European Cooperative Programme for Crop Genetic Resources Networks

ECP/GR

# Report of a Working Group on Medicinal and Aromatic Plants

IPGRI

**First Meeting, 12–14 September 2002, Gozd Martuljek, Slovenia**

D. Baričevič, J. Bernáth, L. Maggioni and E. Lipman, *compilers*

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# PART I. SUMMARY REPORT OF THE MEETING

## Introduction

The newly established Working Group on Medicinal and Aromatic Plants (MAPs) of the European Cooperative Programme for Crop Genetic Resources Networks (ECP/GR) met for the first time in Gozd Martuljek, Slovenia, 12-14 September 2002. The meeting brought together 18 members representing 20 European countries and 12 observers, including Slovenian officials and scientists, representatives from the World Wide Fund for Nature (WWF-UK and WWF-TRAFFIC Europe) and a private consultant from Germany (see list of participants in Appendix IV).

### *Opening of the meeting / Welcome addresses*

**Mihaela Černe**, National Coordinator for ECP/GR in Slovenia welcomed the participants with the following address:

Dear Members of the Working Group on Medicinal and Aromatic Plants, distinguished guests from the Ministry of Agriculture, Forestry and Food (MAFF) and Ministry of Environment and Spatial Planning, and observers,

As National Coordinator for Slovenia I am glad that a new ECP/GR Working Group on Medicinal and Aromatic Plants was established and that Slovenia was chosen to host its first meeting. This is also the first time that Slovenia hosts an ECP/GR Working Group meeting.

Slovenia was invited to join the ECP/GR in 1993 and after 5 years of efforts, Slovenia joined the programme in the last year of Phase V (1998).

I would like to express my sincere thanks to the Ministry of Agriculture, Forestry and Food for the establishment of the National Commission and nomination of a National Coordinator for plant genetic resources (PGR), for the financial support to the Slovenian programme on PGR, and for the contribution to the ECP/GR.

My thanks go also to the ECP/GR coordinators for their useful suggestions and support regarding the formation of the National Commission and plant genetic resources (PGR) programme before Slovenia became an official member of ECP/GR and for continued cooperation since then.

The Slovenian Gene Bank was established in the frame of the late Yugoslav Gene Bank in 1988, although collections of indigenous landraces of bean and cabbage were established before World War II. New collections of maize, bean, cabbage, onion and buckwheat were established after World War II for breeding purposes. Landraces were used as a source material for Slovenian cultivars. Since then only maize accessions were properly stored, but data are available for the bean, cabbage and buckwheat collections for the study of genetic erosion. The national genebank is decentralized. Accessions are kept at three institutions: Biotechnical Faculty of the University of Ljubljana; Agricultural Institute of Slovenia in Ljubljana; and Institute for Hop and Brewery in Žalec. A standardized documentation system is used by all three institutions and also for forest genetic resources.

Collections of medicinal and aromatic plants are kept at the Biotechnical Faculty in Ljubljana and at the Institute for Hop and Brewery in Žalec.

I am very happy that the programme for Slovenian Plant Genetic Resources, which has been financed since 1996 by the Ministry of Agriculture, Forestry and Food, is now expanding and our researchers are well integrated in different ECP/GR Working Groups.

I would also like to express my sincere thanks to the local organizer of this meeting, Dea Baričevič from the Biotechnical Faculty of the University of Ljubljana, who, in collaboration with Prof. Karl Hammer from the University of Kassel, Germany, prepared the proposal for the establishment of a new Working Group on medicinal and aromatic plants. This proposal was submitted to the ECP/GR Steering Committee at its mid-term meeting in St. Petersburg, Russian Federation, October 2001.

I wish all participants a pleasant stay in Gozd Martuljek and fruitful discussions. Welcome to Slovenia!

**Janez Glavač** (Ministry for Agriculture, Forestry and Food of Slovenia) made the following speech:

Ladies and gentlemen, experts and scientists,

I consider it a great pleasure and privilege to welcome you to the first meeting of the ECP/GR Working Group on Medicinal and Aromatic Plants on behalf of the Ministry for Agriculture, Forestry and Food of Slovenia as well as on behalf of our Minister, Mr Franc But.

The MAFF is honoured and very proud that the first meeting of the Working Group on Medicinal and Aromatic Plants is taking place in our country. This is certainly a recognition for our experts and scientists working in the field of plant genetic resources, with modest financial support from the state, but with great enthusiasm.

Slovenia is a small country and therefore we cannot afford to make big mistakes. Lack of attention to the conservation and sustainable use of medicinal and aromatic plants, anywhere in the world, would be a great and unreasonable mistake, with many unpredictable consequences to follow.

Because of the increased global interest in the use of medicinal and aromatic plants and their habitat loss and alteration, the development of a conservation strategy for these important natural resources is a necessity. To reach that objective, we wish you successful work at this meeting as well as in the future.

We would like to wish you also a pleasant stay here in Gozd Martuljek, on the sunny side of the Alps, surrounded with majestic mountains. We hope you will keep a pleasant memory of this particular occasion and of our country.

**Dea Baričevič** also welcomed all Working Group members and observers to the meeting on behalf of the Slovenian genebank and of Lorenzo Maggioni, the ECP/GR Coordinator, who was unable to attend. She indicated that an invitation had been sent to Melpo Skoula, from the MEDUSA Network[1], who expressed interest to keep in contact but was unable to attend the present meeting due to other commitments. Vernon Heywood was also informed of the meeting and considered it an excellent development, and declared himself happy to cooperate with the Group, both personally and on behalf of ICMAP (International Council of Medicinal and Aromatic Plants). IUCN (International Union for the Conservation of Nature), FAO (Food and Agriculture Organization of the United Nations) and ISF (International Seed Federation) were also invited as observers but could not send any representative.

[1] MEDUSA Network for the identification, conservation and use of wild plants in the Mediterranean region (http://medusa.maich.gr/network/).

### *Adoption of the agenda and selection of the Chairperson for the meeting*

Dea Baričevič then presented the meeting agenda, which was accepted by the participants (see Appendix III).

The group agreed that Dea Baričevič and Jenő Bernáth would co-chair the meeting during the first day, taking alternate roles as Chairperson and Vice-Chairperson. It was also agreed that on the second day, Wolfgang Kainz (Austria) and Zlatko Šatović (Croatia) would co-chair the morning sessions, and Eli Putievsky (Israel) and Zlatko Šatović the afternoon sessions.

## Presentations

The presentations given at the meeting are mentioned below in chronological order. Full papers, when available, are published in Part II of this report. The discussions held between the presentations and the ensuing recommendations made by the Group, leading to the elaboration of the workplan, are grouped and summarized in the section *Discussion and Recommendations* (see below, pages 8-11).

### *Conservation of medicinal and aromatic plants (MAPs) – needs and strategy*

#### The role of ECP/GR and IPGRI in MAP conservation

On behalf of the ECP/GR Secretariat, Elinor Lipman provided some background information in the two following presentations:

##### ECP/GR and the Working Group on Medicinal and Aromatic Plants

For the benefit of the many participants who were attending an ECP/GR meeting for the first time, the historical background, overall purpose and current structure of the ECP/GR programme were presented[2], with particular focus on the steps leading to the establishment of the Working Group on Medicinal and Aromatic Plants within the Minor Crops Network:

- The recommendations of the Minor Crops Network Coordinating Group meeting in Turku, Finland, June 1999, were as follows:
  - Propose the establishment of a Working Group on Medicinal and Aromatic Plants to the Steering Committee
  - Dea Baričevič and Karl Hammer to develop a proposal for collaborative action, to be implemented in the context of a Working Group
  - Focal persons, in consultation with relevant experts, to identify crops/species for priority action, on the basis of agreed criteria for prioritization (risk of genetic erosion; economic importance; regional or subregional distribution; traditional knowledge in Europe)
  - Focal persons, in collaboration with experts from different parts of Europe, to compile information regarding: e*x situ* and *in situ* conservation status; distribution and level of utilization; list of experts/institutions; list of ongoing activities; elaboration of a conservation strategy to be proposed for further action.

[2] Update at time of publication: the structure of the Programme has been re-defined by the ECP/GR Steering Committee during its end-of-phase VI meeting held 22-25 October 2003, Izmir, Turkey (see http://www.ecpgr.cgiar.org/Introduction/AboutECPGR.htm).

- The proposal to establish a Working Group was developed by Dea Baričevič and Karl Hammer as recommended and presented successfully at the mid-term meeting of the ECP/GR Steering Committee (14-17 October 2001, St. Petersburg, Russian Federation). The SC recommended that a Working Group on Medicinal and Aromatic Plants be established and that a meeting of the Working Group be held in 2002, considering the recent developments and increasing interest in these species.
- This present meeting is the concrete outcome of these recommendations. The activities listed in the proposal should form the basis for the development of the workplan of the WG on Medicinal and Aromatic Plants (MAPs), now that it is formally established. These activities include:
  - An inventory of natural populations of MAPs
  - A review of the acreage of MAPs under cultivation
  - A review of legal issues for the conservation of MAPs
  - An assessment of the quantities of MAPs collected from the wild and encouragement of the use of cultivated MAPs
  - Preparation of a list of potentially endangered/rare/vulnerable MAP species
  - Developing a strategy for the future development of a MAP conservation strategy
  - Preparation of a proposal for submission to the 6th EU Framework Programme.

The Web pages of the Minor Crops Network and of the Working Group on Medicinal and Aromatic Plants[3] were presented to the participants, who were invited to make comments and suggestions for their improvement.

Other important outcomes of the ECP/GR Steering Committee mid-term meeting in October 2001 were mentioned: in order to develop a strategy for the next Phase (VII), two task forces composed of a few Steering Committee members were established to discuss (1) the impact on PGR of recent developments in science, technology and international policy; and (2) how genebanks might implement relevant international agreements and their impact on their operation. A questionnaire sent to all Working Group Chairs and National Coordinators is being used to sound out opinions on future priorities and mode of operation of ECP/GR, to be defined during the Steering Committee meeting planned for October 2003.

- **IPGRI's contribution to the conservation and sustainable use of medicinal plant species**

This presentation showed how the issue of MAP conservation fits into the global framework in which IPGRI operates. It was illustrated by the example of the IPGRI/IFAD project *"Enhancing the contribution of neglected and underutilized species to food security, and to incomes of the rural poor"*.[4] This project, started in 2001, focuses specifically on MAPs in the Central and West and North Africa (CWANA) region, Andean grains in Latin America and nutritious millets in Asia. The MAP species/genera considered in the CWANA region are the following:

| | | |
|---|---|---|
| *Aloe* spp. | *Cuminum cyminum* | *Mentha piperita* |
| *Boerhavia elegans* | *Flemingia congesta* | *Nigella* spp. |
| *Capparis* spp. | *Glycyrrhiza glabra* | *Origanum syriacum* |
| *Coriandrum sativum* | *Lawsonia inermis* | *Solenostemma arghel* |

[3] http://www.ecpgr.cgiar.org/Workgroups/Med_aromatic/med_aromatic.htm
[4] http://www.ipgri.cgiar.org/nus/docs/IFAD-NUS.doc

Examples of activities carried out by the project include:
- Strengthening or establishing local seed production
- Surveying the distribution of available genetic diversity and of traditional knowledge together with analyses of genetic erosion
- Investigations throughout the production-consumption chain
- Identification of improved agronomic and production procedures and value-adding strategies
- Analysis of market opportunities
- Research on nutritional value of the species, and characterization and evaluation work
- Development of networks
- Studies to identify policy failures

These areas of research might be considered by the WG on MAPs to define its own workplan.

**MAP conservation strategies**

Peter Skoberne (Ministry of Environment, Spatial Planning and Energy of Slovenia) gave an overview of the situation regarding plant conservation, with a detailed review of the legal instruments available at all levels:
- international treaties or conventions, including protocols and recommendations either at global level (RAMSAR Convention, World Heritage, CMS or Bonn Convention, CITES or Washington Convention, CBD, GPA…), regional level (the Bern Convention, the European Landscape Convention…) or sub-regional level (Alpine Convention, Barcelona Convention);
- supranational legislation such as the EU Habitat Directive and Endangered Species Regulation;
- global strategies such as the World Conservation Strategy (IUCN/UNEP/WWF 1980); Caring for the Earth (IUCN/UNEP/WWF 1991); and the Global Biodiversity Strategy (WRI/IUCN/UNEP 1992). Of special interest to this working group are the *Guidelines on the Conservation of Medicinal Plants* (WHO/IUCN/WWF 1993);
- ministerial processes (Environment for Europe, Protection of Forests in Europe)
- national legislation, the essential tool for implementation of these legal treaties and strategies.

Dr Skoberne concluded by saying that it is now time to move from the writing of papers to action. The basic cornerstone of our work should be to respect life and learn from plants, working "from the ground", but we cannot work alone. This is what networks are for!

*The full text of the presentation is included in Part II of this report (pages 14-18*).

**Sustainable use of medicinal and aromatic plants in Europe**

Susanne Schmitt (WWF-UK) and Susan Honneff (WWF-Germany/TRAFFIC-Europe)

*The abstract of the presentation is included in Part II (pages 19-20).*

**Origin of medicinal plants in Central Europe – an ecological approach**

Ernst Schneider (Pharmacist and Biologist, PhytoConsulting)

*The full text of the paper is included in Part II (pages 21-24).*

## *Country reports*

Before the meeting, all Working Group members had been requested to prepare a country report covering the following topics:

- legal protection of MAP species and their natural habitats;
- *ex situ* conservation;
- existence of MAP inventory activities at national level, and if so, type of data recorded;
- review of the acreage of MAPs under cultivation;
- availability of information regarding the use of MAPs by processing industries (respective amounts of material of cultivated origin and of raw materials gathered in the wild);
- experiences in sustainable use of MAPs.

The following reports were presented at the meeting by the respective country representatives: Austria, Bulgaria, Croatia, Cyprus, Estonia, Hungary, Israel, Italy, Lithuania, Macedonia F.Y.R., Nordic countries, Poland, Romania, Slovenia, Turkey, United Kingdom and F.R. Yugoslavia.

