maintained in the national genebank located in the Institute for Plant Genetic Resources "K. Malkov" (IPGR-Sadovo), with a total of 3560 accessions. Since 1999 the increase in the number of entries was mainly due to three collecting expeditions and exchange with other genebanks, botanical gardens and breeding institutions. The collection currently contains 81 grass accessions belonging to 13 genera (*Lolium, Dactylis, Festuca, Bromus, Agropyron, Poa, Phleum, Phalaris, Elymus, Arrhenatherum, Alopecurus, Agrostis* and *Trisetum*) and 223 legume accessions belonging to eight genera (*Medicago, Trifolium, Vicia, Onobrychis, Lathyrus, Lotus, Melilotus* and *Trigonella*) (Table 1).

Table 1. Forage species in the Bulgarian collection (number of accessions)

Genus	Forage collection		Long-term		Bulgarian origin
	2000-2003	Total	2000-2003	Total	(CV = cultivars; L = local; W = wild)
Fabaceae					
Medicago					
perennials	35	461	8	243	
annuals	4	64	-	25	35 (CV); 33 (L); 18 (W); 9 (W)
Trifolium					
perennials	12	318	5	172	14 (CV); 23 (L); 38 (W)
annuals	8	118	-	54	1 (CV); 9 (W)
Onobrychis	11	102	6	73	1 (CV); 15 (L); 7 (W)
Vicia	116	1140	284	1006	4 (CV); 48 (old CV);162 (L); 175 (W)
Lathyrus	34	295	-	339	10 (L)
Lotus	1	57	-	18	1 (CV); 2 (W)
Melilotus	-	8	-	8	2 (W)
Other legumes: *Trigonella, Galega*	2	6	2	2	2 (W)
Total legumes	**223**	**2569**	**305**	**1840**	**102 (CV); 243 (L); 262 (W)**
Poaceae					
Lolium	52	335	7	103	43 (W)
Dactylis	1	237	-	233	1 (CV); 165 (W)
Festuca	2	139	25	121	2 (CV); 23 (W)
Bromus	1	65	-	47	1 (CV); 14 (W)
Agropyron	20	76	10	49	8 (W)
Phleum	3	41	-	10	1 (CV); 2 (W)
Phalaris	-	3		3	-
Poa	2	50		3	2 (W)
Elymus	-	9		5	-
Arrhenatherum	-	20		16	1 (W)
Alopecurus	-	3		-	-
Agrostis	-	7		-	-
Trisetum	-	6		2	-
Total grasses	**81**	**991**	**7**	**592**	**5 (CV); 258(W)**

- **Legumes**

The genus *Vicia* is predominant in the forage legume collection in terms of the number of accessions. Half of the accessions originated in Bulgaria. The originals from Bulgaria which are preserved under genebank conditions are as follow: 4 advanced cultivars, 48 old cultivars, 162 landraces and 175 wild species. The other 138 accessions, mainly landraces and wild accessions, are under medium-term storage and are also maintained in a working collection. Earlier the common vetch was known as a traditional forage crop. Since 1960 the area under this crop has decreased progressively; however interest in growing vetch is now increasing again.

The most important forage crop is alfalfa. All accessions of the *Medicago* collection of Bulgarian origin are under long-term storage: 32 advanced cultivars and breeding lines; 33 landraces; and 18 wild. The foreign materials are mainly cultivars and populations originating from France, Hungary, the Czech Republic, Germany, Italy, Spain and the USA. The collection includes local populations and wild forms of *Medicago* originating from Russia, Kazakhstan and Azerbaijan.

- **Grasses**

The genera *Lolium* and *Dactylis* are the predominant grasses in terms of number of accessions. The material of Bulgarian origin includes wild and semi-wild forms collected in the period 1980-2002. The advanced cultivars (5 only) were donated by the Institute of Forage Crops, Pleven (BGR015) where plant breeding with forages is carried out.

The forage collection also includes some rare and threatened plant species of the Bulgarian flora, belonging to the genera *Medicago, Trifolium, Vicia* and *Lotus* (Table 2).

Table 2. Rare and threatened species of the Bulgarian flora in forage collections

Genus	Species	Storage
Medicago	*Medicago rhodopaea* Vel.	Long-term
	Medicago carstiensis Wulf.	Medium-term
	Medicago falcata L. var. *rhomanica* (Prod.) Schw. et Klink.	Medium-term
	Medicago coronata (L.) Bart.	Long-term
	Medicago littoralis Rhode ex Loisel.	Long-term
	Hymenocarpus circinatus (L.) Savi (syn: *Medicago circinatus* L.)	Medium-term
Trifolium	*Trifolium ligusticum* Balb. ex Loisel.	Medium-term
	Trifolium squarrosum L.	Medium-term
	Trifolium constantinopolitanum Ser.	Long-term
	Trifolium spumosum L.	Medium-term
	Trifolium squamosum L.	Medium-term
Vicia	*Vicia incisa* Bieb.	Long-term
	Vicia amphycarpa Dorth.	Medium-term
	Vicia pisiformis L.	Medium-term
Lotus	*Lotus uliginosus* Schkuhr	Medium-term

Characterization and evaluation

As mentioned above, material of local origin is predominant in our collection. Therefore the main task in the characterization of new samples is to observe their agrobiological characters and their reaction to biotic and abiotic stress. The status of the collected material is recorded as: local populations, old cultivars and wild forms. They are utilized further in the breeding programmes for the creation of new cultivars of alfalfa, vetch, sainfoin, orchard grass, tall fescue and ryegrass. During the last three years 100 accessions of common vetch (*Vicia sativa* L.) and 28 accessions belonging to eight other species of the genus *Vicia* L. were evaluated and described for 38 morphological and economic characters. They are sown both in spring and in autumn. The reaction of different *Vicia* species to below-freezing temperatures and to *Fusarium culmorum* is studied. Sources of resistance are found in some accessions of the following species: *Vicia villosa* Roth., *V. sativa* subsp. *angustifolia* Grufb., *V. grandiflora* Scop., *V. lutea* L., *V. pannonica* Crantz. subsp. *pannonica* and subsp. *striata* (Kitcheva *et al.* 2003).

Regeneration

The regeneration status is presented in Table 3. Urgent regeneration is not needed. Usually only a very limited number of accessions are regenerated upon request from the genebank when the germination rate in the control tests after storage falls below 80%. A total of 32 forage accessions from the base collection were regenerated after more than 10 years in storage.

Table 3. Overview of the developments in the Bulgarian forage collection

	Legumes	Grasses
Regeneration		
2000	120	20
2001	85	15
2002	100	7
Total	305	42
Storage		
Temperature	-18°C	
Moisture of seeds	<5-7%	
Availability of seeds for distribution		
No. of accessions	1670	480
%	65%	48%
Utilization (abroad + Bulgaria)		
2000	37 + 91 = 128	22 + 17 = 39
2001	14 + 63 = 77	14 + 26 = 40
2002	3 + 78 = 81	18 + 5 = 23

Stoyanova (2001) reported that grass seeds preserved in the Bulgarian genebank could survive for more than 11 years with minimal changes in seed viability. Odd results observed in this study are attributed to analyses of empty seeds. In conclusion the results show that changes in seed viability of grasses during the first ten years under genebank conditions could be expected only rarely. For example the predicted safe storage time for a decrease of seed viability by 10% (p_{10}) for *Medicago* seeds is suggested to be 902 years, whereas for *Lathyrus* seeds it is 31 years and for *Vicia sativa* only 27 years (Stoyanova 2001).

The new germplasm bred in research programmes in Bulgaria is accepted as most original because the seeds are produced according to requirements for identity[8] under isolation in the field or in glasshouses.

Documentation

In 2001 the EURISCO project for the compilation of a European PGR database was started. When the project started it was shown in the Bulgarian National Report that our database was separated into three sub-tables with different structures. During the last year the data were entered into a main database table designed according to the multicrop passport descriptors (MCPDs) standards. The Institute for Plant Genetic Resources in Sadovo, in its capacity of national focal point for the national inventory, signed the Memorandum of Understanding regarding collaboration on the development of EURISCO.

The passport data of forage crops are available on-line at http://www.genebank.hit.bg/. Some of the available accessions are presented there.

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[8] 1 = Forage Institute, Pleven (*Medicago,Vicia, Lathyrus, Onobrychis, Lotus, Lolium, Dactylis, Festuca arundinacea, Bromus, Agropyron, Poa, Phleum*).
2 = Institute of Upland and Stock Breeding, Troyan (*Trifolium, Lotus, Lolium, Festuca, Poa, Phleum*).
3 = Institute of Wheat and Sunflower, General Toshevo (*Vicia, Lathyrus*)
4 = Institute of Seed Science and Seed Production, Obrastzov Chiflik-Russe (*Medicago*)

Utilization

Since 1999, 286 accessions of forage legumes and 102 accessions of grasses were distributed upon request. The main users are breeding institutes, farmers and genebanks in Bulgaria and abroad. Our practice includes provision of information according to users' needs. The level of utilization is illustrated by the number of distributed accessions (Table 3). Our analyses show that 1670 accessions of legumes (about 65%) and 480 accessions of grasses (about 48%) are available in the collections. All accessions with Bulgarian origin are available for distribution.

Safety-duplication

There is no duplication of the forage collection abroad. Four institutes (see footnote 8) in Bulgaria carry out breeding programmes with forages and maintain working collections of *Medicago, Trifolium, Vicia, Onobrychis, Lotus, Lolium, Dactylis, Festuca arundinacea, Bromus, Agropyron, Poa* and *Phleum.* Parts of these collections are duplicated in the genebank collection.

Collecting activities

Collecting activities organized by IPGR-Sadovo during the last three years were carried out with the coordination and support of the BSBCP (Bulgarian/Swiss Biodiversity Conservation Programme, Sofia). These include collecting missions in dry and steppe regions of Bulgaria: South Dobrudja (Suha reka, Kaliakra), Thracian Plain and Eastern Rhodope (Yancheva *et al.* 2002). The main goal was to collect seeds and information about wild relative of forages and some rare and threatened species (Table 4). This project has two main goals, which are very important for *in situ* conservation of biodiversity:

- Effect of ruderal plant species on the populations of typical steppe flora in the Kaliakra reserve as a factor limiting the population size;
- Effect of controlled grazing and cutting of the adjacent pastures on the maintenance of biodiversity existing in the reserve (BSBCP 2002).

Table 4. Collecting activities in Bulgaria

No.	Genus / species	No. of accessions
1.	*Medicago sativa*	3
2.	*Medicago falcata* var. *rhomanica*	1
3.	*Medicago sativa* x *falcata*	3
4.	*Onobrychis gracilis*	2
5.	*Onobrychis arenaria*	2
6.	*Onobrychis viciifolia*	1
7.	*Trifolium repens*	5
8.	*Trifolium pratense*	4
9.	*Trifolium hirtum*	1
10.	*Trigonella coerulea*	1
11.	*Vicia sativa*	5
12.	*Vicia narbonense*	3
13.	*Vicia serratifolia*	2
14.	*Vicia villosa*	3
15.	*Vicia lutea*	2
16.	*Vicia peregrina*	1
17.	*Vicia incisa*	1
18.	*Lolium perenne*	6
19.	*Dactylis glomerata*	1
20.	*Bromus inermis*	3
21.	*Agropyron branbzae*	1
22.	*Agropyron pectiniforme*	4
Total		**55**

New approaches in conservation activity

In 2002 a botanical garden was opened at IPGR-Sadovo. It will contain forage plant species from the Bulgarian flora – new sources for plant germplasm use, as well as rare and threatened plants and endemics.

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Grasses, legumes and other forage collections in the Czech Republic

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Collection holders

Three institutions deal with forage collections in the Czech Republic:

1. OSEVA PRO Ltd., Grassland Research Station, Zubří (GRS): grasses including forage, turf and ornamental species
2. Research Institute for Fodder Crops Ltd. Troubsko (RIFC): fodder legumes and other non-leguminous species
3. AGRITEC, Research, Breeding & Services, Ltd., Šumperk: vetches.

Documentation

The collections are documented for passport and characterization/evaluation data in EVIGEZ, the national information system provided by the Research Institute of Crop Production (RICP), Genebank, Prague. The passport data are available on-line (http://www.genbank.vurv.cz/genetic/resources/). The list of genera by accession status and characterization/evaluation data is given in Table 1. Passport data are currently available for 2196 grasses, 1832 legumes and 19 other forages. Descriptive data for 42% of grasses and 56% of legumes have been entered into the information system by the holders of the collections.

The new descriptor list for grasses (Ševčíková *et al*. 2002) and the minimum list of descriptors for vetches implemented recently in the EVIGEZ documentation system (Hýbl and Faberová, unpublished) will enable an increase in the input of descriptive data.

Conservation and utilization

The recent status of conservation and distribution of forage accessions is given in Table 2.

More than 69% of the accessions of seed-propagated grasses and 84% of legumes have been stored in the active or base collection in the Genebank at RICP-Prague and 12 or 2% respectively have been safety-duplicated in the Genebank at the Research Institute of Plant Production (RIPP), Piešťany, Slovakia. About 5% of the collection consists of vegetatively propagated ornamental grasses and grass-like species, maintained in the field collection at GRS-Zubří.

In total 300 accessions of grasses and 215 legumes have been distributed by the Genebank and utilized both in research and plant breeding activities.

Regeneration

The regeneration process has been carried out systematically on grass collections since 1995. A methodology combining spatial isolation between regeneration plots (25 m) and the effect of a surrounding barrier crop of x *Triticosecale* is used for the regeneration of cross-pollinated grasses. A total of 103 accessions including 10 old Czech grass varieties were regenerated in three regeneration cycles at GRS-Zubří. Accessions of fodder legumes are regenerated in technical isolators using an insect pollinator, *Bombus terrestris* at RIFC-Troubsko. Most of the regenerated grasses and legumes represent seed collections in which quantity and quality of seed did not achieve the required standards for entry and subsequent storage in the genebank. Vetches are regenerated in the field plots at AGRITEC-Šumperk. To reduce plant lodging the seed is mixed with a cereal (x *Triticosecale* for winter forms of vetches, spring wheat for spring forms). The seeds are separated after harvest and treated by standard methods prior to storage. Approximately 60% of vetches are stored in the genebank and efforts are being made to complete this activity.

Table 1. Status of the forage collections in the Czech Republic by 31.12.2002 (number of accessions)

Crop/genus	Type of sample					Total passport	Characterization/ evaluation data
	Advanced cultivars	Landraces	Breeders' lines	Wild	Not specified		
Grasses							
Agrostis	44	0	0	13	0	57	16
Alopecurus	7	0	0	6	0	13	9
Anthoxanthum	1	0	0	2	0	3	0
Arrhenatherum	30	0	0	36	0	66	16
Bromus	9	2	0	21	5	37	9
Cynosurus	1	1	0	5	0	7	1
Dactylis	131	2	10	32	1	176	117
Deschampsia	11	0	0	4	0	15	5
Festuca	378	1	12	43	3	437	154
Holcus	3	0	0	4	0	7	0
Lolium	609	4	17	181	2	813	425
Phalaris	12	0	1	1	1	15	7
Phleum	111	0	8	24	0	143	85
Poa	165	0	6	80	0	251	48
Trisetum	4	0	0	9	0	13	4
x Festulolium	13	0	3	0	0	16	12
Others	49	0	26	38	14	127	1
Total grasses	**1578**	**10**	**83**	**499**	**26**	**2196**	**909**
Legumes							
Anthyllis	1	0	0	7	0	8	1
Astragalus	1	0	1	35	0	37	2
Coronilla	2	0	2	23	0	27	2
Galega	2	0	1	0	0	3	3
Genista	0	0	0	18	1	19	0
Chamaecytisus	0	0	0	5	0	5	0
Lathyrus	1	0	1	31	0	33	0
Lotus	26	0	1	27	0	54	25
Medicago	364	6	66	34	3	473	378
Melilotus	13	0	5	46	0	64	13
Onobrychis	7	0	0	11	2	20	9
Ornithopus	1	0	0	1	0	2	1
Trifolium	372	14	69	219	5	679	421
Vicia	271	15	22	80	11	399	168
Others	1	0	1	7	0	9	1
Total legumes	**1062**	**35**	**169**	**544**	**22**	**1832**	**1024**
Other forages							
Malva	0	0	1	1	0	2	0
Phacelia	13	0	0	0	0	13	13
Others	0	1	0	2	1	4	0
Total other forages	**13**	**1**	**1**	**3**	**1**	**19**	**13**

Table 2. Storage of forage accessions by type of collection and distribution by 31.12.2002 (number of accessions)

Crop/genus	Total stored	Active collection	Base collection	Safety-duplication	Distribution
Grasses					
Agrostis	38	37	16	11	13
Alopecurus	11	10	7	5	1
Anthoxanthum	3	3	2	1	0
Arrhenatherum	60	58	20	12	5
Bromus	33	29	16	6	9
Cynosurus	4	4	2	0	1
Dactylis	107	97	40	26	21
Deschampsia	10	9	6	5	2
Festuca	301	294	98	66	47
Holcus	7	5	5	2	1
Lolium	508	493	101	62	101
Phalaris	10	10	2	0	6
Phleum	89	82	28	17	11
Poa	198	180	62	29	66
Trisetum	12	12	8	5	12
x*Festulolium*	11	11	9	2	2
Others	45	36	23	10	2
Total grasses	**1447**	**1370**	**445**	**259**	**300**
Legumes					
Anthyllis	7	1	7	1	0
Astragalus	37	13	24	0	0
Coronilla	27	10	18	1	0
Galega	2	2	0	0	0
Genista	18	5	13	0	0
Chamaecytisus	2	0	2	0	0
Lathyrus	31	5	27	0	2
Lotus	36	6	34	2	6
Medicago	459	391	112	10	89
Melilotus	51	7	46	0	4
Onobrychis	18	6	14	1	10
Ornithopus	2	1	1	0	0
Trifolium	587	367	277	24	94
Vicia	255	195	98	0	10
Others	8	6	2	0	0
Total legumes	**1540**	**1015**	**675**	**39**	**215**
Other forages					
Malva	2	2	1	1	0
Phacelia	13	13	1	0	0
Others	4	3	1	0	0
Total other forages	**19**	**18**	**3**	**1**	**0**

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Forage genetic resources in Estonia

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Advances in plant genetic resources conservation during the last decade in Estonia

In 2002 the Government of Estonia formally approved the national plant genetic resources (PGR) programme. According to the international commitment arising from the ratification of the Convention on Biological Diversity, the main objectives of the PGR programme in Estonia are the preservation and utilization of plant genetic diversity as a local cultural and historical heritage to improve agricultural production and ensure sustainable development.

Until 1993 plant genetic resources for food and agriculture (PGRFA) were preserved at Jõgeva Plant Breeding Institute (PBI) in breeders' working collections. The activities were started with an inventory of those. The most valuable accessions were kept at the N.I. Vavilov All-Russian Scientific Research Institute of Plant Industry (VIR, St. Petersburg) at that time and therefore it was necessary to search for, identify and repatriate the accessions of Estonian origin from VIR and other genebanks. Jõgeva PBI undertook activities for the preservation of plant genetic resources in cooperation with the Nordic Gene Bank (NGB) in 1994. A contract for inventories, preservation and rejuvenation of genetic resources was signed in 1995 between Jõgeva PBI and VIR. To secure *ex situ* material of Estonian origin, Jõgeva PBI and the NGB signed an agreement in 1998, concerning "black box" arrangements whereby NGB agrees to maintain safety-duplicates of the valuable Estonian genotypes.

The genebank of Jõgeva Plant Breeding Institute

The genebank was established in 1999 for long-term *ex situ* conservation of PGRFA. Appropriate procedures for collection, identification, evaluation, characterization, documentation and preservation of accessions were developed.

The genebank participates as a full member in the following international programmes:

1. European Cooperative Programme for Crop Genetic Resources Networks (ECP/GR), coordinated by the International Plant Genetic Resources Institute (since 1999).
2. Collaborative programme between the Nordic Gene Bank and the genebanks of the Baltic States (since 1994). An agreed workplan is carried out in six permanently operating working groups (including forages), whose activities consist of collecting, conservation, characterization and development of computerized databases.
3. EPGRIS project for the creation of central databases covering all European genebanks.

The forage crops collection of Jõgeva PBI was formed on the basis of working collections made by breeders. The old Estonian varieties were missing from their collections. Therefore the computerized databases of VIR, NGB and GCN (Centre for Genetic Resources, Wageningen, the Netherlands) were searched for accessions of Estonian origin. In response to the requests of the genebank of Jõgeva PBI, five lucerne and four red clover varieties of Estonian origin were repatriated from VIR in 2002. Apparently most of the repatriated varieties were missing from the genebank's collection; thus they will be identified. Beside the other crops, the genebank is committed to collect, preserve and evaluate Estonian forage varieties, advanced breeding lines and natural ecotypes adapted to local edaphic and climatic conditions. Advanced cultivars of Estonian origin are fully evaluated and characterized; all relevant information is available. The remaining accessions are partially evaluated. Accessions of advanced cultivars are freely available to all users.

- **Storage conditions**

The genebank of Jõgeva PBI meets the internationally recognized standards (FAO/IPGRI 1994). It has the necessary equipment for preparation of seeds for long-term storage and monitoring of the collection: a seed-processing laboratory, drying room, seed moisture content analyzer, germination cabinet and deep freezers. The equipment for *ex situ* conservation was contributed to Jõgeva PBI within the framework of the Nordic-Baltic project financed by the Nordic Council of Ministries.

All samples are cleaned and graded before drying in a room at a temperature of 15-16°C and air relative humidity of 12-13%. The samples are dried for 4-10 weeks until the required level of seed moisture content (4-6%) is reached. Seeds are packed into laminated aluminium foil bags (4000-10 000 seeds per accession) and distribution bags (100-250 seeds per accession) and stored in deep freezers at -20°C. Distribution bags (5-10 bags per accession) are prepared for germination tests and regeneration as well as for delivery of germplasm to users upon request.

- **Regeneration of the accessions at Jõgeva PBI**

The protocol suggested by Sackville Hamilton *et al.* (1998) has been followed in most respects. Concessions have been made only as regards isolation distances, due to field sizes. Regeneration has been performed either by locating two regeneration plots about 50 m apart or by surrounding the plots with an approximately 25 m-wide stand of winter rye. However, the yearly seeding of a biennial barrier crop could be avoided if the crop were to be replaced by a perennial tall-growing grass, e.g. reed canary grass (*Phalaris arundinacea* L.). This procedure enables the number of accessions of a particular species per regeneration plot to be doubled unless the different species can hybridize. One year after planting, the plants of one accession of a given species are harvested for seed, while those representing another accession of the same species on the same regeneration plot are clipped regularly to prevent heading. The following year the opposite treatments are applied to both accessions. The advantage of this technique is to escape from yearly field preparation and seeding of the barrier crop, and a likely higher seed set of the plants in the second harvest year. The disadvantages are costly grass seed to be used for the isolation and prolonged regeneration cycle of half the accessions.

Status of national forage genetic resources

Nearly the entire national collection of forages is maintained at the genebank of Jõgeva PBI. The total number of 323 accessions consists of 19 grass and 7 forage legume species (Table 1). The samples of 29 cultivars from 15 species, 29 advanced breeding lines from 3 species and the only Estonian landrace of *Medicago falcata* L. are kept in long-term storage; 264 ecotypes representing 15 species of grasses and 5 species of forage legumes are stored at ambient temperature and air humidity. These account for about 80% of all accessions.

Germination capacities of 138 forage accessions deposited between 1989-1998 in short-term storage were determined in 2002. Non-viable accessions (44 altogether) as well as duplicates of lower quality were identified and rejected. Thirty accessions with germination rates higher than 42% were transferred to long-term storage; 64 accessions with germination capacities between 1-41% were passed on to the forage crop department for gradual rejuvenation from 2003 onwards. Germination rates of accessions repatriated from VIR were recorded.

In addition, a minor grass collection in the botanical garden of Tartu University is made up of undeterminable samples of *Alopecurus* sp., *Dactylis* sp., *Festuca ovina* L. and *Trifolium* sp.

Table 1. Forage collection in the genebank of Jõgeva Plant Breeding Institute (number of accessions)

Species	Type of sample				Total
	Advanced cultivars	Breeders' lines	Landraces	Wild or semi-natural ecotypes	
Agrostis tenuis	1			15	16
Alopecurus pratensis	1			2	3
Bromus inermis	1			1	2
Dactylis glomerata	2			40	42
Deschampsia caespitosa				2	2
Deschampsia flexuosa				1	1
Festuca arundinacea				2	2
Festuca ovina				5	5
Festuca pratensis	2			24	26
Festuca rubra	2			29	31
Festulolium		23			23
Koeleria gracilis	1				1
Lolium multiflorum	1	5			6
Lolium perenne	2				2
Lotus corniculatus	1			5	6
Medicago falcata			1	10	11
Medicago media	2			27	29
Phalaris arundinacea	1			7	8
Phleum phleoides				3	3
Phleum pratense	3	1		12	16
Poa compressa				3	3
Poa pratensis	2			37	39
Trifolium hybridum	1				1
Trifolium medium				1	1
Trifolium pratense	4			38	42
Trifolium repens	2				2
Total	**29**	**29**	**1**	**264**	**323**

National collecting activities

The last collecting missions for grasses and forage legumes were performed in Estonian grasslands in 1999 and 1988. As the source material for breeding has run low, three collecting expeditions of forage crop ecotypes were organized in 2002. These were carried out within the framework of the Nordic-Baltic project. Natural and semi-natural grasslands of the western, northern and northeastern parts of the country were visited. Several accessions were collected from the unique plant communities which have formed on the areas of previous Soviet military bases closed to civilians for more than 60 years. Any agricultural intervention was precluded on these habitats at that time. The number of collected accessions per species (designated as wild or semi-natural ecotypes) is indicated in Table 1. The collecting missions resulted in 183 samples of grasses and 81 samples of forage legumes, which will be subjected to either characterization and evaluation or multiplication. Detection of new habitats of *Medicago* sp. in Estonia was another goal of the expeditions. The new accessions will be preserved in long-term storage and examined in the field for the traits that could be introduced to new varieties.

Efforts to improve utilization by breeders

Real value of a genebank does not depend as much on the number of traits characterizing each accession as on the usefulness of the information to germplasm users. The value and utilization efficiency of accessions is determined by the available information on their genetic variability. Thus, a prerequisite of enhanced incorporation of genebank accessions into breeding programmes should be the development of a species-specific optimal set of descriptors, based on the feedback from actual breeders who are looking for a very definite number of useful traits (Loosdrecht *et al*. 1988). Characterization, evaluation and description of the material should focus on desired morphological, biological and agronomical traits that add applicable value to the databases. Special emphasis should be put on collaboration and mutual information exchange with all PGR holders at national and international level.

In order to facilitate access to the information available in the genebank of Jõgeva PBI, the basic structure of the PGR documentation system for the national inventory was developed. All relevant information on *ex situ* collections is documented in a computerized database. Further development of the database is undertaken within the framework of the EPGRIS project.

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Genetic resources of grasses and legumes in France

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Ongoing activities

The main present concern for the French network is to rationalize the sample management of the collection especially regarding the obsolete varieties.

The forage network collection is conserved at the GEVES genebank and represents a small part of the active genebank that holds 28 000 samples from 100 different species. Most of them are marketable varieties or parental lines of commercial varieties and are used in Distinctness, Uniformity and Stability (DUS) studies as part of a reference collection.

Once a bred variety becomes a genetic resource, i.e. is no longer registered on the EC catalogue nor protected, only one sample is necessary. This sample is divided into three sub-samples for active, long-term and safety collections. A procedure was defined to identify the Most Original Sample for each accession of the active collection with emphasis on genetic integrity, seed quality and seed numbers. The focus was to allow future regeneration with no or little genetic drift. At the same time seed samples of small size and low viability were identified and marked for regeneration. In 2002, 22 samples were sown in the nursery and established to be rejuvenated by 8 curators. This programme will go on in 2003 and 2004 and will concern lucerne landraces as well as bred varieties of main fodder crop species.

Future methodology studies

Research on pooling accessions will be carried out in order to optimize the conservation of genes. *In situ* conservation and dynamic management will be used for conservation and breeding purposes.

Documentation

The database of fodder and turf genetic resources will be available on-line (http://www.brg.prd.fr). This database records all passport data of the French national collection.

Probable evolution of the network

In 2001, following an initiative of the French Ministry of Research, the Bureau des Ressources Génétiques (BRG), in cooperation with public organizations, decided to officially support the OCDE Biological Resources Centres (BRCs) Programme. The aim of BRCs is to preserve and develop heritage conservation and to promote access to sustainable, identified and characterized material, through accredited centres.

The fodder and turf species network will turn into a BRC involving all partners of the present network. Quality insurance systems will also be developed.

Fodder crops in the German collection - Developments since 1999

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Introduction

Further to the merger of German genebank collections in order to bring the *ex situ* collections together in one institution, the fodder crops are now held only in the IPK Genebank at Malchow. We are currently recording all forage accessions from Braunschweig.

Table 1 gives an overview of the number of accessions for the eight genera *Trifolium, Medicago, Vicia, Lolium, Festuca, Phleum, Poa* and *Dactylis. Vicia* has the highest number of accessions among forage legumes and *Lolium* among grasses. The German collections include a total of 14 609 accessions.

Table 1. Number of forage accessions in German collections (March 2003)

Genus	Genebank			Total
	Braunschweig	Gatersleben	Malchow	
Dactylis	1101	136	631	1868
Festuca	731	314	1180	2225
Lolium	1546	400	1425	3371
Medicago	508	307	517	1332
Phleum	629	91	419	1139
Poa	155	169	542	866
Trifolium	771	535	380	1686
*Vicia**	220	1902	-	2122
Total	**5661**	**3854**	**5094**	**14609**

* data from last update, 1999

The following part of this report concerns the collection at Malchow. For forage genetic resources from Braunschweig and Gatersleben an inventory will be carried out. All these accessions are stored as active collections. They will be re-arranged in the near future.

The Malchow collection

There were changes in the storage conditions: re-arrangement into active, base and safety-duplicate collections according to FAO/IPGRI Genebank Standards (FAO/IPGRI 1994): the temperature in the active collection is –5°C and in the base collection –20°C. The safety-duplicate collection is stored at –15°C in Gatersleben under "black box" arrangements. There are currently 1938 forage accessions (38% of the whole Malchow collection) stored as safety-duplicates and in the base collection (Table 2). Therefore 62% of the forage collection needs regeneration.

Every year 600-630 accessions of grasses are regenerated according to the guidelines for the regeneration of accessions in seed collections (Sackville Hamilton *et al.* 1998). Owing to the large number of accessions, it is possible to fulfil only the minimum standards for regeneration and to produce unbalanced bulks, both for the active and the base collection.

A total of 4005 accessions are available (Table 2), i.e. 79% of all accessions. The remaining 1089 accessions (21% of the whole collection) require urgent regeneration. Two years will be used for regeneration of the accessions with the highest priority. In order to secure all accessions in the base and safety-duplicate collections, more than 5 years will be necessary for regeneration.

Table 2. Number of forage accessions in active, base and safety-duplicate collections (March 2003, Malchow collections only)

Genus	All accessions	Active collection (*)	Needing urgent regeneration	Base/Safety-duplicate collections	Needing regeneration	
					No.	%
Dactylis	631	426	205	163	468	74
Festuca	1180	875	305	406	774	66
Lolium	1425	1425	0	905	520	37
Medicago	517	386	131	104	413	80
Phleum	419	350	69	259	160	38
Poa	542	380	162	78	464	86
Trifolium	380	163	217	23	357	94
Vicia	-	-	-	-	-	-
Total	**5094**	**4005**	**1089**	**1938**	**3156**	**-**
%	**100**	**79**	**21**	**38**	**-**	**62**

(*) accessions in active collection are available for distribution

Since the last report (1999), 3423 seed samples of forage accessions have been distributed to users (scientists, breeders, NGOs or private users) in Germany and abroad. Seed delivery has tended to decrease in the last two years for various reasons: targeted searches are made in the database via the Internet in order to choose a few accessions; fewer breeding companies and research projects now deal with forage legumes and to some extent also with grasses.

While in the past the maintenance of the collection samples was the main task, more recently evaluation and documentation for users have become increasingly important.

A set of accessions is cultivated as single plants or planted in rows for characterization and evaluation. The focus is on the characterization of new samples and the evaluation of collected Gramineae material, mainly *Poa pratensis* L., for useful traits for breeding.

The passport data of fodder crops (Index Seminum) are available in the on-line database (http://pgrc.ipk-gatersleben.de/malchow/). The database can be searched by species, accession name and country of origin.

The European Central *Poa* Database has been developed in cooperation with the Information Centre for Biological Diversity (IBV) at the ZADI (Zentralstelle für Agrardokumentation und -information = Centre for Agricultural Documentation and Information). It is available on-line (http://www.genres.de/eccdb/poa/) and contains passport data for 3039 accessions of 29 *Poa* species, obtained from 15 institutes in 12 European countries. Searches can be performed on single fields (species, holding institute, country of origin, status of sample and accession name) or by combining multiple criteria (several descriptors) in a user-friendly way. A descriptor list gives additional information about several descriptors, e.g. by entering the institute code, the full organization's name and address for seed requests are provided.

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Status of forage collections in Greece

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The total area cultivated with forage crops in Greece is about 192 430 hectares, i.e. 7.8% of the total arable land of the country. Forage legumes occupy 179 930 ha and forage grasses only 12 500 ha. The most important forage species according to their use and cultivation are as follows (% of total area):

Legumes		
Lucerne	*Medicago sativa* L.	74%
Vetch	*Vicia sativa* L.	15%
Annual clover species	*Trifolium alexandrinum* L. and *Trifolium resupinatum* L.	2.5%
Perennial clover species	*Trifoliun repens* L. and *Trifolium pratense* L.	0.6%
Pea and grasspeas	*Pisum sativum* L. and *Lathyrus cicera* L.	0.3%
Grasses		
Barley	*Hordeum vulgare* L.	5.7%
Rye	*Secale cereale* L.	0.2%
Sorghum	*Sorghum halepense* L.	0.1%
Oat	*Avena sativa* L.	0.4%

Vetch, pea, grasspeas, barley, rye, and oat are cultivated in dry fields in rotation mainly with wheat, while the others are under irrigation in rotation with corn, cotton, sugar beets and other irrigated crops.

In Greece forage collections are conserved mainly in the Greek Gene Bank (GGB) in Thessaloniki and to a smaller extent at the Fodder Crops and Pasture Institute (FCPI) in Larissa and the Cereal Institute (CI) in Thessaloniki. These are all national organizations under the administration of the National Agricultural Research Foundation (N.AG.RE.F), a primary state-funded legal entity of the Ministry of Agriculture.

Fodder Crops and Pasture Institute (FCPI)

FCPI has the national responsibility for the improvement of forage crops, pastures and grain legumes. Breeding forage crops for the creation of new varieties is one of its main tasks. Collection and maintenance of forage genetic material are complementary tasks to support plant breeding projects. Forage collections were established by FCPI starting in 1980 in order to save local populations of the cultivated forages and utilize them in breeding programmes. Forage species had to be collected urgently at the beginning of the 1980s before they were lost due to the release of new varieties to farmers. Priority was given to the regions of the country with developed agriculture where a high level of genetic erosion was expected.

A considerable forage germplasm collection, including about 984 accessions, was collected by FCPI during the period 1980-1985 through collecting missions all over the country. The collected samples were donated to GGB in Thessaloniki (Table 1). Some of these accessions were kept in Larissa under natural room conditions and used for FCPI breeding programmes. Newly bred varieties and cultivars were created from this material and released to farmers, as shown in Table 1.

Since 1985 no serious collecting activities have been carried out by the FCPI owing to lack of resources and also to the fact that forage germplasm material was available from abroad.

The Greek Gene Bank (GGB)

GGB has the national responsibility for plant genetic resource collection, protection and conservation. Medium-term (0° to +5°C) and long-term (-18° to -21°C) storage facilities have a capacity of 80 m³ and can hold approximately 10 000 samples. Forage collections in GGB were also started in 1980. GGB maintains 1150 accessions of forage species in long- and medium-term conditions (Table 1). A database was created to record the germplasm collection in GGB and all the genetic material is documented for passport data and fully computerized in a database using dBase IV. The database currently contains the passport and conservation data. The coverage of evaluation data is still incomplete. There is no safety-duplication at -20°C at present.

Table 1. Forage collections in Greece (number of accessions)

Species	Total	Genebank				
		GGB		FCPI		CI
		Landraces	Wild or semi-natural	Advanced cultivars	Advanced varieties	Advanced cultivars
Agropyron caninum	1	-	1	-	-	-
Agropyron elongatum	4	2	2	-	-	-
Agropyron repens	2	-	2	-	-	-
Agropyrum spp.	16	-	16	-	-	-
Aristella bromoides	4	-	4	-	-	-
Avena sativa	4	-	-	-	-	4
Brachypodium spp.	8	-	8	-	-	-
Briza media	1	-	1	-	-	-
Dactylis glomerata	176	-	150	1	25	-
Ervum ervilia	12	12	-	-	-	-
Festuca arundinacea	64	-	33	1	30	-
Festuca ovina	3	-	2	-	1	-
Festuca spp.	7	-	7	-	-	-
Hedysarum coronarium	2	-	-	1	1	-
Hordeum bulbosum	25	-	25	-	-	-
Hordeum spontaneum	50	-	50	-	-	-
Hordeum vulgare	43	26	1	-	-	16
Lathyrus cicera	20	9	-	4	7	-
Lathyrus ochrus	15	15	-	-	-	-
Lathyrus sativus	21	15	-	1	5	-
Lathyrus spp.	8	5	3	-	-	-
Lolium perenne	82	-	57	1	24	-
Lolium spp.	16	-	16	-	-	-
Lolium multiflorum	1	-	1	-	-	-
Lotus corniculatus	21	2	19	-	-	-
Lupinus albus	3	3	-	-	-	-
Lupinus angustifolius	70	-	70	-	-	-
Lupinus luteus	1	-	1	-	-	-
Lupinus spp.	2	-	2	-	-	-
Medicago arborea	25	-	4	1	20	-
Medicago coronata	1	-	1	-	-	-
Medicago falcata	5	-	5	-	-	-
Medicago lupulina	6	-	6	-	-	-
Medicago minima	1	-	1	-	-	-
Medicago orbicularis	30	-	30	-	-	-
Medicago sativa	50	10	7	8	25	-
Medicago spp. Annuals	54	-	54	-	-	-
Melilotus alba	4	-	4	-	-	-
Melilotus spp.	5	-	5	-	-	-
Onobrychis spp.	3	-	3	-	-	-
Onobrychis viciifolia	12	-	-	11	1	-

Table 1 (cont.). Forage collections in Greece (number of accessions)

Species	Total	Genebank				
		GGB		FCPI		CI
		Landraces	Wild or semi-natural	Advanced cultivars	Advanced varieties	Advanced cultivars
Oryzopsis miliiacea	13	-	13	-	-	-
Oryzopsis spp.	2	-	2	-	-	-
Phacelia tanacetifolia	1	-	-	1	-	-
Phalaris tuberosa	8	-	8	-	-	-
Phleum pratense	2	-	2	-	-	-
Phleum spp.	11	-	11	-	-	-
Pisum sativum	36	21	3	5	7	-
Poterium sanguisorba	13	-	12	1	-	-
Poterium spp.	2	-	2	-	-	-
Sorghum bicolor	2	-	2	-	-	-
Sorghum sudanense	5	-	-	1	4	-
Trifolium alexandrinum	60	-	55	3	2	-
Trifolium angustifolium	2	-	2	-	-	-
Trifolium arvense	9	-	9	-	-	-
Trifolium aureum	2	-	2	-	-	-
Trifolium campestre	13	-	13	-	-	-
Trifolium cherleri	13	-	13	-	-	-
Trifolium dubium	1	-	1	-	-	-
Trifolium echinatum	1	-	1	-	-	-
Trifolium fragiferum	1	-	1	-	-	-
Trifolium hirtum	21	2	19	-	-	-
Trifolium hybridum	1	-	1	-	-	-
Trifolium incarnatum	5	-	5	-	-	-
Trifolium obscurum	5	-	5	-	-	-
Trifolium pratense	56	7	47	2	-	-
Trifolium repens	76	3	71	2	-	-
Trifolium resupinatum	12	-	9	3	-	-
Trifolium scabrum	5	-	5	-	-	-
Trifolium spp.	50	-	50	-	-	-
Trifolium spumosum	3	-	3	-	-	-
Trifolium stellatum	4	-	4	-	-	-
Trifolium striatum	2	-	2	-	-	-
Trifolium subterraneum	1	-	1	-	-	-
Trifolium tomentosum	2	-	2	-	-	-
Trifolium vesiculosum	5	-	5	-	-	-
Trigonella foenum-graecum	2	1	-	1	-	-
Trigonella spp.	2	-	2	-	-	-
Vicia sativa	64	40	2	7	15	-
Vicia spp.	1-	-	1	-	-	-
Total	**1392**	**173**	**977**	**55**	**167**	**20**

GGB = Greek Gene Bank, Thessaloniki
FCPI = Fodder Crops Pasture Institute, Larissa
CI = Cereal Institute, Thessaloniki

Future activities

Collecting activities of forages in Greece are not considered to be complete for some regions, especially the islands, and additional material can be expected.

Other activities planned in the future include:

- completion of the formation of the duplicate set of the collection for long-term storage,
- evaluation of all accessions, and
- creation of evaluation databases.

Status of the national forage crop collections in Hungary

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Status at national level

The institutions listed in Table 1 deal with forage genetic collections in Hungary. Among these, the Institute for Agrobotany in Tápiószele (TABI) belongs directly to the Ministry of Agriculture and Rural Development. For the other institutes (mostly breeding organizations) the Ministry of Agriculture also provides funds for the conservation, regeneration and characterization of those genebank accessions that are included in the National Genetic Resources Database (NGRD). A National Base Collection (NBC) has been created for the safety-duplication of seed-propagated accessions that do not belong to the TABI collections.