Non-attending members (representatives from the Czech Republic, Malta and Portugal) provided contributions for publication in the proceedings.

*All full papers available are included in Part II (see pages 25-142).*

## *Documentation and databases*

### The European Internet Search Catalogue, EURISCO and the EPGRIS project

On behalf of IPGRI, Elinor Lipman presented the EPGRIS project for the Establishment of a Plant Genetic Resources Infra-Structure. This 3-year project (2000-2003) was developed within the ECP/GR Documentation and Information Network and was approved for funding within the Fifth Framework Programme of the European Union. The objective is to establish a European Internet Search Catalogue (EURISCO) with passport information of plant genetic resources maintained *ex situ* in Europe. The catalogue will be frequently updated and publicly accessible via Internet. Initial data sets will be derived from the European Central Crop Databases (ECCDBs); however the project will promote the creation of national inventories, which are planned to become the main source of data. PGR National Coordinators of the large majority of European countries have nominated national inventory focal persons. These people will be invited to attend three subregional meetings, to discuss coordination and standardization of the data flow from the national inventories to the central catalogue. The project partners will also provide technical support to the focal persons and a limited number of training visits to the main European documentation support centres will be arranged. EURISCO is seen as an important European contribution to the Clearing House Mechanism (CHM) of the Convention on Biological Diversity (CBD) and the implementation of the Global Plan of Action (GPA). A Web-based interface will allow easy searching of the European national inventories, in the same way as it is possible today to use SINGER (System-wide Information Network for Genetic Resources) to search the CGIAR collections and GRIN (Germplasm Resources Information System) to search the USDA collections. The catalogue will carry an important minimum set of passport data, frequently and automatically updated from the national inventories. These data are based on the revised version of the FAO/IPGRI *Multi-crop passport descriptor list* (MCPD), finalized in December 2001. The full list is available on-line at http://www.ecpgr.cgiar.org/Databases/Databases.htm.

The project has implications for the central databases that are often the main product of collaboration within the ECP/GR Working Groups. The idea will be to take the workload of collecting the passport data away from the central crop database managers, since these data will become directly accessible from the EURISCO catalogue. On the other hand, ECCDB

managers will be expected to dedicate more time to compile and analyze characterization and evaluation data.[5]

### The Slovenian Gene Bank Database and the National Inventory

Vladimir Meglič (Agricultural Institute of Slovenia)

*The full text of the paper is included in Part II (pages 143-145).*

### Presentation of the MEDPLANT database

Dea Baričevič gave a demonstration of the MEDPLANT relational database, currently under development. The MEDPLANT database was established to hold data on flora, fauna, taxonomy and analysis for medicinal and aromatic plants (MAPs) of Central European and Mediterranean regions. The MEDPLANT system works on relational databases principles: collected data are arranged according to their characters in appropriate databases (systematics, geography, habitats, pedology, phytocoenoses, chemical analyses, varieties/cultivars). Pointers which connect specific data in these databases enable establishing mutual relationships between required data in the network. The objectives/advantages of this first version of the database, which offers user-friendly data management, are:

- rapid and easy input or output of desired/required data;
- the large database dimension enables fast processing of large data volumes;
- MEDPLANT is a multi-user system for a network;
- it is an open system which enables addition of new data types without changing the present structure;
- the MEDPLANT application is open to the addition of processing modules (models). The MEDPLANT graphic module enables the presentation of geographical data (e.g. maps) as well as pictures, and the establishment of their correlations with recorded data.

## *Methodological approaches in MAP conservation and evaluation*

### Molecular tools for determining genetic variability

Branka Javornik (Biotechnical Faculty, University of Ljubljana, Slovenia)

*The abstract of the presentation is included in Part II (page 146).*

### Genetic variability of native populations of oregano in Slovenia

Jelka Šuštar-Vozlič (Agricultural Institute, Slovenia)

*The full text of the paper is included in Part II (pages 147-149).*

[5] Update at time of publication: EURISCO was launched officially at the Final Conference of the EPGRIS Project, 11-13 September 2003, Prague, Czech Republic. A demo version of the catalogue is available at http://eurisco.ecpgr.org/

## Discussion and recommendations

A summary of discussions and recommendations leading to the elaboration of the workplan is presented below.

### *Scope of the Working Group's activities*

- **Preliminary definitions**

#### What are MAPs?

The Group agreed that definitions were needed for "medicinal and aromatic plants" and the following were approved temporarily:

- Medicinal plants are plants used in official and traditional medicine
- Aromatic plants are plants used for their aroma and flavour

#### Geographical origin of MAP species

The WG will consider autochthonous European species (of which the centre of origin is located in Europe) and species which were domesticated in Europe or are well adapted to European ecological conditions.

- **Long-term tasks**

#### Inventory of MAP genetic resources in Europe

Knowledge about available wild genetic resources is rather scarce. An inventory needs to be prepared of MAP distribution and abundance at a European level.

#### Conservation of MAP genetic resources in Europe

According to the country reports presented, a very high proportion of European MAP species on the market is gathered from the wild. The existing legislation at national and European levels does not sufficiently prevent the unsustainable exploitation of MAP natural resources. Only a limited number of species are officially protected but other important MAP species may be endangered. These species must be identified, conserved (*in situ* and/or *ex situ*) and proposed for official protection.

#### Characterization/evaluation of MAPs: development of descriptors at genus level

MAP species differ substantially from other crops (where descriptors are available) due to their content and high variability of secondary metabolites in different environments. The Group agreed that characterization is only possible when descriptors (at the genus level) are defined and that it would be important to prepare a standard list of descriptors to harmonize the data recording methodology, to increase reliability of recorded data and to eventually facilitate data exchange.

#### Development of a central MAP database

The WG agreed that access to and sharing of information on European MAPs would be greatly enhanced by the creation of a MAP database.

- **Coverage**

Given the large number of species concerned, it was agreed that if such a database were to be created it would initially be restricted to only a few priority species identified as defined below (section on short-term tasks).

- **Data format**

It was agreed that the data format should ensure compatibility with the EURISCO catalogue, i.e. FAO/IPGRI *Multi-crop passport descriptors* (second version, Dec. 2001[6]) with the 6 additional descriptors defined for EURISCO.[7]

- **Financial support**

The Group agreed that the creation of the database is not possible without financial support. Funding opportunities from international organizations (FAO, EC, etc.) or pharmaceutical companies which might be interested in MAP genetic resource conservation will be explored by the Chair of the WG.

Ernst Schneider kindly offered to assist in establishing contacts with potential donors from the pharmaceutical industry in Germany and his offer was accepted.

- **Short-term tasks**

#### Election of the Chair and Vice-Chair

Before establishing the workplan for short-term activities, the Group considered it necessary to identify the Chairperson and Vice-Chairperson who would be responsible for ensuring the progress of the workplan and coordination between the WG members until the end of the next meeting of the Working Group. Dea Baričevič was elected as Chairperson and Jenő Bernáth was elected as Vice-Chairperson, both receiving warm congratulations from the Group for their contribution to this first meeting of the WG.

#### Selection of a list of priority species/genera

The Group agreed that it should concentrate its short-term activities on a small number of species/genera of importance to all and considered as needing urgent attention. A list of priority species /genera will be elaborated according to the workplan indicated below.

The methodology developed for these priority species/genera (e.g. inventory of existing material *in situ* and *ex situ*, definition of characterization descriptors, establishment of a database, etc.) will serve as a model for other species and will encourage appropriate activities to be carried out on MAPs at national level.

***Workplan***

1. Each country representative will send a list of 30 species/genera important at national level to the Vice-Chair of the WG by the **end of October 2002**. The list should be accompanied by background information on the selection criteria used.
2. After analyzing the national lists received, the Vice-Chair will select 10 species/genera and will circulate this priority list to all WG members by the **end of December 2002**.[8]

*Note: the identification of the priority species will take in consideration (i) the CITES convention[9]; and (ii) the subregional distribution of the species/genera in order to ensure a balanced geographical distribution.*

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6 FAO/IPGRI's *Multi-crop passport descriptors* (version 2, December 2001) are available on-line (http://www.ipgri.cgiar.org/system/page.asp?frame=catalogue/select.asp).

7 The document *EURISCO Descriptors for uploading information from National Inventories to EURISCO* was distributed at the meeting. The descriptors are also available on-line (http://www.ecpgr.cgiar.org/epgris/Tech_papers/EURISCO_Descriptors.doc).

8 Update at time of publication: the list of priority species has been elaborated and circulated to all members by the Vice-Chair (Appendix I).

9 Convention on International Trade in Endangered Species of Wild Fauna and Flora (http://www.cites.org).

#### Development of MAP descriptors for the priority list

It was agreed that it would be more effective to make use of descriptor lists already available for species which have some degree of similarity to MAPs (e.g. IPGRI's descriptors for *Allium* spp., *Piper nigrum*, *Elettaria cardamomum*). These would be modified in order to fit MAP specificities. To illustrate this, D. Baričevič presented a selection of relevant descriptors taken from *Allium* and cardamom lists. After discussion, the Group agreed on the following workplan:

*Workplan*

1. Dea Baričevič, in consultation with V. Meglič, will prepare a proposal for a list of characterization descriptors, to be circulated to all WG members **before the end of 2002.**
2. After the priority list of species/genera has been identified by the Vice-Chair and validated by the Working Group, D. Baričevič will refine the descriptor list with regard to specific characters of the 10 selected species, and send back the proposed descriptor list to all WG members within two months after reception of the priority list of species/genera.[10]

#### Inventory of MAP populations at national level

On the basis of the priority list species, each member of the WG should make all possible efforts to ensure that an exhaustive survey of natural populations of the selected MAPs is carried out in her/his country, in order to provide information for a European MAP genetic resources inventory.

*Workplan*

At the next WG meeting, WG members will report progress on surveys made in their country and the responsible institute for compiling the European inventory will be defined.

#### Research/training activities

*Recommendations*

- The Working Group encourages collaboration between national programmes working on the same plants/groups of plants. The Chair and Vice-Chair of the WG should facilitate collaboration and coordination of target teams.
- The Working Group recognizes the need for the development of harmonized methodologies/protocols (sampling and conservation techniques, etc.) by the national and international research programmes. The Chair and Vice-Chair of the WG will seek funding from relevant international institutions.
- The Working Group encourages the collectors to provide specimens to the national herbarium collections.

[10] A provisional list of descriptors has been elaborated according to the decisions made at the first meeting of the Working Group on MAPs and will be available on-line at http://www.ecpgr.cgiar.org/Workgroups/Med_aromatic/med_aromatic.htm. This list is adapted from other descriptor lists produced by IPGRI. It is not final but represents a proposal open for discussion at the next Working Group meeting.

### *Next meeting of the Working Group*

Considering the urgency of implementing activities on MAP conservation in Europe, and the number of issues to be addressed, the WG agreed that it would be important to meet again within a short time frame.

*Recommendation*

The Working Group recommends that another meeting be organized **no later than 2004** in order to assess the progress achieved in the implementation of the workplan.

## Conclusion

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

The organizers thanked the participants for their high degree of involvement and valuable inputs, and with best wishes to all for a fruitful implementation of the workplan, the meeting was closed.

# PART II. PRESENTED PAPERS

# General papers

## *Conservation strategies for medicinal and aromatic plants*

***Peter Skoberne***[11]
*Environmental Agency of the Republic of Slovenia, Ljubljana, Slovenia*

### Introduction

This paper gives a broad framework regarding plant conservation, especially in relation to plant use, highlighting international legislation and emphasizing its implementation at the national level.

### Nature conservation and plants

The vital role of plants is usually underestimated in our everyday life. We hardly imagine and recognize their importance as life supporters (transformers of solar energy, oxygen producers, $CO_2$ fixers), their use for food, spices, medicinal purposes, natural resources for timber, oil, fibres, etc.

Exploiting the many possible uses for plants requires wisdom. With new technologies we are inventing new ways of using plants but on the other hand we are losing a lot of wisdom collected within communities closely related to and dependent on nature.

Common sense suggests that we should respect plants and find sustainable ways to use them, as our lives are directly dependent on conserving their diversity.

Thus it is very logical that early conservation ideas in Europe in the second half of the 19th century were connected with plant conservation. For instance, in a part of what is now Slovenian territory, edelweiss (*Leontopodium alpinum*) has been protected since 1896.

Species conservation was later integrated into a broader concept of "biodiversity", a term widely used in biological circles since 1987. Finally, at the Rio Earth Summit (UNCED, United Nations Conference on Environment and Development) in June 1992 the expression "biodiversity" was introduced in the political vocabulary, mostly through the signature of the Convention on Biological Diversity (CBD). But before we take a closer look at this important international treaty, let us have a brief overview of various legal instruments concerning plant conservation.

### Overview of legal instruments

Different legal instruments are available according to geographic scope and basic aim. As threats for plants are not confined within national borders, international legal instruments are essential, especially as the responsibility for species protection is a global one. It is a responsibility of mankind. Major levels are:

- **International treaties (conventions)** including protocols and recommendations

Conventions deal with international problems. Countries commit themselves to common goals and implement them in a coordinated way through national legislation. Regarding

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[11] Address at time of publication:
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geographical scope we are talking about global, regional and sub-regional conventions. The main treaties are presented in Table 1.