Table 1. Hungarian institutions dealing with forage genetic resources and/or related activities

Institution	Crops	No. of accessions
Tápiószele Institute for Agrobotany, Ministry of Agriculture and Rural Development	Grasses = 222 species of 58 genera Legumes = 96 species of 15 genera	5212
Nyíregyháza Research Centre, University of Debrecen	*Lotus corniculatus* L.	5
	Medicago sativa L.	50
	Vicia sativa L.	59
	Vicia villosa Roth	96
Karcag, Research Centre, University of Debrecen	Grass species	86
Szarvas Agricultural Research and Development Institute P.U.C.	*Medicago sativa* L.	99
	Trifolium alexandrinum L.	30
	Trifolium incarnatum L.	35
	Trifolium repens L.	84
	Trifolium repens L. f. *giganteum*	50
	Trifolium resupinatum L.	26
Mosonmagyaróvár Faculty of Agricultural Sciences, University of West Hungary	*Arrhenatherum elatius* (L.) Beauv. ex J. et C. Presl	1
	Bromus pannonicus Kumm. et Sendtn.	2
	Festuca pratensis Huds.	1
	Festuca rubra L.	2
	Medicago sativa L.	22
	Phleum pratense L.	2
	Poa trivialis L.	1
	Trifolium pratense L.	24
	Trigonella foenum-graecum L.	10
Kompolt R. Fleischmann Agricultural Research Institute, St Stephan University	*Medicago sativa* L.	579
	Onobrychis viciifolia Scop.	34
Total		**6510**

Status in TABI

TABI is responsible for the development and maintenance of the Hungarian field crop (including forages) and vegetable genetic resources collection, and, as well as other national and international genebank duties, for the operation of NGRD and NBC.

- **Storage**

Today more than 5000 accessions of 318 grass and legume forage species (Table 2) are stored in the active and base collection chambers of the TABI. The storage temperature is 0°C in the active collection, and -20°C in the base collection chambers, where 1150 accessions are kept.

Table 2. The TABI forage collections according to their degree of originality

Genus	MOS 1	MOS 4	MOS 5	No. of accessions
Gramineae				
Festuca	339	326	19	684
Lolium	90	268	4	362
Dactylis	155	100	11	266
Bromus	77	125	11	213
Poa	129	55	4	188
Agropyron	3	85		88
Aegilops	22	52		74
Phleum	12	53	5	70
Alopecurus	36	16	1	53
Arrhenatherum	26	20	7	53
Elymus	14	29	7	50
Agrostis	17	19	1	37
Puccinellia	25	8		33
Hordeum	3	24		27
Phalaris	10	12	2	24
Holcus	15	5	1	21
Koeleria	13	6		19
Briza	8	6	1	15
Melica	11	4		15
Secale	12	2		14
Brachypodium	11	2		13
Deschampsia	11			11
Eragrostis		5	1	6
Stipa		6		6
Beckmannia	4	1		5
Coix		4		4
Hordelymus		1	3	4
Lagurus		4		4
Pennisetum		3	1	4
Roegneria		1	2	3
28 other genera	7	33	1	41
Leguminosae				
Medicago	273	527	5	805
Trifolium	363	420	15	798
Vicia	161	325	16	502
Lathyrus	81	265	2	348
Lotus	91	79	5	175
Onobrychis	2	93	5	100
Trigonella	1	23		24
Anthyllis	1	12		13
Desmodium		13		13
Tetragonolobus	2	8		10
Melilotus	3	5	1	9
Astragalus		5		5
Coronilla		1		1
Glyceria	1			1
Ornithopus		1		1

MOS 1: likely an Original sample (INSTCODE= COLLCODE or BREDCODE)
MOS 4: 2 or more donations away from MOS (only DONORCODE, no COLLCODE or BREDCODE)
MOS 5: unknown (no DONORCODE, no COLLCODE, no BREDCODE)

- **Documentation**

Management data (passport, characterization and genebank management data) are fully computerized. The degree of completeness of TABI forage databases is shown in Table 3.

Table 3. Data completeness in the databases of the TABI forage crop collections

FAO/MCP Descriptor	Data	Percentage
INSTCODE	5212	100
ACCENUMB	5212	100
COLLNUMB	1467	28
COLLCODE	2022	39
GENUS	5212	100
SPECIES	5212	100
SPAUTHOR	5162	99
SUBTAXA	219	4
SUBTAUTHOR	84	2
CROPNAME	4283	82
ACCENAME	4005	77
ACQDATE	4896	94
ORIGCTY	2972	57
COLLSITE	2075	40
LATITUDE	1920	37
LONGITUDE	1920	37
ELEVATION	0	0
COLLDATE	1911	37
BREDCODE	12	-
SAMPSTAT	4055	78
ANCEST	0	0
COLLSRC	5212	100
DONORCODE	3093	59
OTHERNUMB	5212	100
STORAGE	5212	100

- **Multiplication and regeneration**

The overall volume of field multiplication and regeneration varies from year to year according to changes resulting from introduction or collecting and to the regeneration needs of the genebank in the given period.

For the isolated multiplication of the mainly outcrossing species, 20 to 25 space-isolated nurseries are used on TABI land; there is also a countrywide "backyard garden multiplication system".

- **Characterization and evaluation**

TABI has a universal descriptor list compiled according to the international standards for the grass species. The majority of the grass accessions in the genebank are characterized by 20-25 descriptors. There are also descriptor lists for the most important genera of legumes, but their use is restricted, as is also the evaluation activity in general, because of budgetary and other constraints.

Status of the forage species collection at the Israeli Gene Bank

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The emphasis of the Israeli Gene Bank for Agricultural Crops (IGB) is on collection, regeneration and characterization of local landraces and wild relatives of numerous plants' germplasm including forage species. Part of the forage species collection held by the IGB comprises representatives of barley (*Hordeum* spp.), lupin (*Lupinus* spp.), ryegrass (*Lolium* spp.), lucerne (*Medicago* spp.), clover (*Trifolium* spp.) and vetch (*Vicia* spp.). This collection includes more than 5000 accessions from Israel and worldwide (Table 1). The seeds of each accession are stored, and passport data and important plant characteristics are documented in the IGB database. Most forage genera collected by the IGB are represented by more than one species originating from both Israel and worldwide.

Table 1. Forage collections held in the Israeli Gene Bank (IGB) (number of accessions)

Genus and species	Origin	
	Israel	Worldwide
Agropyrum		
elongatum	0	35
Beta		
vulgaris	66	30
Dactylis		
spp.	1	3
Festuca		
spp.	0	1
Hedysarum		
spp.	1	11
Hordeum		
bulbosum	29	0
spontaneum	2210	0
vulgare	0	48
Lathyrus		
spp.	16	5
Lolium		
multiflorum	38	20
perenne	0	2
rigidum	38	2
Lupinus		
albus	13	23
angustifolius	16	20
luteus	3	38
palaestinus	3	0
pilosus	0	7
varius	2	0
other spp.	3	8
Medicago		
polymorpha	59	30
sativa	8	37
trunculata	5	41
varia	0	40
Onobrychis		
viciifolia	0	69
other spp.	0	3

Table 1 (cont.) Forage collections held in the Israeli Gene Bank (IGB) (number of accessions)

Genus and species	Origin	
	Israel	Worldwide
Phalaris		
tuberosa	14	0
paradox	2	0
brachystachys	1	0
arundinacea	0	1
Pisum		
arvense	0	10
fulvum	9	2
sativum	6	265
Sorghum		
bicolour	6	58
dochana	5	0
halpense	5	0
vulgare	6	12
Tetragonolobus		
maritimus	0	5
palaestinus	3	0
purpureus	7	29
Trifolium		
alexandrinum	47	215
alexandrinum x *berytheum*	2	0
arvense	0	2
berytheum	10	0
campestre	19	4
cherleri	1	19
clypeatum	20	9
constantinopolitanum	1	0
dichroanthum	5	0
echinatum	8	19
glanduliferum	3	20
pratense	0	6
purpureum	35	4
repens	2	17
resupinatum	2	9
salmoneum	1	1
spumosum	4	6
subterraneum	80	193
Trigonella		
berythea	2	0
foenum-graecum	19	0
Urospermum		
picroides	24	0
Vicia		
ervilia	47	8
faba	58	248
sativa	42	189
other *spp.*	121	200
Vigna		
luteola	8	0
Total	**3136**	**2024**

Israel's rich flora is endangered owing to the increases in urbanization, road construction and modern farm practices. Therefore, another important aim of the IGB is to rescue the endangered wild plant species and relatives of the domesticated plants that still exist in their natural habitat. The rescue collection is carried out collaboratively between the Hebrew University of Jerusalem, Rotem–Israel Plant Information Center, Tel Aviv University and the IGB. During the last six years more than 2000 accessions have been collected, representing many plant species including several hundreds of forage accessions (Table 2, Fig. 1). The rescue collection will continue in the future until all high-risk regions are covered.

Table 2. Rescue collections of forage species in Israel

Table 2. Rescue collections of forage species in Israel **Genus and species**	**No. of accessions**
Beta	
vulgaris	11
Hordeum	
bulbosum	28
spontaneum	51
Lathyrus	
aphaca	8
Lolium	
rigidum	37
Lupinus	
angustifolius	15
luteus	2
Medicago	
polymorpha	36
sativa	3
Phalaris	
tuberosa	14
Pisum	
fulvum	
syriacum	
Tetragonolobus	
palaestinus	8
purpureus	1
Trifolium	
berytheum	1
clypeatum	18
dichroanthum	4
palaestinum	22
purpureum	28
Trigonella	
berythea	2
Urospermum	
picroides	22
Vicia	
narbonensis	6
sativa	8
Vigna	
luteola	8
Total	**343**

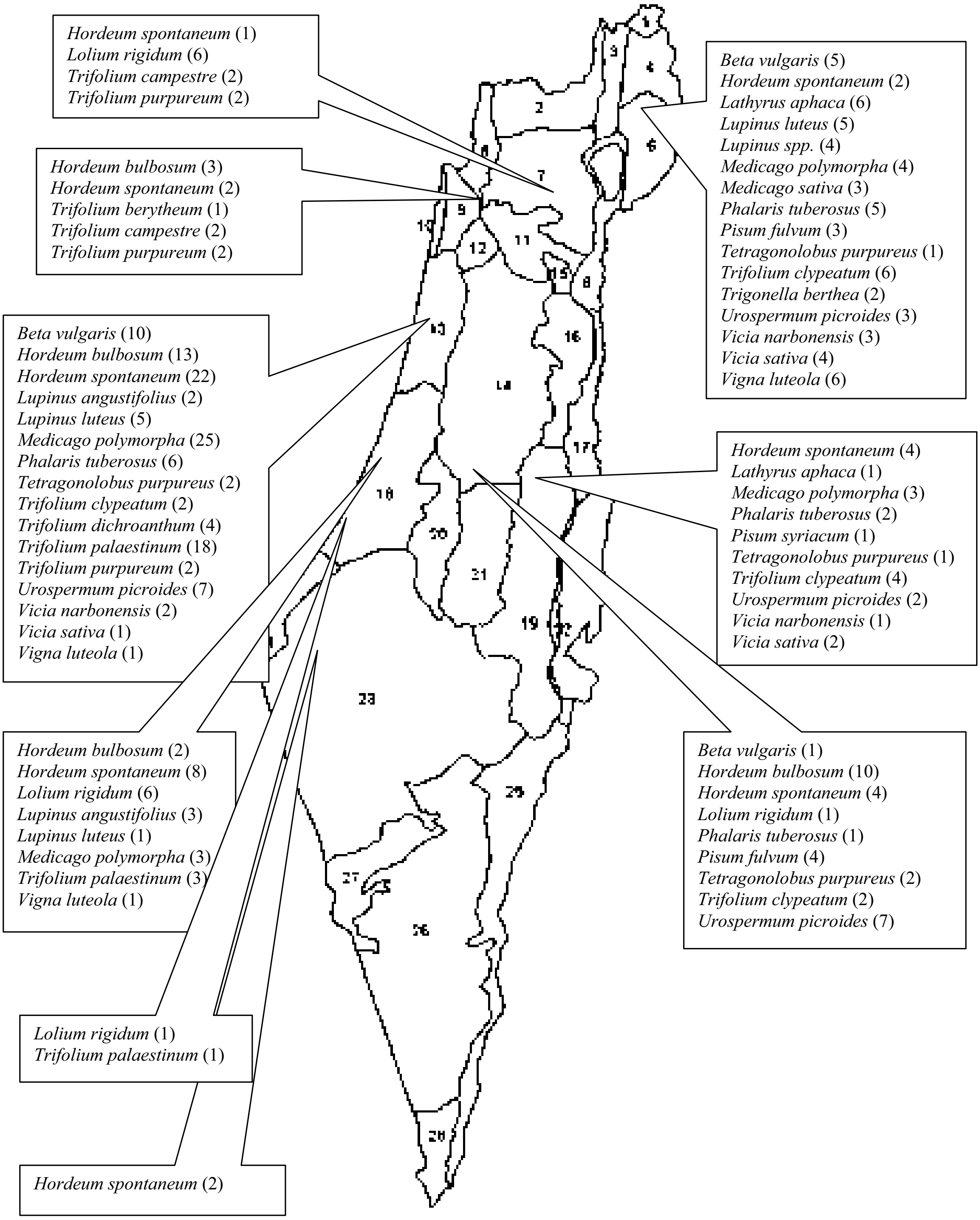

Fig. 1. Forage rescue collections from different regions in Israel carried by the Hebrew University of Jerusalem, Rotem, Tel Aviv University and the Israeli Gene Bank (IGB).

Update on the Italian forage collections

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Accessions are conserved under medium-term storage conditions (4°C, sealed packets). The collection contains a total of 2095 accessions, shown in Table 1.

Table 1. Forage collection held at ISCF

Species	No. of accessions
Lolium multiflorum italicum	3
Dactylis glomerata	5
Festuca arundinacea	113
Agrostis palustri	8
Cynodon dactylon	13
Festuca rubra	8
Lolium perenne	31
Poa pratensis	66
Medicago sativa	97
Trifolium repens	65
T. subterraneum brachycalycinum	717
T. subterraneum subterraneum	934
T. subterraneum yanninicum	35
Total	**2095**

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The only changes in the number of accessions held since November 1999 concern *Onobrychis viciifolia* Scop. (20 new accessions were collected in 1999). The species now totals 181 accessions, including wild populations and landraces. Maintenance conditions are long-term storage (vacuum-sealed aluminium packets, moisture content 5-8%, storage temperature 18°C). Availability of germplasm depends on the quantity of seed stored. We have facilities to multiply the seed upon request. Documentation is computerized; all accessions collected by the Department have minimum passport data.

Note: the working collection is maintained without specific funding. The number of accessions is subject to change.

Genetic resources of forage grasses and legumes in Latvia: status and collecting activities

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Genetic resources of forage grasses and legumes in Latvia consist of registered varieties, valuable breeding material and wild ecotypes.

The majority of the accessions are of Latvian origin. They represent both old and modern varieties as well as advanced breeding lines.

In 1994 a computerized information centre for plant genetic resources (PGR) was established in the Institute of Biology of the Latvian Academy of Science. Since then it has been developed and expanded.

The 32 accessions of forage grasses and legumes from the base collection have been placed in the Latvian Gene Bank for long-term storage (Table 1).

Table 1. List of forage varieties in the Latvian Gene Bank

No.	Species	Variety
1.	*Agrostis gigantea (*syn. *A. alba)*	Priekuļu 15
2.	*Alopecurus pratensis*	Priekuļu 40
3.	*Arrenatherum elatius*	Priekuļu 20
4.	*Dactylis glomerata*	Priekuļu 30
5.	*Festuca pratensis*	Patra
6.	*Festuca pratensis*	Priekuļu 519
7.	*Festuca pratensis*	Rita
8.	*Festuca rubra*	Priekuļu 45
9.	*Lolium multiflorum* var. *westervoldicum*	Uva
10.	*Lolium perenne*	Priekuļu 59
11.	*Lolium perenne*	Spīdola
12.	*Lolium perenne x Festuca pratensis*	Ape
13.	*Medicago varia*	Skrīveru
14.	*Phleum pratense*	Priekuļu
15.	*Phleum pratense*	Priekuļu 2
16.	*Phleum pratense*	Agris
17.	*Poa palustris*	Priekuļu
18.	*Poa pratensis*	Gatve
19.	*Poa pratensis*	Priekuļu 129
20.	*Trifolium hybridum*	Menta
21.	*Trifolium hybridum*	Priekuļu tetraploīds
22.	*Trifolium hybridum*	Priekuļu 26
23.	*Trifolium hybridum*	SK-74
24.	*Trifolium pratense*	Agra
25.	*Trifolium pratense*	Dīvaja
26.	*Trifolium pratense*	Dižstende
27.	*Trifolium pratense*	Priekuļu 66
28.	*Trifolium pratense*	Skrīveru agrais
29.	*Trifolium pratense*	Skrīveru tetra
30.	*Trifolium pratense*	Stendes agrais
31.	*Trifolium pratense*	Stendes vēlais II
32.	*Trifolium repens* var. *giganteum*	Priekuļu 61

In 2005 the Latvian PGR database contains data on 371 accessions of forage grasses and legumes (Table 2).

Table 2. Accessions of forage grasses and legumes in the Latvian PGR database (2005)

Species	No. of accessions
Galega orientalis	14
Lathyrus pratensis	1
Lathyrus sativus	1
Lupinus angustifolius	7
Lupinus polyphyllus	1
Lupinus venustus	1
Medicago falcata	3
Medicago sativa	8
Medicago varia	39
Melilotus albus	2
Trifolium fragiferum	1
Trifolium hybridum	31
Trifolium pratense	154
Trifolium repens	2
Vicia spinosa	3
Vicia villosa	5
Festulolium	2
Agrostis gigantea	1
Alopecurus pratensis	2
Arrhenatherum elatius	1
Bromus inermis	1
Cynosurus cristatus	1
Dactylis glomerata	4
Festuca arundinacea	14
Festuca gigantea	4
Festuca pratensis	28
Festuca rubra	2
Lolium perenne	3
Lolium multiflorum	1
Lolium x Festuca	5
Phalaris arundinacea	1
Phleum phleoides	1
Phleum pratense	7
Poa palustris	3
Poa pratensis	17
Total	**371**

In 2004, the following Latvian accessions of forage grasses and legumes were repatriated from the genebank of the Vavilov Institute (VIR, St. Petersburg, Russian Federation): *Lathyrus sativa* (1 accession); *Lupinus angustifolius* (7); *Medicago sativa* (8); *Lupinus venustus* (1); *Trifolium hybridum* (15); *Trifolium pratense* (24); and *Vicia villosa* (4).

New accessions such as hybrids, wild ecotypes and others are being placed in the Gene Bank every year.

During the last 5 years plant breeders from the Skriveri Research Institute of Agriculture and researchers from the Latvia University of Agriculture (LUA) have jointly organized scientific expeditions to collect accessions of local origin of legume and grass species found in various districts of Latvia, for their conservation, evaluation, description and storage in the Latvian Gene Bank. The main collecting sites are river banks, flood-meadows, old and abandoned meadows and pastures, uncultivated fields, old set-aside land and gravel pits. Since 2000 ten expeditions have been organized, including two at international level, in cooperation with Estonian and Lithuanian scientists. One of these took place in Saarema island in Estonia, where *Medicago varia* and *Medicago falcata* accessions were collected.

Every year after these expeditions the accessions collected under the natural conditions of the Baltic States have been added to the Gene Bank.

Genetic resources of forage grasses and legumes in Lithuania: status and collecting activities

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Status of forage genetic resources in Lithuania

Institution dealing with forage genetic resources
Lithuanian Institute of Agriculture, National Centre of Plant Genetic Resources, Akademija, Kedainiai.

Staff/responsibility
Ausra Gineitaitė - Head of the National Centre of Plant Genetic Resources.
Juozas Labokas - PGR national research project coordinator.
Nijolė Lemežienė - forage grass breeder, curator.

Forage grass and legume breeders are responsible for collecting, multiplication, documentation, evaluation and regeneration.

Storage conditions: long-term storage.

Duplication sites: 28 accessions have been safety-duplicated at the Nordic Gene Bank (NGB), Sweden.

Availability: about 99%.

Documentation status
Passport data are documented according to the Forage Passport Descriptors (Appendix I *in* Maggioni *et al.* 1998). Recently 315 additional accessions of *Dactylis, Lolium, Festuca, Trifolium, Phleum* and *Poa* species were updated. Data were sent to the European Central Forage Databases. The database management system is Microsoft Access®.

Genetic collection of forage grasses and legumes
Since the last meeting of the ECP/GR Working Group on Forages in 1999, the national Lithuanian forage collection has increased by 341 accessions. The collection now consists of 784 accessions of 24 species of forage grasses and legumes, including 465 wild or semi-natural ecotypes, 265 breeders' lines and 54 advanced cultivars.

Evaluation status
Species evaluated are those involved in the forages and turf grasses breeding programmes: *Agrostis tenuis* Sibth., *Dactylis glomerata* L., *Dactylis polygama* Horvat., *Festuca rubra* L., *Festuca ovina* L., *Festuca pratensis* Huds., *Festulolium* hybrids, *Koeleria glauca* Schkuhr., *Lolium perenne* L., *Medicago sativa* L., *Phleum pratense* L., *Poa pratensis* L., *Poa compressa* L., *Poa nemoralis* L., *Trifolium repens* L., *Trifolium pratense* L., etc.

Regeneration and multiplication

Forage grasses are regenerated or multiplied in field plots, isolated by winter rye. Forage legumes are isolated in special netted isolation cages supplied with one small beehive containing Carniolan bees, or by using spatial isolation.

Collecting activities of forage genetic resources in Lithuania, 2000-2002

Evaluation of Lithuanian genetic resources of forages has shown that there are real grounds to search out genetic resources in Lithuania and in other countries with similar climatic conditions since the wild ecotypes collected in various districts have distinguished themselves by a great diversity of morphological characters and agronomic properties within the genera (Kanapeckas *et al.* 2002; Lemežienė and Kanapeckas 2002; Tarakanovas 2002).

The national programme "Genetic resources of cultivated plants" was implemented during the period 1998-2002. One of the most important tasks in this programme was to collect wild or semi-natural ecotypes of forages. During 2000-2002 three expeditions were organized to natural habitats at various geographical locations within seven Lithuanian administrative regions and at one in the Kaliningrad region of Russia (Tables 1 and 2).

The work was started by making inventories of natural meadows in each region.

A total of 564 seed accessions of forage grasses and legumes were collected in 117 natural habitats. Altogether 39 species of grasses and 20 species of legumes were collected. More than 78% accessions belong to the species included in the breeding programmes.

Wild ecotypes of forage grasses and legumes were collected in different geographic locations and districts in different soil conditions:

- on the slopes of hills, in river valleys, and in natural meadows at the edge of forests that have not been touched by any human activity since immemorial times;
- in formerly cultivated meadows and pastures where human interference has been limited for the last 25-30 years.

Accessions were collected as seeds. Passport data were prepared for each accession. The location of the habitat was precisely identified using a Magellan GPS system.

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Table 1. Number of forage and turf grass accessions collected in Lithuania (2000, 2001) and Russia (Kaliningrad region, 2002)

Species	Year			Total
	2000	2001	2002	
Forage grasses				
Agrostis gigantea Roth.			1	1
Agrostis syreistschikovii Smirn.			1	1
Agrostis stolonifera L.			1	1
Agrostis tenuis Sibth.	14		7	21
Alopecurus pratensis L.	1		1	2
Arrhenatherum elatius L.	1			2
Bromus inermis Leysser	3	1	8	12
Calamagrostis lanceolata Roth.		1		1
Cynosurus cristatus L.	2		2	4
Corynephorus canescens L.	1			1
Dactylis glomerata L.	20	19	39	78
Dactylis polygama Horvat.		2	4	6
Deschampsia caespitosa L.			1	1
Festuca arundinacea Schreb.	1	2	16	19
Festuca gigantea L.		2		2
Festuca pratensis Huds.	20	21	31	72
Festuca rubra L.	17	7	13	37
Festuca sabulosa H. Liundb.			2	2
Festuca ovina L.	4	5	10	19
Festuca trachyphylla Hackd.			3	3
Glyceria aquatica Wahlb.	1			1
Helictotrichon pratense L.		3		3
Holcus lanatus L.	1			1
Koeleria glauca Schkuhr.			1	1
Lolium perenne L.	17	12	26	55
Luzula campestris L.		1		1
Nardus stricta L.	1			1
Phalaris arundinacea L.	1		7	8
Phleum pratense L.	16	9	24	49
Phleum phleoides L.		2	1	3
Poa angustifolia L.	1	1	1	3
Poa pratensis L.	16	18	17	51
Poa nemoralis L.	1	1	2	4
Poa compressa L.		1	3	4
Poa palustris L.	1		3	4
Sieglingia decumbens L.	1	1		2
Total	**141**	**109**	**193**	**443**

Table 2. Number of forage legume accessions collected in Lithuania (2000, 2001) and Russia (Kaliningrad region, 2002)

Species	Year			Total
	2000	2001	2002	
Forage legumes				
Anthyllis vulneraria L.	2			2
Astragalus glycyphyllus L.	2			2
Lotus corniculatus L.			1	1
Lotus uliginosus Schkuhr.	1			1
Melilotus albus Med.			2	2
Melilotus officinalis L.			2	2
Medicago sativa L.			15	15
Medicago falcata L.		1		1
Medicago lupulina L.	1		2	3
Onobrychis viciaefolia Scop.		1		1
Trifolium arvense L.	1			1
Trifolium aureum Poll.	1			1
Trifolium campestre Schreb.	1			1
Trifolium fragiferum L.	1			1
Trifolium hybridum L.	3			3
Trifolium medium Grufb.	1	2	4	7
Trifolium dubium Sibth.			1	1
Trifolium montanum L.		2		2
Trifolium pratense L.	10	9	27	46
Trifolium repens L.	12	9	8	26
Total	**36**	**24**	**61**	**121**

Status of forage collections in Macedonia FYR

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Introduction

The Republic of Macedonia (FYR) is a southeast European state situated in the central part of the Balkan Peninsula. It is located between 40°51′16′′ and 42°22′21′′ North and between 20°27′32′′ and 23°02′12′′ East and covers an area of 25 713 km^2. Macedonia is predominantly a mountainous country, cut by large and small valleys, gorges, plateaus and highlands. Altitude ranges from about 60 m at the lowest point to 2764 m at the highest point. It has about 15 mountain ranges higher than 2000 m, but only the Shara Mountain has more than 20 peaks higher than 2500 m. The territory of Macedonia includes 25% pastureland; 25% arable land, meadows, vineyards and orchards; 8% barren land; 37% gazetted (legally established) forestland; 2% lakes; and 3% urban and industrial land.

According to analyses of biodiversity, Macedonia is at the top of the list of states called ″European hotspots″. The great biodiversity of Macedonia is a result of its long historical development. The differentiation of indigenous species, as well as the invasion of plant migrants from other areas, played a significant role in its genesis. Naturally, not all species once living in the area have survived. Many species disappeared due to unfavourable conditions. Therefore, the current biodiversity has to be studied not only from a genetic but also from a historical point of view.

Due to the diversified natural conditions (relief, climate, hydrography and soil), as well as human influence, Macedonia is one of the few countries in Europe to have such a rich diversity of habitats. Numerous water ecosystems (lakes, rivers and streams) support rich shore-line vegetation, and deep gorges are sources of considerable endemism and relicts. At lower altitudes, large agricultural areas, meadows, pastures, and even steppe-like desert terrain are found.

Four phytogeographic regions can be distinguished in Macedonia, characterized by their different climates and altitudes and consequently by their different flora and fauna:

1. sub-Mediterranean region (40% of the Macedonian territory)
2. sub-continental region (37%)
3. sub-humid region (22%)
4. sub-alpine region (1%).

Status of plant genetic resources activities

At the crossroads between Europe and Asia, Macedonia became the home of diverse types of plants in past centuries. Many international and bilateral projects were undertaken in the 1960s and 1970s, most of them with the United States, to collect seeds and plant materials from cereals, forages, fruit and vegetables. These accessions were stored overseas due to the absence of storage facilities at that time in Macedonia. Hundreds of genetic resource accessions (originating in different regions of Macedonia) have been collected, evaluated according to international standards and conserved in genebanks of research institutions in the United States and the former Soviet Union.

The genebank activities for the Republic of Macedonia were initially organized by the headquarters in Belgrade, Former Yugoslavia. For the last ten years, i.e. since the independence of the Republic of Macedonia, the research community has been making efforts to reorganize these activities. For various reasons, mainly inconsistency of governmental policy and instability in the region, very little has actually been achieved to conserve plant genetic resources for food and agriculture. Some activities were undertaken in several small genebank projects for various crops, financed by the Ministry of Agriculture and Ministry of Science. The records were not computerized and the seeds were not stored in cold conditions.

Starting from 1996, genebank activities have been carried out through a Programme for Agricultural Development Support, by the Ministry of Agriculture, Forestry and Water Economy. The national funding varies every year between 5000 and 50 000 US$, depending on the total budget of the Ministry and the current policy. In 1998 the funding was interrupted for one year, due to the Kosovo crisis.

Currently, four institutions work to maintain agrobiodiversity in Macedonia:

- the Institute of Agriculture in Skopje,
- the Institute for Southern Crops in Strumica,
- the Institute for Tobacco in Prilep, and
- the Institute for Rice in Kocani.

After discussions with the National Seed Storage Laboratory in Fort Collins and the Macedonian Institute, the United States Department of Agriculture (USDA) agreed to assist in the repatriation of Macedonian germplasm conserved in the USDA system. The repatriation will probably take several years, depending on the potential of the participating institutes to reproduce the material.

The forage crops collection is maintained only by the Institute of Agriculture in Skopje, Department for Forage Crops, in collaboration with the Faculty of Agriculture, Department for Field Crops and Tobacco.

The Institute has experimental fields (approximately 300 ha) and a cold chamber (2–6°C) for medium-term storage of the active collections. Two additional rooms are under construction: a preparatory room for seed packaging and labelling and a cold chamber (-20°C) for long-term storage of the base collection.

Owing to lack of funds and staffing, progress on forage germplasm collection activities has not been up to our expectations. In general the maintenance activities for forage plants only form part of the existing forage breeding programme. This means that the collections are characterized for the purposes of the programme only. IPGRI descriptors are not being used for the evaluation. Most of the accessions are commercial varieties or lines and cultivars introduced for breeding purposes.

The research work of the Institute of Agriculture in Skopje is especially successful with the crops of highest economic importance (mainly wheat and some vegetable crops). According to the figures of the Federal Commission for the registration of new cultivars, 258 forage cultivars with valuable properties have been approved, distinguishable in three groups:

1. newly created Macedonian forage plants,
2. foreign forage plants, and
3. domestic and domesticated foreign forage plants.

Among these species 103 are perennial grasses, 31 annual legumes, 94 perennial legumes and 30 other forage cultivars.

Starting in 1996, a 3-year genebank project was financed by the Agricultural Development Support Programme. Conservation activities were undertaken for forages, among other crops. The present size of the collection of genetic resources of forage crops, legumes and perennial grasses is shown in Table 1.

The seeds are dried to a moisture content of approximately 5%, then packed in laminated aluminium foil bags and stored under medium-term storage conditions (4°C).

In order to make a proper and unified evaluation of the existing working collection, IPGRI forage descriptors were obtained and translated.

The conservation of perennial grasses and legumes in the Republic of Macedonia is still in its initial phase: the definition of priorities to determine which breeding material and wild species should be collected and stored in the genebank.

Table 1. Forage collection held at the Institute of Agriculture in Skopje

Species	No. of accessions
*Festuca arundinacea**	2
*Medicago sativa**	1
*Onobrychis sativa**	1
*Vicia sativa**	3
Festuca spp.**	32
Total	**39**

* cultivated species; ** wild species

Other genebank activities

As a part of the genebank project activities, contact with the USDA National Seed Storage Laboratory in Fort Collins has been established. USDA maintains a collection of *Medicago* and *Trifolium* originating from different regions in Macedonia (Table 2). Most of them were collected by the Institute of Agriculture in Skopje during 1956-1977.

Table 2. Forage material collected by USDA in the period 1956-1977

Species	No. of accessions
Medicago sativa	4
Trifolium spp.	18
Total	**22**

Taking into consideration the importance of plant biodiversity protection, the Institute of Agriculture submitted a draft National Programme to both the Ministry of Agriculture and the Ministry of Education and Science. This proposal comprises several activities:

- unified inventory of the existing working collections - all holding institutions should have appropriate software based on the IPGRI descriptor lists for data input;
- establishment of an Intranet connection among the holding institutions with a centralized database in order to avoid storage of duplicates;
- collection of landraces and wild relatives;
- on-farm conservation;
- improvement of the connections with similar institutions, starting with the neighbouring countries, where researchers have much more experience and work on similar topics, to promote exchange of seed material, information, etc.;
- training of young researchers and appointment of curators for the priority crops.

Consequently, at the beginning of 2000 a Coordinating Body was established under the auspices of the Ministry of Agriculture. Its aim is to prepare the National Programme and legislation to support the regular functioning and funding of the genebank activities.

Genetic resources activities on forages in the Netherlands

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Introduction

Much emphasis has been given to the broadening of the CGN forage collection during the last three years. Old Dutch varieties of a number of grasses and ecotypes of both grasses and forage legumes have been introduced, regenerated and included in the collection.

At present, the forage collection of CGN includes 886 accessions, comprising 628 accessions of 11 different grass species, and 258 accessions of two *Trifolium* species. Since the last meeting of the ECP/GR Working Group on Forages in 1999, the collection has been increased by 245 new accessions of both grasses and legumes.

In 2000 a project was started to investigate the genetic diversity present in traditional Dutch pastures. For this purpose vegetative material of *Lolium perenne* and *Trifolium repens* was sampled in 2000 from 16 selected old grasslands situated in the different cattle-raising areas of the Netherlands. The collected material was characterized for morphological traits and molecular markers. This project will be briefly discussed and some preliminary results presented.

The collections

- **Grasses**

An overview of the CGN collection of grasses is presented in Table 1. The collection includes 628 accessions. Since the last meeting of the ECP/GR Working Group on Forages in 1999, 189 new accessions of different grasses have been added to the collection. In particular the collections of *Lolium perenne*, *Festuca rubra* and *Poa pratensis* were substantially increased with 98, 42 and 28 accessions respectively. Accessions of *Agrostis* spp. were newly introduced into the collection. Most of the 189 newly introduced accessions were of Dutch origin and in particular, old varieties were included in the collection. The new accessions of *Lolium perenne* included both old varieties and collected ecotypes. The latter group was received from a private Dutch breeding company which collected this material in the period 1970-1976 in mainly old permanent grasslands in the south of the Netherlands. It is the policy of CGN to broaden its forage collection with original Dutch material. Table 1 shows that 77% of the grasses are from the Netherlands.

- **Forage legumes**

At present, the *Trifolium* collection of CGN consists of 258 accessions (Table 2). It includes 162 accessions of red clover (*T. pratense*) and 96 accessions of white clover (*T. repens*). 78% of the collection is of Dutch origin and was largely obtained from CGN collecting missions in the Netherlands and Central Asia (van Soest and Dijkstra 1986, 1998; van Soest and Bas 2000). During two expeditions in Central Asia (Uzbekistan and Kyrgyzstan), organized in 1997 and 1999, ecotypes of both red and white clover were collected at altitudes ranging from 940 to 1790 m asl (van Soest 1998; van Soest *et al.* 1998; van Soest and Bas 2000).

Table 1. Collection of grasses maintained by CGN in 2003 (including material regenerated in 2002 but not yet stored in the genebank)

Species	No. of samples		Origin of the material
	Total	NLD*	
Dactylis glomerata	36	16	Ecotypes from NLD and Hungary
Festuca rubra	55	44	Old varieties from NLD
Festuca pratensis	1	1	Old varieties from NLD
Lolium multiflorum	59	56	Old varieties from NLD
Lolium hybridum	1		Variety from Austria
Lolium perenne	287	203	Ecotypes NLD, old varieties NLD and Europe
Phleum pratense	91	77	Old varieties from NLD and varieties from Europe
Phleum bertolonii	7	7	Old varieties from NLD
Phleum spp.	5		Ecotypes from Uzbekistan
Poa pratensis	80	73	Old varieties from NLD
Agrostis spp.	10	10	Old varieties from NLD
Total	**628**	**487**	

* NLD = samples originating from the Netherlands

Table 2. Collection of forage legumes maintained by CGN

Species	No. of samples		Origin of the material
	Total	NLD*	
Trifolium pratense	162	132	Ecotypes and a number of varieties
Trifolium repens	96	69	Ecotypes and a few varieties
Total	**258**	**201**	

* NLD = samples originating from the Netherlands

Regeneration

Since the last meeting in 1999 approximately 250 accessions of both grasses and forage legumes have been regenerated. With the exception of the apomict *Poa pratensis* all forage species are regenerated in field plots spatially isolated by triticale. The distance between the plots is 50 m. In general the forages are sown in late summer and thereafter kept in an unheated greenhouse during the winter in order to stimulate vernalization. In April of the following year the grasses and clovers are transplanted into isolation plots established in a large triticale field. Approximately 60 plants of each accession are planted, together with other crops, in a large isolation plot of 25-30 m^2. Seeds are harvested in July/August and after cleaning and drying samples are taken for germination tests. Accessions with an initial viability of 80% are included in the collection.

Documentation, storage and utilization

The collections are documented for passport data in the data management system of CGN, called GENIS, based on Oracle software. These data can also be found on CGN's Web site (http://www.genebank.nl).

The material is maintained under long-term storage conditions (-20°C) in the genebank facilities of CGN and is freely available to *bona fide* users. However, before receiving CGN material the user is requested to sign a "Material Transfer Agreement" (MTA). An example of the MTA used by CGN can be found on the Web site.

Table 3 presents an overview of the distribution of forage genetic resources to users over the last 15 years. It shows that, as a result of the additions to the collection during the last three years, the requests and distribution of material have increased significantly. Requests for red and white clovers in particular have increased tremendously. This is because of the organic farmers' desire to include forage legumes in pastures and to make use of the nitrogen-binding effect of these legumes in order to reduce nitrogen fertilization.

Table 3. Distribution of forage accessions by CGN

Forage species	Period 1988-98	Period 1999-2003*	Total
Grasses			
L. perenne	164	21	185
Other grass species	13	61	74
Clovers	15	284	299
Total	**192**	**366**	**558**
% of total distribution	34	66	100

* up to 1 March 2003

Biodiversity in old grasslands in the Netherlands

- **Background**

The Netherlands are situated in the northwestern part of the European-Siberian region of diversity. This region holds only limited biodiversity of our cultivated crops. However, a number of temperate grasses and legumes, important for animal feeding, have their centres of diversity in this region (Zeven and de Wet 1982). Species commonly found in this centre of diversity are the important forage grass *Lolium perenne* L. and the pasture legume *Trifolium repens* L.

In the last decades, practices to optimize fodder production, e.g. reseeding with uniform commercial cultivars and high nitrogen fertilization, have reduced the diversity of most Dutch grasslands. Traditional grasslands are becoming rare in the Netherlands. However, about 50 traditional grassland sites were identified by CGN in 1997 and 1998. These traditional grasslands are located in nearly all provinces where cattle-raising takes place. Some are under organic management. Unique genetic diversity may still exist within these grasslands. As the farmers who manage them cannot guarantee the maintenance of these traditional grasslands, *in situ* conservation is regarded as an option to safeguard the diversity existing within these ecosystems. However, the extent to which traditional grasslands have been affected by the widespread use of uniform commercial cultivars is currently unknown. To determine which grasslands are worth saving and should have priority for *in situ* conservation, data are needed about the genetic diversity present within and among traditional grasslands. In addition to *in situ* measures conserving the entire ecosystem, the relevant species found in these grasslands will be conserved *ex situ* in the genebank.

- **Experimental set-up**

In 2000, vegetative material (splits) of *L. perenne* and *T. repens* was collected in 16 traditional grasslands selected from the previously identified 50 traditional grasslands (Fig. 1). From each of the 16 grasslands 36 splits were selected and planted in the autumn of 2000 in a three-replicate trial at an experimental field in Wageningen. In addition tissue material was collected from each of the 36 plants for DNA analysis. Reference cultivars of both species, particularly varieties which had been widely used in Dutch pastures during the period 1940-1990, were included in the experiments in order to establish the diversity of the old grasslands using comparisons with these reference cultivars. The material in the field was evaluated in 2001 and 2002 for a set of morphological characters, which are also used in the registration of new cultivars. These so-called DUS descriptors are considered to be "distinct, uniform and stable". The molecular characterization was carried out with AFLP markers (Vos *et al.* 1995). The analyses for *L. perenne* started in 2000 and in 2003 all analyses will be completed.

Fig. 1. Location of 16 traditional grasslands in the Netherlands selected for the diversity study.

- **Preliminary results**

Preliminary results obtained for *L. perenne* did not indicate introgression from the reference cultivars into the traditional grasslands (van Treuren *et al.* 2005). Moreover, marked differences were observed between some of the grasslands for both the agromorphological and the ALFP data. For the morphological characters this was particularly true for heading date. The results for *L. perenne* suggested that the Netherlands may be considered a more or less continuous population with some grasslands being affected by selection (van Treuren *et al.* 2005).

Preliminary results of the morphological data in *T. repens* also showed a clear distinction between the reference cultivars and the grassland samples, notably in earliness and characters related to plant vigour. A remarkable result was that the most divergent populations of *T. repens* were collected from the same grasslands as the most divergent populations of *L. perenne.* This may be due to variation of ecological conditions among the traditional grasslands studied until now. So far the combined results do not support large-scale conservation measures for these Dutch grasslands, but rather seem to point to the conservation of a selected group which represents most of the variation.

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Status of the forage collection at the Nordic Gene Bank

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The Nordic Gene Bank has collected forage seeds since 1979. As shown in Table 1 about 70% of the collection is wild material. The material is collected or donated from Denmark, Finland, Greenland, Iceland, Norway and Sweden. Almost 95% of the collection consists of *Agrostis, Dactylis, Festuca, Lolium, Phalaris, Phleum, Poa* and *Trifolium*. NGB stores a total of 3227 accepted forage accessions.

Table 1. Number of forage accessions held at the Nordic Gene Bank (March 2003)

Genus	Type of sample					Total
	Varieties	Local material	Wild material	Breeding material	Unknown	
Agrostis	11	19	184	3	5	222
Alopecurus		11	48	1		60
Anthoxanthum			1			1
Anthyllis			2			2
Bromus	7		30		2	39
Calamagrostis			1			1
Cynosurus	1		1			2
Dactylis	34	21	254	3	1	313
Danthonia			1			1
Deschampsia		1	13			14
Festuca	89	27	512	5	3	636
Holcus			1			1
Lolium	96	1	80	3	2	182
Lotus	1		4			5
Medicago	24	1	3		2	30
Melilotus				3		3
Phalaris	1	4	147	1	1	154
Phleum	39	151	336	5	7	538
Poa	53	9	344		6	412
Trifolium	103	198	282	6	18	607
Vicia		1	2			3
x Festulolium	1					1
Total	**460**	**444**	**2246**	**30**	**47**	**3227**

Repatriation of forage grasses from the Vavilov Institute (VIR)

The Nordic forage working group decided to repatriate Nordic forage grasses from VIR in 2003. The NGB cereal working group has already repatriated some material from VIR. From earlier inventory projects we know that we are still missing some old locally cultivated varieties and we hope that the Scandinavian cultivated and wild material collected by VIR is still present, alive and available.

Status of the safety collection

NGB's safety collection is stored at Svalbard, Norway in an old coal mine. The temperature is stable at around -3/-4°C. NGB has continued its efforts to complete the safety collection, which now contains about 67% of all accepted accessions (Table 2).

The remaining 33% have not yet been sent to Svalbard for various reasons: newly collected material, material with too low germination ability to be accepted in the safety collection, or insufficient amounts of seed.

Table 2. Total number of accepted accessions and number of accepted accessions stored at Svalbard in years 1986, 1998 and 2003

Genus	Total no. of accessions	No. in year			% for 2003
		1986	1998	2003	
Agrostis sp.	222	6	57	149	67
Alopecurus sp.	60		28	35	58
Anthoxanthum	1				0
Anthyllis	2				0
Bromus sp.	39	4	9	29	74
Calamagrostis sp.	1		1	1	100
Cynosurus sp.	2		1	2	100
Dactylis sp.	313	5	73	207	66
Danthonia sp.	1				0
Deschampsia sp.	14		3	8	57
Festuca sp.	636	7	287	478	75
Holcus sp.	1			1	100
Lolium sp.	182	10	153	166	91
Lotus sp.	5		1	3	60
Medicago sp.	30	7	26	26	87
Melilotus sp.	3		3	3	100
Phalaris	154		31	47	31
Phleum sp.	538	7	142	344	64
Poa sp.	412	15	217	328	80
Trifolium sp.	607	64	192	346	57
Vicia sp.	3	4	5		0
x Festulolium	1		1	1	100
Total	**3227**	**129**	**1230**	**2174**	**67**

Utilization of NGB's forage collection

NGB has improved access to information on the stored material via the Web page (www.ngb.se/sesto) from which the following information can be obtained for each accession: taxonomy, culton, quality and quantity of stored seed, evaluation results, regenerations or rejuvenations. It is also possible to generate maps indicating collection site, with longitude, latitude and altitude.

Collecting activities in the Scandinavian countries, 2000-2002

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Introduction

Since its foundation in 1979 NGB has collected seeds in all five Nordic countries. In spite of the 23 collection missions already carried out during the last 24 years we still have some gaps (see Fig. 1). Since the last Working Group meeting in Elvas in 1999, NGB has undertaken four additional collecting missions in the Nordic countries. The result is shown in Table 1. A total of 441 accessions were collected and accepted.

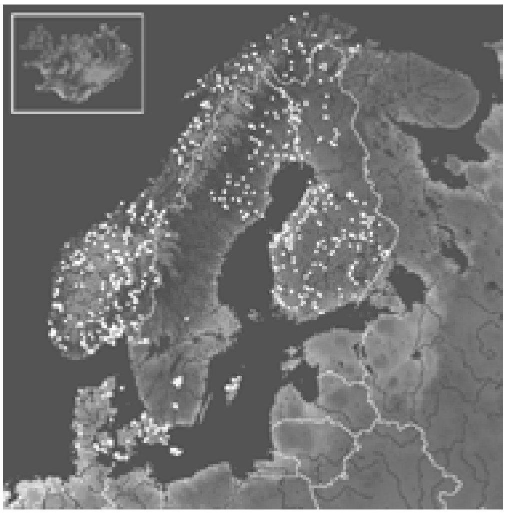

Fig. 1. Each dot on the map indicates a collection site. This map does not include material presented in Table 1.

Table 1. Forage accessions collected 2000-2002 (mainly wild and locally cultivated material)

Species	Country and year of collecting mission (*)				Total
	Norway 2002	Denmark 2001	Finland 2000	Sweden 2001	
Agrostis capillaris	28	13		13	54
Alopecurus pratensis				3	3
Anthyllis vulneraria				2	2
Bromus inermis				3	3
Dactylis glomerata	22	8		11	41
Deschampsia cespitosa				4	4
Festuca ovina	7			5	12
Festuca pratensis	19	1		3	23
Festuca rubra	18	11		10	39
Lolium perenne		10		2	12
Lotus corniculatus		1		3	4
Medicago lupulina				2	2
Medicago sativa				1	1
Phalaris arundinacea	2		79	3	84
Phleum pratense	32	5		8	45
Poa palustris				6	6
Poa pratensis	12	22		6	40
Poa trivialis		2			2
Trifolium hybridum	5				5
Trifolium pratense	20	1		9	30
Trifolium repens	14	7		8	29
Total	**179**	**81**	**79**	**102**	**441**

(*) Collecting sites = Norway: southern areas; Denmark: western part of Jutland; Finland: re-collection of lost *Phalaris* material from the southern and central part of Finland; Sweden: Småland, southern Sweden.

Future collecting activities in the Nordic Countries

The Nordic forage working group has decided to continue the collecting missions to fill all the gaps. This year a collecting mission will be organized in Iceland. NGB still has some areas which are less than adequately covered: most of central Sweden and the northern part of Finland.

Availability of passport data

From the NGB homepage (http://www.ngb.se/sesto) it is possible to search for passport information on collecting site, longitude, latitude, altitude for each accession. Maps can be generated.

Current status and utilization of the forages collections in Poland

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As we stated during the last meeting in 1999, our new activities strongly affect our collection's status. There are now 18 837 forage accessions in the genebank of the National Centre for Plant Genetic Resources of the Plant Breeding and Acclimatization Institute in Radzików, Poland. The grass collection consists of more than 140 species (17 789 accessions) and the legume collection of more than 17 species (1047 accessions). Species with the highest numbers of ecotypes are *Dactylis glomerata* and *Festuca pratensis* for grasses, and *Trifolium* sp. and *Medicago* sp. for legumes. Details of the collections are given in Tables 1 and 2.

The utilization of these collections during the 3-year period 2000-2002 is estimated at 4% (728 accessions) for grasses and 9.6% (101 accessions) for legumes (Table 3).

Additionally, we maintain more than 575 taxa of grasses and 320 taxa of legumes as a living collection in the Botanical Garden of the Plant Breeding and Acclimatization Institute in Bydgoszcz. From 1999 to 2002 more than 150 taxa were added to this living collection (Majtkowski and Majtkowska 2000).

The majority of species in the living collection originate from Europe but some American and Asiatic species are also present. Beside the traditional use (exchange of seed accessions), this collection is a source of information for students or other people wishing to learn more about grasses and legumes.

Table 1. Current status of the collection of forage and related grass species in the Polish Gene Bank

No.	Genus/ species	No. of accessions in						Total
		Advanced cultivars	Breeders' lines	Primitive cultivars, landraces	Semi-natural ecotypes	Wild ecotypes	Status unrecorded / unnamed accession	
1	*Agropyron cristatum*	2						2
2	*Agropyron elongatum*						1	1
3	*Agropyron intermedium*	2						2
4	*Agropyron smithii*	1						1
5	*Agrostis alba*	4				4		8
6	*Agrostis stolonifera*	3			1	17		21
7	*Agrostis tenuis*	18			1	14	1	34
8	*Alopecurus pratensis*	3				6		9
9	*Andropogon gerardi*	2						2
10	*Arrhenatherum elatius*	5				6		11
11	*Avenella flexuosa*						1	1
12	*Avenula pubescens*					1		1
13	*Boissiera squarrosa*					1		1
14	*Bromus benekenii*					1		1
15	*Bromus secalinus*					1		1
16	*Bromus ciliatus*						1	1
17	*Bromus erectus*						1	1
18	*Bromus inermis*	10				93	1	104
19	*Bromus japonicus*						1	1
20	*Bromus lanceolatus*						1	1
21	*Bromus madritensis*						1	1
22	*Bromus marginatus*					1		1
23	*Bromus pannonicus*						1	1
24	*Bromus ramosus*						1	1
25	*Bromus rigidus*						1	1
26	*Bromus scoparius*						1	1

Table 1 (cont.). Current status of the collection of forage and related grass species in the Polish Gene Bank

No.	Genus/ species	No. of accessions in						Total
		Advanced cultivars	Breeders' lines	Primitive cultivars, landraces	Semi-natural ecotypes	Wild ecotypes	Status unrecorded / unnamed accession	
27	*Bromus sewerzowii*						1	1
28	*Bromus squarrosus*						1	1
29	*Bromus tomentellus*						1	1
30	*Bromus willdenowii*						1	1
31	*Calamagrostis arundinacea*						1	1
32	*Calamovilfa longifolia*	1					1	2
33	*Cenchrus echinatus*						1	1
34	*Cynosurus cristatus*	1				26	1	28
35	*Dactylis glomerata*	125				5491	130	5746
36	*Dactylis glomerata aschersoniana*					2	1	3
37	*Dactylis glomerata* subsp. *juncinella*						1	1
38	*Dactylis woronowii*						1	1
39	*Deschampsia caespitosa*	2		1	4	101	7	115
40	*Deschampsia media*					1	2	3
41	*Deschampsia refracta*						2	2
42	*Deschampsia wibeliana*						1	1
43	*Desmazeria rigida*						1	1
44	*Digitaria ischaemum*						1	1
45	*Digitaria sanguinalis*						1	1
46	*Echinaria capitata*						1	1
47	*Echinochloa frumentacea*						1	1
48	*Eleusine indica*						1	1
49	*Elymus canadensis*	1					1	2
50	*Elymus dahuricus*						1	1
51	*Elymus farctus*						1	1
52	*Elymus gmelinii*						1	1
53	*Elymus hispidus*						1	1
54	*Elymus panoramitanus*						1	1
55	*Elymus racemosus*						1	1
56	*Elymus trachycaulus*	1						1
57	*Elytrigia pruinifera*						1	1
58	*Elytrigia pungens*						1	1
59	*Eragrostis pilosa*						1	1
60	*Festuca arundinacea*	44				742	137	923
61	*Festuca arundinacea* subsp. *aspera*					3		3
62	*Festuca arundinacea x Lolium multiflorum*	2						2
63	*Festuca capillata*					5	1	6
64	*Festuca gigantea*					3		3
65	*Festuca heterophylla*	1						1
66	*Festuca nigrescens*					1		1
67	*Festuca ovina*	5				7	1	13
68	*Festuca ovina* var. *duriuscula*	9						9
69	*Festuca pallens*						1	1
70	*Festuca polesica*						1	1
71	*Festuca pratensis*	95				3456	33	3584
72	*Festuca rubra*	59				106	3	168
73	*Festuca rubra* subsp. *arenaria*					4		4
74	*Festuca rubra* subsp. *juncea*					2	1	3
75	*Festuca rubra* subsp. *litoralis*					2		2
76	*Festuca rubra* subsp. *pruinosa*					3		3
77	*Festuca rubra* subsp. *rubra*					1		1
78	*Festuca rubra* var. *fallax*	12						12
79	*Festuca vaginata*						1	1
80	*Gastridium ventricosum*						1	1
81	*Holcus lanatus*					3		3
82	*Hordeum agriocrithon*						1	1
83	*Hordeum distichon*						1	1
84	*Hordeum zeocrithon*						1	1
85	*Hystrix patula*					1		1
86	*Koeleria albescens*					2	1	3
87	*Koeleria caucasica*				2	9	7	18
88	*Koeleria cenisia*						1	1
89	*Koeleria degenii*				2		1	3
90	*Koeleria glauca*				2	1	1	4
91	*Koeleria javorkae*						1	1
92	*Koeleria macrantha*				1			1

Table 1 (cont.). Current status of the collection of forage and related grass species in the Polish Gene Bank

No.	Genus/ species	No. of accessions in						Total
		Advanced cultivars	Breeders' lines	Primitive cultivars, landraces	Semi-natural ecotypes	Wild ecotypes	Status unrecorded / unnamed accession	
93	*Koeleria pyramidata*				5	8	5	18
94	*Koeleria splendens*						1	1
95	*Koeleria vallesiana*				1	5	4	10
96	*Lolium multiflorum*	50					1	51
97	*Lolium multiflorum* var. *westerwoldicum*	15						15
98	*Lolium perenne*	259	1		3	2171	55	2489
99	*Lolium remotum*						1	1
100	*Lolium rigidum*						1	1
101	*Lolium temulentum*						1	1
102	*Lolium x hybridum*	16						16
103	*Melica ciliata*						1	1
104	*Melica jacquemonti* subsp. *canescens*						1	1
105	*Melica jacquemonti* subsp. *jacquemonti*						1	1
106	*Melica transsilvanica*					1		1
107	*Monerma cylindrica*						1	1
108	*Muehlenbergia mexicana*						1	1
109	*Muehlenbergia racemosa*						1	1
110	*Muehlenbergia sylvatica*						1	1
111	*Nardurus maritimus*						1	1
112	*Nardus stricta*						1	1
113	*Panicum capillare*						1	1
114	*Panicum maximum*						1	1
115	*Panicum virgatum*	2					1	3
116	*Phalaris angusta*						1	1
117	*Phalaris arundinacea*	2						2
118	*Phalaris brachystachys*						1	1
119	*Phalaris canariensis*	2						2
120	*Phalaris minor*						1	1
121	*Phalaris truncata*						1	1
122	*Phleum nodosum*					2		2
123	*Phleum phleoides*					1	1	2
124	*Phleum pratense*	99				2416	2	2517
125	*Poa alpina*					1	1	2
126	*Poa araratica*						1	1
127	*Poa badensis*						1	1
128	*Poa bulbosa* subsp.*vivipara*						1	1
129	*Poa chaixii*					1	1	2
130	*Poa compressa*	1				17	1	19
131	*Poa nemoralis*	2				23	1	26
132	*Poa palustris*	4				7	1	12
133	*Poa pannonica* subsp. *scabra*						1	1
134	*Poa pratensis*	88				1360	194	1642
135	*Poa pratensis* subsp. *irrigata*					1		1
136	*Poa stiriaca*					1	1	2
137	*Poa trivialis*	1					1	2
138	*Polypogon monspeliensis*						1	1
139	*Psatyrostachys junceus*	2						2
140	*Puccinellia distans*						1	1
141	*Secale montanum*						1	1
142	*Secale segetale*						1	1
143	*Setaria pumila*						1	1
144	*Setaria verticillata*						1	1
145	*Setaria viridis*						1	1
146	*Sorghastrum nutans*	1					1	2
147	*Trisetum spicatum*						1	1
148	*Vulpia bromoides*						1	1
149	*Vulpia myuros*						1	1
	Total	**952**	**1**	**1**	**22**	**16132**	**681**	**17789**

Table 2. Current status of the small grain legume collection in the Polish Gene Bank

No.	Genus/species	Number of accessions in				Total
		Advanced cultivars	Breeders' lines	Wild ecotypes	Status unrecorded / unnamed accession	
1	*Anthyllis vulneraria*			3		3
2	*Coronilla varia*			1		1
3	*Galega orientalis*			1		1
4	*Lotus corniculatus*	1		128	28	129
5	*Lotus uliginosus*			80		80
6	*Medicago lupulina*	1				1
7	*Medicago sativa*	8				8
8	*Medicago varia*	9	2			11
9	*Medicago* sp.				200	200
10	*Melilotus albus*	1	1	11		13
11	*Onobrychis viciifolia*	4		11		15
12	*Ornithopus sativus*	86		22		108
13	*Trifolium alexandrinum*	1				1
14	*Trifolium hybridum*	3				3
15	*Trifolium incarnatum*	1				1
16	*Trifolium pratense*	84	11	21	1	117
17	*Trifolium repens*	30	1	1	1	33
18	*Trifolium* sp.			269	322	322
	Total	**229**	**15**	**548**	**552**	**1047**

Table 3. Utilization of the forage collection of the Polish Gene Bank

Genus/species	No. of accessions per year			Total
	2000	2001	2002	
Legumes				
Anthyllis vulneraria	2			2
Lotus corniculatus	3		4	7
Medicago lupulina	3		2	5
Medicago sativa L. subsp. *sativa*		5	1	6
Medicago x *varia*	7	3	3	13
Melilotus alba	1	3	5	9
Melilotus officinalis		1		1
Onobrychis viciifolia	2	1	1	4
Trifolium alexandrinum			3	3
Trifolium hybridum	3	3	1	7
Trifolium incarnatum	3	1		4
Trifolium pratense	11	3	1	15
Trifolium repens	4	3	1	8
Trifolium resupinatum	3	3		6
Trifolium sp.			11	11
Total legumes	**42**	**26**	**22**	**101**
Grasses				
Agrostis capillaris		1		1
Agrostis gigantea		1		1
Alopecurus pratensis		1		1
Arrhenatherum elatius		1		1
Bromus sp.	19	1		20
Cynosurus cristatus	1			1
Dactylis glomerata	4	39		43
Festuca arundinacea		4		4
Festuca gigantea	10			10
Festuca heterophylla		1		1
Festuca ovina		5	2	7
Festuca pratensis		1	5	6
Festuca rubra s.l.		21	13	34
Glyceria fluitans			4	4
Koeleria sp.		12		12
Lolium multiflorum		1	3	4
Lolium perenne	30	22	87	139
Lolium westerwoldicum	2			2
Lolium x hybridum	2	1		3
Phalaris arundinacea		1		1
Phalaris canariensis		1		1
Phleum pratense	26	1		27
Poa nemoralis		1		1
Poa palustris		1		1
Poa pratensis	15	14	22	51
Trisetum flavescens			4	4
Other grass species	21	164	163	348
Total grasses	**130**	**295**	**303**	**728**
Grand total (legumes + grasses)	**172**	**321**	**325**	**829**

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Forage collecting activities in Poland

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Introduction

The forage genetic resources programme in Poland was initiated in the early 1970s in the Plant Breeding and Acclimatization Institute. Since then more than 1300 sites have been visited in Poland and more than 19 000 wild forage grass and legume ecotypes were collected as seeds or living plants. The main ideas on how, what and where to collect evolved together with the changing needs of breeders and scientists. During the early years (1972-1989), the main goal was to collect numbers of ecotypes from only a few of the most popular and important forage species, without paying any special attention to site characterization. In the 1990s, to take account of the changing situation in Polish breeding, more attention was paid to species other than typical pasture grasses or legumes. Despite the huge amount of work done, the lack of precise passport data became obvious, especially after the compiling of the European Central Crop Databases of genera *Dactylis* and *Festuca* (Żurek 1997). From 1998 onwards it was decided to focus not only on the collection of plant material but also to collect as many site data as possible.

Collecting methodology

The areas for collecting were chosen mainly during discussions with local people who know the vegetation and agricultural practices in the region to be visited. However, collecting is also done while moving from one place to another (near tracks, paths or roads, alongside ditches, near forests or on the beach).

All sites visited during collecting missions in 2000-2002 were described according to the passport descriptor list composed according to our own practice, which is similar in structure to the Multicrop Passport Descriptor List. Geographical coordinates of each site were recorded using a Garmin 12 GPS receiver (map datum setting - WGS 84). All visible associated plant species were also recorded.

Results

During the last three collecting missions (2000, 2001 and 2002) 110 localities were visited in four provinces of Poland: Zachodniopomorskie, Warmińskomazurskie, Podlaskie and Dolnośląskie (Fig. 1). Most sites visited were more or less similar to typical meadows or pastures. They were extensively grazed or even unmanaged rather than cut. Nearly one-third of all sites were similar to the 'meadow-pasture' type but due to their floristic composition (trees and shrubs, perennial weeds, no traces of usage) they could only be listed as wastelands. During all above collecting missions in Poland 862 seed accessions were collected (685 grasses and 178 legumes) (Table 1).

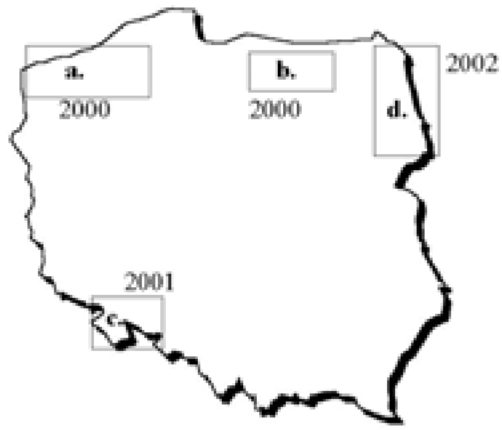

Fig. 1. Forage collecting activities in Poland (2000-2002).
Sites: a = Zachodniopomorskie, b = Warmińskomazurskie; c = Dolnośląskie; d = Podlaskie

Table 1. Forage species collected during missions in Poland (2000-2002) (number of accessions)

No.	Genus/species/subtaxon	Collecting year			Total
		2000	2001	2002	
	Grasses				
1	*Agrostis canina* L.		4		4
2	*Agrostis capillaris* L.	7	9	8	24
3	*Agrostis gigantea* Roth	2	5		7
4	*Agrostis stolonifera* L.	4	2	1	7
5	*Alopecurus geniculatus* L.	1		1	2
6	*Alopecurus pratensis* L.	2	2	5	9
7	*Ammophila arenaria* (L.) Link.	1			1
8	*Anthoxanthum odoratum* (L.) P.B.	2	2	1	5
9	*Apera spica-venti* (L.) P.B.	2	1	1	4
10	*Arrhenatherum elatius* (L.) P.B. ex J. et C.Presl	8	12	5	25
11	*Avena fatua* L.	2	2		4
12	*Brachypodium pinnatum* (L.) P.B.		1		1
13	*Brachypodium sylvaticum* (Hudson) P.B.		2		2
14	*Briza media* L.		4	3	7
15	*Bromus benekenii* (Lange) Trimen		2		2
16	*Bromus carinatus* Hooker et Arn.		1		1
17	*Bromus hordeaceus* L. subsp. *hordeaceus*	2	1		3
18	*Bromus inermis* Leyss	1	2	5	8
19	*Calamagrostis arundinacea* (L.) Roth		5	1	6
20	*Calamagrostis epigejos* (L.) Roth	2	1		3
21	*Calamagrostis villosa* (Chaix) J.F.Gmelin		3		3
22	*Corynephorus canescens* (L.) P.B.	1		1	2
23	*Cynosurus cristatus* L.	6	12	6	24
24	*Dactylis aschersoniana* Graebner	1		1	2
25	*Dactylis glomerata* L.	20	18	15	53
26	*Dactylis glomerata* L. f. *vivipara*	1			1
27	*Danthonia decumbens* (L.) DC.	1	3	3	7
28	*Deschampsia caespitosa* (L.) P.B.	10	18	21	49
29	*Deschampsia flexuosa* (L.) Trin.	2	6		8
30	*Elymus caninus* (L.) L.		2		2
31	*Elymus repens* (L.) Gould		1	2	3
32	*Elymus repens* subsp. *caesium* (J. et C.Presl) Oberd.		1		1
33	*Festuca altissima* All.		2		2
34	*Festuca arundinacea* Schreb.	5	2	2	9
35	*Festuca gigantea* (L.) Vill.	2	5	1	8
36	*Festuca ovina* L.	2	1		3
37	*Festuca pratensis* Huds.	9	16	13	38
38	*Festuca rubra* L.	14	22	24	60
39	*Festuca rubra* L. subsp. *arenaria* (Osb.) Syme	1			1
40	*Festuca* sp.	1	3		4
41	*Festuca tenuifolia* Sibth.		3	1	4

Table 1 (cont.). Forage species collected during missions in Poland (2000-2002) (number of accessions)

No.	Genus/species/subtaxon	Collecting year			Total
		2000	2001	2002	
42	*Glyceria fluitans* (L.) R.Br.	3	3		6
43	*Glyceria maxima* (Hatraman) Holmb.	2		1	3
44	*Holcus lanatus* L.	5	3	3	11
45	*Holcus mollis* L.	4	6	2	12
46	*Holcus* sp.	1			1
47	*Hordelymus europaeus* (L.) C.O.Harz		1		1
48	*Koeleria macrantha* (Ledeb.) Schultes		1		1
49	*Leymus arenarius* (L.) Hochst.	1			1
50	*Lolium multiflorum* Lam.	1			1
51	*Lolium perenne* L.	12	11	16	39
52	*Melica nutans* L.			1	1
53	*Milium effusum* L.	1	1		2
54	*Molinia arundinacea* Schrank			1	1
55	*Nardus stricta* L.			2	2
56	*Phalaris arundinacea* L.	9	4	5	18
57	*Phleum nodosum* L.			4	4
58	*Phleum pratense* L.	16	14	20	50
59	*Phleum pratense* subsp. *bertolonii* (DC.) Bornm.		2		2
60	*Phragmites australis* (Cav.) Trin. ex Steudel	1			1
61	*Poa compressa* L.	1	5	2	8
62	*Poa nemoralis* L.	4	10	1	15
63	*Poa palustris* L.	2	5	7	14
64	*Poa pratensis* L.	15	19	35	69
65	*Poa pratensis* L. subsp. *angustifolia*	1			1
66	*Poa* sp.		1	1	1
67	*Puccinellia distans* (L.) Parl.	1	1		2
68	*Setaria viridis* (L.) P.Beauv.	1		1	2
69	*Trisetum flavescens* (L.) P.B.	1	5		6
	Total grasses	**194**	**268**	**223**	**685**
	Legumes				
1	*Anthyllis vulneraria* L.	1	3	3	7
2	*Astragalus arenarius* L.			1	1
3	*Astragalus glycyphyllos* L.	3	3	2	8
4	*Coronilla varia* L.		2		2
5	*Cytisus scoparius (*L.) Link	2	1		3
6	*Genista tinctoria* L.		2		2
7	*Lathyrus pratensis* L.		5	2	7
8	*Lathyrus sylvestris* L.		2	2	4
9	*Lotus corniculatus* L.	3	7	2	12
10	*Lotus uliginosus* Schkuhr	2	3	1	6
11	*Lupinus polyphyllus* Lindley	2	2		4
12	*Medicago falcata* L.			3	3
13	*Medicago lupulina* L.	2	3		5
14	*Medicago sativa* L.		1		1
15	*Melilotus alba* Medik.	2	2	2	6
16	*Trifolium repens* L.			1	1
17	*Trifolium arvense* L.	2	2	1	5
18	*Trifolium aureum* Poll.	1	6	3	10
19	*Trifolium campestre* Schreb.	1		1	2
20	*Trifolium dubium* Sibth.		1		1
21	*Trifolium fragiferum* L.	1			1
22	*Trifolium hybridum* L.	3	3	3	9
23	*Trifolium medium* L.	6	6	2	14
24	*Trifolium montanum* L.			1	1
25	*Trifolium pratense* L.	10	6	7	23
26	*Trifolium repens* L.	8	11	4	23
27	*Vicia cracca* L.		5		5
28	*Vicia hirsuta* (L.) S.F.Gray	5			5
29	*Vicia sepium* L.	1	2		3
30	*Vicia tenuifolia* Roth.	2			2
31	*Vicia tetrasperma* (L.) Schreber	2			2
	Total legumes	**59**	**78**	**41**	**178**
	Grand total forages	**253**	**346**	**264**	**862**

Conclusions

The most popular pasture grass species such as *Poa pratensis*, *Festuca rubra*, *Dactylis glomerata* or *Phleum pratense* are typical of meadows, pastures and waste places in Poland (Kryszak and Grynia 2001). However, 19 species were noted no more than 3 times. Some of them were quite rare, such as *Hordelymus europaeus* and *Bromus carinatus* (both from Dolnośląskie province) or *Ammophila arenaria* (Zachodniopomorskie province). The average number of grass species per site ranged from 6 (Lubelskie province) to 8.8 (Zachodniopomorskie province). From this province most 'unique' grass species were also recorded. For example *Ammophila arenaria, Elymus arenarius* or *Festuca rubra* subsp. *arenaria* are mostly from the Baltic coast (Falkowski 1982; Zając and Zając 2001a, 2001b; Rutkowski 2002).

It was concluded that site-related data together with the lists of species are of potential value in case of any need for the re-introduction of particular species. For the purposes of genetic resources preservation it is essential to visit not only typical grassland habitats but also other sites to increase species-specific diversity.

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The Romanian forage collection

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The Romanian forage collection consists of 2491 accessions, including 1272 grasses belonging to 54 species and 1219 forage legumes belonging to 41 species. The most important set is represented by *Medicago sativa* (555 accessions), followed by *Vicia faba* (454 accessions) and *Lolium perenne* (290 accessions), while 30 other species are represented by only one accession in the collection. The main holders are the Suceava Genebank with 905 accessions, the Research Institute for Cereals and Technical Plants in Fundulea with 892 accessions, and the Grassland Research Institute in Braşov with 523 accessions.

As regards the origin of samples, the national collection has accessions originating from 60 different countries. Almost 50% (1209) of accessions are of Romanian origin.

The status of the samples is as follows: 991 accessions are traditional varieties, 269 are wild material, while a significant part is represented by advanced cultivars (958 accessions) or breeders' lines (61 accessions). The status of the sample is unknown for 212 accessions.

The National Forage Database is hosted and managed by the Suceava Genebank, and contains passport data which are in accord with FAO/IPGRI Multicrop Passport Descriptors.

Current status of forage collections in Slovakia

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Introduction

The Slovak collection of forage genetic resources consists of 4096 accessions representing 78 species. Two institutions hold the collections of forage legumes: the Research Institute of Plant Production in Piešťany and the Plant Breeding Station Horná Streda. The Plant Breeding Station in Levočské Lúky deals with genetic resources of grasses. The preservation of genetic diversity of forages is tied in with the previous period. The collection, evaluation, multiplication, filling in the passport and descriptive databases, and long-term conservation have remained the fundamental aims.

The implementation of Law No. 215/2001 "Conservation of Plant Genetic Resources for Food and Agriculture", approved by the National Council of the Slovak Republic in May 2001 and ratified 1 July 2001 was an important issue in the field of genepool preservation in Slovakia. The terms for plant genetic resources protection, collection, conservation, sustainable use and maintenance were established by this Act.

Under this Act the Research Institute of Plant Production in Piešťany is entrusted to implement the National Plant Genetic Resources Programme as adopted and governed under the auspices of the Ministry of Agriculture of the Slovak Republic. Moreover, the Institute is responsible for plant genetic resources conservation in the Gene Bank of the Slovak Republic.

Forage legumes

Jarmila Drobná, Curator, *Medicago* spp., *Trifolium* spp., other forage legumes.
Gabriela Antalíková, Curator, *Lupinus* spp., *Lathyrus* spp.
Miroslav Vavák, Curator, *Vicia* spp., *Faba* spp.
Zdeněk Slaměna, Jozef Štefanka, Curator, *Pisum* spp.

The composition of the forage legumes collections by genus is given in Table 1 and further details on the type of sample in Table 2.

Table 1. Forage legumes collections in Slovakia

Genus	No. of species	No. of accessions
Anthyllis L.	3	37
Astragalus L.	3	26
Coronilla L.	2	22
Faba L.	1	157
Lathyrus L.	1	258
Lotus L.	5	102
Lupinus L.	3	76
Medicago L.	10	376
Melilotus Mill.	6	37
Onobrychis Mill.	2	27
Pisum L.	1	228
Trifolium L.	20	612
Vicia L.	2	140
Others	3	4
Total	**62**	**2104**

Table 2. Details of the Slovak forage legumes collections (status at the end of December 2002)

Genus and species	Advanced cultivars	Landraces	Wild	Breeders' lines	Others	Total
Anthyllis jacquinii Kerner	-	-	1	-	-	1
Anthyllis vulneraria L.	1	-	34	-	-	35
Anthyllis taurica Juz.	-	-	1	-	-	1
Astragalus australis (L.) Lamk.	-	-	1	-	-	1
Astragalus cicer L.	11	-	2	1	-	14
Astragalus glycyphyllos L.	-	-	11	-	-	11
Coronilla emeroides Bois. et. Spr.	-	-	1	-	-	1
Coronilla varia L.	1	-	20	-	-	21
Dorycnium sp.	-	-	1	-	-	1
Faba vulgaris L.	116	24	9	1	7	157
Lathyrus sativus L.	6	41	-	211	-	258
Lotus corniculatus L.	24	-	71	1	1	97
Lotus ornithopoides L.	1	-	-	1	-	2
Lotus pedunculatus Cav.	1	-	-	-	-	1
Lotus uliginosus Schkuhr	-	-	1	-	-	1
Lotus sp.	-	-	1	-	-	1
Lupinus albus L.	42	1	-	-	-	43
Lupinus angustifolius L.	7	1	-	3	-	11
Lupinus luteus L.	22	-	-	-	-	22
Medicago arabica (L.) Huds.	-	-	2	-	-	2
Medicago falcata L.	2	-	32	8	5	47
Medicago lupulina L.	1	-	30	-	-	31
Medicago minima L.	-	-	3	-	-	3
Medicago orbicularis (L.) Bartal.	-	-	3	-	-	3
Medicago polychroa Grossh.	2	-	-	-	-	2
Medicago romanica Prodan	-	-	3	-	-	3
Medicago sativa L.	192	3	24	36	3	258
Medicago varia Martyn	16	-	8	-	1	25
Medicago sp.	-	-	2	-	-	2
Melilotus alba Med.	4	-	15	-	-	19
Melilotus dentata (W. et K.) Pers.	-	-	-	-	1	1
Melilotus neapolitanus Ten.	-	-	1	-	-	1
Melilotus officinalis (L.) Pallas	2	-	4	3		9
Melilotus tauricus (M.Bieb.) Ser.	-	-	2	-	-	2
Melilotus sp.	-	-	3	-	2	5
Onobrychis viciaefolia Scop.	19	-	7	-	-	26
Onobrychis sp.	-	-	1	-	-	1
Ornithopus sativus Brot.	-	4	-	-	-	4
Pisum sativum conv. *speciosum*	155	12	-	61	-	228
Trifolium alexandrinum L.	6	-	-	-	-	6
Trifolium alpestre L.	-	-	4	-	-	4
Trifolium angustifolium L.	-	-	1	-	-	1
Trifolium arvense L.	-	-	4	-	-	4
Trifolium aureum Pollich	-	-	19	-	-	19
Trifolium campestre Schreb.	-	-	4	-	-	4
Trifolium caucasicum Tausch	-	-	1	-	-	1
Trifolium dubium Sibth.	-	-	2	-	-	2
Trifolium fragiferum L.	-	-	4	-	-	4
Trifolium hybridum L.	4	-	27	3	-	34
Trifolium hirtum All.	-	-	1	-	-	1
Trifolium incarnatum L.	1	-	-	-	-	1
Trifolium medium L.	-	-	44	-	-	44
Trifolium montanum L.	-	-	18	-	-	18
Trifolium pratense L.	179	-	99	20	9	307
Trifolium repens L.	75	-	74	1	-	150
Trifolium resupinatum L.	6	-	-	-	-	6
Trifolium rubens L.	-	-	4	-	-	4
Trifolium semipilosum Fres.	1	-	-	-	-	1
Trifolium subterraneum L.	-	-	1	-	-	1
Trigonella cretica (L.) Boiss	-	-	1	-	-	1
Vicia sativa L.	122	6	-	7	4	139
Vicia villosa Roth.	1	-	-	-	-	1
Total	**1020**	**92**	**602**	**357**	**33**	**2104**

Evaluation and documentation

The number of accessions evaluated in the period 2000–20002 and the numbers of passport and descriptive data of the forage legumes collections are shown in Table 3. Passport data are available for 56% of the total number of accessions and descriptive data for 35%.

Since the last meeting of the Working Group on Forages the passport data have been updated and completed. In 2000, European database curators transmitted data about 2616 genetic resources of forages belonging to the species and genera of *Poa*, *Lolium*, *Dactylis*, *Festuca*, *Arrhenatherum elatius*, *Trisetum flavescens*, *Agrostis*, *Phleum*, *Trifolium*, *Medicago*, *Lathyrus*, *Vicia* and other forage legumes and grasses.

Table 3. Evaluation and documentation status of the Slovak forage legumes collections

Genus	No. of evaluated accessions (2000-2002)	Passport data	Descriptive data
Anthyllis L.	0	22	0
Astragalus L.	0	22	12
Coronilla L.	0	11	0
Faba L.	18	56	157
Lathyrus L.	0	37	0
Lotus L.	0	69	16
Lupinus L.	19	46	0
Medicago L.	28	325	62
Melilotus Mill.	0	32	6
Onobrychis Mill.	0	20	9
Pisum L.	68	27	228
Trifolium L.	170	446	111
Vicia L.	48	63	140
Others	0	4	0
Total	**351**	**1180**	**741**

Conservation, regeneration and safety-duplication

In 2000-2002, 273 accessions of forage legumes were multiplied, 75 in technical isolation. Safety-duplicates (of accessions stored in the base collection) are located in the Gene Bank of the Research Institute of Crop Production (RICP) in Prague. Table 4 summarizes the storage conditions and need for regeneration of accessions.

Table 4. Storage conditions and regeneration needs of forage legumes accessions

Genus	Base collection -17°C	Active collection +5°C	Working collection(*) +5°C
Anthyllis L.	1	20	17
Astragalus L.	1	15	12
Coronilla L.	1	11	12
Faba L.	27	56	74
Lathyrus L.	2	37	221
Lotus L.	5	63	51
Lupinus L.	0	39	37
Medicago L.	33	217	150
Melilotus Mill.	2	25	17
Onobrychis Mill.	0	3	33
Pisum L.	36	24	168
Trifolium L.	30	397	215
Vicia L.	60	63	17
Others	0	4	0
Total	**198**	**974**	**1024 (51%)***

(*) needing regeneration

Availability and utilization of genetic resources

About 70% of accessions are available in limited quantities on an exchange basis. Since 2000 some 457 accessions of different forage legumes held in RIPP Piešťany have been distributed to breeders, researchers and to other users in Slovakia and abroad (Fig. 1).

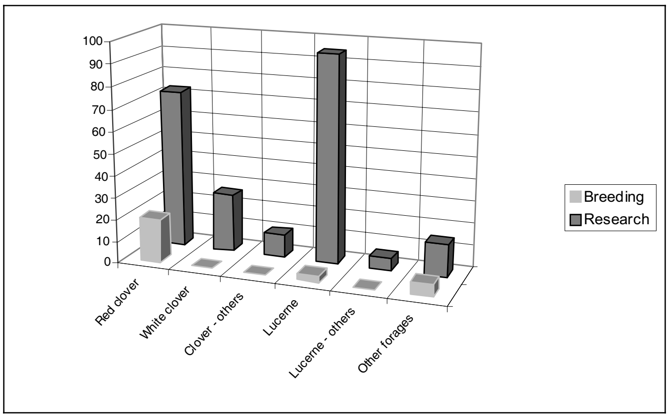

Fig. 1. Utilization of some forage legumes genetic resources in Slovakia.

Grasses

Marek Vitikač, Curator.

The composition of the collections of grasses and details of the types of samples are given in Table 5. Most of these accessions are stored as a working collection. By the end of 2002 there were 90 accessions stored in the active collection and 60 in the base collection in the Gene Bank in Piešťany.

Table 5. Details on the Slovak grasses collections

Genus and species	Advanced cultivars	Wild	Breeders' lines	Total
Agrostis L.	-	6	-	6
Agrostis stolonifera L.	9	11	-	20
Agrostis tenuis Sibth.	9	64	-	73
Alopecurus L.	6	10		16
Arrhenatherum elatius P. Beauv.	10	32	-	42
Briza L.	-	4	-	4
Bromus L.	-	1	-	1
Cynosurus L.	2	9		11
Dactylis L.	48	198		246
Deschampsia caespitosa (L.) P. Beauv.	2	41	3	46
Festuca arundinacea Schreb.	32	6	4	42
Festuca L.	-	22	-	22
Festuca ovina L.	12	42	-	54
Festuca pratensis Huds.	59	517	-	576
Festuca rubra L.	48	39	1	88
x *Festulolium* Aschers. et Graebn.	4	-	-	4
Hierochloe R. Brown	-	1	-	1
Koeleria Pers.	1	1	-	2
Lolium x *hybridum* Hausskn.	12	-	-	12
Lolium multiflorum Lamk.	49	-	-	49
Lolium perenne L.	99	145	10	254
Nardus stricta L.	-	2	-	2
Phleum L.	3	4	-	7
Phleum pratense L.	29	74	4	107
Poa L.	7	24	-	31
Poa pratensis L.	97	146	7	250
Phragmites Adans.	-	1	-	1
Trisetum Pers.	3	22	-	25
Total	**541**	**1422**	**29**	**1992**

Collecting activities at RIPP Piešťany, Slovakia

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The surveying, monitoring and collecting of plant genetic resources is a crucial activity for the preservation of the plant genepool in Slovakia. In recent years the exploration, mapping and collection of wild-growing forages and other plant species have been carried out by several collecting expeditions. In 2000 and 2001 two projects were adopted and implemented under the auspices of the Agreement between the Governments of the Slovak and Czech Republics on scientific and technical cooperation. We took advantage of the opportunity both to participate in and to organize collecting expeditions focusing on a range of plant species on the territories of Slovakia and the Czech Republic.

During our collecting expeditions the Slovak legal regulations in force had to be observed and the relevant FAO principles governing the collection and transfer of plant genetic resources were followed. Technical visits were made to the Slovak National Museum's herbaria before the exploration and collecting expeditions were launched.

Between 2000 and 2002, work continued on the survey, collection and transfer of plant genetic resources samples in Podyjí and Krkonoše National Parks and the Land Protected Areas of Jeseníky and Moravský Kras in the Czech Republic. In the Slovak Republic, collecting expeditions were made to the mountain ranges of Burda, Považský Inovec and Slovenské Rudohorie, the western parts of the TANAP (Vysoké Tatry) and NAPANT (Nízke Tatry) National Parks. In collaboration with the Research Institute of Crop Production (RICP) in Prague-Ruzyne (Czech Republic), we participated in joint Czech-Slovenian collecting expeditions to Slovenia and Croatia (Table 1).

These expeditions collected a total of 1840 plant samples (573, 902 and 365 in 2000, 2001 and 2002 respectively). These include, according to their use and economic importance: 655 grasses, 429 forages, 278 herbs and medicinal plants, 138 legumes, 105 ornamental species, 56 vegetables, 29 oil and industrial crops, 21 cereals, 3 root crop plants and 126 other plant species.

The survey results were used to build a database containing information about the collected plant species samples accompanied by their topographic specifications and GPS coordinates.