**Table 1.** International treaties in the field of nature conservation

| Logo | Name of the treaty | Place, year of signature | Number of parties (year) |
|---|---|---|---|
| | **Global level** | | |
| | Convention on Wetlands of International Importance, especially as Waterfowl Habitat (Ramsar Convention) | Ramsar, 1971 | 130 (2002) |
| | Convention Concerning the Protection of the World Cultural and Natural Heritage (World Heritage Convention) | Paris, 1972 | 161 (2001) |
| | Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES or Washington Convention) | Washington, 1973 | 150 (2000) |
| | Convention on the Conservation of Migratory Species of Wild Animals (CMS or Bonn Convention) | Bonn, 1979 | 70 (2000) |
| | Convention on Biological Diversity (CBD) | Rio de Janeiro, 1992 | 178 (2000) |
| | **Regional level (European)** | | |
| | Convention on the Conservation of European Wildlife and Natural Habitats (Bern Convention) | Bern, 1979 | 45 (2003) |
| | European Landscape Convention | Florence, 2000 | 11 (2004) |
| | **Sub-regional level (concerning Europe)** | | |
| | Alpine Convention | Salzburg, 1991 | 9 (1999) |
| | Convention for the Protection of the Marine Environment and the Coastal Region of the Mediterranean (Barcelona Convention) | Barcelona, 1976 | 21 (2000) |

- **Supranational legislation** (e.g. European Union)

Unlike international treaties that cannot be directly implemented, the legislation of the European Communities has an immediate legal effect on the member states. Most important for plant conservation are:

- *The Habitat Directive* (Council Directive 92/43/EEC of 21 May 1992 on the conservation of natural habitats and of wild fauna and flora – OJ L 206, 22.7.1992) defining a list of plant species (Annex II) that require habitat protection and are part of the NATURA 2000 ecological network, a list of strictly protected plants (Annex IV) and a list of plants that can be utilized in a managed way (Annex V).
- *The Endangered Species Regulation (*Council Regulation (EC) No. 338/97 of 9 December 1996 on the protection of species of wild fauna and flora by regulating trade therein – OJ L 61, 3. 3. 1997) regulates the trade of threatened species.

- **Strategies**

Long-term goal setting and a vision for conservation are set up by strategic documents. Some global documents that were the basis for the preparation of some international conventions (e.g. the CBD) and national activities are mentioned below.

- 1980: World Conservation Strategy (IUCN, UNEP, WWF)
- 1991: Caring for the Earth (IUCN, UNEP, WWF)
- 1992: Global Biodiversity Strategy (WRI, IUCN, UNEP)

Specially focused on medicinal plants are the *Guidelines on the conservation of medicinal plants* (WHO, IUCN, WWF), published in 1993. Selected guidelines, more related to the topic of our meeting, are mentioned below:

- **basic studies:** study traditional knowledge of plant uses, identify medicinal plants, study distribution and ecology;
- **utilization:** wherever possible, to cultivate medicinal plants as the source of supply; ensure sustainable collecting from the wild;
- ***in situ* and *ex situ* conservation** of medicinal plant populations;
- build **public support**, ensure equitable sharing of benefits.

In the framework of the CBD the *Global Strategy for Plant Conservation* was developed in 2002. The overall goals of the strategy are quite ambitious:

- no species of wild flora to be endangered by international trade;
- 30% of plant-based products derived from sources that are sustainably managed;
- the decline of plant resources, and associated local and indigenous knowledge, innovations and practices that support sustainable livelihoods, local food security and health care, to be halted.

These goals were further elaborated at the European level through the Planta Europa Network in the *European Plant Conservation Strategy* (2002) which deals with specific regional aspects, going in some cases beyond global goals, setting clear goals and targets. Target 3.1 is specially related to conservation and use of plants: *"Best practise for the conservation and sustainable use of medicinal plants (and other sociologically important plants) identified and promoted to relevant policy-makers."*

- **Ministerial processes**

Ministerial processes can also serve plant conservation. Most relevant are *Environment for Europe* (meeting of ministers responsible for the environment) and *Protection of Forests in Europe* (ministers responsible for forests).

- **National legislation**

This is the most crucial part of the system of legal instruments as the essential tool for implementation. The success of implementation of international treaties and strategies depends on the quality of national legislation and on the level of liability.

### Convention on Biological Diversity (CBD)

This treaty is the most recent and the most sophisticated of all global conventions in the field of nature conservation.

In June 1987 UNEP convened an *Ad hoc* Working Group of Experts on Biological Diversity to harmonize existing biodiversity-related conventions. At its very first meeting, the Group agreed on the need for a binding international agreement on the overall problem of biological diversity. In May 1988, UNEP established another *Ad hoc* Working Group of Experts on Biological Diversity with a mandate to prepare an international legally binding instrument for the conservation and sustainable use of biological diversity. It was instructed to take into account *"the need to share costs and benefits between developed and developing countries and the ways and means to support innovation by local people"*. In 1991, the *Ad hoc* Working Group evolved into an Intergovernmental Negotiating Committee (INC). The INC held seven working sessions to negotiate and adopt the text of the Convention on Biological Diversity.

Finally on 22 May 1992 in Nairobi the final text of the Convention was agreed (this important day is celebrated every year as "International Day for Biological Diversity").

The Convention was opened for signature on 5 June 1992 during the Rio Earth Summit, where it received over 150 signatures. UNEP then convened an Intergovernmental Committee on the Convention on Biological Diversity which held two meetings to prepare, among other things, the first meeting of the Conference of the Parties (COP), the Convention's supreme body.

The Convention was formally adopted at the UNCED in Rio, entering in force on 29 December 1993, 90 days after the 30th ratification.

The Cartagena Protocol on Biosafety was adopted in 2000.

The expression "biodiversity" is broadly defined in the Convention and encompasses all living forms at genetic, species and ecosystem levels. The Convention has three main goals:[12]

- the conservation of biological diversity,
- the sustainable use of the components of biodiversity, and
- the fair and equitable sharing of the benefits arising from the use of genetic resources.

All three goals are very relevant to the work with medicinal plants.

They are reflected in obligations. It is obvious that they are very broadly defined and the more detailed elaboration is upon the further development of the convention through its bodies (e.g. COP; Subsidiary Body on Scientific, Technical and Technological Advice, SBSTTA) as well as through national implementation.

The **main obligations** are:

- to adopt national strategies, plans or programmes: the main improvement of the CBD is that the responsibility for the biodiversity convention is not restricted to a particular body but everyone is a stakeholder in this process and cannot avoid responsibility. Thus the CBD strategy and its implementation concern all economic sectors and the general public, and is a splendid opportunity to deliver the message of the importance of biodiversity and the need for its conservation;

[12] The text of the CBD is available at http://www.biodiv.org/convention/articles.asp

- to integrate biodiversity conservation concerns into relevant sectors. The CBD formalized in a political sense the changing nature conservation paradigm: a shift from confrontation of nature conservationists and all others (e.g. developers) to their harmonization;
- identification of processes which have significant adverse effects – a possibility for preventive measures and instruments that influence general processes and are not only focused on particular areas or species;
- Clearing House Mechanism (CHM) – an information system for exchange of knowledge worldwide.

**What next?**

The easy part – writing strategies and treaties – is definitely over. We have enough good instruments, so it is time for action. But all action has to be based on personal commitment towards plants: they have to be regarded as living beings that are an integral part of a larger system. They have a role and function in it. We must take this into account when planning to use or actually using plants. The basic cornerstone of our work should be: *respect life and learn from plants!*

## *Sustainable use of medicinal and aromatic plants in Europe* [13]

***Susanne F. Schmitt*[1] *and Susanne Honnef*[2]**
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The trade in European medicinal plants is long established but has been growing rapidly over the past decade. An estimate 150 European medicinal and aromatic plants (MAPs) are threatened in at least one European country by this trade and by habitat loss. The conservation community, concerned practitioners and the more aware consumer are calling for sustainable herbal products. To date only few companies are concerned about sustainable and ethical sourcing. Cultivation is generally seen as the main solution. Indeed, there is much to be said for cultivation both from a commercial as well as conservation point of view. In this paper, however, we would like to emphasize that cultivation cannot be the sole solution in the context of a complex trade in many hundreds of species, predominantly harvested from the wild, providing livelihoods for large numbers of people in numerous European countries. Efforts of domestication should be coupled with measures to achieve sustainable management for controlled wild harvesting.

In this context a number of potential strategies aimed at more sustainable sourcing are highlighted and some of the existing initiatives of WWF-UK and WWF-Germany/TRAFFIC-Europe Germany presented. The relevance of the European Plant Conservation Strategy (http://www.plantaeuropa.org/html/plant_conservation_strategy.htm) and its specific MAP target, imbedded in the wider context of the newly adopted Global Strategy for Plant Conservation of the Convention on Biological Diversity (CBD) (http://www.biodiv.org) are stressed.

We hope that the ECP/GP Working Group on Medicinal and Aromatic Plants takes the points raised into consideration and seeks collaboration with the wider MAP conservation community, interested companies and government bodies.

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## *The origin of medicinal plants in Central Europe – an ecological approach*

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When discussing medicinal and aromatic plants (MAPs), different points of view can be adopted. The genetic science deals with collecting plants and describing the different origins of plant genetic resources, while the pharmaceutical science only considers quality parameters such as identity, purity and the contents of active ingredients. Scientists in the pharmaceutical industry are under pressure from the marketing departments, striving to obtain higher contents, better quality and a distinguishable product. But all are dependent on the agronomist as primary supplier, who is only looking for highest yield.

When discussing MAP genetic resources these different opinions should be considered. And, most importantly, the optimal ecological conditions for plant growth must be known.

### Worldwide origin

To learn about the most important medicinal plants used in Europe, some statistics and a look at the origin of medicinal plants in the pharmacopoeia are necessary. Some figures for the market in medicinal plants used in Germany are given in Table 1. In recent decades the number of medicinal plants in use has decreased and the demand in terms of quantity has increased.

**Table 1.** Number of species used and amounts of medicinal plants imported in Germany

| Year | | No. of species | Amount (t) | References |
|---|---|---|---|---|
| | **Medicinal plants used in Germany** | | | |
| 1985 | Plants used by one company | 400 | | pers. comm. |
| 1986 | BGA Aufruf Nr.16 1986 - medicinal plants in use | 708 | | BGA 1986 |
| 1994 | Commission E monographs - evaluated | 188 | | Blumenthal 1998 |
| 1993 | List of traditional plants | 193 | | BGA 1993 |
| 2001 | Traditional herbal medicinal products | 1045 | | Gaedcke 2003 |
| 2001 | Deutsches Arzneibuch Stand - plant monographs | 75 | | Gaedcke 2003 |
| 2001 | European Pharmacopoeia plant monographs | 71 | | Gaedcke 2003 |
| | **Importation data** | | | |
| 1979 | Amount of herbs imported into Germany | | 27900 | Lange 1997 |
| 1994 | Amount of herbs imported into Germany | | 43900 | Lange 1997 |

BGA = Bundesgesundheitsamt

The worldwide origin of the medicinal plants used in Europe is illustrated in Fig. 1. Most of the species used have their origin in the holarctic region of Eurasia and North America and in the palaeotropic region of Africa.

Examples will be presented from the alpine area of Central Europe, from certain valleys in southern Europe, from tropical West Africa and from the arid zones of Southern Africa.

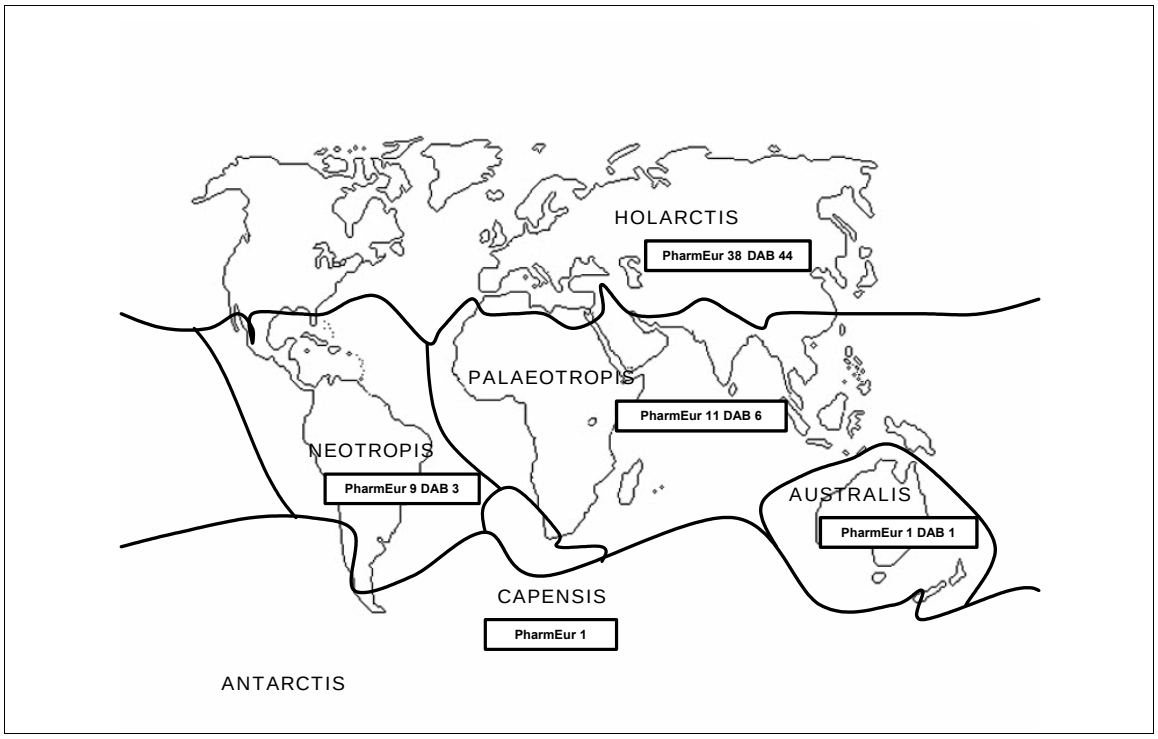


**Fig. 1.** Floristic regions of the world with the number of medicinal plants used in the pharmacopoeia originating in the different regions (PharmEur = European pharmacopoeia; DAB = German pharmacopoeia).

## Ecology of medicinal plants

The ecological parameters influence not only plant growth but also the physiological conditions and in parallel the quality of medicinal properties.

- **Vegetation ecology**

Most plants are adapted to growing in a wide variety of ecosystems, but some are dependent on the particular conditions of their vegetation unit.

The first example is that of the willow species (*Salix* spp.) providing "green aspirin". The quality is described in the European Pharmacopoeia and only a few species contain the high content of salicin required. A special problem with these species is the production of high yield. According to distribution maps of the most important species, *Salix purpurea* L. and *S. daphnoides* Vill. in Central Europe, the first is fairly equally distributed and the latter is found only alongside alpine rivers. Samples with a high salicin content have to be selected from collections along transects through the gallery forests of alpine rivers. Not only is the high content important but also suitability to cultivation. A very good breed of *Salix* eventually failed because of roe deer foraging leading to destruction of the cultivated crop.