Table 1. Collecting activities of the RIPP Piešťany, 2000-2002

Year	Acronym	Country	Region	Participating institutes	Collected species	
2000	SVKBUR00	Slovak Republic	Burda	RIPP Piesťany	Botanical survey	
					Total	**4**
	SVKPOV00	Slovak Republic	Považský Inovec	RIPP Piesťany	Botanical survey	
					Total	**4**
	CZEPOD00	Czech Republic	National Park Podyjí	RIPP Piesťany	Grasses	20
				RIFP Troubsko	Legumes	14
					Total	**34**
	CZEJES00	Czech Republic	Protected Landscape Area Jeseniky	RIPP Piesťany	Grasses	148
				BS Maly Saris	Legumes	61
				RIFP Troubsko	Others	74
				RIPP Prague-Ruzyne	**Total**	**283**
				GBS Zubří		
				KI Ljubljana		
				IHAR Radzików		
				OB IHAR Bydgoszcz		
	SVKROH00	Slovak Republic	National Park Vysoké Tatry	RIPP Piesťany	Grasses	75
				RIFP Troubsko	Legumes	102
				RIPP Prague-Ruzyne	Others	71
				GBS Zubří	**Total**	**248**
2001	CZEKRK01	Czech Republic	National Park Krkonose	RIPP Piesťany	Grasses	35
				RIFP Troubsko	Legumes	30
				GMARI Banska Bystrica	Others	24
					Total	**89**
	SVNPIR01	Slovenia		RIPP Piesťany	Grasses	98
				RIFP Troubsko	Legumes	100
				RIPP Prague-Ruzyne	Others	96
				GBS Zubří	**Total**	**294**
				KI Ljubljana		
	CZEMKRAS01	Czech Republic	Protected Landscape Area Moravský Kras	RIPP Piesťany	Grasses	58
				RIFP Troubsko	Legumes	40
				RIPP Prague-Ruzyne	Others	83
				GBS Zubří	**Total**	**181**
				KI Ljubljana		
				IHAR Radzików		
				OB IHAR Bydgoszcz		
	SVKNTAT01	Slovak Republic	National Park Nízke Tatry	RIPP Piesťany	Grasses	122
				RIFP Troubsko	Legumes	106
				RIPP Prague-Ruzyne	Others	110
				GBS Zubří	**Total**	**338**
				KI Ljubljana		
				IHAR Radzików		
				OB IHAR Bydgoszcz		
2002	HRVISTRA02	Croatia	Istrian Peninsula	RIPP Piesťany	Grasses	74
				RIFP Troubsko	Legumes	74
				RIPP Prague-Ruzyne	Others	133
				GBS Zubří	**Total**	**281**
				KI Ljubljana		
				Univ. of Zagreb		
	SVKGEM02	Slovak Republic	Slovenské Rudohorie	RIPP Piesťany	Grasses	24
				RIFP Troubsko	Legumes	28
				GMARI Banska Bystrica	Others	32
					Total	**84**
Total no. of accessions collected = 1840						

Forages genetic resources in Slovenia

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Introduction
Landscape diversity in the territory of Slovenia is a result of the land's natural characteristics and a long history of human colonization and a variety of different land uses. The main attribute of the diversity is a small mosaic structure and rapidly changing aspects over short distances. Farming adapts to the natural conditions by applying different methods and thus it became the main factor in the development of the Slovenian countryside. It also contributed considerably to the changing face of the landscape. The main bio-regions reflect the diversity of Slovenian landscapes, and the transition zone between the Alps and the other regions is characterized particularly as "the pre-alpine landscapes". Within these regions micro-factors build up the mosaic of small, characteristic and diverse landscape structures. As a result, many different ecotypes of grasses and clovers can be found in the extensive grassland area of Slovenia.

Early projects to collect Slovenian native populations, ecotypes and landraces of agricultural species with the goal of breeding new and improved cultivars were initiated about 35 years ago. In the frame of the former Yugoslavia during the late 1980s a programme started with the task to collect plant genetic resources for the Yugoslav genebank. After the independence of Slovenia, the Slovenian Ministry of Science and Technology financed the establishment of the genebank of vegetables, potato, fodder plants, grasses, clovers, small fruits and grapevine. In 1996 the Ministry of Agriculture, Forestry and Food started financing the Slovene genebank programme with the aim of maintaining, evaluating, regenerating and preserving Slovenian native species, ecotypes, populations and landraces of agricultural plants.

Forages collection
The Slovene genebank system consists of two working collections for forage grass and clover species housed at the Biotechnical Faculty of the University of Ljubljana and at the Agricultural Institute of Slovenia. The whole collection consists of 638 accessions of 20 different forage species (Table 1). The Biotechnical Faculty of the University of Ljubljana holds 91 legume accessions and 39 grass accessions and the Agricultural Institute of Slovenia 224 legume accessions and 284 accessions of grasses.

The collection includes material of forage species of major economic importance. Most of the accessions were collected during short collecting missions throughout Slovenia. In 1999 a cooperative project was initiated with the Czech Gene Bank for several week-long collecting expeditions in Slovenia and Czech Republic. The first joint collecting trip included two northwestern regions of Slovenia: Gorenjska and Primorska.

Characterization and evaluation
Last year we started to evaluate 39 white clover and 13 timothy accessions. White clover hills were evaluated for profuseness of bloom, field pest and disease susceptibility and plant habit. There are fairly large differences in internode length and bloom profuseness between accessions. One accession was free from pests and diseases.

Evaluation of timothy accessions was set up in four replicates, which will help us to assess further some basic characters, as well as herbage yield.

Table 1. Forage species accessions held at the Slovenian genebank

Genus	Wild	Advanced cultivars and breeders' lines
Trifolium	227	3
Medicago	26	5
Vicia	18	
Lotus	13	
Other legumes	23	
Total legumes	**307**	**8**
Lolium	48	2
Festuca	49	9
Phleum	48	1
Poa	13	5
Dactylis	68	1
Agrostis	8	1
Alopecurus	17	
Other grasses	53	
Total grasses	**304**	**19**
Grand total	**611**	**27**

Documentation

In 1999 we started developing an information and database management system for the Slovenian genebank. All forage species accessions are well documented for IPGRI minimum passport descriptors, although some minor gaps exist. So far no characterization/evaluation data are included.

Storage

Seeds dried to a moisture content of ca. 5-7% are stored in glass jars in a vault under medium-term storage conditions at 4°C. We plan to start storing seed samples on a long-term basis as soon as the collection grows large enough to make this economically feasible.

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Activities on forage crop genetic resources in Turkey

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Introduction

Turkey is the centre of origin and/or centre of diversity for a large number of forage species. The plant genetic resources programme was started in 1964 with *ex situ* conservation of plant diversity (Tan 1998). At present, all activities of the Global Plan of Action for the Conservation and Sustainable Utilization of Plant Genetic Resources for Food and Agriculture, including *in situ* and on-farm conservation, are dealt with under the National Plant Diversity Conservation Programme of Turkey.

Survey/collecting

One collecting mission was carried out in northwestern Turkey in 2001. A total of 95 accessions were collected, including 44 *Trifolium*, 3 *Medicago* and 1 *Onobrychis* and 47 Gramineae.

Another 133 accessions, mainly of *Medicago*, *Onobrychis* and *Agropyron,* were collected from natural pastures of the Central Anatolian provinces during 1999-2001 by the Central Research Institute for Field Crops (CRIFC) in Ankara.

Ex situ conservation

The Centre of Plant Genetic Resources with its genebank is situated at the Aegean Agricultural Research Institute (AARI) which is responsible for plant genetic resources activities in Turkey. The Central Research Institute for Field Crops also has storage capacity for safety-duplication of the national base collection.

Storage facilities are available for both long-term and medium-term conservation at AARI's genebank for seed collections. Base collections are kept at –18°C and active collections at 0°C. Collections for temporary storage are kept at +4°C.

In situ conservation

Three projects for the conservation of priority species *in situ* have been initiated, two of which are directly or indirectly connected with *in situ* conservation of forage crops species:

- ***In situ* conservation of genetic diversity**

This project was started in 1993. Its objective is to maintain wild crop genetic resources in their natural habitats. Three areas were chosen as proposed gene management zones (GMZs). Each area contains a number of survey and inventory sites.

1. Kazdağ mountains located in the North-West, representing the Euro-Siberian, Mediterranean and Irano-Turanian elements. Target species chosen are *Castanea sativa*, *Prunus divaricata*, *Abies equitrojona*, *Pinus brutia* and *Pinus nigra*.
2. Ceylanpinar State Farm in the South-East, representing the Mediterranean and Irano-Turanian elements. Target species chosen are *Aegilops speltoides*, *Triticum tauschii*, *Triticum boeoticum*, *Triticum dicoccoides* and *Lens orientalis*.
3. Anatolian Diagonal in South and Central Turkey, representing the Mediterranean, Irano-Turanian and Euro-Siberian elements. Target species chosen are *Aegilops speltoides*, *Lens arvodies*, *Lens orientalis*, *Pisum sativum s.l.*, *Vicia sativa s.l.*, *Vicia johannis*, *Abies cilicica* and *Pinus brutia* (Kitiki and Tan 1988; Sabancı 1999; Tan and Tan 2002).

- ***In situ* (on-farm) conservation of landraces from the transitional zone in Turkey**

The project was initiated in 1999. Socioeconomic and ecogeographical surveys were conducted in the northwestern transitional zone adjacent to the northwestern Black Sea, northeastern Aegean and Central Anatolian regions to determine the distribution of landraces and the socioeconomic status of landrace cultivation. Landraces of hulled wheat, bean, chickpea and lentil were selected as the targets (Tan 2002; Tan and Açikgöz 2002).

- **Ecosystem conservation and management for threatened plant species**

The project was started in 2002. Its objective is the conservation and management of wetlands of steppe ecosystems, the habitats of endangered herbaceous plant species. *Sphaeropyza kotscyana*, *Stipa syreistchikowii* and *Thermopsis turcia* are among the target species which could be used as forage crops (Tan and Açikgöz 2002; Tan *et al.* 2003).

Multiplication and regeneration

The regeneration programme was continued during the period 1999-2001. Regenerated accessions are listed in Table 1. Recently, emphasis has been placed on annual clovers and medics. Original and regenerated/multiplied materials are kept separately at the Gene Bank.

Multiplication of the material collected in Central Anatolia for which the sample amounts were not sufficient was also carried out and their safety-duplicates were sent to AARI.

Table 1. Number of accessions regenerated at AARI since 1999

Genus	1999	2000	2001
Lathyrus	10	21	4
Vicia	29	146	40
Pisum	2	-	4
Trifolium	-	-	20
Astragalus	-	-	22
Medicago	-	-	2
Trigonella	-	-	1

Characterization and evaluation

Characterization and evaluation on *Vicia*, *Festuca*, *Medicago* and *Dactylis* have been carried out by AARI.

Material collected by CRIFC was characterized at the Haymana Research and Production Centre of the Institute for 11 characters for legumes and 9 characters for grasses. Significant variation was observed among and within accessions.

Characterization and evaluation on *Vicia*, *Festuca*, *Medicago*, *Dactylis*, *Bromus*, *Agropyron* and *Astragalus* have been carried out by CRIFC.

Characterization and evaluation on *Vicia*, *Dactylis, Medicago sativa, Onobrychis*, *Trifolium repens* and *T. pratense* have been carried out by the East Anatolia Agricultural Research Institute situated in Erzurum.

Utilization

Breeding programmes for *Vicia*, *Trifolium resupinatum* and *Lolium multiflorum* were initiated in the late 1960s. Based on AARI's forages collections, they have resulted so far in the improvement and registration of 11 cultivars of *Vicia sativa*, *V. villosa*, *V. pannonica*, *Trifolium resupinatum* and *Lolium multiflorum* which are adapted to coastal areas of the Aegean region.

Breeding programmes for *Medicago sativa* and *Sanguiosorba minor* were commenced in the late 1960s and were intensified on *Vicia* in 1988 at CRIFC. Fourteen cultivars of *Medicago sativa*, *Sanguisorba minor*, *Vicia sativa*, *V. villosa*, *V. pannonica*, *V. narbonensis* and *Lathyrus* *sativus* that are well adapted to Central Anatolian conditions have been improved and registered.

Breeding programmes for *Vicia*, *Dactylis*, *Medicago sativa*, *Onobrychis*, *Trifolium repens* and *T. pratense* were started in the early 1990s and are still going on at the East Anatolia Agricultural Research Institute.

Documentation

Data from surveying, collecting, characterization and regeneration of forage crop genetic resources are documented in a central database management system (Tan and Tan 1998).

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International cooperation for collecting missions

Forage collecting expeditions as a contribution to promote international cooperation and improvement of genetic diversity

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Introduction

In order to increase the genetic diversity of conserved forage germplasm and to close gaps in the existing collections (to hinder genetic erosion), a joint German/Irish expedition took place in Ireland in July/August 2002. The partners involved were the Genebank Department of the Institute of Plant Genetics and Crop Plant Research (IPK) and the Department of Crop Science, Horticulture and Forestry of University College Dublin (UCD). The expedition was a "multi-species" mission focusing on grass species which are currently used as forage crops or have the potential to contribute to the genetic improvement of forage crops in northwestern Europe. Particular emphasis was given to collecting wild material and ecotypes from locations that had not been seeded with modern varieties but which have been used for agricultural purposes without being subjected to intensive agricultural management such as fertilizer application or drainage.

Species collected included *Lolium perenne* L., *Festuca* spp., *Dactylis glomerata* L., *Agrostis* spp., *Phleum* spp. and *Poa* spp.

The goals of the expedition were to collect and conserve germplasm from the wild and the cultivated genepools of locally adapted forage species. The collected material will be propagated and will undergo preliminary characterization and/or evaluation. The intention is to make this germplasm freely available for seed exchange with scientists and breeders.

This report outlines the methodology of the collaborative expedition and presents an account of the collected materials.

The collecting expedition

- **Permission to collect**

The Department of Agriculture, Food and Rural Development (DAFRD), Office of the Controller of Plant Breeders' Rights at Leixlip, Co. Kildare is the institution responsible for granting the permit for the collecting expedition. Permission in the form of an "Agreement for the Acquisition of Material (AAM)" was obtained from DAFRD allowing the IPK Genebank, in association with Prof. Edward J. Walsh of University College Dublin (UCD), to carry out plant exploration in Ireland and to collect germplasm for crop improvement (see Annex I, pp. 105-106).

- **Conducting the expedition**

The expedition was planned with Dr V. Conolly, Teagasc, and Prof. E. Walsh, UCD. Exploration and collecting were carried out by IPK personnel accompanied by one or more of the Irish collaborators. Collecting was carried out in a diverse range of habitats (varying for soil type, drainage status, weather and agricultural use) located in the eastern (Counties Dublin and Kildare), midland (County Offaly), western (Counties Roscommon, Galway, Clare and Limerick) and southwestern (Counties Kerry and Cork) regions of Ireland. The travel route of the expedition is illustrated in Fig. 1.

Fig. 1. Travel route of the Ireland expedition.

Information on the location of old grassland and on where to collect ecotypes was generally provided by local people. The contribution and assistance of extension personnel from Teagasc was especially valuable. In many cases they introduced us to local farmers who were in a position to provide detailed information on grassland use and management at collecting sites. The collecting sites were identified on maps (1:50 000). Longitude and latitude data were obtained with a global positioning system (GPS) receiver. Altitude was determined using an altimeter. All data were recorded on a prepared site collecting sheet. Representative soil samples were taken at most collecting sites (20 samples per site with a soil corer set to a depth of 100 mm). These were subjected to laboratory analysis (pH: minimum 5.07 and maximum 8.69; carbon: minimum 3.42% and maximum12.30%; and sulphur content: minimum 0.046% and maximum 0.187%).

- **Germplasm collection, increase and distribution**

In accordance with the AAM, seed and vegetative material (tillers of single plants) were collected. It was not possible to collect seed in every case, because seed was not always ripe. A total of 266 accessions were collected (Table 1).

Table 1. Material collected by the joint German/Irish expedition in Ireland, 2002

Genus/species	No. of accessions
Lolium perenne L.	59
Festuca sp.	53
Dactylis glomerata L.	23
Agrostis sp.	37
Phleum sp.	6
Poa sp.	29
Other	59
Total	**266**

Each seed sample was divided in two parts, one for the Irish Genebank and the other for IPK. The seed samples will be used for true seed increases and first characterization (IPK) and evaluation (IPK, UCD, NUI (National University of Ireland)-Maynooth

In the case of vegetative material, the German partners will multiply it in Germany during 2003 and a portion of the seed harvested from it will be forwarded to the Irish Genebank.

When characterization and evaluation have been completed, the valuable new germplasm, together with its description, will be recorded in the plant genetic resource collections of IPK and Ireland.

- **Plans for future collecting**

There are some regions in Ireland (e.g. the Aran islands) where germplasm of grasses that are local ecotypes peculiar to that particular region can still be found. The reasons for this are varied: good adaptation to local edaphic and climatic conditions or special qualities and uses. Such germplasm would be interesting for genebanks. Furthermore, the southeastern region of Ireland (e.g. Counties Waterford and Wexford) was not included in the 2002 collecting expedition reported here. There is, therefore, need for further collecting of wild material/ecotypes in these regions.

Annex I. Agreement for the Acquisition of Material (AAM) between IPK Gatersleben and DAFRD Ireland

Agreement for the Acquisition of Material – (AAM)

between
Department of Agriculture, Food and Rural Development
Ireland
represented by
[redacted]
- hereinafter referred to as **DAFRD** -
and
IPK
Institut für Pflanzengenetik und Kulturpflanzenforschung
(Institute of Plant Genetics and Crop Plant Research)
Corrensstraße 3,06466 Gatersleben
Germany
represented by its Acting Director, [redacted],
and its Administrative Director [redacted]
- hereinafter referred to as **IPK** -

Preamble

1. The FAO Commission on Genetic Resources for Food and Agriculture is presently engaged in drawing up guidelines for the future exchange of germplasm to be used by the food and agricultural industries. Current negotiations centre on the adaptation of the International Undertaking on Plant Genetic Resources to the Biological Diversity Convention. Depending upon the results of these negotiations, the terms of this agreement are subject to future modification such that they comply with the norms to be established by the FAO's Revised International Undertaking with regard to Plant Genetic Resources and with other international norms agreed upon. The German government actively participates in these negotiations with the aim of ensuring that any future regime will facilitate the exchange and use of these world resources of inappreciable value, and the just and equal sharing of benefits derived from the commercial or other uses of the germplasm.
2. IPK is a governmental, non profit organisation, devoted to research on genetic resources. The genetic resources kept by the IPK Gene Bank are freely available and IPK will comply with the issues of the Agenda 21.

§ 1 Subject Matter of this Agreement

Subject matter of this agreement is a joint expedition to sites of Grasses species (ecotypes, land-races, old culture forms) in diverse regions of Ireland (to be organised by joint collaborators in Ireland) and the collection of samples of grasses germplasm. This Agreement regulates also the further use of the collected material.

§ 2 Terms for plant exploration

1. The collection will take place in various sites throughout Ireland (with the permission of all relevant land owners) between July 20th and August 10th, 2002.
2. Taxa which are to be collected, mainly grass species like *Festuca, Poa, Lolium, Dactylis, Phleum, Agrostis* ...
3. The collection will proceed in a way that does not endanger natural populations. In particular, only mature seeds will be collected, and in some cases tillers of plants in quantities compatible with the sustainable maintenance of the populations.
4. The group of scientists will consist of [redacted] (curator of the oil seeds and fodder plants) and [redacted] (assistant of the working group: Resources Genetics and Reproduction) both of IPK,a representative of DSV (Deutsche Saatveredelung Lipstadt-Bremen GmbH), [redacted], a representative of University College Dublin, Department of Crop Science, Horticulture and Forestry, [redacted], (who will liase with [redacted], Teagasc, Oak Park), a representative of Trinity College Dublin, Department of Botany, [redacted], who will also provide information and the necessary contacts for the success of the expedition.
   A representative of DAFRD may be present at any time.
5. Responsibility for the proposed project and collection vests with [redacted] of IPK. However, any necessary permits in accordance with the relevant laws of Ireland for visiting the collecting sites (especially if there are nature reserves) and for the collecting itself will be organised and obtained by [redacted]. It must be ensured, that the plant-material (seeds and living material) can be exported to Germany and transported directly by the German scientists at the departure.

§ 3 Reports and Costs

1. Participants of the project will send to the DAFRD within 3 months a report of the collecting, mentioning the collected taxa, their location and other relevant information.
2. The expedition will be financed in its entirety by IPK (including travelling costs and expenses payable to the Ireland representatives).

§ 4 Property in collected material and usage of collected material

1. The collected material will be shared between Ireland (National Genebank, for the Ireland National Collection) and the Gene Bank at IPK in equal parts for its safe preservation and investigation at both institutions.
2. Accessions collected will be multiplied, characterised and evaluated at the IPK Gene Bank and IPK External Station Malchow, for research, training and breeding by IPK and external Station Malchow staff. The information obtained from characterisation and evaluation of collections in Germany will be provided periodically to Ireland (DAFRD, UCD, TCD).
   Information obtained from similar work carried out in Ireland shall be made available to IPK, Germany.
3. No rights, in particular intellectual property rights, that limit access to and/or utilisation of the plant genetic resource collected or any material essentially derived from it shall be claimed by IPK. Any publication or intellectual property rights obtained by IPK on results using material collected will mention the origin of the material.
4. The IPK Gene Bank may pass on the material to third parties for the purposes of research, examination, training, presentation and breeding.
5. The IPK Gene Bank will only make the material accessible to third parties after these have made an obligation in writing to the IPK Gene Bank
   a) not to register any intellectual property rights to the unaltered material provided to them. This does not affect the right to register intellectual property rights to breeding results, provided these do not restrict the right of third parties, including the material provider, to use the unaltered material for the purposes of breeding;
   b) to name in publications, compiled with the use of the plant material provided, the IPK Gene Bank as the supplier of the material and of any possible data and research results;
   c) to provide the IPK Gene Bank with any new characterisation and evaluation data that is made available to them, as a rule three years after receipt of the material, and to concede the right free of charge to the cultivated plant bank to unrestricted simple use, particularly for storage in public databases, duplication and dissemination of this data unrestricted to time and place.

§ 5 Disclaimer

The DAFRD accepts no responsibility for the identity, quality, health, purity (genetic or mechanical), the phytosanitary state, objectionability or other properties of the material released.

§ 6 Miscellaneous

1. Any alteration, modification, amendment and completion of this agreement may be done in a written form only. If any clause or part of this agreement is determined to be unenforceable or nugatory by a court or tribunal of competent jurisdiction to make such a determination, the remainder of this agreement shall remain in full force and effect. In the case that the unenforceability or invalidity of a clause rests on its material, spatial or temporal extent the clause shall be valid in its largest and widest enforceable extent possible.
2. Both parties agree to replace an unenforceable or nugatory clause or part of this agreement by a new clause to be negotiated in good faith which covers the economic content of the invalid clause as far as possible. This may also be applicable in the case of a supplementary interpretation of the agreement.
3. This agreement shall be governed and construed in accordance with German Law and the parties hereto submit to the jurisdiction of the German Court which is locally competent in Gatersleben.

On behalf of the IPK:

Authorised signature   Date: 04.06.02

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[redacted]   [redacted]

On behalf of the Department of Agriculture, Food and Rural Development, IRELAND.

Authorised signature   Date: 22.05.02

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Joint collecting mission VIR/NGB in Karelia, 2001

Louise Bondo
Nordic Gene Bank (NGB), Alnarp, Sweden

The objective of the collecting mission was to collect local germplasm samples of crop plants and their wild relatives in the Karelian Republic (seeds of vegetables, cereals, forage grasses, fruits and berries).

The following participants took part in this joint mission:

- from VIR: Dr V.F. Chapurin, Department of Fodder Crops; Dr T.V. Buravtseva, Department of Grain Legumes; and Dr I.V. Seferova, Department of Grain Legumes;
- from NGB: Dr Seija Leena Lehtinen and Dr Pentti Kaarlo Matias Alanko, both NGB representatives from Finland.

The route of the exploration was as follows: St. Petersburg → Lodeynoye Pole → Olonets → Suojarvi → Sortavala → Lahdenpohja → Pitkyaranta → Petrozavodsk → Kondopoga → Medvezhyegorsk → Pudozh → St. Petersburg.

The mission took place from 14 August to 2 September 2001. Vegetation types covered included meadow areas, sandy beach, gardens, roadsides, lake shores and ditches.

A total of 133 accessions from 31 species were collected (Table 1).

Table 1. List of samples collected in Karelia in 2001

Species	No. of samples		Species	No. of samples
Agrostis gigantea	1		*Phalaris arundinacea*	1
Agrostis tenuis	4		*Phleum pratense*	12
Allium fistulosum	1		*Poa pratensis*	5
Allium sativum	1		*Rheum* sp.	1
Alopecurus pratensis	2		*Rosa acicularis*	1
Aronia mitschurinii	1		*Rumex acetosa*	10
Carum carvi	5		*Trifolium aureum*	2
Dactylis glomerata	17		*Trifolium hybridum.*	3
Echinocystis lobata	1		*Trifolium medium*	2
Festuca arundinacea	3		*Trifolium pratense.*	11
Festuca pratensis	2		*Trifolium repens*	2
Lathyrus maritimus	2		*Vicia cracca*	13
Lathyrus palustris	3		*Vicia sepium*	10
Lathyrus pratensis	12		*Vicia* sp.	1
Lupinus polyphyllus	1		*Vicia sylvatica*	1
Melilotus albus	2			
Total = 133				

Availability of material: the material is stored at NGB. After the forage working group accepts the material, it will be multiplied and distributed to all interested parties.

Plant collecting expedition in the Pyrenees mountains, Spain 2001

Ian D. Thomas[1], Ken H. Chorlton[1], Dewi Bowen[1] and Federico Fillat[2]

[1] *Genetic Resources Unit, Institute of Grassland and Environmental Research (IGER), Aberystwyth, United Kingdom*

[2] *Instituto Pirenaico de Ecologia, Consejo Superior de Investigaciones Científicas (CSIC), Jaca, Spain*

The Pyrenees run 430 km north-west to south-east across the neck of land separating France from Spain between the Atlantic Ocean and the Mediterranean Sea. The highest point is Pico d'Aneto at 3404 m. The Pyrenees are composed of slates, schists, limestone and granite.

The Atlantic influence penetrates southward across the low peaks of the Western Pyrenees while the Eastern Pyrenees are influenced by the Mediterranean.

The people of the Pyrenees traditionally have depended on agriculture for their livelihood. Typical Mediterranean products such as wine, vegetables and fruit predominate in the Eastern Pyrenees, while in the Western and Central Pyrenees, with their abundant rainfall, potatoes, sweet corn and forage crops are grown. Livestock management involves the seasonal movement of flocks of sheep and cows up and down the mountains, using the meadows of the valley bottoms and the alpine pastures of the higher altitudes, depending on snow cover.

Few young people have been willing to settle into the traditional agricultural system. Gradually, the less fertile areas have been deserted, and the landscape has been colonized by scrub or planted with trees. Local breeds of sheep and cows have been superseded by imported breeds, which are less well adapted to the climate and topography.

A search of the ECCDBs for all *Lolium* and *Trifolium repens* collections in Spain and Portugal showed that there were very few collections from the Pyrenees. Indeed there were no *T. repens* collections documented at all.

The Pyrenees are a diverse region with great variation in altitude, aspect, soil type, climate and agricultural practice. In addition, the traditional agricultural system is gradually changing. This combination of factors encouraged us to carry out a collecting expedition in the region.

The Institute of Grassland and Environmental Research, Aberystwyth, UK and the Instituto Pirenaico de Ecologia, Jaca, Spain carried out a joint plant collecting expedition in the Pyrenees from 21 May to 1 June 2001.

The plant collecting expedition had two main objectives:

- Collection of *Lolium* and *Trifolium* populations as vegetative units and their return to IGER (for seed regeneration) with no losses.
- Collection from a wide range of distinct ecological niches. Populations were collected therefore from a range of altitudes, aspects and slopes, macro and micro climates, soil types and agricultural and non-agricultural management strategies.

Seed was produced from the collected populations in 2002 and it is hoped to begin characterization and evaluation in 2003.

The Pyrenees provided us with material from a very wide range of habitats and management zones in a relatively small area. We would now like to extend our collecting activities further west into the wetter, more Atlantic-influenced climate of the Cordillera Cantabria, particularly the Picos de Europa, and we are planning to collaborate with colleagues from Oviedo University later this spring [2003].

Forage collecting activities - International cooperation (Poland, Slovakia, Czech Republic, Slovenia)

Grzegorz Żurek[1], Wiesław Podyma[2], Jan Schmidt[1], Pavol Hauptvogel[3], Jarmila Drobná[3], Vojtěch Holubec[4], Magdalena Ševčíková[5] and Vladimir Meglič[6]

[1] *Plant Breeding and Acclimatization Institute, Botanical Garden, Bydgoszcz, Poland*

[2] *Plant Breeding and Acclimatization Institute, National Centre for Plant Genetic Resources, Radzików, Poland*

[3] *Research Institute of Plant Production (RIPP), Piešťany, Slovakia*

[4] *Genebank Department, Research Institute for Crop Production (RICP), Prague-Ruzyne, Czech Republic*

[5] *OSEVA PRO Ltd., Grassland Research Station (GRS), Zubří, Czech Republic*

[6] *Agricultural Institute of Slovenia, Ljubljana, Slovenia*

During international collecting missions (2000-2001) 54 localities in Slovakia (Sudety Mts. and Lower Tatra Mts.) and the Czech Republic (Moravský Kras and Jeseníky) were visited and a total number of 500 accessions were collected (Table 1).

The most frequently collected species belonged to the genera *Festuca*, *Poa* and *Agrostis* for grasses and *Trifolium*, *Medicago* and *Vicia* for legumes.

The elevation of all visited localities was higher than 300 m and in some cases reached 1500 m or more (Lower Tatra Mts.). Some of the sites visited were located on limestone or rocky soils and were typical mountain pastures or subalpine meadows.

The Moravský Kras was a quite specific region with limestone soils. The number of species noted and specific vegetation type were quite different from those of the other regions visited. Species such as *Bromus erectus, Festuca valesiaca, Trifolium alpestre* or *Lembotropis nigricans* are quite specific to alkaline soils.

Table 1. Forage species collected during international missions (2000-2001)

No.	Genus/species	Collecting year		Total per species
		2000	2001	
Grasses				
1	*Agrostis alpina*	1		1
2	*Agrostis canina*	1		1
3	*Agrostis capillaris*	8	7	15
4	*Agrostis gigantea*	4	3	7
5	*Agrostis rupestris*		2	2
6	*Agrostis stolonifera*	1	3	4
7	*Alopecurus pratensis*	3	3	6
8	*Anthoxanthum alpinum*	2		2
9	*Arrhenatherum elatius*	5	11	16
10	*Avenella flexuosa*	6	3	9
11	*Brachypodium pinnatum*		3	3
12	*Brachypodium sylvaticum*	1		1
13	*Briza media*	4		4
14	*Bromus benekenii*	2	2	4
15	*Bromus erectus*		2	2
16	*Calamagrostis arundinacea*	2	1	3
17	*Calamagrostis* sp.	1		1
18	*Calamagrostis villosa*	4	1	5
19	*Cynosurus cristatus*	2	4	6
20	*Dactylis glomerata*	9	11	20
21	*Dactylis glomerata* subsp. *slovenica*	1		1
22	*Dactylis polygama*		1	1
23	*Deschampsia caespitosa*	9	8	17
24	*Elymus caninus*		2	2
25	*Elymus repens*		1	1
26	*Festuca altissima*	1	1	2
27	*Festuca arundinacea*		3	3
28	*Festuca gigantea*	2	1	3
29	*Festuca nigrescens*		2	2
30	*Festuca ovina*		1	1
31	*Festuca pratensis*	11	12	23
32	*Festuca rubra*	6	12	18
33	*Festuca rupicola*		2	2
34	*Festuca supina*	3	2	5
35	*Festuca tenuifolia*		1	1
36	*Helictotrichon pratense*		1	1
37	*Helictotrichon* sp.		2	2
38	*Holcus lanatus*		1	1
39	*Holcus mollis*	1		1
40	*Hordelymus europaeus*		2	2
41	*Koeleria macrantha*		2	2
42	*Koeleria* sp.		2	2
43	*Lolium perenne*	6	10	16
44	*Melica ciliata*		1	1
45	*Melica transsilvanica*		1	1
46	*Melica uniflora*		1	1
47	*Milium effusum*	2	1	3
48	*Molinia arundinacea*		1	1
49	*Molinia caerulea*	1		1
50	*Nardus stricta*	1		1
51	*Phalaris arundinacea*	1	1	2
52	*Phleum alpinum*	4		4
53	*Phleum phleoides*		2	2
54	*Phleum pratense*	8	8	16
55	*Phleum pratense* subsp. *bertolonii*		1	1
56	*Poa alpina*	1		1
57	*Poa angustifolia*		1	1
58	*Poa annua*	1		1
59	*Poa chaixii*	1	1	2

Table 1 (cont.). Forage species collected during international missions (2000-2001)

No.	Genus/species	Collecting year		Total per species
		2000	2001	
60	*Poa compressa*	2	5	7
61	*Poa nemoralis*	6	3	9
62	*Poa palustris*	1	3	4
63	*Poa pratensis*	7	10	17
64	*Poa riphaea*	1		1
65	*Poa* sp.	1	1	2
66	*Sesleria caerulea*		1	1
67	*Sesleria* sp.		1	1
68	*Setaria pumila*		1	1
69	*Trisetum flavescens*	4	8	12
Total grasses		**138**	**177**	**315**
Legumes				
1	*Anthyllis vulneraria*	3	10	13
2	*Astragalus glycyphyllos*	1	6	7
3	*Coronilla varia*		5	5
4	*Lathyrus latifolius*		1	1
5	*Lathyrus niger*		2	2
6	*Lathyrus pratensis*	6	5	11
7	*Lathyrus sylvestris*	1	1	2
8	*Lembotropis nigricans*		1	1
9	*Lotus corniculatus*	7	14	21
10	*Lupinus polyphyllus*		2	2
11	*Medicago falcata*		8	8
12	*Medicago lupulina*	4	8	12
13	*Medicago sativa*		3	3
14	*Medicago varia*		6	6
15	*Melilotus alba*	1	5	6
16	*Melilotus officinalis*		2	2
17	*Melilotus* sp.		1	1
18	*Onobrychis viciifolia*		2	2
19	*Trifolium alpestre*		2	2
20	*Trifolium aureum*	2	3	5
21	*Trifolium campestre*	1		1
22	*Trifolium hybridum*	4	3	7
23	*Trifolium medium*	1	4	5
24	*Trifolium montanum*		7	7
25	*Trifolium pratense*	6	9	15
26	*Trifolium repens*	13	5	18
27	*Vicia cracca*	5	7	12
28	*Vicia faba*		1	1
29	*Vicia hirsuta*		1	1
30	*Vicia sepium*	4	2	6
Total legumes		59	126	185
Total forages (grasses + legumes)		**197**	**303**	**500**

Minimum standards for regeneration

ICONFORS: Improving germplasm conservation methods for perennial European forage species

Maurice Hinton-Jones, Kenneth H. Chorlton, Ian D. Thomas, Athole H. Marshall and Mervyn O. Humphreys

Institute of Grassland and Environmental Research (IGER), Aberystwyth, United Kingdom

The Project aims to acquire the genetic and economic knowledge required to improve seed multiplication methodologies for genebanks which maintain *ex situ* seed collections of perennial European forage species. The results and conclusions will also be generally applicable to all genebanks, particularly to seed collections of outbreeding species. Experiments on gene flow within and between plots will also have broader relevance to *in situ* conservation of biodiversity and will help to inform the continuing debates about the risks of GM crops.

Research background

Poor conservation of plant germplasm collections has been highlighted as a global problem (FAO 1999), both by the CBD (Convention on Biological Diversity) in article 9, and by the CGRFA (Commission on Genetic Resources for Food and Agriculture) in its report on the SWPGR (State of the World's Plant Genetic Resources). In consultation with governments and genebanks worldwide, the CGRFA has developed a GPA (Global Plan of Action for the Conservation and Sustainable Use of Plant Genetic Resources for Food and Agriculture) (FAO 1999), in which it details recommended actions to improve the effectiveness of genebanks. ICONFORS addresses two of the recommended actions of the GPA: Activity 5 "Sustaining existing *ex situ* collections" and Activity 6 "Regenerating threatened *ex situ* accessions".

Within Europe, nearly 100 000 populations of forages are conserved as seed samples (accessions) in genebanks, for possible use by plant breeders in reconstituting native pastures and to facilitate research in a broad range of areas such as functional genomics and conservation genetics (Marum *et al.* 1998.). Seeds cannot be stored indefinitely: they gradually senesce even in the best storage conditions. It is estimated that over 20 000 of the accessions are now in urgent need of rejuvenation through a cycle of seed multiplication. Establishing a standardized Europe-wide protocol and plan for rejuvenating these accessions has been a priority activity of the Forages Network of the ECP/GR (European Cooperative Programme on Crop Genetic Resources Networks) (Sackville Hamilton *et al.* 1997; Sackville Hamilton 1998; Maggioni *et al.* 1998; Sackville Hamilton *et al.* 1998). However, our attempts until now have been hindered by the realization that we do not understand the genetics and economics of regeneration well enough to establish appropriate standards (Sackville Hamilton *et al.* 1998). Here, we describe the steps which we are taking to improve our understanding of this area of knowledge.

Partners

1. Institute of Grassland and Environmental Research, Plas Gogerddan, Aberystwyth, Ceredigion, UK, SY23 3EB
2. The Norwegian Crop Research Institute, Løken Research Station, 2940 Heggenes, Norway
3. Oseva PRO Ltd., Grassland Research Station Roznov-Zubří, 756 54 Zubří 698, Czech Republic

4. Centre for Genetic Resources, The Netherlands, Plant Research International B.V. (PRI), Wageningen University and Research Centre, PO Box 16, 6700 AA Wageningen, The Netherlands
5. Estação Nacional de Melhoramento de Plantes, Apartado 6, 7351 Elvas, Portugal
6. DLF-Trifolium Research and Development, PO Box 19, DK-4660 St. Heddinge, Denmark

The challenge

Perennial forage species are mostly outbreeders. Each sample of seed is a genetically variable mixture of genotypes (Sackville Hamilton and Chorlton 1997)). A new generation of young healthy seed is produced by growing a sample of the original and allowing the plants to pollinate each other. The new generation will be genetically different from the original (Sackville Hamilton and Chorlton 1997; Sackville Hamilton 1998; Sackville Hamilton *et al.* 1998), for three reasons: genetic drift, genetic shift and contamination.

Such genetic changes are directly counter to the objectives of genebanks, namely to conserve genetic variation intact. We cannot totally eliminate change. How can we minimize it?

The situation is complicated by economics. We work with limited budgets. If we try too hard to maintain genetic stasis, we reduce the number of accessions that we can process, and so we may reduce the effectiveness of the genebank. Conversely, if we relax regeneration standards too much in order to increase the number of accessions processed, we also reduce the effectiveness of the genebank. We need to seek and adopt the optimum compromise between **quality** of regeneration for each accession and the **quantity** of accessions regenerated (Sackville Hamilton and Chorlton 1997; Sackville Hamilton 1998; Sackville Hamilton *et al.* 1998).

Options for regeneration

Numerous options are available for regeneration, although their genetic and economic implications are often poorly understood (Sackville Hamilton and Chorlton 1997). The major options being considered in ICONFORS are:

- **Environment used for regeneration.** How important is it to regenerate each accession in the "right" place? How important is geographical proximity to the location where the accession was originally collected?
- **Regenerate in open field plots or in isolation chambers?** How effectively can pollen contamination be prevented in open field plots? How economic are isolation chambers compared with field plots?
- **Isolation distance.** If accessions are regenerated in open field plots, how far apart should the plots be to achieve adequate isolation?
- **Harvest plants of one accession together or separately?** Separate harvesting enables a reduction in shift and drift, e.g. by taking a "balanced bulk" (mixing an equal number of seeds from each mother plant) but takes more work. Is this additional effort worthwhile?
- **Number of plants used.** Using more plants as parents helps to reduce genetic drift by the "founder effect", but increases the time and cost per accession and therefore may reduce the total number of plants that can be regenerated. Is there an optimum compromise? Some regeneration options, such as using balanced bulks, require additional investment per plant - how much does this affect the optimum number of plants?

Expected achievements of the Project

We expect that the project will eliminate (or at least develop the technology required to eliminate) many of the major remaining gaps in knowledge about the genetic and economic implications of different approaches to multiplying seed of germplasm collections of perennial forage species in Europe (Sackville Hamilton *et al.* 1998). We will do this for two wind-pollinated grasses (perennial ryegrass and meadow fescue) and an insect-pollinated legume (white clover).

Project Workpackages

The project contains six experimental workpackages:

- WP1 and WP2 aim to assess the variation in seed production between plants within populations while providing a comprehensive assessment of economic factors.
- WP3 forms the second phase of trial work carried out in WP2 and attempts to estimate the broad sense heritability of within-plot variation in seed production and of genotype × year interactions.
- WP4 and WP5 assess patterns of pollen-mediated gene flow within plots and the resulting variation in pollination between plants within populations.
- WP6 aims to estimate the effects of species and multiplication site on pollen contamination in field regeneration plots.

Progress to date

Workpackage 1

This workpackage aims to estimate the effect of the site of multiplication on the cost of multiplication, the magnitude of variation in seed production among plants within plots, and the rank order for seed production of genotypes within populations, and the dependence of these on species and adaptation of the population being multiplied.

The initial phase of WP1 is nearing completion (details are included in the following section). Seeds of 10 native populations of *Lolium perenne* and *Trifolium repens* were sown in the U.K. in controlled environmental conditions in order to provide enough cloned plantlets of both species for distribution to partners. Management protocols were devised and distributed to all partners, establishing "common" field regeneration methodologies. This included data collection, assessment of optimum harvest time and techniques for the subsequent drying and threshing of samples for seed weights.

At each of 4 sites, 10 *L. perenne* and 10 *T. repens* populations in 2 replicate regeneration plots containing 7 x 7 = 49 plants, with 0.5 m and 1 m spacing respectively within a plot and a 30 m distance between adjacent plots, were planted within a barrier cereal crop appropriate to each environment. Post-harvest, samples were dried, threshed and stored in desiccators. Seed samples were weighed and data have been collated by IGER for statistical analysis.

Results thus far

Preliminary statistical analysis has been carried out on seed yield per genotype across populations and sites. From initial results, there are some substantial replicate differences at certain sites. This could cause complications in interpreting the results and formulating recommendations for future regeneration. There are also significant differences in seed yield obtained from the same genotype in different sites. Partners are currently completing an economic analysis of the costs associated with the different processes of seed regeneration. This will enable the cost of regeneration to be quantified if seed production were to be carried out on a contractual basis in future. Comparisons of the cost of regeneration with statistical analysis on the magnitude of variation of seed production should provide a valuable insight for future regeneration methodologies.