Another example of a famous medicinal plant dependent on very special conditions of its vegetation unit is Spanish arnica (*Arnica montana* L. subsp. *atlantica)* that only grows in Galicia, northern Spain. Because of its special chemical composition and lower allergenic potency the Spanish origin is preferred. The species is under threat in its natural habitat because of changing ecological conditions. Decades of wild crafting were not harmful to the species but the improvement of the marshy meadows along the typical rias valleys preferred by the plant caused disappearance of the species.

- **Centres of origin and diversity**

The ecological conditions in the centre of origin and diversity of the species are very important. Only limited information is available about these centres for medicinal plants, and in some cases it will be difficult to identify these owing to the long history of cultivation.

Rosella, a favourite herbal beverage in Europe brewed from the red flower calyx of *Hibiscus sabdariffa*, has its centre of origin and diversity in the Sahel area in western Africa, the so-called "non-centre A2" according to Harlan (1971). *Hibiscus sabdariffa* L. var. *ruber* is not only used as herbal tea but also, in its place of origin, in a traditional sauce with millet dishes. The species is better known as the source of the fibre kenaf.

For further use as a beverage or for other medicinal or aromatic uses the variability of the calyx is important. As usual in the centre of diversity, this variability can be found, and even the colour of the calyx is very variable.

- **Physiological ecology**

The quality of the medicinal properties of a plant depends on the contents of secondary metabolites. Plant metabolism and the physiological conditions of the plant are influenced by ecological parameters. The very harsh conditions of arid zones are best to demonstrate this influence on active ingredients of medicinal plants.

A very suitable example is devil's claw (*Harpagophytum procumbens* DC.) growing in the bush savannah of the Kalahari in southern Africa. Because of severe wild crafting the species was considered as endangered. Due to its adaptability in terms of seed germination the plant is able to survive under hard conditions and has a good regeneration rate. The secondary root tuber is used as herbal remedy in rheumatic diseases. The main trait of interest is the level of harpagoside – one of the active ingredients. When testing many single plants it was found that the genetic variation in harpagoside content is exceeded by the influence of many environmental and other factors: precipitation, nutrients (phosphorus is a limiting factor in the Kalahari), age of mother plant, age of secondary tuber and competition with other plants. Removing the competition of other plants on vegetation-free plots results in an astonishing increase in the growth of *Harpagophytum procumbens*. Influenced by higher precipitation in recent decades, the growth of other bushes in the Kalahari increased competition and this may be an important reason for the endangering of *Harpagophytum*.

## Conclusions

The points to consider when discussing future cooperation in collecting and describing MAP genetic resources are summarized below.

From the scientific point of view, the pharmaceutical/food quality of plant material is most important. The ecological conditions of the original habitat and genetic properties are the basis of the variation in quality. To select proper agricultural conditions for cultivation it is necessary to know the ecological tolerance of the species. Interdisciplinary scientific research may be necessary to collect the necessary data.

For the agronomist, high yield and good prices are the most important criteria. He must ensure good management for quality, for which he will depend on scientific knowledge.

The future legal conditions are also very important. In the future herbal market there will be several categories of products: medicinal plant products, traditional herbal remedies and products called food supplements with functional properties.

As mentioned above (Table 1) the number of species used as MAPs has fallen in recent years. With the legal framework for food supplements with functional properties that will be established in future years the number of species used will increase once more. All scientists involved in MAP research will have to cope with this challenge.

The solution must be an interdisciplinary effort of pharmacognosy, ecology and plant genetics to cooperate in building up a knowledge database of MAP ecological requirements.

It is highly recommended to elaborate pharmacognostic and ecological fact sheets for medicinal plants, and the data should be included in the characterization descriptors for MAP genetic resources.

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# Country reports

## *Medicinal and aromatic plants in Austria*

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### Introduction - General information on the Austrian collections

In 2002 the former Federal Office of Agrobiology became part of the Austrian Agency for Health and Food Safety and was renamed "Austrian Agency of Health and Food Safety – AGES Linz". The new agency is a merger of 19 Austrian institutes dealing with human healthcare, food analysis and agriculture. It belongs to the ministries of Health, Environment and Agriculture. The reason for the establishment of this agency was to encourage synergy, reducing costs for the government.

The Austrian seed collections are well documented as far as the state genebanks are concerned. The *Index Seminum Austriae,* survey of the seed collections in Austria according to the multicrop passport descriptor format, is available on-line.

These seed collections are held in the following institutions:

- Organizations funded by the district government:
  - Genebank Tyrol, maintaining the oldest cereal collection in Austria, established in the 1920s, which consists mainly of cereals grown in the alpine regions.
  - Landesversuchsanlage Wies/Styria, maintaining "special crops" such as medicinal plants, spices, traditional oil plants (e.g. Styrian oil pumpkin) and vegetables. They serve as experimental stations under Styrian climatic conditions.
- Organization funded by the federal government:
  - The Austrian Agency for Health and Food Safety, maintaining the largest collection in Austria with mainly cereals for the pre-alpine and pannonical climate areas, a traditional poppy collection, and a medicinal plant collection started in 1898.
- Private organizations
  - Arche Noah Association, with a large vegetable collection, but no detailed passport information published so far. Data are not included in the Index Seminum Austriae yet. This will be changing in the near future.
  - Several local associations maintaining traditional fruit trees in ex situ arboreta.

Details of the collections (excluding the cereal collections) are given in Table 1.

**Table 1**. The Austrian collections

| | Crops | No. of accessions | Origin |
|---|---|---|---|
| Landesversuchsanlage Wies/Styria | special crops, vegetables | 260 | Austrian |
| AGES-Linz | special crops | 250 | Austrian/German |
| | vegetables | 700 | Austrian *Phaseolus* |
| | medicinal plants, spices* | 590 | European |
| | poppy** | 150 | Austrian landraces |

* This collection was set up in 1898 with importance as a spice-tea store and has increased in importance since the renewed interest in natural products in the 1980s.

** This collection was set up in 1968 with the main task of collecting the last existing landraces of cereals and poppy. In 2003 the genebank collections of the Vienna and Linz institutes were concentrated in Linz.

### Legal protection

The *Red list of endangered species* is updated every few years.

### Sustainable use of medicinal and aromatic plants (MAPs) in Austria

Over 80% of the different species of medicinal plants cultivated in Austria are selections of wild plants, but are mainly distributed as cultivars. Most of these selections are very old and their origin can no longer be verified. Advanced cultivars are rather rare.

The cultivation and processing are carried out by:

- seed companies
- agricultural cooperatives working as seed companies
- agricultural associations of farmers.

### Use of MAPs

MAPs are used mainly for the production of:

- teas (of herbs and fruits)
- single herbs or mixtures of herbs in small packages (e.g. *Petroselinum* or *Carum*)
- foodstuffs with a bacteriostatic effect (*Thymus vulgaris, Origanum vulgare*) used as dry matter directly; this is increasingly important for the large number of farmers using organic/biodynamic methods in Austria.

Only the production and packaging of medicinal and aromatic plants is done in Austria. For the processing/extraction itself, semi-processed products are exported to Switzerland or Germany.

Production of poppy should be mentioned separately because of its high volume of 1000 to 1500 tonnes per year, grown on an area of over 1000 ha. It is only used in traditional food prepared in home kitchens and in the pastry industry. There is no production of poppy syrup for medicinal processing at all in Austria, because of its low alkaloid content under the prevailing climatic conditions.

## *Medicinal and aromatic plant diversity in Bulgaria – protection, collection, study, use and conservation*

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The geographic location of Bulgaria, with its central European, Mediterranean and East Asian flora, as well as the very rugged terrain, determine the rich plant species diversity (Bondev 1995). Data summarized from different studies showed that of the known 3567 higher plant species, 750 are medicinal plants. There are different views on the number of commonly used species. According to some authors this number is 150 (Stoeva 2000), while according to others it is 248 (Hardalova *et al.* 1994). The number of medicinal and aromatic plants (MAPs) is probably much higher if we include the species classified only as aromatic. Among the MAPs used, 85% are wild plants and only 15% are introduced or local cultivars.

### Legal protection of MAP species

The legal protection of MAPs was established in 1936 by passing a law prohibiting or limiting the collection of flowers and herbs in certain regions.

The increased trade in medicinal plants from native habitats led to the *Law for Medicinal Plants*, passed in 1941 and regulating the protection of 63 species (Hardalova *et al.* 1994).

Current legal protection is provided by the *Law for Nature Protection* (LNP) (1967) regulating the listing of species with protected status. According to the last normative papers of 2001, a special regime is imposed for 79 species, including 44 protected, 24 banned for use and 11 with limited use.

The use of medicinal plants from forest territories is regulated by the *Forest Law* and is controlled by the Forest Office at the Ministry of Agriculture, Forestry and Agrarian Reform.

The Ministry of Environment and Waters is in charge of the management and control of the national natural resources (forests excepted) in the protected territories. A different regime of protection has been imposed on 3.5% of the total area of the country, and 82% of the protected territory (nature reserves and national parks) received nature protection status corresponding to categories I and II defined by IUCN (Mihova 1994). Besides nature reserves and national parks, MAPs are also preserved in 49 protected territories, specified for definite medicinal plants: *Leucojum aestivum* L., *Paeonia peregrina* Mill., *Glycyrrhiza glabra* L., *Galanthus nivalis* L., *Juniperus communis* L., *Sideritis taurica* Steph., *Inula helenium* L. and *Primula veris* L. (Hardalova *et al.* 1994).

A new *Law for Medicinal Plants* (LMP), regulating the sustainable use of natural resources and biodiversity protection has been passed on 7 April 2000 (Official Gazette 29, Sofia).

Another basis for active protection is the *Red Book of Bulgaria* which includes 38 medicinal plant species according to Hardalova *et al.* (1994).

### *Ex situ* collections

The main activities of the Institute of Plant Genetic Resources (IPGR) in Sadovo are focused on the protection and conservation of plant resources as a source of germplasm enrichment (Koeva 1998). Part of this general programme is related to MAP resources (Angelova *et al.* 1994; Varbanova and Dimitrova 2000).

The main lines of study include: (i) collecting plants for their diversity; (ii) creation and enrichment of collections; (iii) evaluation; and (iv) conservation. They are detailed below.

- **Collecting plants for their diversity**

MAP collecting is funded by IPGR, through national and international projects.

MAP collecting in Sadovo started as early as 1902, but significant activities related to genetic resources started in 1983 with the establishment of international exchanges with botanical gardens, research institutes, firms, etc. (Guteva *et al.* 1998).

Active collaboration and germplasm exchange are currently maintained with the Research Institute for Roses, Aromatic and Medicinal Plants in Kazanlak, the Agricultural University in Plovdiv, the Institute of Botany in Sofia, as well as with non-governmental organizations (NGOs).

The collection of wild accessions started in 1993 and is carried out through collecting missions in different regions of the country. A total of four joint projects with the Committee of Forests, the Ministry of Education and Science, the Bulgarian/Swiss Biodiversity Conservation Programme (BSBCP) and 13 collecting missions in the country were carried out during the period reported.

- **Creation and structure of the collections**

The strategy for the creation of collections is to achieve:
- Species diversity
- Intraspecific diversity
- Diversity of plants with protected status
- Ecogeographic diversity.

The *ex situ* MAP collection in IPGR is represented by 461 accessions distributed in 24 families. The highest species diversity is found in the families Lamiaceae (28 species) and Asteraceae (15). Other families include from 1 to 7 species (Table 1).

**Table 1.** Structure of the *ex situ* field collection

| No. | Family | No. of species | No. of accessions | | | |
|---|---|---|---|---|---|---|
| | | | Total | Wild | Foreign | Bulgarian (local cultivars and populations) |
| 1 | Amaryllidaceae | 1 | 23 | - | 23 | - |
| 2 | Araceae | 1 | 1 | 1 | - | - |
| 3 | Asteraceae | 15 | 292 | 18 | 252 | 22 |
| 4 | Boraginaceae | 1 | 3 | 3 | - | - |
| 5 | Caryophyllaceae | 7 | 9 | 5 | 4 | - |
| 6 | Cistaceae | 2 | 2 | 2 | - | - |
| 7 | Crassulaceae | 3 | 3 | 3 | - | - |
| 8 | Fabaceae | 3 | 3 | 3 | - | - |
| 9 | Ericaceae | 2 | 2 | 2 | - | - |
| 10 | Gentianaceae | 1 | 1 | 1 | - | - |
| 11 | Hypericaceae | 1 | 2 | 2 | - | - |
| 12 | Iridaceae | 6 | 12 | 10 | 2 | - |
| 13 | Lamiaceae | 28 | 83 | 69 | 5 | 10 |
| 14 | Malvaceae | 1 | 1 | 1 | - | - |
| 15 | Onagraceae | 2 | 2 | 2 | - | - |
| 16 | Paeoniaceae | 2 | 3 | 3 | - | - |
| 17 | Primulaceae | 2 | 2 | 2 | - | - |
| 18 | Ranunculaceae | 4 | 4 | 4 | - | - |
| 19 | Resedaceae | 1 | 1 | 1 | - | - |
| 20 | Rosaceae | 4 | 4 | 4 | - | - |
| 21 | Rubiaceae | 1 | 1 | 1 | - | - |
| 22 | Rutaceae | 1 | 1 | 1 | - | - |
| 23 | Scrophulariaceae | 5 | 5 | 5 | - | - |
| 24 | Solanaceae | 1 | 1 | 1 | - | - |
| **Total** | | **95** | **461** | **143** | **286** | **32** |

According to the plant material origin, MAPs are divided into two groups – cultivated and wild:

- **Cultivated species**

This group includes local and foreign cultivars and populations and is represented by 318 accessions. Among the introduced MAPs obtained by exchange with 15 countries, those from European countries are predominant, with the Netherlands and Russia being the most represented. Seven countries are represented by 1 to 5 accessions only. The highest share in the collection is that of foreign accessions (252) of the family Asteraceae, including mainly species with ornamental qualities: *Calendula officinalis* L., *Tagetes* sp., *Narcissus* sp. The cultivated Bulgarian MAP species are represented by 10 cultivars and 22 populations.