Other workpackages

Data pertaining to WP2 are currently being compiled and should be completed before the end of April 2003 in preparation for analysis, which will also include an economic breakdown for each regeneration process.

Work on WP4 continues in order to develop the technology required for paternity identification in WP5.

In WP6, screening of *Trifolium repens* progeny seed from the 2002 harvest continues and will be completed by the end of June 2003.

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Experiences with the new regeneration strategy at the Nordic Gene Bank

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Introduction

The forage working group at NGB decided in 1999 to improve the quality of the regeneration and rejuvenation procedures for forage seed in the Nordic Gene Bank. The new NGB protocol was based on the recommendations from Sackville Hamilton and Chorlton (1997). The forage working group at NGB has followed the new protocol since 1999, so we now consider it worth while to evaluate our experiences.

As shown in Table 1, each regenerated accession must give at least 20 000 living seeds and each rejuvenated accession must give at least 40 000 living seeds. It is much more difficult to decide on exact guidelines for the minimum germination ability. In general it is preferred that the germination ability should not be below 75-80%.

Table 1. Number of seeds in each collection

Collection	Purpose	No. of seeds
Active	Distribution, evaluation, characterization, regeneration, germination tests and other purposes	19 500
Base	Conservation and rejuvenation	20 000
Safe	Conservation	500
Total		**40 000**

We assume that it will take about 30 years before germination falls below the established lower limit for germination ability. During these 30 years enough seeds must be harvested to cover all purposes mentioned in Table 1.

Sources of seeds to be regenerated

The Nordic forage working group has decided to distinguish between regeneration of the active collection, rejuvenation of the base collection and multiplication of collected seeds. The active collection and newly collected accessions are regenerated by bulk harvest of at least 30 plants. The base collection is rejuvenated by single plant harvest of at least 49 plants and the compiling of a balanced bulk.

Table 2 shows which collections can be used as sources of seeds for the regeneration of the active collection or rejuvenation of the base collection.

Table 2. Choice of source collections for regeneration of the active collection or rejuvenation of the base collection

		Receiving collection	
		Active	**Base**
Source collection	**Active**	Regeneration	Unacceptable
	Base	Rejuvenation	Rejuvenation

Before the regeneration/rejuvenation procedure can start, the accession must fulfil the following criteria: the total number of living seeds in active and base collections must be lower than 1000 or the germination ability must be below 50%.

Experiences with the new protocol

As expected, it is not cost-free to work according to the new protocol, which involves more administration at NGB, higher costs for NGB and is more time-consuming. The higher costs reduce the number of accessions that can be regenerated or rejuvenated each year. But the main problem or bottleneck is the limited availability of qualified labour at the field stations. Since it is more time-consuming to follow the new protocol, it takes more of our limited resources: qualified workers' time and money.

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Sackville Hamilton, N.R. and K.H. Chorlton. 1997. Regeneration of accessions in seed collections: a decision guide. IPGRI/FAO Handbooks for Genebanks 5. International Plant Genetic Resources Institute, Rome, Italy.

Core collections

*Report of the subgroup "*Medicago sativa *core collections"*

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A bibliographic search on the development of core collections carried out at the beginning of 2003 yielded 79 records. Among them only 10 concerned forages and only one the *Medicago sativa* complex (Basigalup *et al.* 1995). About 1100 perennial *Medicago* Plant Introductions (PIs) collected from 47 countries were classified into 18 geographical groups based on passport data from the USDA Germplasm Resources Information Network (GRIN) System. Eight methods for developing a 200-entry core were compared by non-parametric procedures with the original PI collection. The methods included multivariate procedures, random and/or directed selection of entries, and a totally randomly selected core. Two methods were the best for designating a core collection: one method combined cluster analysis based on principal components within each geographical group with random selection of entries within each cluster; the other method was direct selection of entries within each geographical group. The former needs a complete data set and extensive computer facilities, whereas the latter can utilize incomplete data sets and does not require computer facilities.

The above findings underline the need to carry out more extensive studies on the criteria to develop alfalfa core collections. The criteria developed for other species cannot be directly applied to this species because of its autotetraploid nature. Furthermore, the extent of genetic variation in this forage species, so important for temperate regions, is still poorly understood and should be better elucidated. Genetic markers based on PCR techniques (AFLP, SAMPL, S-SAP, etc.) which yield reproducible and reliable results at a relatively low cost could easily be used for the purpose.

However, at the present state of knowledge, the selection of accessions within geographical groups appears to be the most reliable, convenient and time-saving method for developing a European *Medicago sativa* core collection.

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Basigalup, D.H., D.K. Barnes and R.E. Stucker. 1995. Development of a core collection for perennial *Medicago* plant introductions. Crop Sci. 35:1163-1168.

Study of genetic diversity between and within ryegrass populations of the ECP/GR collection by means of AFLP markers

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Introduction

Management of genebanks depends on good categorization of the material in the collection. In recent years, genebank holders often make use of additional genetic analysis based on molecular markers, the better to describe the genetic variation available in their collections. Although not cheap, the use of molecular characterization of the material can be an alternative to extensive field evaluation. In this study we wanted to compare genetic characterization by the use of AFLP markers with large-scale field evaluation of the *Lolium* ECP/GR collection. Although set up as an introductory study, as only a limited number of genotypes per accession (10 plants) was analyzed – even to describe properly the genetic variation in ryegrass varieties, which are assumed to be more narrow than natural populations, 30 to 50 plants need to be analyzed –, this effort required a substantial study because of the large number of accessions involved.

Material and methods

- **Plant material, DNA isolation, AFLP reactions and PAGE**

Within the ECP/GR collection (162 accessions under trial in 1996 and 1997 at the Belgian testing site), 80 accessions (Table 1) were selected. Per accession, 10 individual plants were sampled; leaf material was cut, immediately immersed in liquid nitrogen and lyophilized. The DNA isolation was based on a CTAB method (Doyle and Doyle 1987). Leaf material (±200 mg fresh weight) was freeze-dried and ground (Culatti micromill, Prosep) and 1 ml extraction buffer was added: Tris-HCl 100 mM pH 8.0, EDTA 20 mM, NaCl 1.4 M, PVP 1%, $Na_2S_2O_5$ 0.5 M, CTAB 2% and β-mercaptoEtOH 0.4%. After 90 minutes incubation at 60°C, 500 µl chloroform/isoamylalcohol (24/1) was added, the tubes were shaken during 5 min. and centrifuged for 10 min. at 7000 rpm. The DNA was picked up with a Pasteur pipette after precipitation by adding 900 µl isopropanol to the supernatant. Then it was washed twice with 76% EtOH + 10mM NH_4OAc, dried and resolved in H_2O. The DNA concentration was defined by comparing with a standard series on 1.5% agarose gels 1x TAE. AFLP reactions (Vos *et al.* 1995) were run on an ABI Prism 377 DNA Sequencer using the commercially available kit for fluorescent fragment detection (Perkin-Elmer 1995). *Eco*RI and *Mse*I were used for DNA digestion. Selective amplification was done using 5 fluorescent labelled *Eco*RI-*Mse*I primer combinations with 6 selective bases: *Eco*RI-AGG + *Mse*I-CTT, *Eco*RI-AGC + *Mse*I-CTT, *Eco*RI-ACC + *Mse*I-CAT, *Eco*RI-ACC + *Mse*I-CAA, *Eco*RI-AAC + *Mse*I-CAT.

Morphological and agronomic data were collected on the Belgian test site during 1996 and 1997 according to the guidelines provided.

Table 1. Identification of the ECP/GR accessions included in the genetic diversity study

	Accession	Country	No.	Code
1	A001	Belgium	554	Bel554
2	A002	Belgium	750	Bel750
3	A003	Belgium	751	Bel751
4	A004	Belgium	757	Bel757
5	A005	Belgium	891	Bel891
6	A006	Germany	GR 5028/94	Ger5028
7	A007	Germany	GR 5029/94	Ger5029
8	A014	Germany	GR 5041/94	Ger5041
9	A015	Germany	GR 5042	Ger5042
10	A016	Germany	GR 2859/94	Ger2859
11	A025	Spain	6	Sp6
12	A026	Spain	7	Sp7
13	A028	Spain	9	Sp9
14	A029	Spain	10	Sp10
15	A030	Switzerland	ABY-BA 12219.00	Swit19
16	A031	Switzerland	ABY-BA 12220.00	Swit20
17	A032	Switzerland	ABY-BA 12221.00	Swit21
18	A033	Switzerland	ABY-BA 12222.00	Swit22
19	A034	Czech Republic	ABY-BA 12275.00	Tsjech75
20	A035	Czech Republic	ABY-BA 12276.00	Tsjech76
21	A036	UK	ABY-BA 9790.94	UK790
22	A037	UK	ABY-BA 9791.94	UK791
23	A038	UK	ABY-BA 9792.94	UK792
24	A044	UK	ABY-BA 9819.94	UK819
25	A051	UK	ABY-BA 9960.94	UK960
26	A059	Romania	1:3035 - Jibert	Rom1
27	A061	Romania	3:3028 - Beclean Pesomes	Rom3
28	A065	Romania	7:3026 - Viseul De Sus	Rom7
29	A066	Romania	8:3085 - Stupini	Rom8
30	A069	Romania	11:3091 - Sighisoara	Rom11
31	A073	Romania	15:3326 - Simleul Silvaniei	Rom15
32	A074	Hungary	IV-51-166 Kadarkut	Hun66
33	A077	Hungary	IV-51-173 Martonvasar	Hun73
34	A079	Hungary	IV-51-177 Bakonybank	Hun77
35	A081	Hungary	IV-51-181 Asvanyrard	Hun81
36	A083	Hungary	IV-51-183 Dunatetetlen	Hun83
37	A085	Hungary	IV-51-185 Kigyos	Hun85
38	A087	Poland	130533	Pol0533
39	A089	Poland	130878	Pol0878
40	A093	Poland	131712	Pol1712
41	A097	Poland	132079	Pol2079
42	A099	Poland	132126	Pol2126
43	A101	Bulgaria	92 E1	Bul1
44	A102	Bulgaria	92 E6	Bul6
45	A105	Bulgaria	92 E10	Bul10
46	A106	Bulgaria	92 E13	Bul13
47	A107	Bulgaria	92 E14	Bul14
48	A109	Greece	M-15140	Gr5140
49	A110	Greece	M-15478	Gr5478
50	A111	Greece	M-16181	Gr6181
51	A112	Greece	M-16185	Gr6185
52	A113	Greece	M-17226	Gr7226
53	A116	Italy	3407 Balvano Cluster B	Ita07
54	A117	Italy	3408 Sepino Cluster B	Ita08
55	A118	Italy	3409 Rieti Cluster C	Ita09
56	A119	Italy	3410 Papasirero Cluster C	Ita10
57	A120	NGB	4090 Valinge (Swe)	Swe4090
58	A121	NGB	4266 16-7-62-2 (No)	No4266
59	A122	NGB	5040 MH8309230101 (DK)	Dk5040

Table 1 (cont.). Identification of the ECP/GR accessions included in the genetic diversity study

	Accession	Country	No.	Code
60	A123	NGB	8476 MH8505080201 (DK)	Dk8476
61	A124	The Netherlands	CGN 15662	Neth2
62	A126	The Netherlands	CGN 15664	Neth4
63	A127	The Netherlands	CGN 15665	Neth5
64	A128	The Netherlands	CGN 15666	Neth6
65	A130	The Netherlands	CGN 15668	Neth8
66	A131	Ireland	81401	Ire1401
67	A133	Ireland	81414	Ire1414
68	A134	Ireland	81624	Ire1624
69	A135	Ireland	81629	Ire1629
70	A137	Ireland	82240	Ire2240
71	A138	France	010111	Fra0111
72	A144	France	010316	Fra0316
73	A149	France	010604	Fra0604
74	A153	France	010769	Fra0769
75	A154	France	010879	Fra0879
76	A159		011104	Fra1104
77	A163			Arion
78	A164			Frances
79	A165			Talbot
80	A166			Vigor

- **Statistical analyses**

Filters for AFLP marker selection were set (De Riek *et al.* 1999) for marker signal intensity and marker frequency: average signal peak height of a marker >75 and frequency of a marker in the whole data set higher than 0.15. Two kinds of parameters were applied to calculate the genetic variation in the data set: (1) the genetic conformity as expressed by different similarity/distance coefficients; and (2) the molecular variance as obtained from the Analysis of Molecular Variance (AMOVA; Excoffier *et al*. 1992). This kind of analysis took a Euclidean distance matrix between individual plant genotypes as an input. AMOVA also generates a population pairwise Phi-statistic (Phist) for which the probability is tested by permutation analysis. Calculation of Nei genetic distance and standard errors, construction of dendrograms (UPGMA) and bootstrapping were performed by Dispan (Nei 1972, 1978; Ota 1993). Calculation of other similarity coefficients, construction of dendrograms (UPGMA) and principal coordinate analysis (PCO) were performed by the modules SIMIL, CLUSTER and PCOORD of the "R package" (Legendre and Vaudor 1991) and with the MVSP package (Kovach Computing Services, UK).

Results and discussion

Scoring of the AFLP patterns for the 800 individual plants using the automatic scoring settings as defined yielded a dataset with 737 markers. Two types of data analysis were applied: on the marker frequency data, taking into account the average presence of each marker into an accession, and on the individual plant data.

- **Data analysis based on marker frequencies per accession**

Starting from marker frequency data, taking all plants of an accession as a group, Nei standard genetic distances and Euclidean distances were calculated. Fig. 1 shows the relationships revealed between the accessions for the Nei standard genetic distance (Euclidean distances gave an almost identical dendrogram: data not shown). Bootstrapping (100 datasets) was applied to test the reproducibility of the grouping below each node; bootstrap values are also indicated on the dendrogram. Although we observed a slight tendency towards groupings of accessions belonging to the same geographical area, these clusters are not sustained by high bootstrap values. Only accessions grouped at the lower genetic distances sometimes attain reliable bootstrap values.

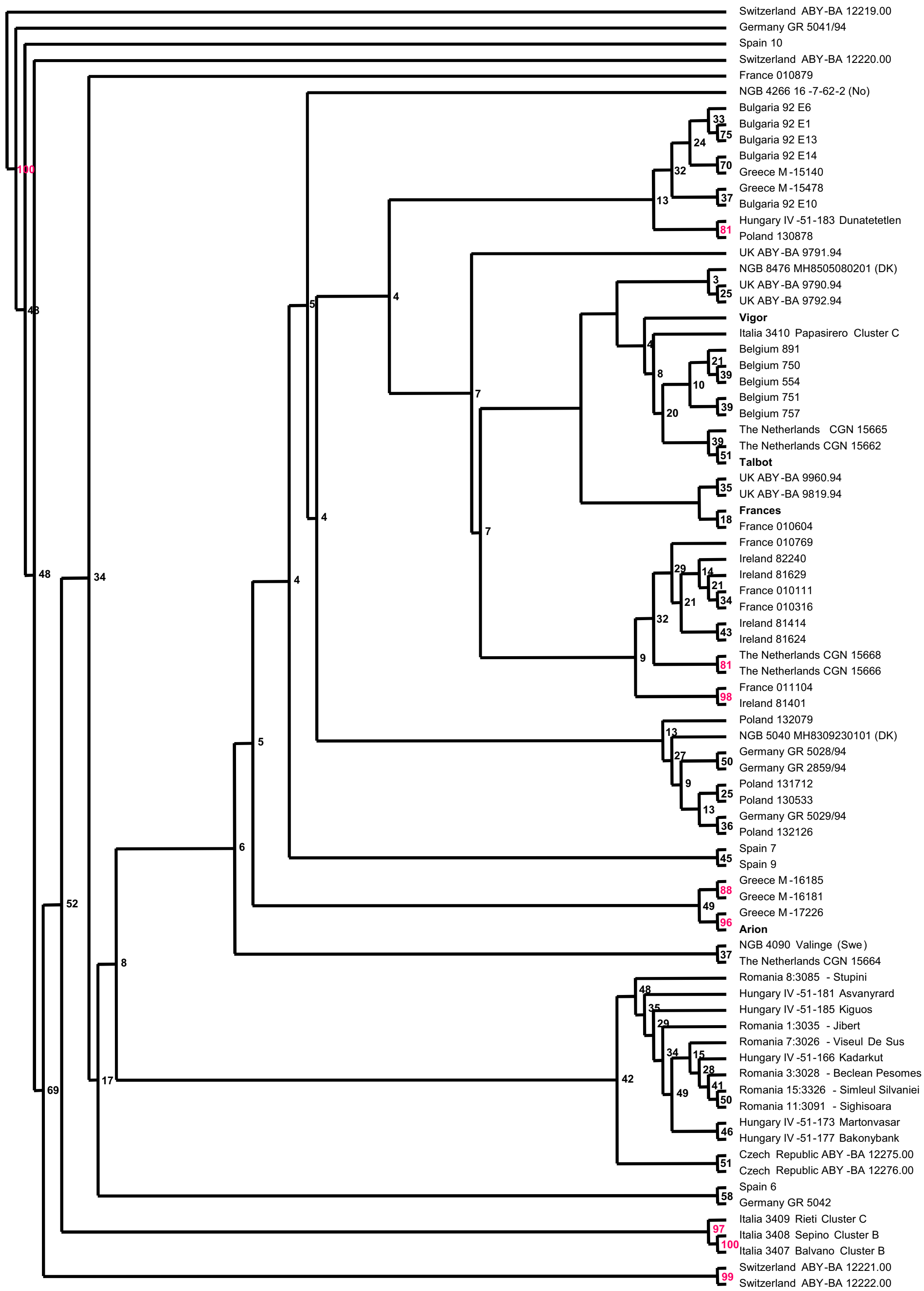

Fig. 1. Ranking of the standard genetic distance between accessions (UPGMA clustering) with indication of bootstrap values on the dendrogram nodes (100 replicated datasets). The branch lengths do not reflect the actual genetic distances.

To reveal better the underlying genetic structure, principal coordinate analysis (PCO) was applied on the matrix of genetic distances. A biplot showing the first and second axis, revealing respectively 7.2% and 12.3% of the variation, is shown in Fig. 2. From the plot it can be observed that axis 1 merely separates East European accessions from West European ones; axis 2 in a similar way separates North from South European accessions. In this way, a number of regional groups can be identified: two East-Central-European clusters (Romania, Hungary and Czech Republic; Bulgaria and Greece); a German-Polish group; a group of Southern Europe (Italy, Spain); a Nordic group; and a broad West-European group (Belgium, the Netherlands, Ireland, France and the UK). The reference varieties tend to cluster with the West-European group.

As a conclusion, it can be stated that ranking of the populations by PCO plots and dendrograms revealed a genetic structure that corresponds to the geographical localization of the populations. However, the low bootstrap values indicate low redundancy within the dataset, making this grouping rather loose and much influenced by the actual composition of the AFLP dataset.

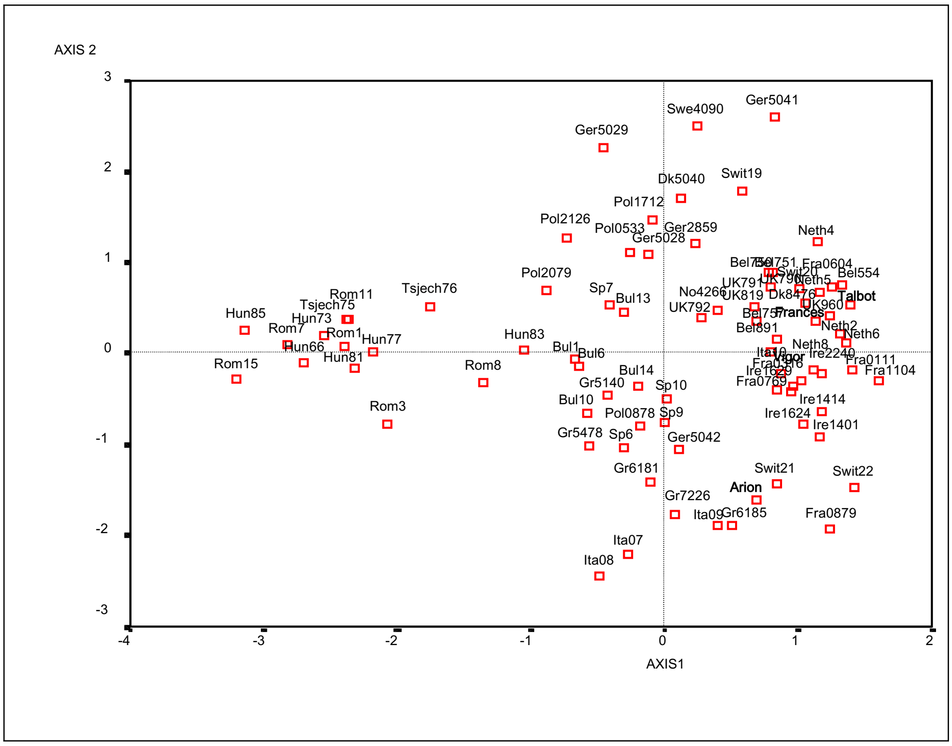

Fig. 2. PCO biplot based on the Euclidean distance between marker frequency of the different accessions.

- **Analysis of Molecular Variance, based on individual plant data**

A low redundancy in the dataset, as revealed by the low bootstrap values when calculating genetic distances based on marker frequency data, can be indicative of considerable variation within the accessions. This can be studied by applying analysis of molecular variance (AMOVA, Schneider *et al.* 2000). AMOVA was applied (Schneider *et al.* 2000) on the Euclidean distance matrix between individual genotypes to attribute the distribution of genetic variation to different sources (breeding programmes, varieties and seed lots).. AMOVA is a statistical tool originally developed for population genetics (Excoffier *et al.* 1992) and is based on the scheme of analysis of variance. The AMOVA procedure provides a general framework for the analysis of population genetic structure based on any distance matrix (Euclidean distances were used) between individual plant genotypes.

AMOVA allows testing genetic structures with up to two levels, e.g. (1) accession and (2) geographical location. A grouping according to geographical location was based on the coordinates of the collecting sites (kindly provided by R. Sackville Hamilton, IGER). In Fig. 3, the group structure for geographical regions is given. The AMOVA procedure then computes the different variance components: between geographical regions, within regions but between accessions, and within accessions. Table 2 lists the results: only 2.54% of the total variation in the dataset can be attributed to differences between geographical regions, 3.65% of the variation corresponds to differences between accessions (within regions) but the major part of the variation (93.80%) is based on differences within populations.

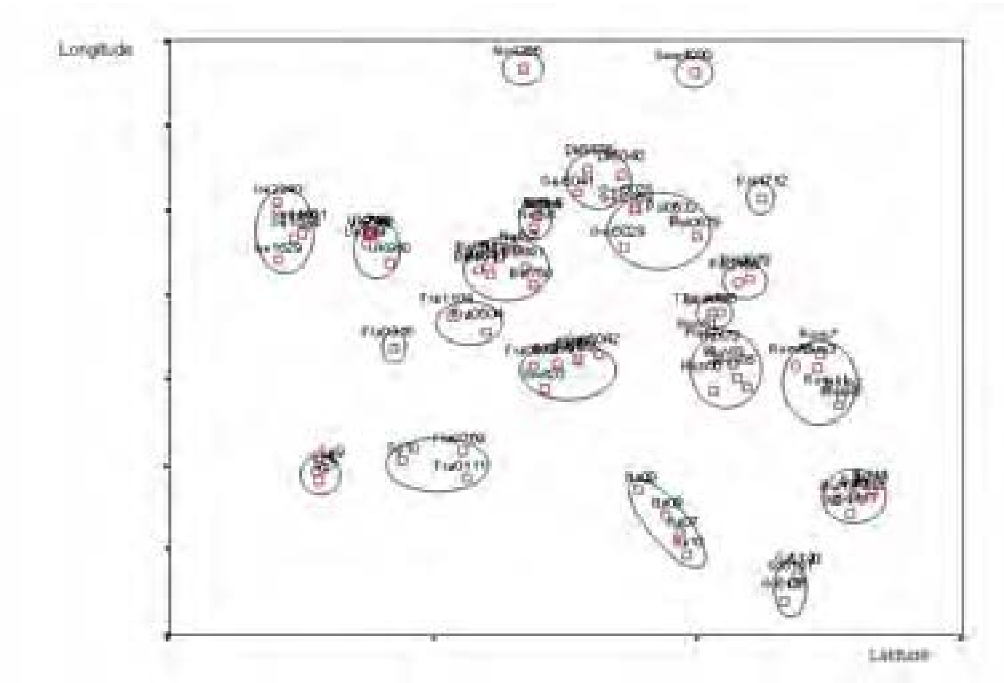

Fig. 3. AMOVA grouping based on geographical region.

Table 2. AMOVA structure and results

Source of variance	Degree of freedom	Sum of squares	Variance components	Percentage of the variance explained
Between regions	16	4319.942	3.01544**	2.54
Between accessions within regions	52	7964.010	4.33323**	3.65
Within accessions	596	66528.600	111.25184**	93.80
Total	**666**	**78812.552**	**118.60050**	

** Significant at the 0.05 level, evaluated by 1000 permutations

In addition, AMOVA generates a population pair-wise F-statistic (comparable to e.g. the Nei genetic distance calculated from marker frequency data) for which the probability is tested by permutation analysis. Although this approach is computationally more complicated than the use of marker frequency data, it also takes the variation within the group of genotypes into account. The corresponding dendrogram is shown in Fig. 4. Although different in details of the clustering, the general structure as revealed by Figs. 1 and 2 is maintained.

From the AMOVA analysis it can be concluded that because the major part of the genetic variation has to be attributed to differences between individual plants, any groupings based on overall genetic distances are apt to be very unreliable. Nevertheless, the genetic differentiation between populations and geographical regions does exist but is more or less blurred by the vast genetic variation within the accessions.

- **Comparison with morphological and agronomic data**

Evaluation of the morphology and the agronomic performance of the different accessions was the major goal of this ECP/GR trial. Therefore, it is tempting to look for correspondences between the classical field evaluations and the genetic analysis performed. Nevertheless, the ways of handling data are very different: field trials end up with analysis of variance and significant differences for a certain set of traits, whereas genetic analysis takes a multivariate approach. To enable a comparison, the typical trait-by-trait analysis on the field data was transformed into a kind of distance matrix by counting the number of significantly different parameters between a pair of accessions. For this purpose, the data generated by the field trials of 1996 and 1997 were taken through a conventional ANOVA (each year was considered separately; two management systems and two replicates per year) and significant differences were tested using LSD-intervals. In total, 40 parameters (observation × year) were taken into account. This matrix of differences was also turned into a dendrogram (Fig. 5). Three major clusters can be discriminated: the first groups the accessions from Belgium and the Netherlands with 'Talbot' and 'Vigor'; the second merely groups the accessions from France, Italy and Ireland with 'Frances', and a third groups the East-European inputs. Although only rather approximately, the major structure of the genetic analysis as represented by Figs. 1, 2 and 4 is reflected in this dendrogram. The dendrogram based on the field data seems to maintain a better geographical separation than the one based on AFLP data.

- **Conclusions**

Ranking of the populations by PCO plots and dendrograms reveals a genetic structure that corresponds to the geographical localization of the populations. However, this grouping is rather loose. The major part of the genetic variation has to be attributed to differences between individual plants (as revealed by AMOVA analysis), causing any groupings based on overall genetic distances to be rather changeable. Nevertheless, the genetic differentiation between populations and geographical regions exists and is real, but is more or less blurred by the vast genetic variation within the accessions. Comparison with the morphological analysis indicates that both morphology and genetic analysis refer to a common background, although the view of it, as reconstructed by both assessment techniques, is rather fuzzy. Similar studies using AFLP markers to reveal genetic distances within e.g. breeding pools or essentially derived varieties (EDVs) have indicated the huge internal genetic variation within varieties and selected genepools of ryegrasses (Roldan-Ruiz *et al.* 2000, 2001). Only in the case of EDV varieties, which are by definition very narrow, can a good correlation between a classification based on AFLP markers and morphological data be observed. This high internal genetic variation within groups of morphologically similar plants is partly due to the nature of ryegrasses, which are highly heterozygotic self-incompatible cross-pollinators, but can also be caused by the type of marker used for these studies. Because AFLPs are randomly generated, dominant markers, they tend to represent the non-coding part of the plant genome. Therefore they can only disclose functional differences, e.g. those caused by selection towards a certain ecological niche, when they are genetically closely linked to the genes involved. Gene-based markers are thought to be better adapted to this molecular approach.

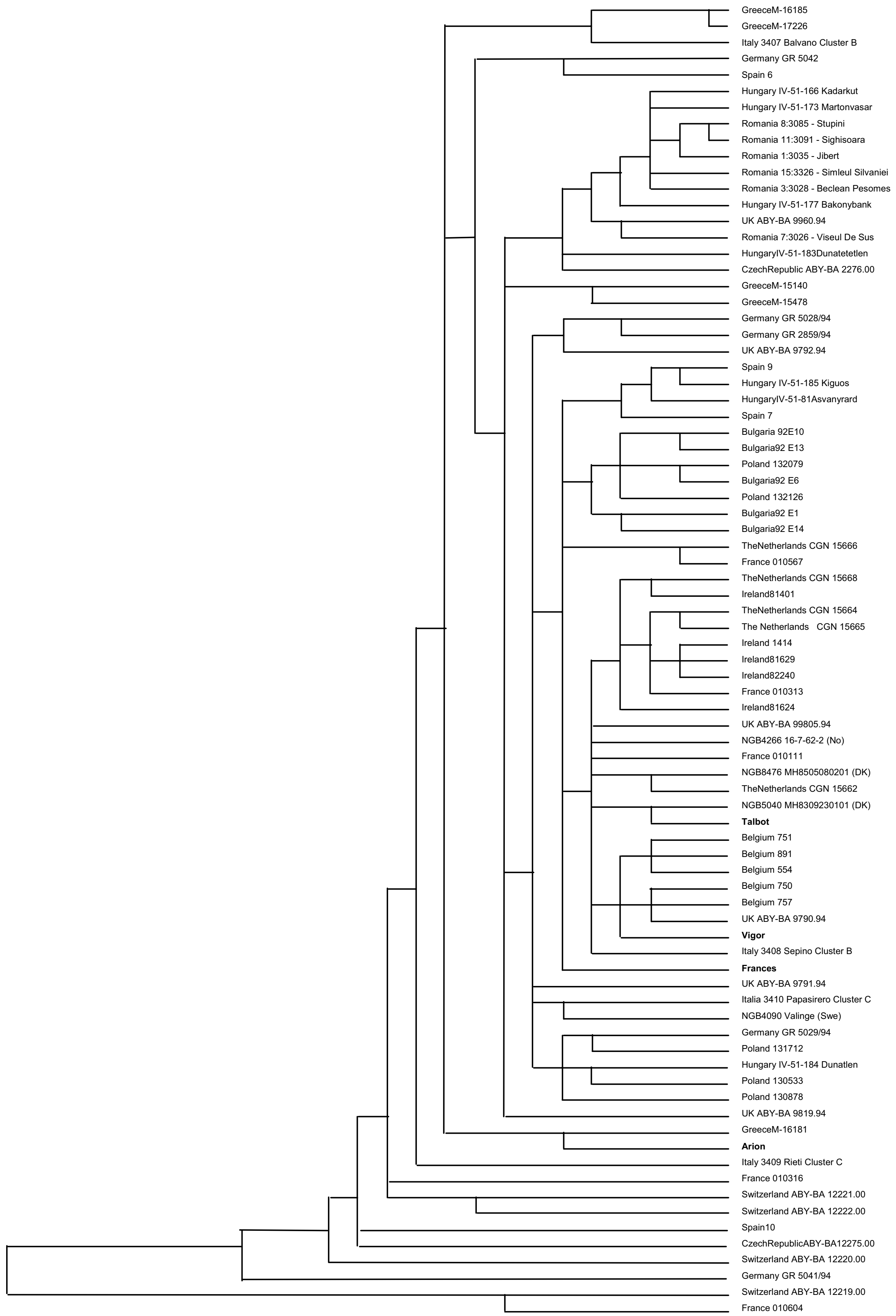

Fig. 4. Ranking based on the Phi statistic taken from the AMOVA analysis (UPGMA clustering; branch lengths are representative).

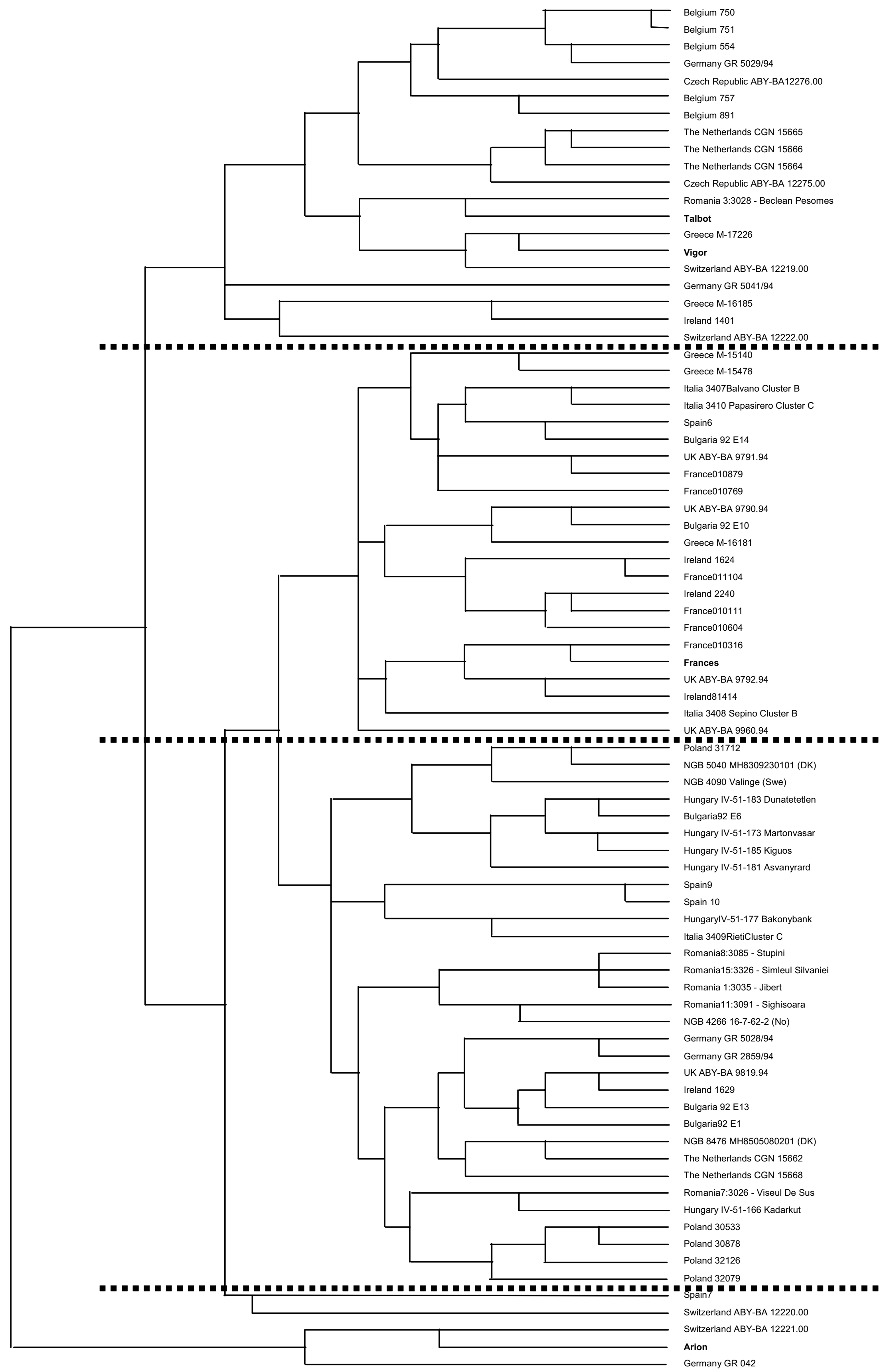

Fig. 5. Ranking based on the agronomical/morphological differences between accessions (UPGMA clustering).

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The European Lolium perenne *core collection in the Botanical Garden of the Plant Breeding and Acclimatization Institute, Bydgoszcz, Poland*

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Introduction

During the meetings of the ECP/GR Working Group on Forages in 1991 and 1994 it was decided to develop a European core collection for perennial ryegrass (*Lolium perenne* L.) ecotypes (IBPGR 1993; Gass *et al.* 1995). It was developed in 17 European countries holding ryegrass collections. One hundred and fifty-four ecotypes with four control varieties ("main" core collection) were planted, cultivated and evaluated according to the methodology given by Sackville Hamilton *et al.* (1997, 1998).

The main goal of the evaluation of the core collection in Bydgoszcz was to examine the variation of ryegrass ecotypes under Polish conditions, using a wider range of traits than is normally used to assess the "main" core collection.

Materials and methodology

- **"Main" core collection**

Seeds of 154 accessions were provided by curators of *Lolium perenne* collections from 17 European countries. In the spring of 1995 seeds of ecotypes, together with control varieties ('Arion', 'Frances', 'Talbot' and 'Vigor') were sown to produce seedlings which were transplanted into small pots and grown from March to June in a cold greenhouse. In July the young plants were planted in the field in 4 replicates, in 1.5 m-long rows with 7 plants at 25 cm intervals and 75 cm between the rows in a split-plot design. Two replicates were subjected to conservation management, with a cut when the last accession had 50% anthesis. The other two replicates were subjected to frequent cutting management (to simulate periodic grazing) with cuts every 3-5 weeks, as appropriate for the evaluation site.

The evaluation protocol was based on the IPGRI descriptor list with small changes (Sackville Hamilton *et al.* 1997, 1998). The following scores were noted:

- heading tendency: tendency to produce inflorescences in the year of sowing (on a scale of 1 = none to 9 = high);
- winter damage: estimated % of dead tillers (on a scale of 1 = minimum to 9 = maximum),
- heading date: number of days from 1 April to emergence of the inflorescence;
- plant habit: tiller angle (on a scale of 1 = horizontal/prostrate to 9 = vertical/erect);
- leaf blade width at heading (on a scale of 1 = very narrow to 9 = very wide);
- bulks: in conservation management = green matter yields (in kg per plot); in frequent cutting management = visual estimation of yield (on a scale of 1 = very low to 9 = very high);
- rust infestation (on a scale of 1 = no rust to 9 = all plants completely infected);
- aftermath heading: intensity of heading after second and third cut in conservation management (on a scale of 1 = none to 9 = numerous heads).

Under the conservation management regime, cuts were adjusted to the developmental phase (i.e. earliness) and were made at five different dates for the first cut (28.05.96, 03.06.96, 21.06.96, 25.06.96 and 01.07.96), once for the second cut (01.09.96) and once for the third cut (15.10.96). Estimations of yield under frequent cutting management were made ca. 6 times per month from 15.05.96.

Winter 1996-97 damage affected the majority of the collection so strongly that further evaluation would not have been worthwhile. We then decided to redefine the core collection on the basis of the results from 1995 and 1996 and to make a more precise evaluation on a "sub-collection". Cluster analysis (UPGA and Euclidean distance) was performed on 22 characters and a sub-collection of 28 ecotypes from 9 clusters was selected. The methodology of selection of sub-samples for further analysis was similar to that of Charmet *et al.* (1990) and Casler (1995).

- **Sub-collection**

Control varieties were the same as in the "main" core collection and two Polish varieties were added: 'Nadmorski' and 'Arka'.

Seeds from the selected accessions were sown in August 1997. Young seedlings were planted in pots in the hot bed and planted out in the field at the end of September 1997. Six replications were used (3 replicates in the frequent cut and 3 replicates in conservation management) with 10 plants per replicate. Evaluation and measurements were carried out in 1998-1999.

Apart from traits similar to those assessed in the "main" collection, additional evaluations were done on:

- young seedlings in pots: number of leaves (4 weeks after planting in pots), number of tillers and seedling dry matter weight (grams per seedling at 8 weeks after planting) and average dry matter of tiller (total plant dry matter weight/number of tillers);
- plants in the field: aftermath heading in the frequent cut treatment, plant height and length of stem leaf at the beginning of heading in conservation management;
- dry plants (cut at the heading phase): for the three longest stems per plant the following traits were measured: height, length of inflorescence, number of spikelets per inflorescence, number of florets per spikelet, length of spikelet (mm), number of spike branches, intensity of spike branching (on a 1-9 scale where 1 = no branching, 3 = branching only on a few spikes, 5 = branching on some spikes, 7 = numerous branching on few stems, 9 = very numerous branches on numerous stems), distance between florets in spikelet and distance between spikelets in spike.

Analysis of variance (one-way ANOVA) was carried out on 71 characters. To describe the variation of the selected ecotypes under different conditions, the results from the "main" core collection were compared to results from the sub-collection. For sub-collection results, Pearson correlation coefficients (r) were calculated for the same traits evaluated in different years. If r was high (>0.6) mean values from both years were accepted for further calculations. Further, principal component analysis (PCA) with varimax rotation and cluster analysis (UPGA and Euclidean distance) were carried out.

Results

- **"Main" core collection**

For ecotypes from the "main" core collection analyzed on the basis of country of origin, differentiation for all 20 traits was significant for ecotypes from France, Great Britain and Switzerland. The lowest trait differentiation was observed for ecotypes from the Czech Republic (4 traits), Bulgaria (6) and Greece (7).

Cluster analysis selected nine groups of ecotypes. Clusters were characterized as follows:

1. the earliest ecotypes, with a high percentage of heading in the sowing year and high aftermath heading, high rust infestation after the second cut;
2. no heading in the sowing year, low winter damage and good yield, highest value of aftermath heading after the second cut;

3. early heading, narrowest leaves and prostrate habit;
4. high green matter yield estimated in first cut in frequent cutting management, no heading in sowing year, high rust infestation of the third regrowth, good yields especially at spring cuts;
5. low percentage of heading in the sowing year and at the second and third regrowths, good yields of autumn cuts;
6. low winter damage and good yields, wide leaves and erect habit;
7. good yields, latest ecotypes with a low percentage of heading in the sowing year;
8. highest values of winter damage and lowest yields, prostrate habit and highest percentage of heading in the sowing year;
9. no heading in the sowing year, rather late, prostrate habit, high rust infestation of the third cut.

As a result of cluster analysis on country mean values the following groups were selected (Fig. 1): first group: one element = Bulgaria (n=6); second group: Mediterranean countries (Italy, Spain and Greece, n=17); third group: one element = Switzerland (n=4); fourth group: rest of Europe (n=130). In the last group some subgroups also appeared: Czech Republic and Poland; Germany, Belgium and the Netherlands; Great Britain and France.

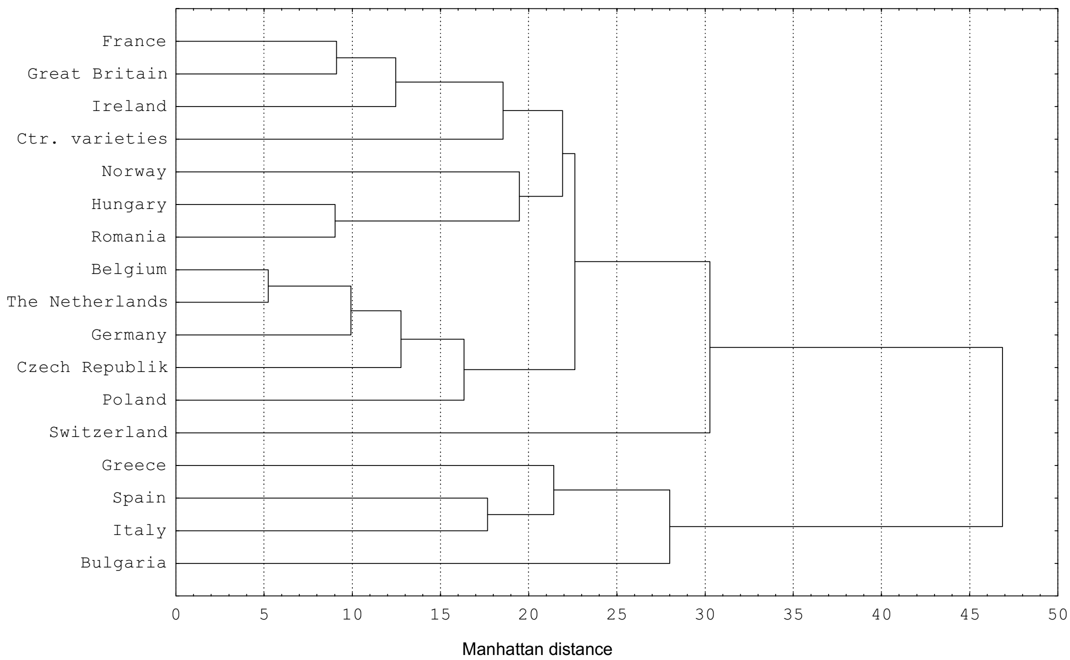


Fig. 1. Results of cluster analysis on country means (main core collection).

- **Sub-collection**

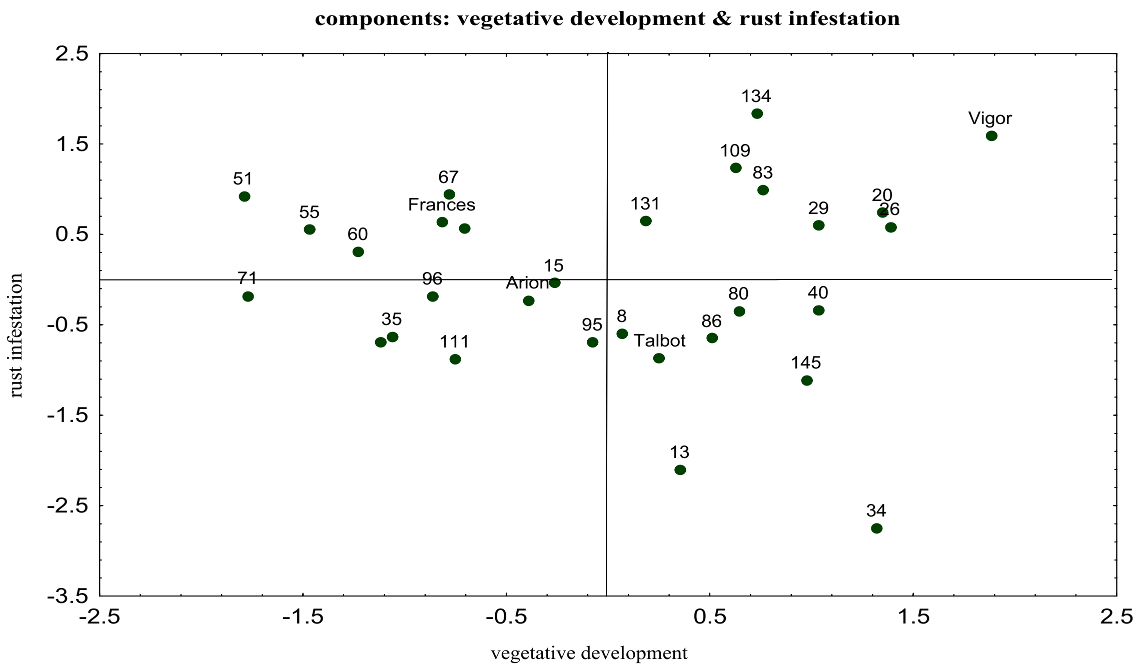
As compared to control varieties, ecotypes from the sub-collection showed higher values of the coefficients of variation for: dry matter weight per stem, winter damage, fresh matter yield of the last conservation cut, yields in frequent cutting, aftermath heading in conservation management in 1996, third and fourth regrowth in frequent cut, rust infestation in 1996 and in frequent cutting in the sub-collection, intensity of spike branching, plant habit, length of spikelet in 1998, average distance between florets in spikelet and width of leaves in 1996 and 1999.

Principal component analysis selected five components:

1. the first component accounted for 19.3% of variation and was mostly related to vegetative development. It was negatively correlated with the following traits: aftermath heading in both treatments and number of florets in spikelet in 1999. Positive correlation was found for the number of days to heading;
2. the second component (yield performance) accounted for 22.4% of variation and was strongly positively correlated with yields from both treatments (especially for first cuts), height of plants at heading, length of leaves and inflorescences. Negative correlation was found between the second component and winter damage, especially in the first year of the experiment;
3. the third component (rust infestation) accounted for 9.8% of total variation;
4. the fourth component was correlated with width of leaves and length of spike, number of florets in spikelet in 1998;
5. the fifth component, accounting for 8.7% of variation, was highly correlated with seedling dry matter weight, average dry matter of tiller and winter damages in 1999.

When plotting them over first and second and first and third components (Fig. 2) it appears that ecotypes nos. 26 and 29 from Belgium, ecotypes no. 83 from Netherlands and 134 from Switzerland had low rust infestation, high yield ability and low ability to produce flowering stems (high vegetative development). The highest yield ability was noted for ecotype 51 from Hungary, and the lowest yield for ecotype 67 from Spain. Variety 'Vigor' displayed the lowest ability to produce flowering stems (highest vegetative development) as opposed to ecotypes from Spain and Hungary (ecotypes 71 and 55, respectively). The ecotype from Switzerland (134) had lower rust infestation as opposed to ecotype 34 from Sweden. Except for 'Vigor', variation of all ecotypes in the sub-collection was wider than that of the control varieties.

For the seven clusters shown in Fig. 3 the mean values for selected traits were calculated. Ecotypes in clusters were characterized as summarized in Table 1.



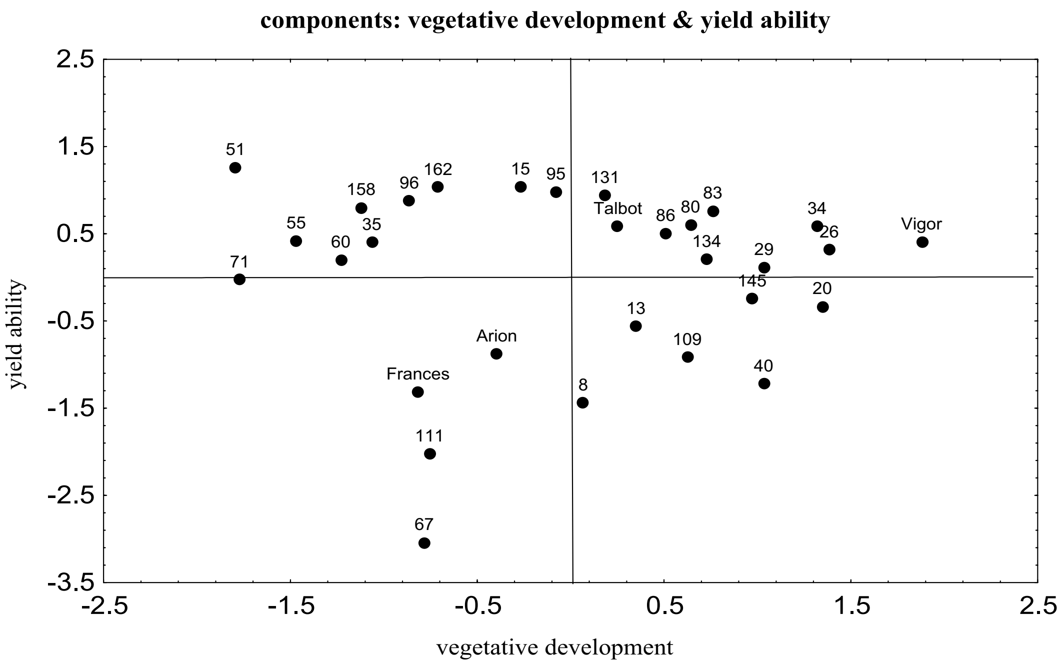


Fig. 2. Principal component analysis (sub-collection).
Figures and names near dots on graphs = no. of ecotypes and names of varieties.

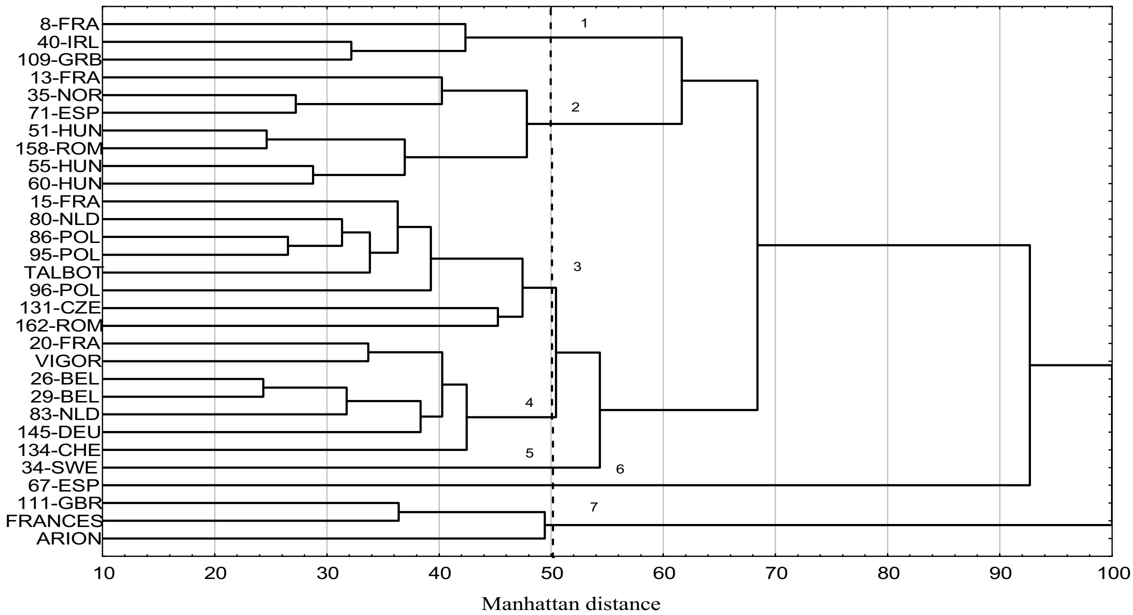


Fig. 3. Results of cluster analysis (sub-collection).
Figures inside graph (1-7) = cluster number (see also Table 1)

Table 1. Composition of clusters and characteristics of the ecotypes in the sub-collection

Cluster no.	Ecotypes	Characteristics
1	ecotypes from western Europe	low yield ability, high winter damage and low dry matter of seedlings
2	ecotypes from Hungary and one from Romania, Norway and France	low dry matter yield of seedling, low winter damage, high yield ability, highest yield of fresh matter in early spring cut, high tendency to aftermath heading at second regrowth
3	variety 'Talbot', three ecotypes from Poland and one from France, the Netherlands, Czech Republic and Romania	high yield ability and low winter damage
4	variety 'Vigor', two ecotypes from Belgium and one from France, the Netherlands and Germany	high dry matter yield of seedlings, good yielding, late heading and highest plant at heading
5	one ecotype (no. 34) from Sweden	high yield ability in frequent cut management, wide and long leaves, highest number of florets per spikelet, high intensity of spike branching and low dry matter of seedlings
6	one ecotype (no. 67) from Spain	high yield ability in frequent cut management, very low plants during heading, short and wide leaves
7	varieties 'Arion' and 'Frances', one ecotype (no. 111) from Great Britain	very early heading, high winter damage, high aftermath heading in regrowth after the second cut, short and narrow leaves, short inflorescence, low intensity of spike branching and high seedling dry matter

Discussion and conclusions

Winter damage affected all characters examined. It was confirmed by observations made on the perennial ryegrass core collection in Norway and in the Czech Republic (Sackville Hamilton *et al.* 1997). A different reaction was observed in Great Britain and Germany where practically no winter damage was observed and even winter yields were measured, but ecotypes from the Czech Republic and Norway yielded low. Winter damage is therefore related to the origin of the ecotype and place of examination, with lowest damage in the region closest to the original collecting site of the ecotype.

Variation of winter damage in the second year was lower for conservation management than for frequent cutting. It may have been due to a weaker condition of the plants resulting from cuts made in the later phases compared to those under a frequent cut management regime.

Yield analysis indicated that ecotypes from northeastern and eastern Europe (especially from Romania) were of higher yield potential in spring cuts. Similar results were obtained for Romanian ecotypes by Tyler *et al.* (1984) and Charmet and Balfourier (1991). Differences in yield were observed for different cuts and management regimes. But, as mentioned before, winter damage most affected yield. High, negative correlation was calculated between first cuts and winter damage for the "main" core collection (Schmidt and Kaszuba 1997). However, the decreasing value of the correlation coefficient in later cuts could be the effect of the high regeneration ability of ecotypes.

Observations made by Sadowski *et al.* (1997) on rust infestation in the "main" core collection indicated *Puccinia coronata* Corda as the causal agent of 60% of rust visible symptoms and *Puccinia graminis* Pers for the rest of the symptoms. The highest differentiation of ecotypes by their degree of rust infestation symptoms was observed in summer, at the initial stage of the disease. Chorlton and Thomas (1987) obtained similar results when studying the susceptibility of ryegrass ecotypes to *Puccinia coronata* infestation. It is worth noting that in our experiment none of the tested ecotypes showed lower infestation than the Polish variety 'Arka'.

Aftermath heading was negatively correlated with the number of days to heading and it was also found that ecotypes originating from northern Europe produced lower numbers of heads. This is probably due to the different life strategies of ecotypes originating from different regions (Lorenzetti *et al.* 1971; Breese and Tyler, after Breese 1989). Ecotypes from Romania showed high values of aftermath heading after the first cut, as also found by Charmet and Balfourier (1991).

The key role of origin in overall ecotype performance, as found in our study, was also suggested by Lorenzetti *et al.* (1971), Balfourier and Charmet (1991), Charmet *et al.* (1993), Loos (1994), Casler (1995) and Amin and Thomas (1996).

Preliminary observations on mean heading dates and winter damage recorded in other institutions cooperating in the perennial ryegrass core collection (Sackville Hamilton *et al.* 1998) as well as conclusions from other researchers (Charmet *et al.* 1990; Elgersma 1990a, 1990b; Charmet and Balfourier 1991; Solberg *et al.* 1994) strongly support the multi-site evaluation of collections. It is especially recommended for traits of low heritability, with high dependence on environmental conditions. Therefore, to obtain more complete information on the nature of variation of European perennial ryegrass ecotypes and its interaction with environment, it is necessary to discuss similar results from other countries.

The above results strongly support the development of core collections for other species stored in European genebanks.

Further conclusions from the evaluation of the perennial ryegrass core collection in Bydgoszcz, Poland are as follows:

- it is possible to indicate the following regions of origin of the ecotypes of the tested species in Europe: southern Europe, western Europe, northern-central Europe and Romania and Hungary;
- none of the tested ecotypes showed better yields than control varieties; however a few ecotypes appeared to have advantages for particular traits (better winter performance, faster spring regrowth, etc.);
- the high differentiation of ecotypes examined indicates the existence of many botanical varieties and intermediate ecotypes;
- traits associated with inflorescence morphology (distance between florets in spikelet and distance between spikelets in spike, length of spikelet) seem to be the most stable traits, in contrast the most variable traits were aftermath heading, tendency to produce inflorescences in the year of sowing and green matter yield of the first cut.

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On-farm conservation

On-farm conservation in Finland

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The Finnish on-farm conservation project was presented by Merja Veteläinen at the meeting in Elvas, 1999. During the last four years six landraces have been registered: five red clover varieties ('Sirppilahden Johanna' from Muuruvesi, 'Vesilahtelainen' from Vesilahti, 'Lahtua' from Orivesi, 'Perttuli' from Tuovilanlahti, and 'Turunen' from Lammasaho) and one winter rye variety ('Eelis-Antti' from Kiuruvesi). The clover accessions are bred for fodder and they have good winter hardiness.

Reference: Communication with Kaarina Paavilainen, Plant Production Inspection Centre, Seed Testing Department, Finland.

On-farm conservation of forage landraces in Central Italy

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Introduction

Conservation has been defined as a system of resource management which yields the greatest benefit to present generations without impairing benefits for future generations (IUCN/UNEP/WWF 1980).

It is proposed that landraces should be preserved for future generations because they harbour a diversity of interesting traits for future breeding work and for developing new farming systems and moreover, reflect the cultural identity of certain groups of people (Altieri and Merrick 1987; Brush 2000). Since many landraces have already completely disappeared, in recent years some initiatives have been undertaken to preserve these genetic resources at the regional and inter-regional level.

Until recently, germplasm conservation of crop landraces, as well as of their wild relatives, relied on *ex situ* methods (i.e. the conservation of biological material outside its natural habitat, UNCED 1992), mostly in germplasm banks. More recently *in situ* (on-farm) conservation (i.e. the conservation of biological diversity in its natural habitat) has been proposed as a conservation strategy which allows evolutionary processes to continue rather than being halted as occurs to a large extent in *ex situ* conservation (Frankel *et al.* 1995; Maxted *et al.* 1997). The need to conserve and sustainably use biodiversity with special reference to *in situ* conservation of crop plants has also been emphasized by the Convention on Biological Diversity (CBD) Agenda 21, the Global Plan of Action (GPA) and other influential international documents. In on-farm conservation, the farmer or farmers' communities, who are considered to be the stewards and managers of the agroecosystem, are the actors (i.e. they are the subjects actually carrying out conservation). They should be the ones to maintain landraces in the field, under the conditions found in the area of cultivation. This means that not only are natural selection pressures at work, but the selective pressures (deliberate or not) applied by farmers also come into play. For example, a farmer uses the agronomic techniques he/she considers the most appropriate in that year, or he/she may select a different colour of seed. On-farm conservation is not an open-air museum of conservation. Policy makers and local institutions should facilitate farmers' activity by implementing measures to support on-farm conservation.

Before implementing on-farm conservation activities, knowledge of the situations which lead individuals to agree to take part in on-farm conservation is required. Few data are available about human motvation behind on-farm conservation in Europe (Negri *et al.* 2000; Nowosielka and Podyma 2001; Negri 2003).

This paper reports data relative to on-farm conservation of forage landraces in central Italy.

Materials and methods

- **Exploration and collection missions. Collection of information**

Forage species were collected by DBVBA (Dipartimento di Biologia Vegetale e Biotecnologie Agroambientali) during several collecting missions undertaken since 1981.

In some cases collection was made possible through the help of local extension services, while most visits were randomly organized. Farmers were approached in a friendly manner, the reason for the visit was explained and an interview followed during which information on the presence of materials which had been reproduced on-farm over at least one farming generation was gathered. This study is solely based on this material. Information about the farm (name and age of farmers, farm extension, crops grown), farm management and on adaptative and agronomic traits of landraces found, as well as information relating to crop use and social context were collected. A key question during the interviews carried out in the last decade was: "Why do you keep growing landraces?"

Passport data relative to altitude, longitude, latitude and habitat were also recorded. Data were collected using purposely developed forms. The farmers were also requested to fill in a form when donating germplasm to the bank. Once collected, the landraces entered the conservation procedure of the DBVBA.

- ***Ex situ* germplasm preservation, exchange and characterization**

Seed were first cleaned, dehydrated to an average moisture content of 7-8%, vacuum-sealed in aluminium packets and stored at -18°C in a freezer, with an accession number. Some seeds were used in programmes of characterization, evaluation and breeding: molecular markers showed the existence of genetic differences among and within landraces of a given species in the collection (Russi *et al.* 1997; Veronesi *et al.* 1997).

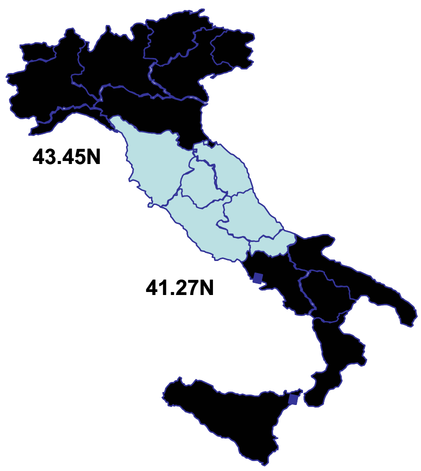
Passport data relative to the collected materials are stored in a relational database according to IPGRI and FAO recommendations (Lipman *et al.* 1997; Maggioni *et al.* 1998). Since we do not have any institutional duty to distribute germplasm, it is freely donated to all *bona fide* users as small samples, provided that enough seed remains for long-term storage.

Results

- **Landraces found**

One hundred and fifteen landraces belonging to different species (*Medicago sativa* 67, *Onobrychis viciifolia* 35, *Hedysarum coronarium* 5, *Trifolium incarnatum* 5, *Trifolium squarrosum* 3) were found on-farm in central Italy, an area characterized by quite diverse pedoclimatic conditions (Fig. 1).

No grass forage landraces were found in the area. Detailed information on collecting sites and on farmers maintaining landraces on farm can be found in Negri (2003); the following results are drawn from those data.



Average annual rainfall (mm)	Average temperatures (°C)			soil pH
	Maxima of the coldest month	Annual	Maxima of the hottest month	
550 - 1500	-4 - +8	9.5 - 15.7	24.5 - 31.9	5.5 - 8.0

Fig. 1. Range of climatic constraints and soil pH in the collection area (sea level - 1000 m asl).

- **Where landraces are preserved on farm**

M. sativa landraces can be found almost everywhere, from sea level to 1000 m asl., on average farm elevation was 451.5 m; *O. viciifolia* is generally found in inland mountainous areas from 190 to 1300 m asl: on average farm elevation was 749.1 m. The other species are sporadically found at low elevation. My next comments only refer to the most frequently found species.

- **Who preserves landraces**

Forage landraces are generally grown under modern agricultural techniques by farmers of average age equal to 60.9 and 56.8 years old for *Medicago sativa* and *O. viciifolia* growing farmers, respectively. Farm dimensions are quite large by Italian standards: 72.2 and 65.8 ha for farms where *M. sativa* and *O. viciifolia* can be found, respectively.

- **Main destinations of the landraces found**

The forage is mainly used on the farm (98.6%) for both species. A different situation exists for the seed. *M. sativa* seed is mostly used on the farm (83% of recorded cases) while in 17% of recorded cases it is sold on the local market. A significantly lower percentage (56%) of *O. viciifolia* seed is used on the farm, while 42 and 2% of it is sold on the local and larger markets, respectively (Fig. 2).

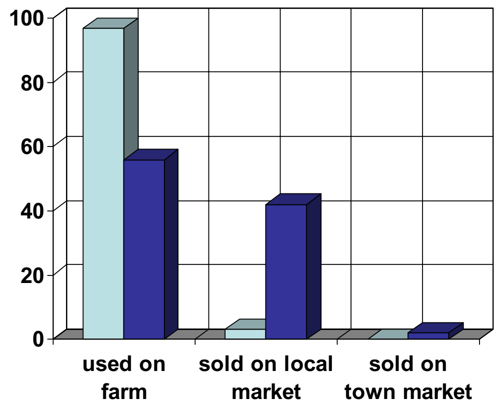


Fig. 2. Main destinations of the seed of *M. sativa* and *O. viciifolia* landraces in central Italy.

- **Why landraces are conserved on farm**

Better persistence and productivity than modern varieties and adaptability and good productivity under difficult or harsh condition are the reasons given by the farmers for continuing to grow landraces.

Discussion and conclusions

Despite the agronomic value of landraces as declared by the farmers, problems for continued on-farm conservation exist.

Social problems seem to be leading to progressive genetic erosion. Rather few people, many of them old, remain in the countryside. They often declared that they felt unable to carry on their farm activities any longer.

The younger people who live in the country are often employed part-time in agriculture and often find it more convenient to buy the seeds from the market rather than to reproduce them. Seed harvesting, cleaning and conditioning require time and sometimes the appropriate equipment, which is not always present on the farm. Lack of skill is another constraint in reproducing seed, since younger farmers often lack the practical know-how that was generally available and used in the past. This makes it difficult to increase landrace cultivation or, even more troubling for plant genetic resource conservation, to continue cultivation.

In addition, the seed legislation is a major constraint to on-farm conservation of *M. sativa* landraces in the future. Commercialization of *M. sativa* seed of landraces has been allowed up to now, but only varieties will be commercialized from this year. At present, farmers can only reproduce landraces for their own use on the farm.

A different situation exists for the other legume forage crops since they can be commercialized without the status of variety.

If Directive 98/95 allows forage landrace conservation on-farm, how to implement it is still matter of discussion. A fierce opposition by the local seed industry exists toward the implementation of this Directive. As a consequence of all these problems, most landraces appear to be condemned to extinction if concrete actions for their safeguard are not rapidly taken.

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On-farm conservation/improvement of forages in Norway

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The best way to preserve forage genetic resources is to use them. Up until the 1950s many landraces of timothy and red clover existed in Norway. The landraces were, through generations of regeneration by local farmers, adapted to the local climatic and management practices. Almost all of these landraces were lost before we saw the value of preserving them. Our idea is to recreate/produce new landraces by restarting the processes that created them. We want to create landraces that are adapted to a variety of climatic and management practices. The project will start by producing three broad genepools in timothy (*Phleum pratense*), meadow fescue (*Festuca pratensis*) and red clover (*Trifolium pratense*), the three most used forages in Norway. We will use seeds of all the accessions in the Nordic Gene Bank, including some commercial cultivars. These will be intercrossed in two generations followed by one generation of seed multiplication in different parts of Norway.

A suitable mixture of the three species will be made and the seed distributed to selected farmers in different parts of Norway. As a general rule the farmers will harvest their meadow for two production years and harvest seed on part of the meadow the third year. The farmer will use this seed to establish a new meadow with the appropriate mixture. A seed sample will be stored from each generation and location under long-term storage conditions for further reference. The project will start in 2003.

Research activities

Cyprus: collection and evaluation of local germplasm for forage production

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Introduction

Many forage species, both annual and perennial, have been introduced and tested in Cyprus under rain-fed conditions. The results indicated that the introduced material was not always successful here and it was therefore decided to give more emphasis to the use of local genetic material for improving fodder and pasture crops. Priority was given to *Medicago*, *Vicia, Hordeum, Dactylis, Cynodon* and various other grass species for forage/turf purposes.

Materials and methods

All the promising genetic material collected from different parts of the island at various times was multiplied, and when sufficient seed was available, was evaluated in replicated trials at two or three locations for several parameters. The parameters tested were date of flowering, plant height at flowering, dry matter yield, dry matter content, nitrogen yield and content, digestible organic matter yield in herbage dry matter and percentage digestibility. In many cases this genetic material was also used in the breeding programmes of the Institute.

Results and discussion

- ***Vicia* species**

In 1997 in cooperation with FAO (Regional Office for the Near East), which partially financed the collection and evaluation of the most important forage crops, 16 accessions of *Vicia* sp. were collected for evaluation. The seed of each accession was sown in autumn 1997 for multiplication and evaluation. Six of them were taken forward for further evaluation in replicated trials at two locations during the growing seasons 1999-2000 to 2001-2002. The results so far indicate that the dry matter yield of these six accessions ranged from 5256 kg to 6102 kg dry matter/ha, the differences being significant (Table 1).

Table 1. Dry matter and digestible organic matter yield of the best six local accessions of *Vicia sativa* (mean of five environments, 1999-2000/2001-2002)

Accessions	Dry matter yield (kg/ha)	Digestible organic matter yield (kg/ha)
Sel 2556	6013	3791
Sel 96 (Paphos)	5446	3342
Sel 97 (Statos)	5522	3443
Sel 98 (P. Pedi)	5465	3467
Sel 99 (Achilleas)	6102	3761
Local (Local)	5256	3286
S.E. ±	228	230

- ***Hordeum* species (wild barley)**

In Cyprus, wild barley behaves as a pasture crop (Hadjichristodoulou 1988), and it was thought that with proper management it might be used for pasture development. Wild barleys, *Hordeum spontaneum* and *H. agriocrithon* (natural outcrosses of *H. spontaneum* with *H. vulgare)* are found in abundance in the Mediterranean region and are distinguished from *H. vulgare* by their brittle rachis, shrunken kernels and seed dispersing mechanisms. Due to these characteristics both species of wild barley are able to regenerate naturally, except where overgrazing occurs. Wild barley also has a certain level of seed dormancy, thus safeguarding the survival of the species. Taking advantage of the pasture characteristics of wild barleys, Hadjichristodoulou (1995) established pastures in Cyprus to test the performance of these crops and their crosses with *H. vulgare* under grazing conditions. In these trials it was shown that there were no adverse effects on crop growth when the herbage was grazed by sheep two or three times, depending on weather conditions (particularly rainfall) from mid-December to mid-April. However, by the end of April the crop must be left to go to seed. After seed maturity the dry herbage can also be grazed (July). By using this procedure, reseeding is not required. Since barley is not a nitrogen-fixing crop, ample amounts of nitrogen fertilizer are necessary for maximizing forage production. Research work is now under way to study the possibility of using mixtures of wild barley with either medics or with *Vicia amphicarpa* (a species that produces seed both above and below ground), so that the legume component will mainly provide nitrogen, and the cereal, the herbage production. Results so far show that wild barley is very aggressive and suppresses the legume component in the mixture. Therefore, research is being re-directed to study the performance and methods of management of wild barley in pure stands.

- ***Medicago sativa* (lucerne)**

Lucerne is considered to be the most nutritious and profitable perennial forage crop grown in Cyprus. Since the results from the introduction and testing of new varieties were disappointing, it was decided in 1984 to select the most productive ones. Twenty-nine populations were evaluated in replicated trials for four years. The results showed that there were big differences among locally selected populations for the various parameters examined. The dry matter yield over the whole experimental period (May 1985–December 1988) ranged from 68 t/ha to 116 t/ha, while herbage yield of the check (local variety) was 108 t/ha.

It appears, therefore, that selection of local germplasm holds more promise in the search for improved material than the introduction of foreign varieties (Droushiotis 1994). Following this conclusion, the four best accessions were taken forward for further evaluation in larger plots for their persistence and other agronomic parameters. In the same experiment one genotype of *Poterium sanguisorba* is also included. It is important to note that a selection is under way from the local variety to find genotypes resistant to drought stress. The results so far are satisfactory.

- ***Avena* species**

The seeds of ten *Avena* species (oats) accessions collected from several parts of Cyprus in 1995 were multiplied and evaluated in replicated trials during the period 1996-1997 to 2001-2002. Two commercial forage oat varieties, 'Mulga' and 'Algerian', served as controls. The forage yield of the accessions tested ranged from 6646 to 8673 kg dry matter/ha. The yield of the best accession, 'Dromolaxia', was 7121 kg dry matter/ha while those of the commercial varieties 'Algerian' and 'Mulga' were 7867 kg and 8673 kg dry matter/ha respectively (Table 2). Differences were significant. The evaluation continues.

Table 2. Dry matter and digestible organic matter yield of *Avena sativa* accessions collected in Cyprus (mean of seven environments, 1996-1997 to 2001-2002)

Accessions	Dry matter yield (kg/ha)	Digestible organic matter yield (kg/ha)
Menico	6646	3958
Latsia	6730	4093
Kokkinotrimithia	6955	3982
Dromolaxia	7121	4284
Mesoyi	6944	4448
Polis	7044	4451
Algerian	7867	5086
Mulga	8673	5173
S.E.$^{\pm}$	260	261

- ***Dactylis* species**

The seeds of 30 genotypes, 9 collected from Cyprus and 21 collected from Greece during 1997, were sown in plots of 0.9 m^2 each at the end of 1997. Observations so far confirm that there are promising genotypes for forage production among the populations under evaluation. Ten populations were taken forward during 2001-2002 for further evaluation in replicated trials at one location. The results so far are satisfactory. The dry matter yield of the various populations tested ranged from 149 kg to 567 kg dry matter/ha (Table 3). Special emphasis is given to genotypes resistant to drought stress. The evaluation continues.

Table 3. Dry matter yield (kg/ha) of *Dactylis glomerata* populations tested at Saittas, 2001-2002

Population	Dry matter yield (kg/ha)
Perrevia	388
Pera–Pedi	253
Pissouri	307
Yiolou	434
AS-03	245
Monastiri Phthiotidos	245
Itea Fokidos	222
Ayii Theodori	307
Larissa	149
Syn-53	567
S.E.$^{\pm}$	112

- ***Cynodon dactylon***

This species abounds in Cyprus and under favourable conditions it can produce herbage abundantly. However, its main interest is as a turf grass. Several accessions have been collected from all over the island and are being evaluated. Results to date are very encouraging.

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Screening of tall oatgrass and yellow oatgrass germplasm held in the European Central Crop Databases

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Introduction

Tall oatgrass and yellow oatgrass are widespread in the floristically rich communities of semi-natural mesophytic meadows of the *Arrhenatheretalia* order. Tall oatgrass – *Arrhenatherum elatius* (L.) Beauv. ex J. et C. Presl – occurs in highly productive meadows of the *Arrhenatherion* alliance from lowlands to hilly uplands (to about 1400 m) and flourishes on roadsides and banks throughout Europe and western Asia. Yellow oatgrass – *Trisetum flavescens* (L.) Beauv. – extends through Europe to temperate Asia from lowlands to mountains (to about 2300 m). It is a typical representative of the communities of the *Polygono-Trisetion* alliance found in poor mountain meadows. Both species are bred and cultivated as forage grasses, especially in central Europe (Austria, the Czech Republic, Germany, Hungary, Poland, Slovakia and Switzerland). Utilization of tall oatgrass has also spread to southern Europe (Italy, Slovenia). The number of varieties is relatively low in comparison with other grass species. The awned nature of the seeds causes difficulties in seed production, therefore the new plant breeding strategy has focused on producing awnless types of varieties.

The European genetic resources of tall oatgrass and yellow oatgrass are recorded in the European *Arrhenatherum* and *Trisetum* Databases managed by OSEVA PRO Ltd., Grassland Research Station (GRS) at Zubří, Czech Republic. The databases contain passport data on 254 and 86 accessions of tall and yellow oatgrass, respectively, belonging to 15 institutes (Ševčíková 2000) and are available via the Internet as follows:

- *Arrhenatherum* Database: http://www.ecpgr.cgiar.org/databases/Crops/arrhenat.htm
- *Trisetum* Database: http://www.ecpgr.cgiar.org/databases/Crops/Trisetum.htm

The aims of the project funded by the Grant Agency of the Czech Republic were (a) to analyze the variability of European wild populations and varieties recorded in the European Databases and maintained in the European genebanks and institutes; and (b) to obtain information on gaps in coverage of European areas.

Materials and methods

Seed samples of 140 and 46 accessions of tall and yellow oatgrass, respectively, were obtained from 14 European institutions holding these species. The original seed was tested for thousand-seed-weight (TSW) and seed viability in laboratory germination tests. Analysis of relative DNA content from leaf tissue by a flow cytometer (Partec) using fluorescent dye (DAPI) and ploidy determination were carried out in the laboratory of the Plant Breeding Station Hladké Životice Ltd. Viable accessions of tall and yellow oatgrass (128 and 24 accessions respectively) were characterized and evaluated as spaced plants in field plots with 3 replicates and a split-plot design at Zubří (360 m asl, average annual rainfall 903 mm and annual average temperature 7.6°C). Seven spaced plants per accession and replication were planted in rows, where distances within and between rows were 0.25 m and 0.45 m, respectively.

The following characters were measured in 2000-2001:

- heading date: time of inflorescence abundance, measured from 1 April, when the tips of 3 inflorescences are just visible on single spaced plants (1 = very early (<26 days); 2 = very early to early (26-32 days); 3 = early (33-39 days); 4 = early to medium (40-46 days); 5 = medium (47-53 days); 6 = medium to late (54-60 days); 7 = late (61-67 days); 8 = late to very late (68-74 days); 9 = very late (>74 days);
- morphological data at the beginning of flowering: stem length (mm) from soil surface to top of inflorescence; inflorescence length (mm); number of leaves on the stem; flag leaf length and width (mm); second leaf length and width (mm);
- abundance of inflorescences: visual estimate of the number of inflorescences per plant at heading (1 = none or very low; 3 = low; 5 = intermediate; 7 = high; 9 = very high);
- dry matter yield (g/plant): annual dry matter yield harvested in three-cut management;
- aftermath heading: visual estimate of the number of inflorescences per plant at aftermath heading (1 = none or very low; 3 = low; 5 = intermediate; 7 = high; 9 = very high);
- regrowth: scored visually after cutting (1 = very slow, 3 = slow; 5 = intermediate; 7 = rapid; 9 = very rapid).

To measure variation and correlation of selected variables, the coefficient of variation (*C*) and the Pearson correlation coefficient (*r*) were used. The characterization variables (morphological data, TSW and heading date) were cluster-analyzed by using the UNISTAT Statistical Package (hierarchical cluster analysis, distance measure: Euclid, linking method: average between groups).

Results

The germination tests mostly showed a very good seed quality for stored tall oatgrass (95.6% viable accessions with mean germination of 71.1% and mean germination time of 6.1 days), while only 57.8% viability was found in accessions of yellow oatgrass with mean germination 58.5% and mean germination time of 10.4 days. TSW ranged from 2.100 to 4.521 g and from 0.203 to 0.328 g and was characterized by *C* = 14.1% and 11.4% in tall and yellow oatgrass, respectively.

The viable accessions of tall oatgrass represented germplasm from 18 countries of origin (in 22 accessions, the country of origin was indicated as unknown), whereas only 8 countries of origin were represented in yellow oatgrass (Table 1).

Table 1. Country of origin (FAO codes) and number of evaluated accessions (ecotypes/cultivars)

Arrhenatherum elatius																	
ARM	AUT	CZE	DEU	EST	GBR	GEO	HUN	ITA	KAZ	LVA	POL	PRT	RUS	SVK	SVN	UKR	YUG
1/1	2/-	13/4	12/8	-/1	2/2	-/1	18/1	1/1	-/1	-/2	8/7	1/-	2/1	6/2	2/1	1/2	-/2
Trisetum flavescens																	
	BGR	CZE	DEU				HUN				POL		ROM	SVK	SVN		
	1/-	5/5	-/3				2/-				2/-		1/-	-/2	3/-		

The ploidy level was determined as diploid (2n = 28) in all accessions of both species (Ševčíková *et al.* 2001).

The time of inflorescence emergence, measured from 1 April, varied from 37.8 to 60.4 days l in tall oatgrass and from 42.8 to 61.3 days in yellow oatgrass. In the collection of tall oatgrass, the majority of the accessions were found in the group of “early to medium” heading date (56% of accessions). The plants of yellow oatgrass emerged later and 63% of accessions were classified in the "medium" category. In both species, the abundance of inflorescences with scores ranging from 6.5 to 9.0 was higher and more uniform than for the aftermath heading, which had scores ranging from 1.5 to 7.0. The time of inflorescence emergence, with C = 9.3 and 8.3%, and the abundance of inflorescences, with C = 6.1 and 7.9% in tall and yellow oatgrass respectively, appeared to be the most consistent characters, while aftermath heading was the most variable (C = 31.6 and 30.2%).

The dry matter yield per plant achieved a mean annual production of 114.5 g and 73.0 g and ranged from 62.8 to 184.6 g and from 40.0 to 111.4 g in tall and yellow oatgrass, respectively; its variability was characterized by C = 19.0 and 26.4%. The regrowth after cutting was evaluated using a 9-point scale and varied from 3.5 to 8.5 in tall oatgrass (C = 15.7%) and from 4.7 to 6.6 in yellow oatgrass (C = 9.2%).

The length (C = 17.8 and 15.0%) and the width of the flag leaf (C = 15.8 and 13.0%) were the most variable among the morphological characters, while the length of the inflorescence (C = 9.9 and 7.6%) and the length of the stem (C = 11.6 and 11.0%) were less variable in both tall and yellow oatgrass. Lower variability was found for almost all characters in the subset of cultivars in comparison with ecotypes, especially in the case of yellow oatgrass.

In tall oatgrass, 16 significant positive correlations were found between morphological characters. No correlations were found between heading date, TSW and other characters except for number of leaves and second leaf length both correlated with heading date. These positive correlations predicate better foliage development in late types (Table 2).

Table 2. Correlation matrix for selected characters in the collection of tall oatgrass

No.	Variable	1	2	3	4	5	6	7	8
1	Stem length								
2	Inflorescence length	0.38 **							
3	Number of leaves	0.72 **	0.00						
4	Flag leaf length	0.23 **	0.39 **	-0.07					
5	Flag leaf width	0.14	0.24 **	-0.08	0.60 **				
6	Second leaf length	0.38 **	0.50 **	0.18 *	0.79 **	0.54 **			
7	Second leaf width	0.32 **	0.27 **	0.13	0.45 **	0.90 **	0.52 **		
8	1000-seed-weight	0.14	0.08	0.13	-0.04	-0.18	-0.05	-0.08	
9	Heading date	0.11	0.08	0.30 **	-0.06	0.04	0.20 *	0.12	-0.15

In yellow oatgrass, 14 significant positive correlations were found between morphological characters. A negative correlation between the heading date and the length of stem indicates shorter growth in the later-heading accessions. No correlation was found between TSW and other characters (Table 3).