- **Wild species**

This group is represented by 143 accessions from nine floristic regions of the country. Most MAPs have been collected from the North and South Black Sea coast, followed by Strandza, East Rhodope Mountains, and the hilly Tundza river plain. A rich species diversity of steppe origin is also represented. This group includes 23 families, the highest diversity being shown by the family Lamiaceae (28 species). A rich species diversity is collected from the genera *Salvia (S. verticillata* L., *S. aethiopis* L., *S. sclarea* L., *S. pratensis* L., *S. officinalis* L., *S. nutans* L., *S. austriaca* Jacq., *S. virgata* Jacq., *S. pinnata* L.) and *Mentha (M. aquatica* L., *M.* x *piperita* L., *M. pulegium* L., *M. spicata* L., *M. viridis* L.*)*. Collecting from different habitats contributed to the achievement of intraspecific diversity in *Inula helenium, Symphitum officinale, Thymus* sp., *Teucrium chamaedris, Hypericum perforatum, Mentha viridis* L., *Salvia verticillata* and *S. pratensis.*

A total of 32 wild species with different degrees of vulnerability and protection have been collected and conserved in *ex situ* collections (Table 2). Twenty-two of those are registered in the *Red Book of Bulgaria*, with 12 being categorized as rare, 4 as threatened and 6 as endemic.

Under the *Law for Nature Protection* (LNP), 17 MAPs have been included in the collection, 6 of which are protected, 5 are banned for use and 6 are allowed limited use.

- **Evaluation**

The MAP *ex situ* field collection is studied in three stages: preliminary evaluation, complex evaluation and special evaluation.

Preliminary evaluation is compulsory for the introduced accessions and is performed in the quarantine field, where accessions are assessed for quarantine diseases.

Complex evaluation is an important source of information to be recorded in databases and is required for seeds that will be placed under long-term storage in the national genebank. It involves biological, morphological and economic characteristics.

A special evaluation is made for some of the accessions which have undergone complex evaluation. It is performed in accordance with the specific lines of use – breeding, trade, biogarden establishment, etc. (Varbanova *et al.* 2003). Trait collections were created for *Tagetes* sp. and *Calendula officinalis* L., for use as a source of information in breeding programmes.

In wild accessions, special attention is paid to the adaptability of species to the new growing conditions and the establishment of suitable propagation methods.

The evaluation of species adaptability comprises biological, morphological and economic traits and is carried out in the habitats and conditions of Sadovo (Peeva *et al.* 1995; Varbanova *et al.* 1995, 1997; Dimitrova and Varbanova 1995; Dimitrova *et al.* 1997) (Table 3).

**Table 2.** Wild MAPs with protected status in the *ex situ* collection

| No. | Species | Red Book of Bulgaria | | | Law for Nature Protection | | |
|---|---|---|---|---|---|---|---|
| | | Rare | Threatened | Endemic | Protected | Banned | Limited |
| 1 | *Rubia tinctorum* L. | | | | | + | |
| 2 | *Symphytum officinale* L. | | | | | | + |
| 3 | *Inula helenium* L. | | | | | + | |
| 4 | *Glycyrrhiza glabra* L. | | + | | + | | |
| 5 | *Origanum vulgare* subsp. *hirtum* (Link) Ietswaart | | | | | + | |
| 6 | *Gypsophila trichotoma* Wend. | + | | | | | |
| 7 | *Gypsophila tekirae* Stefanov | | | + | | | |
| 8 | *Acorus calamus* L. | | | | + | | |
| 9 | *Iris aphylla* L. | | | | + | | |
| 10 | *Iris suaveolens* Bois et Reut. | | | + | | | |
| 11 | *Rhododendron ponticum* L. | | + | | | | |
| 12 | *Cistus salvifolius* L. | + | | | | | |
| 13 | *Artemisia pedemontana* Balbbs. | | + | | | | |
| 14 | *Achillea clypeolata* Sibth. et Sm. | | | + | | | |
| 15 | *Adonis vernalis* L. | | | | | | + |
| 16 | *Althaea officinalis* L. | | | | | + | |
| 17 | *Atropa bella-donna* L. | + | | | | | + |
| 18 | *Paeonia peregrina* Mill. | | | | | | + |
| 19 | *Salvia pinnata* L. | | | | + | | |
| 20 | *Ruta graveolens* L. | + | | | + | | |
| 21 | *Alchemilla asteroantha* Rotbm. | + | | + | | | |
| 22 | *Erica arborea* L. | + | | | | | |
| 23 . | *Satureja coerulea* Janka. | | | + | | | |
| 24 | *Sedum acre* L. | | | | | | + |
| 25 | *Helichrysum arenarium* (L.) Moench | + | | | | + | |
| 26 | *Saponaria stranjensis* D. Jord. | | | + | | | |
| 27 | *Centaurium erythraea* Rafin. | + | | | | | |
| 28 | *Onanthus maritimus* (L.) Hoffm. et Link | + | | | | | |
| 29 | *Adonis wolgensis* Stev. | + | | | | | |
| 30 | *Primula vulgaris* Huds. subsp. *sibthorpii* (Hoffm.) Sm. et Forest | + | | | | | + |
| 31 | *Primula veris* L. | + | | | | | |
| 32 | *Galanthus nivalis* L. | | + | | + | | |
| **Total** | | **12** | **4** | **6** | **6** | **5** | **6** |

**Table 3.** Adaptability of wild MAPs to cultivation

| No. | Species | Adaptability | | |
|---|---|---|---|---|
| | | Very good | Good | Low |
| 1 | *Inula helenium* | + | | |
| 2 | *Iris aphylla* | + | | |
| 3 | *Gypsophila trichotoma* | + | | |
| 4 | *Gypsophila tekirae* | + | | |
| 5 | *Gypsophila paniculata* | | + | |
| 6 | *Clinopodium vulgare* | | + | |
| 7 | *Althaea officinalis* | + | | |
| 8 | *Mentha longifolia* | | + | |
| 9 | *Symphitum officinale* | | + | |
| 10 | *Paeonia tenuifolia* | | + | |
| 11 | *Rubia tinctorum* | + | | |
| 12 | *Origanum vulgare* subsp. *hirtum* | + | | |
| 13 | *Adonis vernalis* | | | + |
| 14 | *Rhododendron ponticum* | | | + |
| 15 | *Vaccinium arctostaphyllos* | | | + |
| 16 | *Cistus incanus* | | | + |
| 17 | *Cistus salvifolius* | | + | |

To study the propagation abilities of some wild species (difficult to propagate, valuable, with protected status, commercial, etc.) traditional and *in vitro* methods are applied (Dimitrova *et al*. 1994, 1996, 2001). The results are used to create *ex situ* field collections and *in vitro* collections, as well as for commercial propagation.

Many MAPs have ornamental value and can be successfully grown in home biogardens. The collection of Sadovo includes more than 35 MAP species with ornamental qualities. Of high ornamental value are the species *Calendula officinalis, Artemisia* sp., *Iris* sp., *Adonis vernalis, Althaea officinalis, Oenothera biennis, Inula helenium, Paeonia peregrina, Salvia* sp., *Tagetes* sp.

- **MAP conservation**

MAP species are conserved *ex situ* (field collections, genebank seed collections and *in vitro* collections), and *in situ* (Stoyanova *et al.* 1998).

- ***Ex situ* conservation** (Table 4)

A total of 196 accessions are conserved under **field conditions**. Two collections are maintained: a working collection and an *in vivo* conservation collection. The working collection includes 75 accessions. Besides conservation, studies and propagation of accessions are performed to produce seed material for the genebank. The *in vivo* collection contains 121 vegetatively propagated accessions.

The national **genebank** of IPGR in Sadovo contains 266 seed accessions of 27 MAP species, kept in three types of collections: base, active and exchange collections. The base collection provides long-term conservation of 152 accessions at a temperature of –18°C for a period of 15-20 years. The active collection contains a total of 114 accessions under mid-term storage conditions, at a temperature of 6°C.

An alternative method for conservation and storage of MAPs is the creation of ***in vitro* collections**. Sixteen wild MAP species are propagated and stored *in vitro* at IPGR-Sadovo.

**Table 4.** *Ex situ* conservation of MAP species

| Family | Total | No. of accessions | | | | |
|---|---|---|---|---|---|---|
| | | Genebank | | Field | | *In vitro* |
| | | Long-term | Medium-term | *In vivo* collection | Working collection | |
| Amaryllidaceae | 23 | - | - | 23 | - | - |
| Araceae | 1 | | | 1 | | |
| Asteraceae | 292 | 146 | 96 | 12 | 38 | - |
| Boraginaceae | 4 | - | 1 | 1 | 1 | 1 |
| Caryophyllaceae | 12 | 2 | 3 | 2 | 2 | 3 |
| Cistaceae | 3 | - | - | 2 | - | 1 |
| Crassulaceae | 3 | - | - | 2 | 1 | - |
| Fabaceae | 4 | - | - | 2 | 1 | 1 |
| Ericaceae | 2 | 1 | - | - | 1 | - |
| Gentianaceae | 2 | - | - | - | 1 | 1 |
| Hypericaceae | 3 | - | - | - | 2 | 1 |
| Iridaceae | 12 | - | 2 | 10 | - | - |
| Lamiaceae | 88 | - | 11 | 57 | 15 | 5 |
| Malvaceae | 1 | - | - | - | 1 | - |
| Onagraceae | 2 | 1 | - | - | 1 | - |
| Paeoniaceae | 3 | - | - | 2 | 1 | - |
| Primulaceae | 2 | - | - | - | 2 | - |
| Ranunculaceae | 4 | - | 1 | 1 | 2 | - |
| Resedaceae | 2 | - | - | 1 | - | 1 |
| Rosaceae | 4 | 1 | - | 2 | 1 | - |
| Rubiaceae | 1 | - | - | 1 | - | - |
| Rutaceae | 3 | 1 | - | - | 1 | 1 |
| Scrophulariaceae | 5 | - | - | 1 | 4 | - |
| Solanaceae | 2 | - | - | 1 | - | 1 |
| **Total** | **478** | **152** | **114** | **121** | **75** | **16** |

- ***In situ* conservation**

Within the framework of a Bulgarian/Swiss Biodiversity Conservation Programme (BSBCP) project, a programme for *in situ* conservation was implemented. Ecogeographic investigations in the Kaliakra nature reserve and the habitats of *Adonis vernalis* and *Paeonia tenuifolia* were conducted. During the 5-year investigation period, information about the status of populations was collected (Varbanova *et al.* 1998, 2001). Databases for the biodiversity in the Kaliakra nature reserve and its adjacent territories were created and a management plan was developed (Angelova and Varbanova 2002).

## Use and trade

Herb collecting in Bulgaria has long-established traditions (Mladenova 2000). Bulgarian medicinal plants are considered to be among the highest quality plants in the world owing to the specific soil and climatic conditions. Annually, from 12 000 to 15 000 tonnes of herbs are collected and processed in the country. Wild plants account for 70% of the total herb production while 30% are cultivated; 60-70% of the MAPs collected are intended for export, the other 30-40% remaining in the country as raw material for pharmaceutical and cosmetic industries, for the preparation of medicated and common teas, spices, etc. Of the wild plant species, the largest quantities exported are of *Mentha* sp., *Melissa officinalis, Lavandula vera, Hypericum perforatum, Matricaria chamomilla, Lamium album, Rosa canina, Tilia* sp. Bulgaria is one of the biggest MAP exporters in Europe, Germany being the major consumer. Recently, exports to Italy and Spain have also increased.

## Conclusion

The great MAP diversity in Bulgaria, the traditions and the trade activities for MAP collecting determine the main priorities in the conservation of MAP genetic resources:

- Effective legislative protection at the national level;
- Clear strategy of IPGR for MAP collecting, study and conservation;
- Coordinated activities and interrelations of IPGR as a national coordinator with related institutions, research institutes, NGOs, etc.;
- International collaboration, established and enriched through projects and programmes;
- Development of a database and participation in research on biodiversity conservation (Angelova *et al.* 1996; Angelova and Varbanova 2002).

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## *Legal protection, conservation and cultivation of medicinal and aromatic plants in Croatia*

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### Legal protection of medicinal and aromatic plants (MAPs) and their natural habitats

The *Law on Nature Protection* (Official Gazette 'Narodne novine' Nos. 30/94 and 72/94) is the basic Croatian law regulating the issues of biological and landscape diversity preservation. The principal *in situ* biological conservation method is the establishment of protected natural areas. Pursuant to the *Law on Nature Protection*, approximately 7.5% of the Croatian State territory is protected in this manner. This Law establishes 8 spatial protection categories (national park, park of nature, strict reserve, special reserve, monument of nature, protected landscape, park-forest and park architecture monument). All protected areas are subject to very strict biodiversity protection measures (Radović 2000).

Apart from the protected areas, certain plant species (44) are also protected pursuant to the *Law on Nature Protection* (Box 1). Five out of these 44 species (in **bold**) are traditionally used in folk medicine although it is not likely that overexploitation for medical purposes is the main reason of their rareness. However, overexploitation of related species such as *Gentiana lutea* L. and *Paeonia officinalis* L. has led to genetic erosion of these plants in some environments.

The Ministry of Environmental Protection and Physical Planning is currently working on a Rule Book that would regulate protection of all plant species on the Red List in Croatia. The *Red Data Book of Plant Taxa of the Republic of Croatia* (Šugar 1994) provided a list of 401 nationally threatened plant species classified as extinct (2), possibly extinct (2), endangered (87), vulnerable (85), rare (214), and indeterminate (11). According to the *Red Data Book* there are 17 plant species that are endangered, *inter alia*, due to their collecting as medicinal plants (Box 2). Nevertheless, other factors such as habitat loss or modification (drainage works, dam building, clearing of land for agriculture, road construction, mining) as well as small population size and collecting for decorative purposes seems to be more important in most cases.

According to Article 36 of the *Law on Nature Protection* the Ministry's approval should be obtained for gathering plants (and their parts) that are not protected by the present or other laws if this is done for processing, commercial or trading purposes. The *Rule Book on Collecting Plants in the Wild* which regulates the gathering, trade and export of particular wild medicinal plants is still in preparation. Upon request, the Ministry issues permissions and sets quotas for the gathering of particular wild medicinal plants. In 1991, nine companies have obtained permissions for wild-collecting of 87 plant species (Box 3) and a total quota for the quantities to be wild-collected was set to 108 886.50 kg. The quantities were set according to the state of the natural population for the different species on the basis of research made by specialists from the Faculty of Science and the Faculty of Pharmacy and Biotechnology. Data on actual quantities gathered upon permissions are not complete.

In the framework of the project "Biological database and geographic information system" (main researcher: Toni Nikolić, Faculty of Science, University of Zagreb), the *Flora Croatica: Index Florae Croaticae* and *Flora Croatica* Database have been developed. They are continuously updated, supplemented and modified regarding the qualitative structure of Croatian flora (Nikolić 1994, 1997, 2000). However, the survey of the status of MAP natural resources still has to be done.

**Box 1.** List of 44 protected plant species in Croatia

1. ***Anacamptis pyramidalis* (L.) L.C.M. Richard**
2. *Anthyllis barba-jovis* L.
3. *Aquilegia kitaibelli* Schott
4. *Arbutus andrachnoides* Link
5. *Betula pubescens* Ehrh.
6. *Centaurea ragusina* L.
7. *Cephalanthera damasonium* (Mill.) Druce
8. *Cephalanthera longifolia* (L.) Fritsch
9. *Cephelanthera rubra* (L.) L.C.M. Richard
10. *Convolvus cneorum* L.
11. *Cypripedium calceolus* L.
12. *Daphne blagayana* Freyer
13. *Daphne cneorum* L.
14. *Daphne laureola* L.
15. *Degenia velebitica* (Degen) Hayek
16. *Doronicum orientale* Hoffmann
17. *Eranthis hyemalis* (L.) Salisbury
18. *Eryngium alpinum* L.
19. *Fritillaria meleagris* L.
20. ***Gentiana acaulis* L.**
21. *Gentiana clusii* Perr. & Songeon
22. ***Gentiana lutea* L. subsp. *symphyandra* (Murbeck) Hayek**
23. *Ilex aquifolium* L.
24. *Leontopodium alpinum* Cassini var. *krasense* (Derg.) Hayek
25. *Lilium bulbiferum* L.
26. *Lilium carniolicum* Bern. ex Koch
27. *Lilium martagon* L.
28. *Nigritella nigra* (L.) Reichenbach fil.
29. ***Paeonia mascula* (L.) Mill. subsp. *mascula***
30. *Pinus mugo* Turra
31. *Platanthera bifolia* (L.) L.C.M. Richard
32. *Platanthera chlorantha* (Custer) Reichenbach & Moessler
33. *Polygala chamaebuxus* L.
34. *Primula auricula* L.
35. *Primula kitaibeliana* Schott
36. *Primula wulfeniana* Schott
37. *Quercus coccifera* L.
38. *Rhododendron hirsutum* L.
39. *Ruscus hypoglossum* L.
40. *Scopolia carniolica* Jacuin
41. *Sibiraea croatica* Degen
42. *Styrax officinalis* L.
43. *Taxus baccata* L.
44. ***Trollius europaeus* L.**

**Box 2.** List of 17 plant species endangered by wild collecting as medicinal plants

1. *Achillea clavenae* L.
2. *Anacamptis pyramidalis* (L.) L.C.M.Rich.
3. *Arctostaphylos uva-ursi* (L.) Spreng.
4. *Arnica montana* L.
5. *Crataegus transaplina* Kern.
6. *Digitalis grandiflora* Mill.
7. *Gentiana acaulis* L.
8. *Gentiana symphyadra* (Murb.) Fritsch (syn. G. *lutea* L. subsp. *symphyandra* Murbeck)
9. *Iris illyrica* Tomm
10. *Ophrys fusca* Link
11. *Orchis laxiflora* Lam.
12. *Orchis mascula* (L.) L.
13. *Paeonia mascula* (L.) Mill.
14. *Paeonia officinalis* L.
15. *Scopolia carniolica* Jacq.
16. *Silybum marianum* (L.) Gaertner
17. *Trollius europeaus* L.

**Box 3.** List of 87 wild-collected species for which permission was granted in 1991

1. *Abies alba* Mill.
2. *Achillea millefolium* L.
3. *Aesculus hippocastaneum* L.
4. *Agropyron repens* (L.) PB.
5. *Alchemilla vulgaris* L.
6. *Allium ursinum* L.
7. *Althaea officinalis* L.
8. *Anthyllis vulneraria* L.
9. *Arctium lappa* L.
10. *Arctostaphylos uva-ursi* (L.) Spr.
11. *Artemisia absinthium* L.
12. *Artemisia vulgaris* L.
13. *Asarum europaeum*
14. *Bellis perennis* L.
15. *Berberis vulgaris* L.
16. *Betula pendula* Roth.
17. *Calendula arvensis* L.
18. *Calluna vulgaris* (L.) Hull.
19. *Capsella bursa-pastoris* (L.) Med.
20. *Castanea sativa* Mill.
21. *Centaurium erythraea* Rafn.
22. *Centaurium umbellatum* L.
23. *Ceratonia siliqua* L.
24. *Chelidonium majus* L.
25. *Cichorium inthibus* L.
26. *Colchicum automnale* L.
27. *Corylus avellana* L.
28. *Crataegus monogyna* (Poir.) DC.
29. *Crataegus oxycantha* L.
30. *Daucus silvestris* L.
31. *Equisetum arvense* L.
32. *Euphrasia officinalis* L.
33. *Filipendula ulmaria* (L.) Maxim.
34. *Fragaria vesca* L.
35. *Fumaria officinalis* L.
36. *Galium verum* L.
37. *Geranium robertianum* L.
38. *Glechoma hederacea* L.
39. *Hedera helix* L.
40. *Helichrysum italicum* (Roth.) Mill.
41. *Humulus lupulus* L.
42. *Hypericum perforatum* L.
43. *Juniperus communis* L.
44. *Lamium album* L.
45. *Malva silvestris* L.
46. *Melilotus officinalis* (L.) Pall.
47. *Melissa officinalis* L.
48. *Ononis spinosa* L.
49. *Origanum vulgare* L.
50. *Petasites hybridus* (L.) G.
51. *Phyllitis scolopendrium* (L.) Newm.
52. *Plantago lanceolata* L.
53. *Plantago major* L.
54. *Polygonum aviculare* L.
55. *Potentilla erecta* (L.) Rauschel.
56. *Prunus spinosa* L.
57. *Pulmonaria officinalis* L.
58. *Quercus robur* L.
59. *Rhamnus frangula* L.
60. *Rosa canina* L.
61. *Rosmarinus officinalis* L.
62. *Rubus fruticosus* L.
63. *Rubus idaeus* L.
64. *Rumex acetosa* L.
65. *Salix alba* L.
66. *Salvia officinalis* L.
67. *Sambucus ebulus* L.
68. *Sambucus nigra* L.
69. *Satureja montana* L.
70. *Solidago serotina* Ait.
71. *Solidago virgaurea* L.
72. *Symphytium officinale* L.
73. *Taraxacum officinale* Web.
74. *Teucrium montanum* L.
75. *Thymus serpyllum* L.
76. *Tilia cordata* Mill.
77. *Tilia platyphyllos* Scop.
78. *Tussilago farfara* L.
79. *Urtica dioica* L.
80. *Vaccinium myrtillus* L.
81. *Valeriana officinalis* L.
82. *Verbascum* sp.
83. *Vinca minor* L.
84. *Viola odorata* L.
85. *Viola tricolor* L.
86. *Viscum album* L.
87. *Vitex agnus-castus* L.

### *Ex situ* conservation of MAP species

*Ex situ* conservation of MAP species is carried out at the Pharmaceutical Botanic Garden "Fran Kušan"; Faculty of Pharmacy and Biochemistry, University of Zagreb; and Croatian Bank of Plant Genes, Faculty of Agriculture, University of Zagreb.

- **Pharmaceutical Botanic Garden "Fran Kušan"**

This garden was established in 1947, and since then it is one of the few European botanical gardens specialized in growing medical plants. The collections include about 3000 taxa which are intended for morphological, systematic and ecological studies for scientific, practical and educational purposes and needs and also for making collections. This garden publishes a *Delectus seminum* since 1958, and since 1962 the publication *Informationes Botanicae*.

- **Faculty of Pharmacy and Biochemistry**

Ongoing projects include:

- "Research on medicinal plants of Croatia" (main researcher: Zdenka Kalođera),
- "Pharmacobotanical and chemotaxonomical investigation of medicinal plants" (main researcher: Željan Maleš), and
- "Medicinal plants - biologically active compounds and QSAR" (main researcher: Marica Medić-Šarić).

- **Croatian Bank of Plant Genes**

The MAP collection was established in 1998. Accessions are held in classical *ex situ* maintenance facilities (cold chamber 75 $m^3$; medium-term seed storage at +4°C; seed samples in paper bags and glass jars). Currently, the collection covers more than 900 accessions of about 180 MAP species. Most accessions represent wild material of Croatian origin. Foreign accessions were obtained from commercial seed companies (Austria, Germany, Hungary, Italy, the Netherlands, Russia, USA) as well as from other genebanks (Austria, Czech Republic, Germany, Lithuania, Poland, Slovakia, USA) and botanical gardens (Italy, Germany).

Collecting of MAP species is set as one of the priorities of the Croatian Bank of Plant Genes and a number of collecting missions are organized each year. In 2002 an international collecting mission (Croatia, Czech Republic, Slovakia and Slovenia) was carried out in Istria with the aim of collecting medicinal and aromatic plants and forage legumes and grasses. In collaboration with the Central Slovene Genebank for Plant Genetic Resources at Ljubljana a collecting mission to Kvarner islands (Krk, Cres, Rab, Pag) is planned for September 2003.

A MAP multicrop collecting form was developed to standardize collecting information. Collected information includes: accession identification (taxonomy, distinguishing morphology, etc.); collecting site information: physical description (latitude, longitude, elevation, topographic landform, soil characteristics, etc.), vegetation description (local vegetation classification, dominant vegetation, etc.); and assessment of overall risk of genetic erosion (abundance, spatial pattern, diversity status, causes of biodiversity loss, overexploitation status, etc.).

The main problem in the characterization and evaluation of the collected material is the lack of suitable descriptor lists. A descriptor list for basil species (*Ocimum* spp.) based on Hiltunen and Holm (1999) has been developed and the characterization of basil accessions has been carried out (Karlović 2002). Descriptor lists are currently being developed for marshmallow (*Althaea officinalis* L.), helichrysum (*Helichrysum italicum* (Roth) G. Don), St John's wort (*Hypericum perforatum* L.), oregano (*Origanum vulgare* L.), sage (*Salvia*

*officinalis* L.), winter savory (*Satureja montana* L.) and Dalmatian pyrethrum (*Tanacetum cinerariifolium* (Trev.) Schultz Bip.).

These activities have been carried out as part of two projects at the Faculty of Agriculture, University of Zagreb: "Croatian Bank of Plant Genes" (main researcher: Ivan Kolak), and "Genetic variability of medicinal and aromatic plants" (main researcher: Zlatko Šatović).

Characterization and evaluation of germplasm samples in plant genebanks is traditionally based on morphological traits, but recently modern conservation programmes include the analysis of genetic variability using molecular genetics techniques. Currently, molecular genetics research activities include phylogenetic studies of basils (Šatović 2002) and investigations on the genetic structure of Croatian sage populations. In collaboration with the Agricultural Institute of Slovenia, Ljubljana, studies on the genetic variability of oregano populations have been carried out.

### Cultivation of MAP species

According to the Croatian Bureau of Statistics (CBS 2002), MAPs are cultivated on an area of about 2000 ha (yearly average between 1997 and 2001: 2146 ha). The share of MAP production amounts only to 0.16% of the area of arable land and gardens (or 0.07% of total agricultural land). About 80% of the total cultivation area of MAPs is on family farms and the rest is owned by agricultural companies.

Major cultivated MAP species are: German chamomile (*Chamomilla recutita* (L.) Rauschert), peppermint (*Mentha* x *piperita* L.), lavender (*Lavandula angustifolia* Mill.), sage (*Salvia officinalis* L.), marshmallow (*Althaea officinalis* L.), pot marigold (*Calendula officinalis* L.), fennel (*Foeniculum* vulgare Mill.) and St John's wort (*Hypericum perforatum* L.). Since there are no statistical data available on the production of specific crops, it is very difficult to give a correct estimation.

The dominant cultivated MAP is German chamomile. Its area of cultivation is estimated to amount to more than 3000 ha and this is clearly in contradiction with official statistics. It is cultivated on more than 800 family farms concentrated in three Croatian counties (Virovitičko-podravska, Osječko-baranjska, and Koprivničko-križevačka). Nearly all the farmers have contracts with wholesalers who supply them with seeds and technical information and buy the harvested crop. There are three main wholesalers who buy almost the entire production (Duhanprodukt, Pitomača; Jan Spider, Pitomača; Agristar, Osijek). The average yield of dry chamomile flower is about 600 kg/ha. More than 90% of dried chamomile flowers is exported.

MAP cultivation on family farms is usually a marginal activity and only a few farms specialize in MAP production. Most of the farmers produce crops on the basis of contracts with wholesalers or industry. The trade is dominated by a few wholesalers who purchase plant material from cultivators and also from collectors. There are 15 registered wholesalers dealing with MAPs. The harvested MAP material is delivered to different kinds of industries, such as the pharmaceutical, cosmetic or food industries, or exported. The main buyers of MAP material in Croatia are Pliva (Zagreb) and Frack (Zagreb).