Table 3. Correlation matrix for selected characters in the collection of yellow oatgrass

No.	Variable	1	2	3	4	5	6	7	8
1	Stem length								
2	Inflorescence length	0.58 **							
3	Number of leaves	0.45 *	0.20						
4	Flag leaf length	0.35	0.65 **	0.18					
5	Flag leaf width	0.73 **	0.54 **	0.29	0.43 *				
6	Second leaf length	0.59 **	0.72 **	0.39	0.92 **	0.48 *			
7	Second leaf width	0.72 **	0.66 **	0.31	0.40	0.92 **	0.52 *		
8	1000-seed-weight	-0.08	0.03	0.17	0.37	-0.11	0.34	-0.01	
9	Heading date	-0.57 **	-0.11	-0.21	-0.02	-0.40	-0.12	-0.29	0.34

Morphological data, TSW and heading date were cluster-analyzed and the accessions with a full data set were grouped into 6 and 4 clusters in tall and yellow oatgrass, respectively. The results of cluster analysis are given in Tables 4 and 5 as mean data for cluster members.

Table 4. Results of hierarchical cluster analysis in the collection of tall oatgrass

Cluster	Cases	Stem length (mm)	Inflorescence length (mm)	No. of leaves	Flag leaf Length (mm)	Flag leaf width (mm)	Second leaf length (mm)	Second leaf width (mm)	TSW (g)	Heading date (days from 01.04)
1	23	1139.4	214.6	3.3	130.0	5.9	198.6	8.4	3.45	45.5
2	15	962.4	205.5	2.9	103.5	5.0	174.2	7.5	3.48	45.4
3	15	1045.2	220.8	2.8	117.1	5.5	185.3	7.8	3.41	47.6
4	10	1345.0	232.7	3.7	118.2	5.6	196.6	8.8	3.70	47.6
5	46	1247.2	222.5	3.6	118.1	5.6	196.6	8.6	3.55	47.2
6	1	824.6	199.9	3.1	89.8	4.8	151.4	7.1	3.17	44.5
Total		1145.7	218.3	3.3	117.7	5.6	191.2	8.3	3.51	46.6
s_M		11.75	1.90	0.04	1.85	0.08	2.17	0.10	0.05	0.38

s_M = standard error of the mean

Table 5. Results of hierarchical cluster analysis in the collection of yellow oatgrass

Cluster	Cases	Stem length (mm)	Inflorescence length (mm)	No. of leaves	Flag leaf length (mm)	Flag leaf width (mm)	Second leaf length (mm)	Second leaf width (mm)	TSW (g)	Heading date (days from 01.04)
1	1	466.9	121.6	2.7	78.2	3.6	97.9	4.3	0.27	56.3
2	4	696.7	150.5	3.1	130.1	6.5	159.1	7.1	0.28	48.8
3	17	800.8	151.9	3.2	115.1	6.7	154.6	7.6	0.28	47.0
4	1	918.5	173.1	3.1	135.1	7.6	177.0	8.5	0.26	47.3
Total		771.6	150.9	3.2	117.4	6.6	153.6	7.4	0.28	47.7
s_M		17.36	2.34	0.05	3.60	0.17	4.00	0.19	0.01	0.69

s_M = standard error of the mean

A different cluster classification was found in some cultivars which were identical for the name of the accession but which originated from different holding institutes. To compare such cases, the radar graphs were constructed by using transformed values for each character analyzed. The comparison of characters of identically and distinctly classified accessions of tall oatgrass is given in Figs. 1 and 2.

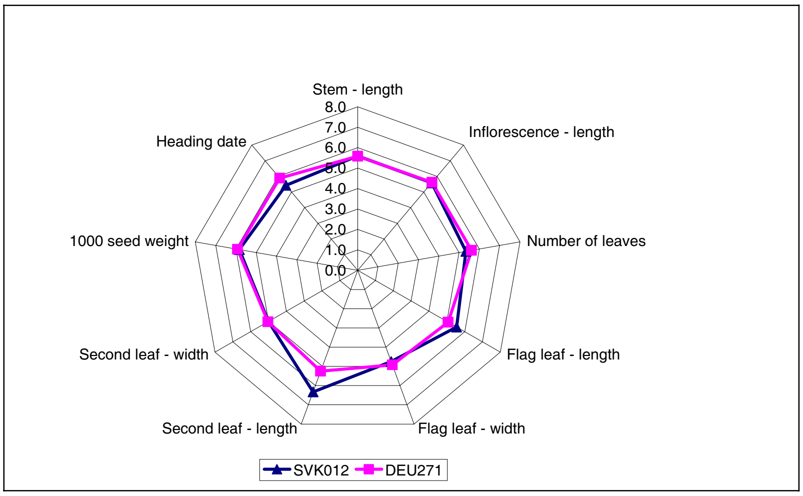

Fig. 1. Comparison of two accessions of cv. Levočský (*A. elatius*) in the same cluster.

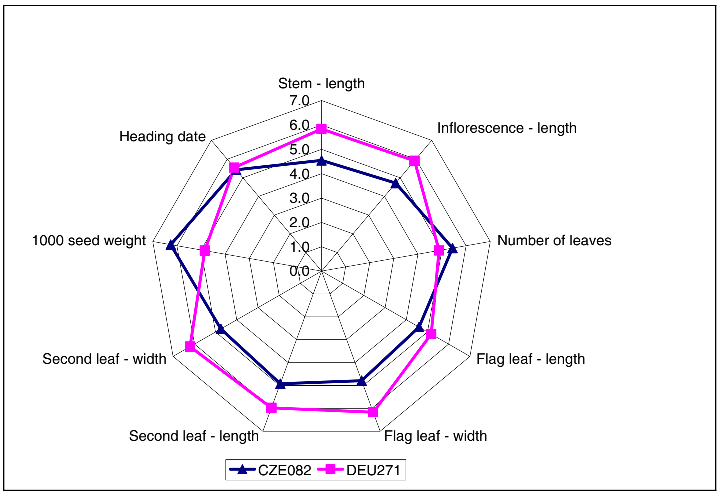

Fig. 2. Comparison of two accessions of cv. Rožnovský (*A. elatius*) in distinct clusters.

Conclusions

The results provide important information on the screening of stored European germplasm of tall and yellow oatgrass. The observed variability shows that there is wide genetic diversity in wild populations of tall oatgrass. However, there are still gaps in coverage of European areas. No ecotypes from countries such as Switzerland (located in the area of its cultivation), the Scandinavian countries, France and others are recorded as being held by any European institute. The viable collection of yellow oatgrass is too small and is not representative enough, and collecting further wild material would be useful in typical areas for the species, e.g. in Alpine countries. Concerning the bred material, the morphological characterization of some cultivars held by different European genebanks under an identical accession name showed differences in their phenotypes and therefore a possible misidentification or genetic changes in some accessions of both species.

Acknowledgements

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Evaluation of important traits as the basis for an assessment of Poa *genetic resources for breeding purposes*

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Introduction

The genus *Poa*, with about 500 species (Giussani 2000), is one of the largest genera of the Poaceae family and shows high ecological and reproductive diversity. Spread worldwide because of its high morphological variability and adaptability, the origin of the facultative apomict *Poa pratensis* L. (Kentucky bluegrass) is believed to be Eurasia.

As a forage and meadow grass, Kentucky bluegrass combines a number of agronomically valuable traits like cold and drought resistance, persistence, multiple cutting tolerance, resistance to trampling and good density of sward, which favour its utilization in different areas. The capacity of Kentucky bluegrass to reproduce sexually also makes it a valuable source for breeding programmes.

The aim of our research is to gain a comprehensive description of the variability in a major part of the European *Poa pratensis* ecotypes/wild material. This will be achieved by summarizing field data, analyzing reproductivity patterns and generating molecular fingerprints. The latter will be conducted with the help of the AFLP technique (Vos *et al.* 1995) and represents the first molecular analyses of diversity within the species *Poa pratensis*. These data will be used to characterize the surveyed material, and in combination with morphological and cytological data, will cover the topics of putative differences in degrees of diversity in relation to the place of origin, of stable inheritance of traits between generations and of the modes of reproduction, i.e. the presence of pure sexual or apomictic types or their combination, as well as providing the first clues on the influence of the environment on this character.

Material and methods

For more than 1600 ecotypes and wild accessions of European origin (for countries of origin and number of accessions, see Table 1), a first round of evaluations was carried out at Boldebuck, Bornhof, Malchow/IPK (Institute of Plant Genetics and Crop Plant Research), and Malchow/NPZ (Norddeutsche Pflanzenzucht). The largest number of accessions originated from Poland, Germany, Norway and Iceland.

Over a 2-year period, the entire collection of material was evaluated for 27 morphophysiological traits (an overview of the investigated traits – vegetative, generative and disease susceptibility traits – is given in Table 2). Here, three breeding companies – Deutsche Saatveredlung (DSV), Norddeutsche Pflanzenzucht (NPZ) and Saatzucht Steinach (SZS) – participated along with the Malchow branch station of IPK Gatersleben.

The evaluation was carried out on blocks with 10 single plants per accession. For statistical analysis of the field data, all ecotypes were grouped according to their country of origin, and the frequency of the individual scores for the accessions from each country was calculated. Using the software NTSYS-PC, the diversity between the accessions was calculated according to Nei and is presented in an UPGMA (unweighted pair group method using arithmetic averages) phenogram.

Table 1. Countries of origin of the material

Country (ISO country code)	No. of accessions
BEL	2
CHE	16
CSK	15
CZE	2
DEU	116
DNK	43
ESP	7
EST	3
FIN	4
FRA	14
GBR	3
GRL	24
HRV	15
HUN	83
ISL	50

Country (ISO country code)	No. of accessions
ITA	1
LTU	43
MNG	30
NLD	50
NOR	57
POL	936
PRT	1
ROM	5
RUS	48
SLO	12
SUN	20
SVK	8
SWE	41
TUR	1
YUG	9

Table 2. Traits evaluated in the examined *Poa* material

Vegetative traits
Growth type
Leaf width
Leaf colour
Homogeneity in vegetative state
Mass
Plant density
Offshoot development
Status before winter
Status after winter
Spring growth
Biomass
Growth after cutting

Generative traits
Uniformity in generative state
Shoot growth
Flowering
Height after flowering

Disease susceptibility
Rust
Powdery mildew
Leaf necrosis
Red leaf tips
Others

Results and discussion

The phenogram resulting from evaluations at IPK-Malchow (Fig. 2; mainly accessions from Poland, northeastern European countries, Slovenia and Switzerland) groups 525 ecotypes from 15 countries into two major groups. The first group is subdivided into two sub-clusters, with one sub-cluster with southern European material (Switzerland, Slovenia), while the other sub-cluster contains accessions from central/eastern European countries (Denmark, Poland, Slovakia, Romania). The second major group, which contains mainly material from the northern European countries (Iceland, Norway, Sweden, Greenland, and also the United Kingdom), shows a rather high similarity between accessions of Norwegian, Swedish and Icelandic origins. The material from the remaining four countries (Hungary, Portugal, Finland and Italy) does not form a distinct cluster and shows the highest distance from the other countries: this might be caused by the low number of accessions evaluated from these countries.

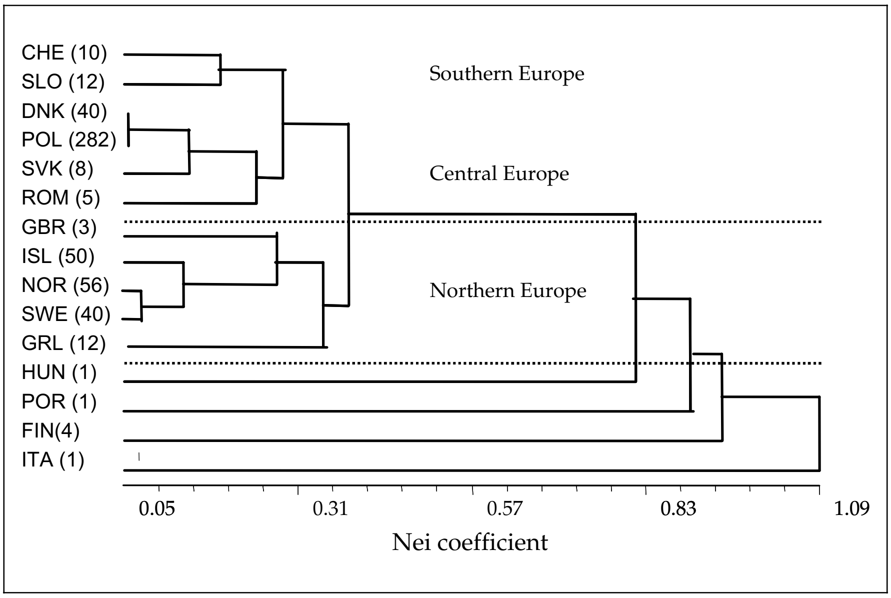

Fig. 2. UPGMA phenogram of 525 *Poa pratensis* ecotypes from 15 countries, based on the evaluation of 24 morphological traits (data from IPK-Malchow).

Concerning putative correlations between the variability of morphophysiological traits and geographical origin, a significant grouping of the ecotypes is observed, especially regarding susceptibility to rust and panicle formation: accessions from northern European countries suffer less from rust than those from central and southern Europe (Fig. 3). Here, the highest variability for this trait is exhibited in central European material, which has scores from 9 to 1. The ecotypes from southern European countries are very susceptible to rust, which can be explained by the physiological properties or requirements of the fungus.

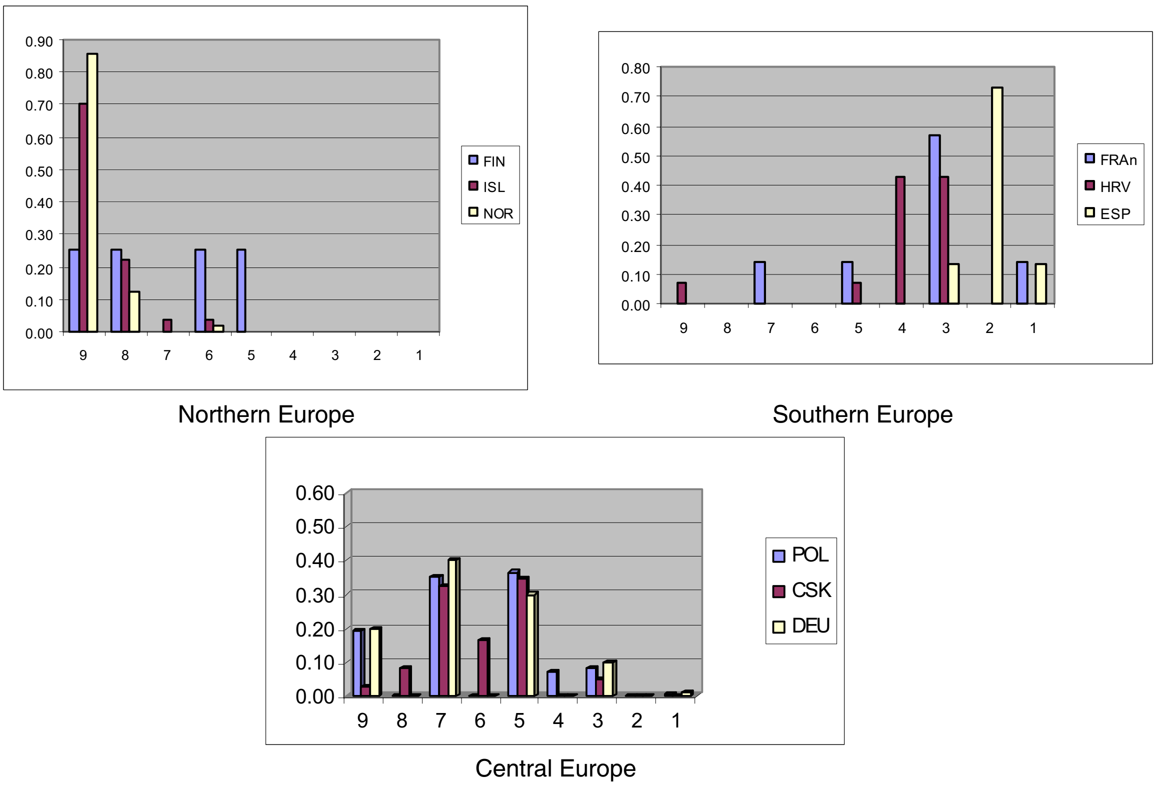

Fig. 3. Evaluation data on rust resistance, grouped by geographic region.

A similar tendency was observed for time to panicle formation (Fig. 4): again there are differences based on region of origin, with the latest types coming from Finland and Iceland.

In general, it can be concluded that in Kentucky bluegrass, the investigation of 27 traits seems sufficient for characterizing and grouping the ecotypes.

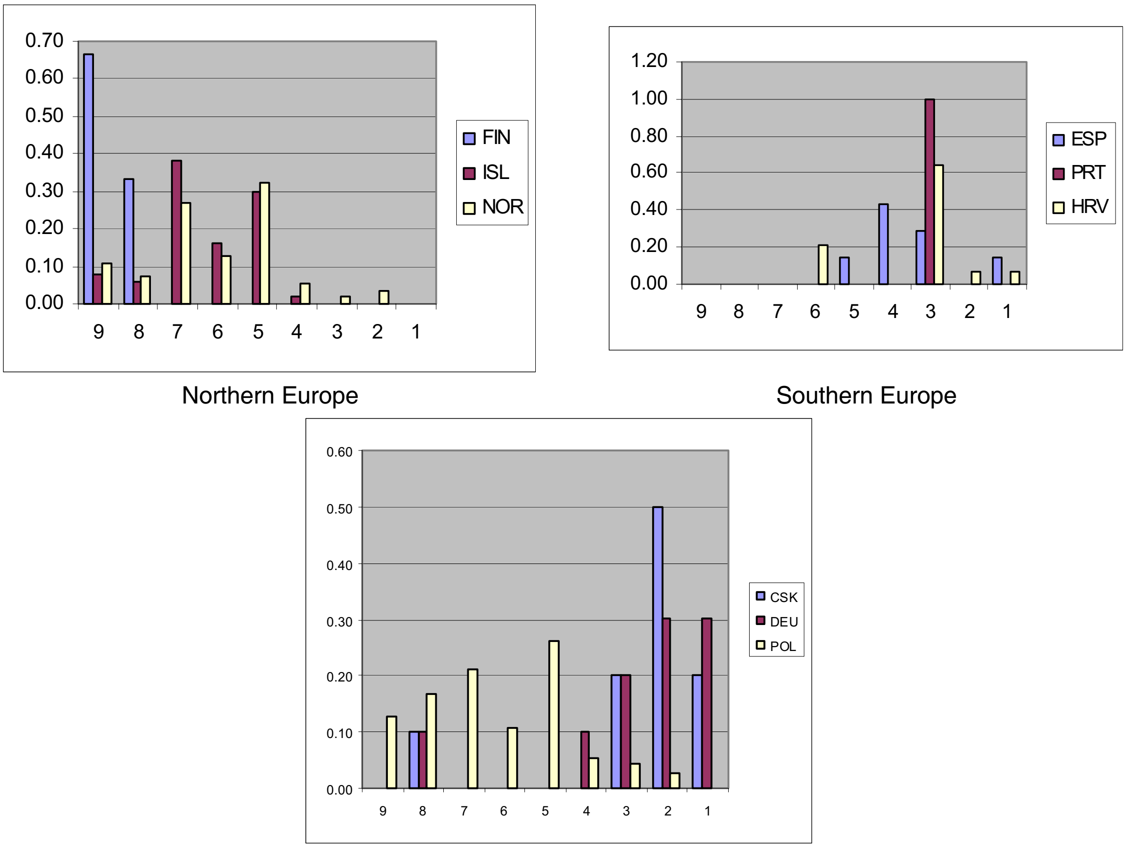

Fig. 4. Panicle formation evaluation data, grouped by geographic region.

Further steps of analysis

The application of molecular markers is aimed at enabling estimation of the genetic diversity which will be independent of the environment. Leaf material from the clones of all mother plants selected for a secondary evaluation was collected and DNA was isolated and quantified. In parallel to this, the selection of AFLP primer combinations was started. 247 Pst/Mse and 183 Eco/Mse primer combinations were tested in different PCR (polymerase chain reaction) conditions. Usually, high quantities of PCR products were observed, but by increasing the number of selective bases in the amplification reactions, a selection of suitable Pst/Mse combinations was achieved.

In addition, the offspring will be analyzed using Flow Sorter Seed Screens to distinguish possible pure apomicts from sexually reproducing *Poa* types. Together with results from ploidy analyses of offspring which diverge from their mother plants, these results will be investigated in combination with the results from AFLP and the field.

Acknowledgements

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Study of quantitative features in Phalaris arundinacea *accessions in two different environments in Romania*

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Introduction

Reed canarygrass, *Phalaris arundinacea* (syn. *Typhoides arundinacea; Digraphis arundinacea*) represents an important source for the diversification of fodder plants in Romania. *Phalaris arundinacea* is characterized by its adaptability to moisture, high productivity and poor forage quality. The interaction of the constitutive elements of the cultivated species in different environmental conditions was studied, in order to increase the diversity of the species and also to increase the cultivation area:

- the first study area, characterized by low precipitation, is the Banat Plain – the study was carried out in the Didactic Station of the University of the Agricultural Sciences and Veterinary Medicine of Banat Timişoara (90 m alt.);
- the second area is located in the Barsa Depression – the study was carried out in the Grassland Research and Development Institute, Braşov (600 m alt.).

Materials and method

The material consisted of 1000 genetically identical clones collected from the Danube Delta and near the lowlands up to 1100 m altitude in mountain zones of spruce fir forests which have been cultivated in these two regions in two experimental fields since 2000. The pedological and climatic features of the Banat Plain region are characterized by a chernozemic soil type, yearly rainfall of 500-550 mm and average temperature 10.5°C; and the Braşov area by chernozemic soil, rainfall 700-800 mm and average temperature 7.8°C.

We studied the timing of flowering data, plant height, foliar surface, bunch surface and dry matter yield (q) in the two field collections. Significant morphological variability was shown. Correlations and determination coefficients between the quantitative features were also studied. Statistical measurement methods were used.

Provenances of *Phalaris arundinacea* 'Premier' cultivated in Timişoara were used as the control population for all other populations cultivated in the experimental trial at Braşov.

Results and discussions

For plant height, we found that only the 'Szarvasi' variety ranked below the control 'Premier' cultivated in Timişoara. The highest significantly positive differences from the control were observed for 'Populatie Romaneasca' and 'Populatie Sampetru'. Some provenances were close to the control such as 'Maxier Timişoara' and 'Populatie Bod'.

For dry matter production we observed that the same 'Szarvasi' variety ranked below the control. The 'Bod BV' population was very close to the control. The highest significantly positive differences from the control were for 'Sampetru' and 'Arpasi' provenances.

The climatic conditions of the Braşov area in comparison with Timişoara had a great influence on the plant surface, the control having a smaller plant surface.

Results obtained for the foliar surface ranked the control 'Premier Timişoara' between the four other Braşov populations and 'Szarvasi', 'Populatie Romaneasca', 'Populatie Geoagiu' and 'Populatie Topolovat'. The closest to the control were 'Populatie Moara Domneasca' and 'Populatie Podu Olt'.

The regression between plant height and dry matter production gives the following results: in Braşov the maximum dry matter production was for plants between 140 and 200 cm in height, and in Timişoara, for plants between 60 and 80 cm in height.

The same relations are illustrated for foliar surface and dry matter production; the correlation indices were significantly higher in Timişoara (r=0.71) as compared to Braşov (r=0.30).

Results of the regression between plant height and plant surface are as follows: the highest value for plant surface was for plants between 120 and 200 cm in height in Braşov and for plants between 172 and 178 cm in height in Timişoara.

Conclusions

Genetically identical material may behave differently under different climatic conditions, especially as regards production characteristics. For plant height, dry matter production and plant surface the Timişoara trial ranked below that of Braşov, with the exception of 'Szarvasi'. There was a wide range in the distribution for the character "foliar surface". The study of interrelations between these characters shows how they are related to yield values. The maximum level of dry matter production was reached by plants of 140-200 cm height in Braşov and 60-80 cm plant height in Timişoara.

Safeguarding a unique collection of former Swiss red clover landraces

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Introduction

Red clover (*Trifolium pratense* L.) landraces used to be maintained and propagated on-farm by Swiss farmers in the 19th and 20th centuries. By consciously harvesting the seed in the last year of stand of a 3- to 4-year ley, the farmers improved the persistence of these landraces which became known under the name of 'Mattenklee', meaning "clover of the leys" (Boller 2000). Numerous "farm landraces" developed, some of which were produced and marketed locally on a modest scale. However, most of the farm landraces were maintained only for the purpose of re-sowing on the farm of origin, often for decades. This tradition was abandoned for the most part in the 1970s. In 1971 and 1972, the former red clover breeder of Reckenholz, Bruno Nüesch, collected samples of about 100 landraces which still existed. After a preliminary agronomic evaluation (Nüesch 1976), the original samples, some of which were of doubtful seed quality, were stored at low temperature without regeneration.

An investigation in 1999 revealed that the germination of about two-thirds of the samples had dropped below 10% and it was recognized that many were in urgent need of regeneration (Table 1). Moreover, the landraces had never been described systematically in terms of morphological or agronomic characters. We therefore decided to completely regenerate and evaluate the collection with the help of a federal grant received from the National Plan of Action for maintenance and sustainable use of plant genetic resources for food and agriculture (NAP). The work was carried out in collaboration between a public research station (FAL Reckenholz) and the seed industry represented by the association of Swiss seed and plant traders (VSSJ).

Table 1. Germination in spring 1999 of original seed samples from the 1971-1972 collection of 'Mattenklee' landraces stored at FAL Reckenholz

% germination	No. of landraces
0 to 5	33
5 to 10	36
10 to 20	20
20 to 40	14
>40	2
Overall average: 10.3%	**105**

Materials and methods

- **Regeneration**

Plots of 25 m² were established by using the original seed samples of 1971-1972 in a rapeseed field in 1999 and 2000, and in a maize field in 2001 at Neuenegg (Bern Canton). The site was chosen because it lies near the centre of the greatest abundance of farm landraces represented in the collection. The plots were arranged in 1.5 m-wide alleys separated by 15 m of rapeseed/maize crop. Within the alleys, the plots were at least 33 m apart. This arrangement allowed for about 30 landraces to be regenerated per year.

For about one-third of the samples, 20 g viable seeds (between 51 and 182 g total seeds, depending on germination) were available for sowing directly with a plot seeder. For the remaining samples of poorer seed availability, 200 to 220 seedlings were raised in the greenhouse. A seed quantity that was expected to yield 400 seedlings, judging by its germination and thousand-grain-weight, was treated with 4 g/kg Thiram 80. If the percentage of hard seeds exceeded that of germinated seedlings, the seed was scarified before the treatment. Seeds were then evenly distributed in seed boxes (40 x 60 x 5 cm). To avoid direct contact between seeds, no more than 15 g seeds were sown in one box. These precautions were taken to minimize loss of seedlings due to seedborne diseases. In most cases, sufficient seedlings emerged; if not, a second batch of seedlings was raised. When they had developed about three true leaves, seedlings were transplanted to 54 unit quickpot trays and grown on for at least two months. They were then transplanted into the field at 30 x 40 cm distances. A bulk seed harvest was taken from each plot with a plot harvester in late summer of the second year after sowing.

- **Evaluation of morphological and agronomic characters**

A choice of compulsory UPOV characters was observed or measured on 50 to 60 spaced plants per accession at Zürich-Reckenholz. These plants were also evaluated for persistence over three years. Additionally, row trials with 3 replications were observed for 3 years in one location. Yield and persistence were evaluated in standard plot trials with three replications in one to three locations, depending on seed availability. The entire material was split into subsets, each sown or planted in 2001, 2002, 2003 and 2004. First results are available for the subset established in 2001.

Results and discussion

- **Regeneration**

Seed yield varied greatly among the three harvest years 2000, 2001 and 2002, and among the landraces. We aimed to obtain 300 g viable seeds per landrace for storage and evaluation. This objective was achieved for 45 landraces (Table 2). The poor yield in 2002 was due to a greatly delayed harvest because of bad weather conditions. Game deer also caused some damage in all years.

A high degree of hardseedness was observed for many samples. However, the sum of germinated seedlings and hard seeds mostly exceeded 80%, which was considered satisfactory for long-term storage. Seed multiplication of landraces with insufficient seed yield or quality is being repeated, starting in 2002 and continuing 2003 to 2004. By the end of 2004, we hope to successfully safeguard 80 to 90% of the original stock of landraces.

Table 2. Seed yield and quality of harvests 2000 to 2002 from regeneration of 'Mattenklee' landraces

Harvest year	Plots established	Seed yield per 25 m^2-plot		Germination			No. of landraces with >300 g viable seeds
		mean	std. dev.	% germinated seedlings	% hard seeds	% total(*)	
2000	32	677	398	65.5	27.2	85.2	26
2001	32	488	399	67.6	12.5	78.5	19
2002	32	56	62	79.4	13.9	92.8	0

(*) % germinated seedlings +% hard seeds, except fractions >20%

- **Evaluation of morphological characters**

The first evaluation year 2002, on 33 landraces, revealed significant differences for flowering date, stem length, leaflet length and width and mildew susceptibility. Cluster analysis of these results separated most landraces clearly from the 'Mattenklee' cultivar 'Milvus' (Fig. 1).

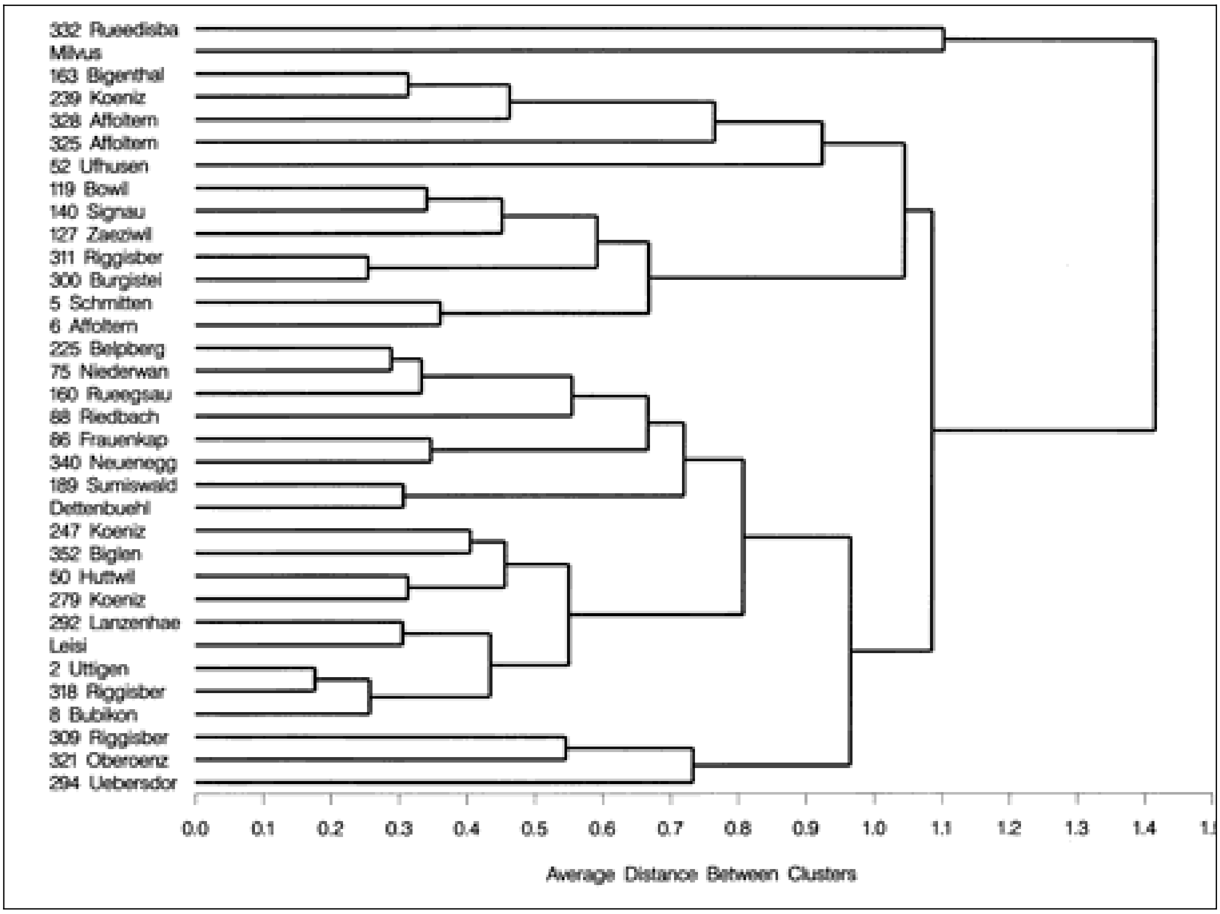

Fig. 1. Cluster analysis of morphological data and mildew resistance observed on spaced plants of 33 landraces and the cultivar 'Milvus'. Deviation from the mean over all accessions was weighted with least significant differences before analysis.

With few exceptions, the landraces were earlier flowering, shorter and more susceptible to mildew than 'Milvus'. Grouping by morphological characters was not related to the geographic origin of the landraces. For example, landraces 309, 311 and 318, all originating from the same small community Riggisberg, were each allocated to a different main cluster. Conversely, one of them (318) was grouped very closely with two landraces that originated from farms 7 and 236 km apart, respectively. These preliminary findings need to be corroborated by the evaluation of a larger number of landraces. A study on the basis of molecular marker diversity confirmed that 'Mattenklee' landraces form a diverse group of accessions clearly distinct from current 'Mattenklee' cultivars (Kölliker *et al.* 2003). In this study, it was also found that clustering on the basis of AFLP markers was not related to the geographic origin of the landraces.

- **Relationship between original seed quality, success of regeneration and morphological characters**

Germination of the original seed sample was positively correlated with seed yield in those plots that had been sown with a plot seeder in 1999 (harvest 2000, Table 3). Apparently, a linear increase in sowing density was insufficient in this case to compensate for poor germination. No correlation between germination and seed yield was observed in the other years.

Table 3. Correlation between seed yield and germination of original seed sample in sown and planted plots harvested 2000 to 2002

Harvest year	Method of plot establishment	Seed yield (g/plot)	Coefficient of correlation (r) between seed yield and germination of original sample (*p* for r≠0)
2000	Planting	650	n.a.
	Sowing	678	0.35 (*0.05*)
2001	Planting	503	0.10 (*0.64*)
	Sowing	450	-0.11 (*0.76*)
2002	Planting	56	-0.19 (*0.31*)

Failure to raise sufficient seedlings for transplanting resulted from extremely poor germination only. From all except one sample which had shown at least 2% germinated seedlings in an official ISTA test, it was possible to raise the desired number of 200 plants. Even seven samples with 0 to 1% germination yielded enough plants for transplanting. Once established, these planted plots yielded as much seed as those sown with the plot seeder, irrespective of their original germination (Table 3). Seed quality (germination and purity) tended to be better for the planted vs. the field sown plots.

Poor germination might lead to a shift in the population structure because long-term seed survival could be linked with other plant characters. Indeed, there was a trend for germination of the original seed sample to be correlated negatively with flowering time (Table 4). This would indicate that genotypes that survived long seed storage would tend to flower later than the population mean. More data are needed to confirm and interpret this trend.

Table 4. Correlation between original seed germination or seed yield during regeneration and morphological characters of 32 'Mattenklee' landraces

Character	Coefficient of correlation, r (with *p* for r≠0) between	
	Character and original germination	Character and seed yield
Date of flowering	0.32 (*0.07*)	-0.12 (*0.51*)
Stem length	0.14 (*0.43*)	-0.32 (*0.07*)
Leaflet length	0.04 (*0.83*)	-0.18 (*0.31*)

- **Evaluation of agronomic characters**

Results for the first full harvest year of the first series of trials indicate that most farm landraces have an inferior yield potential as compared to current cultivars of the 'Mattenklee' type. Annual dry matter yield of the 20 farm landraces averaged 91% of that of the 'Mattenklee' cultivar 'Milvus', ranging from 86 to 101%. 'Dettenbühl' was the only landrace that outyielded 'Milvus' slightly: formerly, it used to be produced on a modest scale for the local market, and had been recommended officially in the 1960s and 1970s. These preliminary results must be complemented by the second full harvest year, decisive for the expression of the 'Mattenklee' character of persistence.

Outlook

The primary goal of safeguarding an important part of the remaining pool of Swiss red clover landraces can probably be achieved within the next two years. This unique material will be made publicly available. We plan to complete morphological description and agronomic evaluation of the collection to assess its value as a potential genetic resource for further breeding. The results will be complemented further by molecular marker studies, and by a study of performance of some landraces at their sites of origin. These studies will be useful for making a choice of landraces to be included in a breeding programme, and may result in the identification of obsolete duplicates in the collection.

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*Evaluation of orchardgrass (*Dactylis glomerata *L.) populations collected in Turkey*

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Introduction

Orchardgrass (*Dactylis glomerata* L.) is a perennial forage grass and has significant potential for grazing and hay production. Orchardgrass is widespread in southern and central Europe, the Mediterranean, Anatolia and the Middle East. Harlan (1983) stated that orchardgrass is an important forage crop in Europe and Mediterranean gene centres that include Turkey. Orchardgrass is tolerant to grazing and cutting, has early spring growth and a good regrowth capacity (Genckan 1983; Açikgöz 1995).

Orchardgrass with good adaptability grows commonly across Turkey. Some studies showed that orchardgrass genotypes responded differently to various climatic conditions and found a wide range of variation in yield characters (Tosun and Sagoz 1994; Tukel and Hatipoglu 1994; Avcioglu *et al*. 1999).

Genetic variation is an important source for plant improvement: thus making use of plant genetic resources with a wide range of variation is essential. Therefore, genetic resources of plants such as orchardgrass should be collected, evaluated and maintained for current and future uses.

The aim of this study was to evaluate the orchardgrass populations collected from different parts of Turkey.

Material and method

Thirty-two orchardgrass populations were collected from various parts of Turkey. Collecting sites are shown in Fig. 1.

Seeds were sown in pots and transplanted to the experimental field of the Aegean Agricultural Research Institute in 2001. The following characters were measured during 2002:

- plant height (cm): distance from soil surface to top of panicle;
- green yield (g/plant): weight of plants harvested 5 cm above the soil level at heading;
- dry matter yield (g/plant): weight of samples used to determine fresh yield after oven-drying at 78°C for 48 hours;
- tillering capacity: scaled visually after cutting (1 = very few tillers; 9 = very numerous tillers);
- vegetative growth habit: expressed as the mean angle of tillers from the vertical in the following spring after establishment year (1 = prostrate; 4 = erect);
- growth habit at inflorescence: expressed as the mean angle of tillers from the vertical (1 = prostrate; 4 = erect);
- time of 50% inflorescence emergence: time in the year following establishment when 50% of the spaced plants reached inflorescence emergence (1 = very early; 3 = early; 5 = intermediate; 7 = late; 9 = very late);
- abundance of inflorescences: visual estimates of the number of inflorescences per plant at heading (1 = very low; 3 = low; 5 = intermediate; 7 = high; 9 = very high);
- statistical analyses: minimum, maximum, and mean values of populations for each character were calculated and data on productivity characters such as green and dry matter yields and plant height were statistically analyzed using analysis of variance.

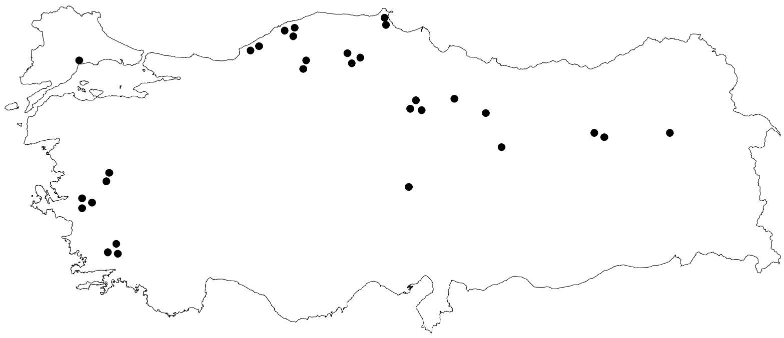

Fig. 1. Collecting sites of orchardgrass populations in Turkey.

Results and discussion

Mean, minimum and maximum values of characters observed are given in Table 1. Based on analysis of variance, the differences between populations were significantly important for all characters. These results are in agreement with the findings of some previous work (Tosun and Sagsoz 1994; Tukel and Hatipoglu 1994).

Table 1. Mean performances and minimum and maximum values of orchardgrass populations

Character	Mean	Minimum	Maximum
Green weight yield (g)	1387	731	2383
Dry matter yield (g)	365	208	519
Plant height (cm)	23	11	44
Tillering capacity*	5.14	3.2	7.3
Vegetative growth habit*	2.81	2.1	3.7
Growth habit at inflorescence*	2.17	1.1	3.0
Time of 50% inflorescence emergence*	4.73	3.0	6.7
Abundance of inflorescences*	4.45	2.7	6.0

*Visual score

Dry matter frequencies of populations are given in Fig. 2. Fifteen out of 32 populations (i.e. 47% of the total material) were above the mean value of 365 g.

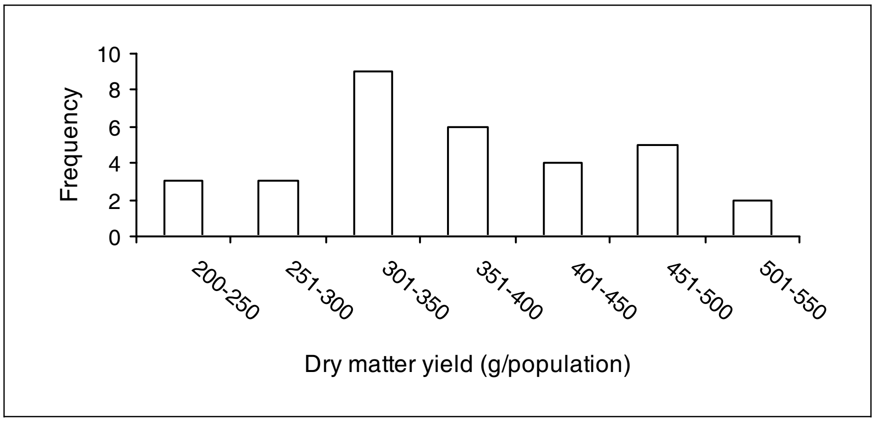

Fig. 2. Dry matter yields of orchardgrass populations.

This study shows that there is a wide range of variation between *Dactylis* populations for the characters examined. Variation is a very important source for the success of plant breeding. Therefore utilization of these accessions in plant breeding programmes will be beneficial.

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Evaluation of the agronomic value of perennial fodder legumes in Georgia

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Introduction

For the last two centuries Georgia has not been self-sufficient for animal feed. Average roughage production from hay lands, arable lands, pastures, grain mills and other byproducts covers only 60% of the country's livestock physiological diet requirements, especially in plant albumins. Therefore, an increase in the production of local forages in both the arid and humid environments is necessary to satisfy the present and future needs of the livestock industry. Many legume forage species, both annual and perennial, have been tested in Georgia under very diverse environmental conditions. The ecosystems and landscapes of Georgia change over small distances from high mountains with glaciers and eternal snow (up to 5068 m asl, peak Shkhara) to subtropical landscapes at the Black Sea coast (the sea coastal area extends for 315 km) and steppes and semi-deserts in the southeast. In such different climatic and soil conditions we collected over 2000 well-adapted landraces of fodder legumes. The next stages were the establishment of a genebank for *ex situ* conservation, *in situ*/on-farm conservation, multiplication and testing of the collected material.

The results indicated that the tested material was not always successful and, therefore, it was decided to place more emphasis on the use of the local genetic material for improving fodder and pasture crops. Priority was given to *Medicago, Onobrychis, Trifolium* and other forage species.

Material and methods

All the promising genetic material collected during 1998-2002 from different parts of the country at various times was multiplied in three typical regions and when sufficient seed was available, was evaluated in replicated trials at three locations for several parameters. With this local material we have tested the legume nursery obtained from the CGIAR international centres (ICARDA, ILRI, IPGRI, ICRISAT, IITA, etc.). The parameters tested were date of germination, flowering, plant height at flowering, biological and dry matter yield, dry matter content, nitrogen yield and content, digestible organic matter yield in herbage dry matter and percentage digestibility, level of biological nitrogen fixation. In many cases this germplasm was also used in the breeding programmes of the Georgia Agrarian State University (GASU) and neighbouring countries.

Results and discussion

- ***Medicago* species**

Alfalfa is well known in Georgia as the oldest (above 5000 years) forage crop and is grown as indigenous populations (Korakhashvili 1991). Testing of *Medicago* species to evaluate their potential role in Georgian agriculture began in the 1960s using local and some introduced material and has been intensified since 1990. The aim was to explore the possibility of using alfalfa in a cereal forage legume rotation, always with five fields (three fields of alfalfa), or as a permanent self-regenerating or partially reseeded pasture for the marginal lands (in this case we used alfagraze as a control). The main conclusion of the tests is that alfalfa was neither successful in rotations with cereals on arable land nor for pasture improvement on marginal lands, but was very successful with corn and soybeans. The main reasons for this failure were the extremely slow growth of all alfalfas during winter time, resulting in severe weed competition and late availability of forage for grazing; the much lower dry matter yield compared to that of other legumes; and the unsatisfactory regeneration of alfalfas for establishing a good pasture stand in the following seasons, even in the case of alfagraze. Recent studies at the international institutions have shown that locally selected medic germplasm shows more promise that any introduced varieties. Medic collecting was also done in Georgia in 1993 by American scientists (Prof. Carl S. Hoveland, Department of Crop and Soil Sciences, University of Georgia). In 1998-2002 two new varieties of alfalfa (alfagraze) were released. The advantage of these varieties is their resistance to saline soils and drought. Some agronomic values of the tested alfalfa material are presented in Table 1 (no. 10 corresponds to one of the above-mentioned new varieties).

Table 1. Agronomic values of the tested alfalfa material (*Medicago* spp.)

No.	Species/variety	Organic matter yield (t/ha)	Nitrogen fixation (kg/ha/year)
1	*hemicycla*	2.27	28.8
2	*minima*	2.81	31.7
3	*glutinosa*	4.17	69.3
4	*djavakhetica*	4.88	77.8
5	*orbicularis*	2.23	38.3
6	*lupulina*	2.17	44.3
7	*coerulea*	5.82	81.0
8	*falcata*	3.34	58.3
9	*sativa*	6.12	102.8
10	*sativa* cv. alfagraze	5.42	86.7

- ***Onobrychis* species**

Some scientists (Korakhashvili and Bolkvadze 1992; Agladze and Korakhashvili 1998) state that the motherland of *Onobrychis* is Georgia. There are 3 local genotypes and above 40 varieties of *Onobrychis* have been tested in our collection in various soil and climate conditions for the improvement of grasslands as well as for the forage rotation in low rainfall areas of the country. *Onobrychis* has high drought tolerance, high nutritive quality, high biological nitrogen fixation, generally good pest resistance, and the longest productive season of any perennial legume in dry environments. Since low precipitation is a serious problem in eastern Georgia, the potential for *Onobrychis* as a pasture plant has been limited. A major limitation of *Onobrychis* for pasture use has been its lack of persistence under continuous grazing (Korakhashvili and Agladze 2000). Breeding and selection for drought-tolerant *Onobrychis* on the base of local varieties has made some progress. This new species of *Onobrychis* has the potential to improve livestock production on perennial pastures in the dry regions. Table 2 shows some agronomic values of the tested material.

Table 2. Agronomic values of the tested *Onobrychis* material

No.	Species	Organic dry matter yield (t/ha)	Nitrogen fixation (kg/ha/yea)r
1	*transcaucasica*	2.12	17.2
2	*sativa*	4.78	48.3
3	*vaginalis*	3.31	41.4
4	*viciifolia*	2.83	38.2
5	*kachetica*	4.12	51.7
6	*meschetica*	4.07	46.0
7	*iberica*	3.81	43.5

- ***Trifolium* species (clovers)**

Observations in Georgia have shown that some genotypes of *Trifolium* behave as a main pasture crop and it may be used for pasture development. All species of clover are able to regenerate naturally, except where overgrazing occurs. Even though clovers are highly desirable for nitrogen fixation and improving forage nutritive quality, they are not generally an important component in humid region pastures of west Georgia. In a recent survey by pasture extension specialists (Agladze and Korakhashvili 1998), the percentage of pasture containing fair-to-good stands of clovers were rated as about 30% in the northeast, 24% in the north-central region, and only 16% in the south. The instability and lack of dependability of clovers is an important reason for the reliance upon grass-N pastures, especially in the southern regions of Georgia. White clover, the most widely planted feed legume in the humid areas, lacks drought tolerance and is highly susceptible to a variety of insects, diseases and nematodes (Hoveland 1989). There are three types of white clover: large, intermediate and small. Unfortunately, no improved persistent cultivars of this species for stressful environments are available. Red clover is more productive for about 3 years in the west of Georgia (Cubero and Moreno 1979). In the lower-lying areas of the west, stands persist for only 1 to 2 years (Didebulidze and Plachter 2002). Various diseases, insects and nematodes contribute to short stand life. Many naturalized varieties and strains of clovers are well adapted to the lower western part of the country; they are prolific re-seeders and usually act as annual plants. Table 3 gives some of the agronomic values of the tested *Trifolium* material.

Table 3. Agronomic values of the tested *Trifolium* material

No.	Species	Organic dry matter yield (t/ha)	Nitrogen fixation (kg/ha/year)
1	*repens*	1.67	24.7
2	*pratense*	3.88	66.3
3	*fragiferum*	1.58	21.4
4	*neglactum*	1.42	22.7
5	*ambiguum*	1.83	20.8
6	*alexandrinum*	2.08	37.4
7	*incarnatum*	1.89	29.0

Conclusion

The selection of local germplasm holds more promise in the search for improved material than the introduction of foreign varieties of alfalfa, *Onobrychis* and clover. Following this recommendation, the best 3 species will be promoted for future evaluation in larger plots for their persistence and other agronomic parameters. The fodder species alfalfa, *Onobrychis* and clover can be cultivated on an area covering about 184 000 ha of pastures and meadows, as well as 62 000 ha of arable lands in rotations. With the transition of our country to a free market economy with sustainable agriculture, the surface under these forage legumes will increase nearly threefold. Increasing the area cultivated under these crops depends greatly on improvements in seed production, which is still the main cause of the slow development of forage production.

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The use of phenological data for the estimation of global warming "fingerprints" on forage grasses

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Introduction

Evaluation of germplasm is, in the broad sense, the description of the material in collections. Since the beginning of genetic resources activities, numbers of descriptor lists with detailed descriptions of evaluation methodology have been accepted and published. In the course of genetic resource evaluation and characterization, numerous qualitative and quantitative traits were described, assigned to each plant accession and finally, together with passport data, entered into crop-specific databases.

But are there any unexpected advantages arising from large and highly specialized databases? In the case of the databases of the grass collection stored in the Botanical Garden of Plant Breeding and Acclimatization Institute this seems to be the case. The first evaluation data came from field collections of grass ecotypes in 1973. Since then, each year hundreds of grass ecotypes have been grown in similar soil and climatic conditions and evaluated with similar methodology (OECD 1971). Such long-term phenological records are of great value for climate "fingerprinting" determination (Parmesan and Yohe 2003; Root *et al.* 2003).

Phenological phases of plants such as heading or flowering are mainly driven by environmental factors. Among these are pests, diseases, competition, soil properties, genetics, age and, most importantly, weather conditions (Menzel and Fabian 1999; Menzel 2000). The most interesting feature of phenology is that changes in seasonal plant activities from year to year may be a sensitive and easily observable indicator of changes in the biosphere (Chuine *et al.* 1998; Schwartz 1998; Fabian and Menzel 1999; Menzel and Fabian 1999; Chuine *et al.* 2000; Menzel 2000; Sparks *et al.* 2000; Menzel *et al.* 2001). At the end of the last century there was an emerging recognition that phenological recording can be especially useful in environmental monitoring and it has gained the UK government approval as an indicator of climate change (Sparks *et al.* 2000).

The response in phenology to global warming depends on the nature of the change in climate and is variable among species: 75% of European tree and shrub species showed no significant trends while others showed significant negative trends (phenology advanced) or positive trends (phenology delayed) in response to global warming (Chuine *et al.* 2000).

Surveys have been made concerning global meta-analyses (Parmesan and Yohe 2003; Root *et al.* 2003), European tree and shrub species (Fabian and Menzel 1999; Menzel and Fabian 1999; Chuine *et al.* 2000; Menzel 2000; Sparks *et al.* 2000; Menzel *et al.* 2001; Kramer *et al.* 2000), and few wild-growing perennial species (Cenci and Ceschia 2000; Sparks and Carey 1995; Sparks *et al.* 2000) but little is known about perennial forage species. However, long-term observations of some phenological events for forage species exist in databases as an element of characterization of plant genetic resources (Faberová and van Hintum 1995; Horváth and Szabo 1997). Among all the traits observed and recorded in forage grass collections (heading and flowering date, height of plants, length and width of stem leaf, length of inflorescence, etc.), the heading date is probably the single most important character which determines the growth rhythm of plants (Tyler *et al.* 1987).

The aim of the work presented here is to describe long-term trends in mean heading date for Polish ecotypes of three forage grass species.

Material and methods

- **Plant data**

Phenological records for 11 589 ecotypes of three perennial forage grass species were used: Kentucky bluegrass (*Poa pratensis* L.) - 2475 ecotypes; orchard grass (*Dactylis glomerata* L.) - 6005 ecotypes; and meadow fescue (*Festuca pratensis* Huds.) - 3109 ecotypes. Data for native Polish ecotypes were selected and used for the analysis.

Ecotypes were grown in one place – the Botanical Garden of the Plant Breeding and Acclimatization Institute, Bydgoszcz, Poland (53°10'28"N, 018°02'49"E). Each year since 1973, a set of ecotypes collected during missions in the previous year has been planted in a spaced-plant nursery. A 3-replicate design was used with 30 plants per ecotype (10 plants per one plot). Phenology, i.e. date of heading, was always noted 2 years after planting.

For statistical calculations "date of heading" was expressed as the number of days from 1 April to the moment when on 30% of spaced plants of a particular ecotype "emerged" tillers were visible (not less than 3 tillers per plant) (OECD 1971). Emerged tillers were noted when part of the head became visible above the sheath of the flag leaf. Extreme values were omitted from each year's data set.

Gaps in heading records were mostly due to the different results of the various collecting missions. Missions carried out in the 1970s were never species-oriented but rather site-oriented. This resulted in different numbers of ecotypes of particular species collected in the various sites sampled in different years. Between 1973 and 2002 *Poa pratensis* was evaluated over 24 years, *Dactylis glomerata* over 17 years and *Festuca pratensis* over 16 years.

- **Temperature data**

The Jones temperature data set (Jones *et al.* 2001) was used as the reference temperature data. It contains annual temperature time series data at a grid-box resolution of 5° latitude by 5° longitude over the period 1880-2000. Annual temperatures are expressed as *departures from normal temperature*, i.e. means of the 1961-1990 reference period. The Jones data set is a product of the Climate Research Unit of the University of East Anglia and the Hadley Centre of the United Kingdom Meteorological Office and is available via Internet. For this report data from the grid-box covering most of Poland (between 50°-55°N and 15°-20°E) were selected.

- **Statistical calculations**

All statistical procedures were performed using the STATISTICA package (v. 5.0, StatSoft Inc. 1997). The normality of distribution of all phenological records was tested with Kolomogrov-Smirnov ($K\text{-}S_d$) test. Heading time was related to year by means of linear regression analysis. The significance of trends was tested by the F-test.

Results

Distribution of phenological data used for these analyses followed a normal distribution. This was proved by the $K\text{-}S_d$ test: for *Dactylis glomerata* and *Festuca pratensis* $K\text{-}S_d = 0.119$, and for *Poa pratensis* $K\text{-}S_d = 0.125$, with a probability $P>0.2$ for all species.

Trends for mean heading date (Fig. 1) were all negative and significant for *Poa pratensis* ($t = -2.6$, $R^2 = 23.8\%$, $P = 0.01$) and *Dactylis glomerata* ($t = -2.6$, $R^2 = 32.9\%$, $P = 0.02$) but not significant for *Festuca pratensis* ($t = -1.6$, $R^2 = 13.3\%$, $P = 0.14$). For temperature data a significant positive trend ($t = 2.11$, $R^2 = 14.2\%$, $P = 0.04$) was calculated.

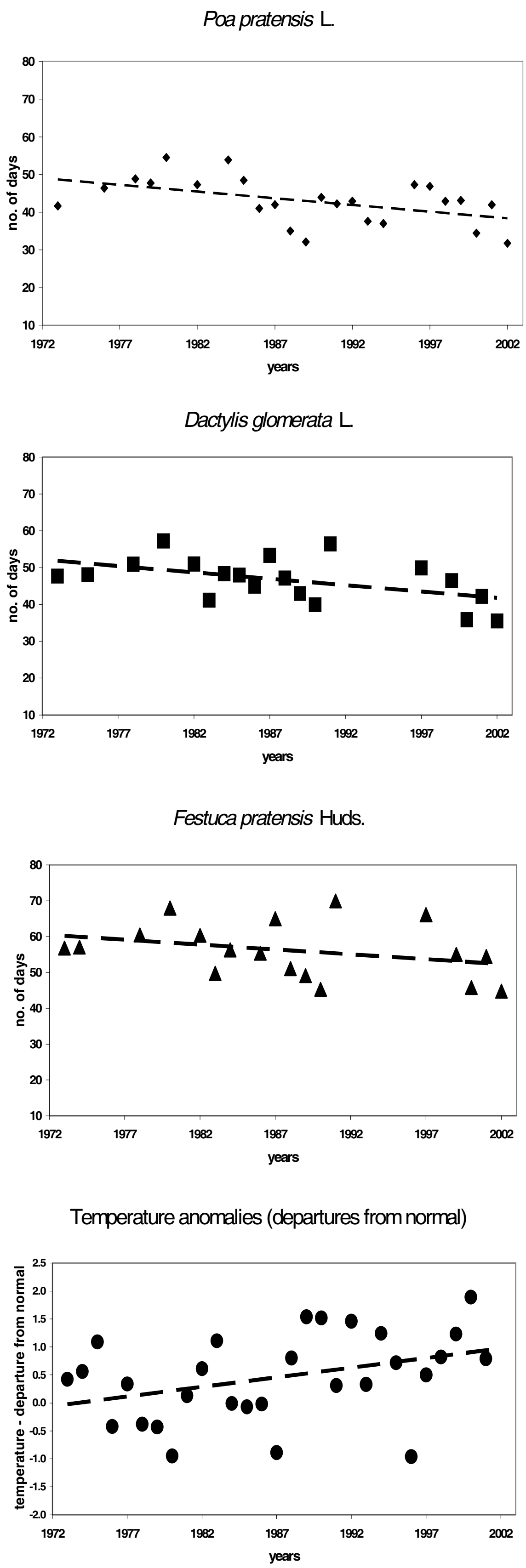

Fig. 1. Trends of mean heading dates and temperature anomalies.

Averaging heading data for given grass species revealed a negative trend of -0.35 day per year (it was the same for *Poa pratensis* and *Dactylis glomerata* but for *Festuca pratensis* it was -0.27 day per year). It is equivalent to an advance of mean heading time of 10 days over 29 years for *Poa pratensis* and *Dactylis glomerata* and of 8 days for *Festuca pratensis,* along with a mean temperature increase of 1°C. Correlation coefficients calculated between mean heading time and temperature data were all negative and statistically significant (for *Dactylis glomerata* $r = -0.81$, $P = 0.00$; for *Festuca pratensis* $r = -0.77$, $P = 0.00$; and for *Poa pratensis* $r = -0.64$, $P = 0.01$). It is therefore clear that along with increasing air temperature, the mean heading date of the above grass species was advanced.

Discussion

It is a well-known fact that air temperature is closely related to forage grass development (Cooper 1959). Different species can show marked differences in their responses to rising temperatures, water stress and increasing CO_2 concentrations (Jones 1997). The latest report by the Intergovernmental Panel on Climate Change (IPCC) predicts a 1.4-5.8°C average increase in the global surface temperature over the period 1990 to 2100 (Luo *et al.* 2001).

Results from our work clearly support the statement that changes in plant behaviour will not only happen in the near future but that they have happened already. In general, over the last three decades, the growing season in Europe has been lengthened by 10.8 days on average (Menzel and Fabian 1999). Global meta-analyses documented significant mean advancement of spring events by 2.3 days per decade (Parmesan and Yohe 2003).

Along with extending the growing season and the faster heading of forages, the total pasture period for animals will also be extended. Yields are likely to be increased most frequently in areas where the temperature already approximates to the optimum for crop growth. However, further increase in temperature will reduce yields by decreasing C gain and accelerating crop development (Polley 2002). Faster seed development may result also in less seed predation (Sparks *et al.* 2000).

Little is still known about the long-term effect of climate change on plant populations. As concluded by Luo *et al.* (2001), ecosystems in a future, warmer climate might acclimatize more rapidly than the current ecosystems do. On the other hand, periodic growth of some species such as *Polygonum bistorta* will put it at a competitive disadvantage relative to plants that can respond to a lengthened growing season (Starr *et al.* 2000). There may well be a general conclusion that all the factors of climate change will have a direct effect on plants, but the duration of these effects and their impact at the level of population and ecosystem is still relatively unknown. The synergism of several pressures (one of which is global warming) is likely to prove to be the greatest challenge to plant conservation in the 21st century (Root *et al.* 2003).

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APPENDICES

Appendix I. Description of the regeneration standards used for forage species

Information compiled by E. Willner and B. Boller according to "Regeneration protocol", pp. 170-183 *in* Sackville Hamilton *et al.* (1998)[9].

Data were provided by the following members of the Working Group on Forages: A. Ghesquiere (Belgium), Y. Guteva (Bulgaria), M. Ševčíková (Czech Republic), E. Willner (Germany), V. Negri (Italy), N. Lemežienė (Lithuania), L. van Soest (The Netherlands), L. Bondo (Nordic countries), J. Drobná (Slovakia), M. Murillo Vilanova (Spain) and B. Boller (Switzerland) (see contact details in list of participants, Appendix V).

For some items of the regeneration procedure, the preferred/acceptable values had been indicated as follows :

Item of regeneration procedure	**Preferred**	**Acceptable**
site		
greenhouse/cabins (yes/no)	yes	
field/cages (yes/no)		
field/isolation with other crops (yes/no)	no	yes
insects as pollinator		
natural population / *commercial* product		
insect species (specify if known)		
crop used as isolation (specify; n.a. for cages)		
isolation distance between plots (n.a. for cages)		
length (m)		>50 m
width (m)		>50 m
plants per accession (*number*)	100	30
distance between single plants	0.20 m	
scoring of traits:		
time of flowering (yes/no)		
others (specify)		
selection in accessions (yes / no) (elimination of other crop plants and weeds)	yes	
harvesting		
once / several (times)		
as *balanced / unbalanced* bulk	balanced	unbalanced
drying		
in *dry room / greenhouse /* using *drying equipm.*	dry room	drying equipm.
threshing and cleaning		
manual / with machines	manual	with machines
final drying		
temperature, relative humidity (specify)		
final moisture content	3-7%	
viability testing before storage (yes/no)	yes	
seed packaging and storage		
base and duplicate collection	per plant	bulk
active collection	balanced bulk	bulk
information management	IT based	

[9] Sackville Hamilton, N.R., K.H. Chorlton and I.D. Thomas. 1998. Guidelines for the regeneration of accessions in seed collections of the main perennial forage grasses and legumes of temperate grasslands. Pp. 167-183 *in* Report of a Working Group on Forages (L. Maggioni, P. Marum, R. Sackville Hamilton, I. Thomas, T. Gass and E. Lipman, compilers). Sixth meeting, 6-8 March 1997, Beitostølen, Norway. International Plant Genetic Resources Institute, Rome, Italy.

Regeneration standards used for forage species – Belgium, Bulgaria, Czech Republic, Germany

	DvP, Belgium		IPGR, Bulgaria		OSEVA, GRS, Czech Rep	RIFC, Czech Rep	IPK-Malchow, Germany	
Item of regeneration procedure	**for grasses**	**for legumes**	**for grasses**	**for legumes**	**for grasses**	**for legumes**	**for grasses**	**for legumes**
site								
greenhouse/cabins (yes/no)	no	no	no	yes	no	no	no	yes
field/cages (yes/no)	no	no	no	yes	no	yes	no	yes
field/isolation with other crops (yes/no)	yes	yes	yes	no	yes	yes	yes	no
insects as pollinator								
natural population / *commercial* product		natural		commercial		natural		commercial
insect species (specify if known)		bumble bees		bumble bees		bumble bees		bumble bees
crop used as isolation (specify; n.a. for cages)	rye		rye		rye or triticale	rye or other	rye	n.a.
isolation distance between plots (n.a. for cages)								
length (m)	15	1000	15		20	200	20	n.a.
width (m)	15	1000	18		20		20	n.a.
plants per accession (*number*)	100	100	49	30-50	49	15-100	49	15-30
distance between single plants	0.4 m	0.7 m	0.25 m		0.25 m	0.15 m	0.25 m	0.3 m
scoring of traits:								
time of flowering (yes/no)	yes		yes	yes	yes	yes	yes	yes
others (specify)	morphol. traits	shape, colour			descriptor list	descriptor list	morphol. traits	morphol.
selection in accessions (yes / no) (elimination of other crop plants and weeds)	yes	yes	yes	yes	yes	yes	yes	yes
harvesting								
once / several (times)	once	once	several	several	several	several	several	several
as *balanced / unbalanced* bulk	unbalanced	unbalanced	unbalanced	unbalanced	unbalanced	unbalanced	unbalanced	unbalanced
drying								
in *dry room / greenhouse /* using *drying equipm.*	drying equipm.	drying equipm.			greenhouse	in lab	dry room	dry room
threshing and cleaning								
manual / with machines	with machines	with machines	manual		with machines	with machines	with machines	with machines
final drying								
temperature, relative humidity (specify)			20°C, 20%		23°C, 3%	23°C, 3%	21°C, 30%	21°C, 30%
final moisture content					4-6%	4-6%	<8%	< 8 %
viability testing before storage (yes/no)	yes	yes	yes		yes	yes	yes	yes
seed packaging and storage								
base and duplicate collection	bulk	bulk	bulk		bulk	bulk	bulk	bulk
active collection	bulk	bulk	bulk		bulk	bulk	bulk	bulk
information management			IT based	IT based	IT based	IT based	IT based	IT based

Regeneration standards used for forage species – Italy, Lithuania , Netherlands, Nordic countries

	DBVBA, Italy		**LIA, Lithuania**		**CGN, Netherlands**		**NGB, Nordic countries**	
Item of regeneration procedure	**for grasses**	**for legumes**	**for grasses**	**for legumes**	**for grasses**	**for legumes**	**for grasses**	**for legumes**
site								
greenhouse/cabins (yes/no)	yes	yes	no	yes	no	no	no	no
field/cages (yes/no)	yes	yes	no	yes	yes	yes	no	yes
field/isolation with other crops (yes/no)	no	no	yes	no	yes	yes	yes	no
insects as pollinator								
natural population / *commercial* product								yes
insect species (specify if known)	*Apis mellifera*	*Apis mellifera*		*Apis mellifera*				
crop used as isolation (specify; n.a. for cages)		n.a.	rye	n.a.	Triticale, wheat		rye	n.a.
isolation distance between plots (n.a. for cages)								
length (m)		n.a.	15-20	n.a.	40	40	100	n.a.
width (m)		n.a.	15-20	n.a.	50	50		n.a.
plants per accession (*number*)	50	50	49-64	25-35	70 (min.25)	70 (min.25)	49	49
distance between single plants	0.5-0.6 m	0.5-0.6 m	0.25 m		0.25 m	0.5 m	0.3-0.5 m	0.3-0.5 m
scoring of traits:								
time of flowering (yes/no)	no	no	sometimes	sometimes	no		no	no
others (specify)								
selection in accessions (yes / no) (elimination of other crop plants and weeds)	yes	yes	yes	yes	yes	yes	yes	yes
harvesting								
once / several (times)	several		several	several	by hand	by hand		
as *balanced / unbalanced* bulk	unbalanced	unbalanced	unbalanced	unbalanced	balanced	balanced	balanced/ unbalanced	unbalanced
drying								
in *dry room / greenhouse* / using *drying equipm.*	yes	yes	greenhouse		dry room			
threshing and cleaning								
manual / *with machines*	yes	yes	with machines		yes	yes	manual / with machines	manual/ with machines
final drying								
temperature, relative humidity (specify)	yes	yes	20°C, 10-15%		15°C, 15%			
final moisture content					5%		5%	5%
viability testing before storage (yes/no)	yes	yes	yes		yes	yes	yes	yes
seed packaging and storage								
base and duplicate collection	yes	yes	bulk		laminated foil bags		bulk	bulk
active collection	yes	yes	bulk				bulk	bulk
information management	yes	yes	IT based		IT based		IT based	IT based

Regeneration standards used for forage species – Slovakia, Spain, Switzerland

	BSLL, Slovakia	BSHS, Slovakia	RIPP, Slovakia		JDE, Spain		FAL, Switzerland	
Item of regeneration procedure	**for grasses**	**for legumes**	**for legumes**	**for legumes**	**for grasses**	**for legumes**	**for grasses**	**for legumes**
site		*Faba*, *Pisum* sp., *Vicia*	*Lupinus* sp.	clover, alfalfa, other forage legumes	no data	annual *Medicago*		
greenhouse/cabins (yes/no)	no	no	no	no		no	no	no
field/cages (yes/no)	no	no	no	yes		no	no	no
field/isolation with other crops (yes/no)	yes	yes	farmers field/no	no		no	yes	yes
insects as pollinator								
natural population / *commercial* product				yes				natural
insect species (specify if known)				*B. terrestris*				bumble bees
crop used as isolation (specify; n.a. for cages)	wheat, barley, maize	wheat		*B. lapidarius*			rye	peas
isolation distance between plots (n.a. for cages)								
length (m)		6 m		n.a.			15	15
width (m)		1 m		n.a.			15	15
plants per accession (*number*)	30-50	360	50-100	15-50		150-200	100	100
distance between single plants	0.5 m	0.1 m	0.4 m	0.25 m		0.15 m?	0.3-0.5 m	0.3-0.5 m
scoring of traits:								
time of flowering (yes/no)	yes	yes	yes	no		yes	yes	no
others (specify)	descriptor list	7 traits	descriptor list			morphol. traits	earliness	
selection in accessions (yes / no) (elimination of other crop plants and weeds)	yes	yes	yes	yes		yes	yes	yes
harvesting								
once / several (times)				once/several*		by hand	once	once
as *balanced / unbalanced* bulk	unbalanced	unbalanced	unbalanced	unbalanced		unbalanced	unbalanced	unbalanced
drying								
in *dry room / greenhouse /* using *drying equipm.*	greenhouse	greenhouse	greenhouse	greenhouse		dry room	drying equipm.	drying equipm.
threshing and cleaning								
manual / *with machines*	with machines	with machines	with machines	manual/ with machines*		manual/ with machines	with machines	with machines
final drying								
temperature, relative humidity (specify)	20°C, 15%	20°C, 15%	20°C, 15%	20°C, 15%		20°C, 15%	35°C	35°C
final moisture content	4-6%	4-6%	4-6%	4-6%			6%	6%
viability testing before storage (yes/no)	yes	yes	yes	yes		yes	yes	yes
seed packaging and storage								
base and duplicate collection	bulk	bulk	bulk	bulk			bulk	bulk
active collection	bulk	bulk	bulk	bulk		bulk	bulk	bulk
information management	IT based	IT based	IT based	IT based		IT based	IT based	IT based

*according to species

Appendix II. Statistical analysis of the IGER European *Lolium* Core Collection Trial

R.N. Edmondson
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Overview by HRI Biometrics

The data set supplied by IGER comprises a large number of core accessions of *Lolium* grass tested at a large number of European sites during a trial commencing in 1995 and lasting for at least three years. Two management systems, conservation or frequent cutting, were tested for each accession at each site. A number of records were made on each accession at each site in at least two years of the trial, the main records being:

i. Heading tendency
ii. Winter damage
iii. Combined bulk yields
iv. Leaf width
v. Growth habit
vi. Inflorescence abundance
vii. Aftermath heads
viii. Disease scores
ix. Emergence dates

We have not yet had time to fully examine the data but a preliminary inspection has shown that the records at different sites are not fully consistent. For example, at some sites individual plants were scored whereas at other sites only plot mean scores were recorded. Most of the sites recorded score data for the measured attributes but at least for the UK site, actual bulk yields were recorded as well as bulk yield scores. For at least one site (Germany), heading tendency was recorded as a binary variable where each plant was given a binary score of 0-1 while at the Belgium site, equivalently, heading tendency was recorded as a binomial count out of 28. However, at most other sites, heading tendency was recorded as a score variable on the range 0-9 either for individual plants or as a mean of plots of 7 plants.

The disease score records involved assessments at different times at different sites and sometimes involved different diseases although only a single overall disease score was recorded.

We think that for at least for two sites, Ireland and Spain, the trial began a year later than at other sites. We have not yet looked at each site in detail but it appears that not every accession was tested at every site.

Proposed analysis

Stage 1

In view of the complexity of the data, we propose that the analysis should be regarded as a two-stage process. In the first stage, we propose to make a statistical analysis of variance of the data from each site individually. We would aim to provide a full comparison of the accessions within each site individually and would aim to use standardized methods for each site so that the results from each site would be comparable across sites. This would allow relative comparisons to be made across sites using tabulated data sets with appropriate standard errors and statistical tests. Where there are comparable records, we would aim to compare the two management systems and to compare the consistency of accession comparisons between the management systems within sites. If feasible, we would aim to include the repeated records over years within the same analysis for each site so that the consistency of the records over years could be compared.

During this stage of the analysis, we would not attempt to make any formal combined analysis across sites or to make any Genotype x Environment analysis using site or environmental variables as explanatory variates nor would we attempt any modelling of the different disease incidence rates at the different sites.

We would expect this first stage of the analysis to provide a valid statistical analysis of each site individually and we would expect to complete our report for this stage of the analysis by late summer 2003.

Stage 2

After completion of stage 1 of the analysis, we would then discuss the results with IGER scientists and we would consider the options for further analysis of the data. Depending on the outcome of Stage 1, there could be the possibility of making a full or partial multi-site analysis of the data using cluster analysis methods or other multivariate or multi-dimensional methods to mode site interaction effects. However, the feasibility and utility of the full Genotype x Interaction analysis of this data set cannot be assessed at present and we do not think we can plan a second stage until the basic analysis outlined for stage 1 has been completed.

We therefore propose that we make an agreement to complete stage 1 of the analysis as outlined above to the satisfaction of the IGER scientists involved and then consider further whether we wish to proceed to the stage 2 analysis.

Appendix III. Acronyms and abbreviations

AARI	Aegean Agricultural Research Institute, Turkey
AFLP	amplified fragment length polymorphism
ARI	Agricultural Research Institute, Nicosia, Cyprus
BRG	Bureau des ressources génétiques, France
BSHS	Breeding Station Horná Streda, Slovakia
BSLL	Breeding Station Levočské Lúky, Slovakia
CBD	Convention on Biological Diversity
CCDB	Central Crop Database
CGN	Centre for Genetic Resources, The Netherlands
CGRFA	Commission on Genetic Resources for Food and Agriculture (FAO)
CPRO-DLO	Centre for Plant Breeding and Reproduction Research, The Netherlands
CRF	Centro de Recursos Fitogenéticos, Madrid, Spain
CRIFC	Central Research Institute for Field Crops, Ankara, Turkey
CSIC	Consejo Superior de Investigaciones Científicas, Spain
DAFRD	Department of Agriculture, Food and Rural Development, Ireland
DBVBA	Dipartimento di Biologia Vegetale e Biotecnologie Agro-ambientali, Università degli Studi, Perugia, Italy
DEFRA	Department for Environment, Food and Rural Affairs, United Kingdom
DvP	Department of Plant Genetics and Breeding, Melle, Belgium
EC	European Community
ECCDB	European Central Crop Database
ECP/GR	European Cooperative Programme for Crop Genetic Resources Networks
ENMP	Estação Nacional de Melhoramento de Plantas, Portugal
EPGRIS	European Plant Genetic Resources Information Infra-Structure
EU	European Union
EUCARPIA	European Association for Research on Plant Breeding
EURISCO	European Internet Search Catalogue
FAL	Swiss Federal Research Station for Agroecology and Agriculture
FAO	Food and Agriculture Organization of the United Nations, Italy
FCPI	Fodder Crops and Pasture Institute, Larissa, Greece
GASU	Georgia Agrarian State University, Tbilisi, Georgia
GEVES	Groupe d'étude et de contrôle des variétés et des semences, France
GIS	Geographic Information System
GRIN	Germplasm Resources Information Network (USDA)
GRS	Grassland Research Station, Zubří, Czech Republic
HRI	Horticulture Research International, UK
IDG	Istituto del Germoplasma, Bari, Italy
IGB	Israeli Gene Bank for Agricultural Crops, Bet-Dagan, Israel
IGER	Institute of Grassland and Environmental Research, UK
IHAR	Plant Breeding and Acclimatization Institute, Poland
IPGR	Institute for Plant Genetic Resources "K. Malkov", Sadovo, Bulgaria
IPK	Institut für Pflanzengenetik und Kulturpflanzenforschung, Germany
ISCF	Istituto Sperimentale per le colture foraggere, Lodi, Italy
IUCN	International Union for the Conservation of Nature (now The World Conservation Union)
JDE	Junta de Extremadura, Spain
LIA	Lithuanian Institute of Agriculture, Kedainiai, Lithuania

LUA	Latvia University of Agriculture
MCPD	multicrop passport descriptor
MOS	most original sample
MTA	Material Transfer Agreement
NAGREF	National Agricultural Research Foundation, Greece
NGB	Nordic Gene Bank, Sweden
NGO	non-governmental organization
PBI	Plant Breeding Institute, Jõgeva, Estonia
PCR	polymerase chain reaction
PGR	plant genetic resources
PGRFA	plant genetic resources for food and agriculture
RAPD	random amplified polymorphic DNA
RICP	Research Institute of Crop Production, Prague, Czech Republic
RIFC	Research Institute of Field Crops, Fushë-Krujë, Albania
RIFC	Research Institute for Fodder Crops Ltd., Troubsko, Czech Republic
RIPP	Research Institute of Plant Production, Piesťany, Slovakia
SAMPL	selective amplification of microsatellite polymorphic loci
SIDT	Servicio de Investigación y Desarrollo Tecnológico, Badajoz, Spain
S-SAP	sequence-specific amplification polymorphism
UCD	University College Dublin, Ireland
UNCED	United Nations Conference on Environment and Development
UNDP	United Nations Development Programme
UNEP	United Nations Environment Programme
UPGA	unweighted pair group analysis
UPGMA	unweighted pair group method using arithmetic averages
UPOV	Union pour la Protection des Obtentions Végétales, Switzerland
USDA	United States Department of Agriculture
VIR	N.I. Vavilov Research Institute of Plant Industry, Russian Federation
WIEWS	World Information and Early Warning System
WWF	World Wildlife Fund (now The Global Conservation Organization)
ZADI	Zentralstelle für Agrardokumentation und -information, Germany

Appendix IV. Agenda

Eighth Meeting of the ECP/GR Working Group on Forages
10-12 April 2003, Linz, Austria

Wednesday 9 April 2003
Arrival of participants

Thursday 10 April 2003

8:30 – 9:30	**Introduction** • Welcome on behalf of the Austrian Minister of Agriculture (*H. Wögerbauer, 15 min.*) • ECP/GR briefing (*L. Maggioni, 15 min.)* • Working Group on forages: Past Chairman's report (*P. Marum, 15 min.*) • Aims and schedule of Meeting (*B. Boller, 15 min.*)
9:30 - 10:30	**European Central Forages Databases** • Progress in development of a new, relational central database for European crop genetic resources (EURISCO): implications for forage databases *(IPGRI, 10 min.)* • Major advances in development of individual forage crop databases - *Trifolium subterraneum* and annual *Medicago* species (*M. Murillo, 10 min.*) - *Arrhenatherum* and *Trisetum (M. Ševčíková, 10 min.)* - *Dactylis* and *Festuca (W. Majtkovski, 10 min.)*
10:30 – 11:00	*Coffee break*
11:00 – 12:30	**Reports on status of National Collections and collecting activities:** *10 min. contributions from: Albania, Bulgaria, Croatia, Czech Republic, Estonia, France, Germany, Greece, Israel, Lithuania*
12:30 – 14:00	*Lunch*
14:00 – 15:00	**Reports on status of National Collections and collecting activities, continued:** *Macedonia FYR, Netherlands, Nordic countries, Poland, Romania, Slovakia, Slovenia, Turkey*
15:00 – 15:30	*Coffee break*
15:30 – 17:00	**International cooperation** • Reports of 5-10 min. on larger scale collecting activities including partners of different countries: - Forage collecting expeditions as a contribution to promote international cooperation and improvement of genetic diversity *(E. Willner, 10 min.*) - Joint collecting mission VIR/NGB in Karelia, 2001 *(L. Bondo, 10 min.)* - Plant collecting expedition in the Pyrenees Mountains, Spain/France 2001 *(I. Thomas, 10 min.)* - Forage collecting activities—international cooperation (Poland, Slovakia, Czech Republic and Slovenia) *(G. Żurek, J. Drobná, M. Ševčíková and V. Meglič, 10 min.)* • Implications of the International Treaty on Plant Genetic Resources for Food and Agriculture for international collecting missions and exchange of material *(IPGRI, 10 min)*

Friday 11 April 2002

8:30 - 10:00	**Sharing of responsibilities** • Definition and identification of Most Original Samples (MOS) • Designation of responsible institutions for evaluating/maintaining/regenerating an accession • Safety-duplication
10:00 – 10:30	**Reconsidering minimum standards for regeneration** • Status of current EU-funded research project ICONFORS: Improving germplasm conservation methods for perennial European forage species *(I. Thomas, 15 min.)* • Experiences with implications: - Experiences with the new regeneration strategy at the Nordic Gene *Bank (L. Bondo, 15 min.)*
10:30 – 11:00	*Coffee break*

11:00 – 11:45	**Core collections** • Report of the *Medicago* "core collections" subgroup established at Elvas in 1999 *(V. Negri, 5 min.)* • Progress in analysis of the evaluation of the European *Lolium* core collection: Genetic diversity between and within ryegrass populations of the ECP/GR collection by means of AFLP markers *(A. Ghesquiere, 10 min.)* • Experiences with establishment of national or regional core collections: - *Lolium perenne* core collection - results and conclusions from Poland *(W. Majtkovski, 10 min.)* • How should we proceed with the concept of core collections? Need for a European vs. national approaches, choice of species, and definition of the "core".
11:45 – 12:30	**On-farm / *in situ* conservation** • Landraces: concepts and approaches to promote maintenance, use, or re-creation of landraces: - On-farm conservation in Finland *(L. Bondo, 10 min.)* - On-farm conservation/improvement of forages in Norway *(P. Marum, 10 min.)* • Ecotypes: concepts and approaches to identify sites for *in situ* conservation and to ensure their protection. *Short contributions (5-10 min.) are welcome*
12:30 – 14:00	*Lunch*
14:00 – 15:30	**Research activities** • Current/recently terminated activities. - Cyprus: collection and evaluation of local germplasm for forage production *(D. Droushiotis, 10 min.)* - Screening of tall oatgrass and yellow oatgrass germplasm held in the European Central Crop Databases *(M. Ševcíková, 10 min.)* - Assessment of *Poa* genetic resources for breeding purposes by evaluation of important traits *(E. Willner, 10 min.)* - Study of *Phalaris arundinacea* accessions in Romania *(T. Marusca, 10 min.)* - Safeguarding a unique collection of former Swiss red clover landraces *(B. Boller, 10 min.)* - Evaluation of *Dactylis glomerata* (orchard grass) *(H. Özpinar, 10 min.)* • Update on the Framework 6 Programme and the EC 1467 Regulation of the European Union *(IPGRI, 10 min.)* • Planning cooperative research activities and ways to generate funding • Possibilities of funding forage genetic resources projects as part of national "Plans of Action" to implement the "Global Plan of Action", following the Rio Convention on Biodiversity
15:30 – 16:00	*Coffee break*

Saturday 12 April 2002

Drafting of the report. For those not involved a technical tour will be offered by the local organizers	
14:30 – 17:00	**Conclusion** • Presentation of the report and adoption of recommendations • Decision about the future of the Network Coordinating Group and priorities for the next phase of ECP/GR • Closing remarks
Social dinner in the evening	

Sunday 13 April 2002

Departure of participants

Appendix V. List of participants[10]

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[10] This list was updated according to latest information available at time of publication. The current composition of the ECP/GR Working Group on Forages can be found on the ECP/GR Web site, constantly updated (see http://www.ipgri.cgiar.org/networks/ecpgr/Contacts/ecpgr_wgfg.asp).

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