Trade figures obtained from the Croatian Chamber of Economy are based on the tariff heading "Medicinal plants and spices". Unfortunately, tariff codes do not discriminate between the wild or cultivated nature of the material. Moreover, sometimes they do not refer to actual plant species (or plant parts), but to a mixture of dried herbs that may include a number of different species.

The annual volume of MAP material exported from Croatia in 2001 was 1856 t. German chamomile was the most important export commodity and it accounted for more than 60% (1207 t) of overall MAP exports. Other important species include peppermint (46 t), sage (21 t), marshmallow (12 t) and fennel (11 t). Since all these species are regularly produced in

Croatia it seems that the export is largely based on cultivated material. MAPs are exported mainly to European countries (Bosnia and Herzegovina, Yugoslavia, Slovenia, Austria, Germany, Sweden, Italy, Macedonia, etc.) and USA. However, the annual volume of MAP material imported into Croatia in 2001 amounted to 2463 t.

Since 1999, Croatian MAP producers and processors are organized as the "Affiliation of Medicinal Herb Producers and Processors at the Croatian Chamber of Economy". The main aim of the Affiliation is to encourage cooperation in this sector and to strengthen links between research institutes, private companies and government agencies. The Affiliation is also involved in making regulations and legislation affecting production, processing, quality control, and trade of MAP species and products.

MAP producers and processors generally agree that marketing opportunities do exist but there are many problems to be solved and increased governmental support is clearly needed. The main problems are the lack of commercially manufactured specialized machinery, lack of suitable storage and drying facilities, lack of commercially available seeds of high quality (high-yielding cultivars) and lack of knowledge on crop-specific management (especially fertilizer requirements: lack of approved herbicides/pesticides) and harvesting techniques. Specific investments are often essential to start production and the return on investment is less predictable than for traditional crops.

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# *Genetic resources of medicinal and aromatic plants in Cyprus with emphasis on the selection, evaluation and management of* Origanum dubium

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## Introduction

Cyprus is the third largest island in the Mediterranean with an area of 9251 km$^2$ and includes four distinct topographical areas. The climate is intense Mediterranean with wet, changeable winters from November to March, and hot, dry summers from May to September, separated by short spring and autumn seasons. The vegetation consists mainly of coniferous forest, the maquis, the garigue and batha. A total of 1907 taxa have been recorded as native or naturalized and 376 taxa as cultivated. Of the native taxa 141 are endemic (Della 1999a, 2000). A number of aromatic, medicinal and other useful plants are being exploited in Cyprus, e.g. *Origanum dubium*, *Salvia fruticosa*, *Sideritis perfoliata*, *Matricaria recutita*, *Urtica urens*, *Plantago coronopus* subsp. *commutata*, *Mentha spicata* subsp. *spicata*, *Rosmarinus officinalis*, etc. Among the wild shrubs which are partly used for their aromatic fruits are: lentisk, *Pistacia lentiscus* L. ("schinia", "schinnos") and *Pistacia terebinthus* L. (terebinth, "trimithkia"). The aromatic fruit of *Pistacia atlantica* Desf. ("tremithos"), the mastic-producing tree, which is used for making pies, was used in the past for oil production. *Laurus nobilis* L. (laurel) is gathered or grown for its aromatic leaves and fruit. The leaves are used as aromatics and the oil from its fruit in cosmetics. An interesting plant of economic importance is *Rhus coriaria* L. (sumach, "roudhi", "soumatji"), an erect or spreading shrub of about 2 m in height, which grows on stony mountainsides and in vineyards above 600-1800 m. The leaves of *Rhus coriaria*, an industrial plant rich in tannin, are collected every year and exported. *Crataegus azarolus* L. (azarole, "mosphilia"), a small round-headed tree, is grown on rocky mountainsides, by road sides or by field margins, where it is often planted. It is found from sea level to 1200 m altitude. Its fruit is gathered and eaten fresh or used to make home-made jams. It is used also in industry. The carob tree, *Ceratonia siliqua* L., the "charoupia" or "teratsia" of the Cypriots, which is grown in the wild, is also cultivated for its ripe fruit. It grows on dry hillsides in the garigue and in coastal and sub-maritime maquis from sea level to 700 m. It is widely cultivated in lowland areas (Della 1999b, 1999c).

*Ex situ* collections are conserved in the National Genebank (Della 1997). National legislation protects the forest, rare endemic plants, national forest parks or nature reserves, as well as very old trees. A framework law has recently been ratified for the protection of nature. Although aromatic plants have always been grown in Cyprus, their cultivation has increased in recent years due to their use in international cuisine and to their association with better health. It is fortuitous that the agroclimatic conditions in Cyprus are very suitable for a wide range of aromatic crops. Aromatic and medicinal plants are grown at the Athalassa (near Nicosia) Government Nursery (Department of Agriculture) for evaluation and utilization. A number of the above species are already grown on a commercial scale.

Research work at the Agricultural Research Institute includes studies of plant population density, cutting height, selection of different genotypes of oregano, experiments with pre- and post-emergence herbicides in a number of aromatic plant species, and irrigation experiments to study the effect of irrigation on the yield and quality of oregano and sage.

## Selection of local genotypes of oregano

The aim of this study is to select among local genotypes of oregano the best clone for yield and quality. It has been observed that the populations of oregano were not uniform for many parameters such as flowering time, type of flowers (single or double), colour of leaves,

size of leaves, plant size, growth habit (erect/prostrate), and scent (strong/weak). These differences, particularly in flowering time, create difficulties at harvest, resulting in yield and quality loss, since oregano is normally harvested at full bloom. Oregano is a cross-pollinated species, and hence not genetically uniform. Because of its genetic heterogeneity it is possible to select better genotypes for agronomic and quality traits. Twenty-one genotypes collected from various parts of Cyprus were multiplied vegetatively and planted at Saittas in replicated trials for evaluation. Seed of the best genotypes will be produced in isolation and will be bulked to produce a synthetic stock. The results so far showed that there are large differences in yield and other parameters among the various genotypes tested. Total dry matter yield (leaves and flowers) during the last two years (2000-2001) ranged from 4190 kg/ha to 8670 kg/ha, while the oil content ranged on average from 3.3% to 5.85%.

**Plant population density of oregano**

The aim of this experiment is to determine the optimum population density of oregano plants. The treatments comprised nine combinations of three row spacings (60, 90 and 120 cm) and three within-row spacings (30, 45 and 60 cm). Two experiments were established in spring 1997, one at Saittas, which lies at an altitude of approximately 600 m above sea level, and the other at Zygi, located at sea level. During the whole experimental period from 1997 to 2001 there were 8 cuts at Saittas and 6 at Zygi. The total dry matter yield (leaves and flowers) at Saittas and Zygi was 22 100 kg/ha and 9000 kg/ha, respectively. The highest yield at Zygi during the experimental period was obtained from plants spaced 60 x 30 cm giving 5.5 plants/$m^2$ (10900 kg/ha) and the lowest from plants spaced 90 x 60 cm giving 1.8 plants/$m^2$ (7450 kg/ha), SE ± 727.0. At Saittas there were also significant differences among treatments in the first three years, with the plant density of 5.5/$m^2$ being the best in 1997, while the 3.7/$m^2$ was best in 1998 and 1999. Thereafter, differences disappeared because the plants grew large enough to cover the whole plot area.

**Effect of cutting height on the yield of oregano**

Height of cut is important for yield and persistence. A tall stubble leaves a photosynthetic area that provides additional carbohydrates for regrowth after cutting. Also, by adjusting the cutting height it may be possible to get more cuts per year. Three cutting heights, 8, 13, and 18 cm from the soil surface were tested. Two experiments were established in spring 1997, one at Saittas and the other at Zygi. During the whole experimental period from 1997 to 2001 there were 8 cuts at Saittas and 6 at Zygi. The total dry matter yield of leaves and flowers over the experimental period at Saittas and Zygi was 24330 kg/ha and 10100 kg/ha, respectively.

Cutting at a height of 18 cm from the soil surface at Saittas produced the highest yield, 26 500 kg/ha whilst a height of 8 cm gave the lowest, 22 300 kg/ha. Cutting at 13 cm was intermediate. At Zygi there were no significant differences among the three cutting heights.

**Control of weeds**

Weeds are a serious problem in aromatic and medicinal crops especially where the crop is irrigated. In a series of preliminary experiments initiated in 1996 at the Agricultural Research Institute several pre-emergence herbicides were screened for their suitability and effectiveness in 15 aromatic plant species, i.e. *Foeniculum vulgare, Hyssopus officinalis*, *Laurus nobilis*, *Lavandula angustifolia*, *Melissa officinalis*, *Mentha piperita*, *M. viridis*, *Ocimum basilicum*, *Origanum dictamus*, *O. dubium*, *O. majorana*, *Rosmarinus officinalis*, *Salvia fruticosa*, *Sideritis scardica* and *Thymus vulgaris*. The herbicides selected were further tested in field trials. Trials that started in autumn 1996 with *Lavandula*, *Salvia* and *Origanum*, were completed in 1998. The results showed that among the residual herbicides tested, chlorthal dimethyl, oxadiazon

and oxyfluorfen, applied post-planting to crops at 7.5 kg, 0.75 kg and 0.75 kg a.i./ha. respectively, were suitable for the above three crops (Vouzounis 2000).

## Irrigation

- **Oregano**

Four amounts of water were tested with drip-irrigated Oregano at Zygi. Irrigation in 1999 started in mid-April, ending in mid-November, and was applied every two weeks early in the irrigation season and weekly during the summer and autumn months. The amounts of water tested were 165, 290, 415 and 540 mm, equivalent to 40, 70, 100 and 130% of the estimated irrigation requirement, respectively. The crop was harvested in late May, but due to high summer temperatures regrowth was prevented, irrespective of irrigation treatment. Total fresh yield (stems, leaves, and flowers), marketable air-dried yield (leaves and flowers), oil content, and oil yield increased with increasing amount of water applied up to 415 mm (Metochis 2000).

- **Sage**

Four amounts of water applied by drip irrigation were tested with sage, also at Zygi. Irrigation in 1999 started in mid-April ending in mid-November and was applied every 2 weeks early in the irrigation season and weekly during the summer and autumn months. The amounts of water tested were 125, 215, 310 and 405 mm, equivalent to 40, 70, 100 and 130% of the estimated irrigation requirement, respectively. The crop was harvested in late March, mid-June and early December. Annual fresh yield (stems, leaves, and flowers), marketable air-dried yield (leaves and flowers), and oil yield decreased when seasonal irrigation was less than 310 mm (Metochis 2000).

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## *Current status of the collection of aromatic, culinary and medicinal plants in the Czech Gene Bank in Olomouc*

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The genetic resources of aromatic, culinary and medicinal plants are maintained in the Gene Bank in Olomouc, which is now part of the Czech national genebank for agricultural crops. Since 1994 it has been, from an organization point of view, affiliated to the Research Institute of Crop Production (RICP) in Prague-Ruzyne. The bases of the germplasm collections were constituted at the Research Institute of Vegetables Growing and Breeding (RIVGB) in Olomouc, which was established in 1951 and ceased operating in 1994. The station in Olomouc now maintains genetic resources of all vegetable species usually grown in Central Europe (genera *Allium, Beta, Brassica, Capsicum, Cucumis, Cucurbita, Daucus, Lactuca, Lycopersicon,* etc.), and a wide range of aromatic, culinary and medicinal plants. Within the ECP/GR, the Gene Bank in Olomouc is responsible for holding the international field genebank for vegetatively propagated long-day *Allium* species.

Collecting and growing medicinal and culinary plants have a long tradition in the Czech Republic. These activities were supported by intensive breeding based on selection from rich collections of genetic resources. In spite of a relatively limited growing area (the biggest production areas are, with the exception of mountains, in Moravia, along the river Elbe, around the towns of Litoměřice, Teplice, Pardubice and Hradec Králové, and in South Bohemia), the importance of medicinal and culinary plants is based on their specific quality.

### The collection of aromatic, culinary and medicinal plants

This collection consists of 973 accessions representing 78 species, including 286 accessions that are vegetatively propagated and 687 propagated from seed. The species composition of this collection is shown in Table 1. New accessions are obtained from botanical gardens, private seed companies, research institutes and international and local collecting missions.

**Table 1.** Species composition of the aromatic, culinary and medicinal plants collection in Olomouc

| Species | No. of accessions |
|---|---|
| *Carum* L. | 277 |
| *Origanum* L. | 67 |
| *Ocimum* L. | 44 |
| *Satureja* L. | 42 |
| *Lavandula* L. | 41 |
| *Plantago* L. | 41 |
| *Salvia* L. | 37 |
| *Mentha* L. | 35 |
| *Calendula* L. | 34 |
| *Anethum* L. | 24 |
| *Althaea* L. | 22 |
| *Foeniculum vulgare* Miller | 19 |
| *Leonurus* L. | 19 |
| *Hyssopus* L. | 16 |
| *Thymus* L. | 14 |
| *Malva* L. | 13 |
| *Melissa* L. | 12 |
| *Ruta* L. | 12 |
| *Achillea* L. | 11 |
| *Hypericum* L. | 11 |
| *Verbascum* L. | 11 |
| Other species (*Agrimonia* L., *Anchusa* L., *Anthemis* L., *Valeriana* L., *Verbena* L.) | 171 |
| **Total** | **973** |

The aromatic, culinary and medicinal plant accessions of the Gene Bank Olomouc are grown in the field at Holice. Cross-pollinated species (e.g. *Satureja, Origanum, Carum*) are grown in isolation cages. Every year 50 accessions are regenerated on average. Regeneration of the collection was started in 1994. The whole collection will be regenerated during the next 6 years. The harvested seeds are stored in boxes at –20°C. Work on genetic resources complies with the rules of the National Programme on plant genetic resources conservation and utilization in the Czech Republic.

The evaluation of aromatic, culinary and medicinal plants includes a basic morphological description. Chemical analyses (essential oil content) are made on important species (e.g. *Carum, Lavandula, Origanum, Ocimum*).

Passport data are fully processed and computerized. Evaluation data are gradually recorded and the development of a suitable database is in progress.

## *Medicinal and aromatic plants in Estonia*

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### Introduction

The flora and vegetation of Estonia are both very interesting for their biological diversity. Despite its small surface area, Estonia has two different biogeographical subdivisions. Geologically, eastern Estonia can be characterized by its sandstone bedrock and western Estonia by its limestone bedrock. Therefore different soil types occur in these areas. The composition of plant communities of western Estonia is, in turn, under the influence of the more maritime climate. These conditions give rise to very different plant communities. About half of Estonia is covered by forest, over 20% by mires, and about 20% by meadows. The indigenous flora of Estonia includes approximately 1500 vascular plants. More than 500 plant species are situated on the margin of their distribution area: 121 on the northern, 128 on the northeastern, 45 on the eastern, 56 on the southeastern, 27 on the southern, 11 on the southwestern, 15 on the western and 52 on the northwestern margins.

### Legislation

The *Law of Natural Objects under Protection* (passed in 1994, supplemented in 1998) establishes the terms for the protection of both plant communities and plant species. Almost 10% of the country's area is subject to various levels of protection. The strict regime of protection applies to over 1% of the whole area, and is planned to cover up to 5% of the mainland by 2010. The above-mentioned law has established the following categories of protection:

- Conservation areas
  - national parks, meant for the protection of typical landscapes and the biological diversity of ecosystems (Lahemaa – northern Estonian plant communities; Karula – southern Estonian plant communities; Soomaa – the communities of mires and bogs; Vilsandi – littoral communities);
  - wildlife conservation areas, meant for the protection of rare species and species in danger of extinction;
  - landscape conservation areas (parks, arboreta, botanical gardens), usually small;
  - programmed areas, in which monitoring, research work and educational activities are carried out in accordance with the programme;
- Protected natural individual objects (live or lifeless natural objects of high scientific, historical or cultural value (e.g. single trees, rocks and stones, waterfalls, caves, etc.);
- Species, fossils and minerals under protection.

Endangered plant species are divided into three categories according to the protection regime:

1. The first category includes 22 protected species of high scientific value (relicts, species with a narrow area of distribution and those on the margin of an area), very rare plants (1-5 sites) and clearly endangered species (Kukk 1999).
2. The second category (145 species) consists of rare and endangered species and species of scientific value, for which the threat is not as acute as for the species in the first category. They include endemic species, relicts from earlier climatic periods or species situated on the margin of their distribution area. The following species used as medicinal plants belong to this category: *Taxus baccata* L., *Jovibarba globifera* (L.) J. Parnell (syn. *J. sobolifera* (Sims) Opiz), *Prunus spinosa* L., *Rubus arcticus* L., *Hedera helix* L., *Helichrysum arenarium* (L.) Moench and *Orchis* spp.

3. The third category of protected species covers 41 quite common species that are endangered for various reasons. These include many decorative, medicinal and edible plants. The following medicinal species are listed: *Huperzia selago* (L.) Bernh. ex Schrank. et Mart., *Lycopodium clavatum* L., *Daphne mezereum* L., *Myrica gale* L., *Allium ursinum* L., *Colchicum autumnale* L. and *Orchis* spp.

### *In situ* and *ex situ* conservation of medicinal and aromatic plants

Given the rather large coverage of protected areas in Estonia, the habitats of numerous medicinal plants enjoy a fairly good level of protection. In case of necessity it is possible, besides protecting the species under observation, to protect the habitat also by setting up small conservation areas, thus making the protection of plant communities considerably more effective.

Regarding *ex situ* conservation, the collection of medicinal and aromatic plants (MAPs) in the experimental garden of the Pharmacy Institute at the University of Tartu has a leading role. The collection is essential to the research and studies of the pharmacy students. Regrettably, an adequate MAP seed bank is still lacking. The botanical gardens of Tartu and Tallinn have organized to a certain degree the *ex situ* conservation of medicinal plants, kept in the Department of plant systematics, and a collection of useful plants. In the Nigula Wildlife Conservation area, *ex situ* conservation has been arranged for 760 indigenous forms of *Oxycoccus palustris* Pers. as well as for natural forms of *Vaccinium vitis-idaea* L. and *Rubus arcticus* L. at the University of Agriculture.

In Estonia, the following measures are taken for *in situ* and *ex situ* conservation:

- Further development and unification of laws;
- Land use restrictions in the areas under protection;
- Arranging support systems and paying compensation in the areas under protection;
- Claims for damages in case of violation of the law.

### Use of MAPs

Estonia has long traditions in using medicinal and aromatic plants. Even now one can find considerably more herbal drugs and tea blends in our chemists' shops than in most European countries. Some of the drugs are gathered in the wild. These include mainly common, widely spread species. Along with the restoration of land ownership within the last decade, a number of farmers have started growing indigenous MAP species. On the average, about ten species are grown but there are also farms where 30 or more species are grown. Statistical data about MAPs gathered in the wild and grown on-farm are lacking since no reporting or monitoring system has been established yet.

Regarding medicinal plants, thorough research was done in Estonia in the 1980s on the resources and sustainable use of two forest plants: *Arctostaphylos uva-ursi* (L.) Spreng. and *Vaccinium vitis-idaea* L. A possible annual use was calculated on the yearly gain in growth of the biomass that guaranteed a continuous renewal of resources. Based on the findings, distribution maps of the resources were drawn up.

### Conclusion

To summarize, owing to long-term traditions of nature conservation in Estonia, the status of *in situ* protection of MAPs is considerably better than that of *ex situ* conservation.

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## *Perspectives and achievements in genetic conservation of medicinal and aromatic plants in Hungary*

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### Tradition and future of the utilization of medicinal plants growing wild in Hungary

The first reports of collection and small-scale cultivation of medicinal and aromatic plants (MAPs) in Hungary are found in the books written by monks settled in the country in the Middle Ages, arriving from the Mediterranean regions (Italy, France). Furthermore many medicinal and aromatic plants, especially members of the Lamiaceae family, were introduced into the Carpathian valley in these early times. However, till the end of the 19th century, MAP production was carried out on the "family scale" only. Intensification of production (both collecting and cultivation) started in the first years of the 20th century, when the processing of the plant raw material including oil distillation also started. In spite of the political and economic strife of the former century the MAP sector became a successful branch of the Hungarian economy (Bernáth 1994). Some of the national products made from either collected or cultivated species became known as special Hungarian products ("Hungaricum") highly valued on the world market (*Chamomillae flos*, *Basilici herba* and *folium*).

When evaluating the future of the worldwide utilization of natural habitats it should be noted that about 90% of the 1200-1300 MAP species which are sold in EU countries are of wild origin and originate in developing countries (Lange 1996). In Hungary about 30-35% of the total MAP production is based on the natural flora. This means that about 10 000-15 000 t dry biomass is produced from the wild, from about 120-130 different species (Németh 1997). This places Hungary in an intermediate position considering the European situation as a whole. In western European countries there is practically no collecting, while in eastern and southern Europe the proportion of drugs gathered in the wild often reaches 100%. International data show that for instance in England collecting is restricted to occasional self-consumption, while in Spain about 50% and in Albania 100% of the total drug production comes from the wild. A feature of the Hungarian MAP sector is that the spectrum of collected species is rather wide. In most countries the collecting of some particular species dominates: *Thymus* in Spain, *Origanum* in Turkey and Italy, while collecting of *Salvia* and *Tillia* spp. in Albania provides an important export crop. According to statistical data the number of medicinal plants collected in Hungary and either exported or found on the domestic market is relatively high (Box 1).

The worldwide reduction in the number of collected MAPs drew attention to the fact that the damage to plant communities over the "theoretical losses" could result in a large decrease in profit. The protection of many territories used for collecting MAPs in the past, as well as the extension of the protection to individual species resulted in an enormous limitation in MAP production. The importance of MAP preservation was recognized by international organizations as well as Hungarian authorities. It was recognized that the protection and development of plant communities based on scientific analysis and activity, including gene conservation, can contribute to sustaining or even increasing the production capacity of the indigenous flora.

**Box 1.** List of medicinal and aromatic plants of great importance collected in Hungary

**Medicinal plant drugs produced in natural ecosystems (collected species)**

***Achilleae* herba***
*Anserinae* herba
*Apii* herba
*Asarae* herba cum radix
*Asperulae* herba
***Bardanae* radix***
*Betonicae* folium, herba
*Betulae* folium
*Bursae pastoris* herba
*Calcatrippae* flos, herba
*Cerefolii* herba
***Chelidonii* herba***
*Cichorii* herba, radix
*Clematitis* herba
*Coryli* folium
***Crataegi* fructus, summitas***
*Cynodon dactylon* rhizome
***Cynosbati* fructus***
*Echii* herba
***Equiseti* herba***
*Eupatorii* herba
*Euphrasiae* herba
*Fagopyri* herba
*Farfarae* flos, folium
*Filipendulae* herba
*Fragariae* folium
*Frangulae* cortex
*Fraxini* folium
*Fumariae* herba
*Galegae* herba
*Galeopsidis* herba
*Galli* herba
*Gei* rhizome
*Glandes quercus* testae pulvis
*Graminis* rhizome
*Hederae* herba
*Helianthemi* herba
*Herniariae* herba
***Juniperi* fructus***
*Lamii* albi flos, herba
*Lepidii* herba
*Linariae* herba
*Menthae aquaticae* herba
*Mori* folium
*Myrtilli* folium, fructus
*Ononidis* radix
*Papaveris rhoeas* flos
*Plantaginis lanceolatae* folium
*Plantaginis majoris* folium
*Polygalae* herba
*Polygoni* herba
*Polygoni hydropiperis* herba
*Populi* gemma
*Primulae* flos, folium, radix
*Pruni* spinosae flos, fructus
*Pulmonariae* folium
*Quercus cerris* folium, lichen
*Rhei* rhizome
*Robiniae* flos
*Salicis* cortex
***Sambuci* flos*,** fructus
*Sedi* herba
*Serpylli* herba
*Sideritidis* herba
***Solidaginis* herba***
*Sorbi aucupariae* fructus
*Stellariae* herba
***Taraxaci* folium, herba, radix***
***Tiliae* flos, folium***
*Tormentillae* rhizome
***Urticae* folium***
*Verbenae* herba
*Veronicae* herba
*Vincae* herba
*Violae odoratae* folium
*Violae tricoloris* herba
*Virgae aureae solidaginis* herba
***Visci* stipes***

**Medicinal plant drugs produced in both natural and agrarian systems (collected and/or cultivated species)**

*Absinthii* herba
*Agrimoniae* herba
*Althaeae* flos, folium, radix
*Armoraciae* radix
*Amygdalae* dulcis farina
*Centauri* herba
***Chamomillae* flos***
*Cotinius* folium
***Hyperici* herba***
*Inulae* radix
*Leonurii cardiacae* herba
*Malvae silvestris* flos, folium
*Marrubii* herba
*Meliloti* flos, herba
*Millefolii* flos, herba
*Origani* herba
*Saponariae albae*
*Saponariae officinalis* herba, radix
*Tanaceti* flos, herba
*Verbasci* flos

* = about 10–500 tonnes collected per year

**Regions of Hungary specialized in MAP collecting and/or cultivation**

The traditional regions of utilization of the indigenous flora and the main cultivation areas of MAPs are shown in Fig. 1.

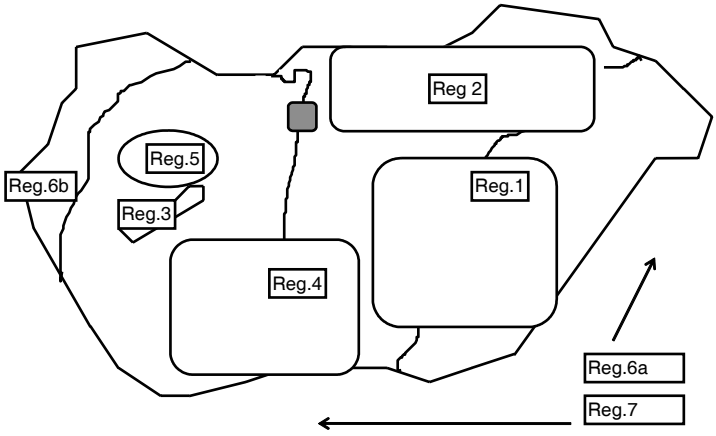


**Fig. 1.** Traditional regions for utilization of indigenous flora and main cultivation areas of medicinal and aromatic plants in Hungary:

Reg. 1 = Great Plain and East Tisza river region
Reg. 2 = North-central mountainous region of Hungary
Reg. 3 = Balaton highland
Reg. 4 = South Hungary
Reg. 5 = Bakony and its surroundings
Reg. 6a = Region suitable for "spring poppy" cultivation
Reg. 6b = Northern part of Transdanubia suitable for "autumn poppy" cultivation
Reg. 7 = Region of plant species which can be cultivated all over the country

- **Traditional regions of utilization of the indigenous flora**

The regional specialization in Hungary for utilization of the indigenous flora dates back to the beginnings of the 1920s. This specialization took place spontaneously in response to various biological, economical and social factors (Bernáth 1994). As a result of spontaneous specialization during the past 80-90 years, well-defined production areas have appeared, which have had a great influence on the effectiveness of the medicinal plant sector in the past and may also affect its future development.

- **Region 1**

The natural occurrence of *Matricaria recutita* in Hungary is one of the model examples illustrating the regional specialization for utilization of species of the indigenous flora (Fig. 1). From a biological point of view, chamomile grown in saline habitats was regarded as high quality. This was first based on empirical observations and later confirmed by sophisticated chemical and pharmacological analysis. From a socioeconomical point of view the development of this region was promoted by the abundance of labour locally. According to the data of trading companies, as many as 15 000-20 000 people are involved in gathering chamomile flowers at harvest time, even today. Because of the short duration of the chamomile harvest and processing, the regional activity had to be complemented by utilization of other MAPs indigenous to the region (*Juniperus communis, Achillea millefolium, Gypsophila paniculata, Crataegus* spp., *Rosa* spp., etc.).

- **Region 2**

The development of special regions for utilization of indigenous plant flora was obvious in the north-central mountainous region of Hungary (Fig. 1) since the beginning of the 20th century. In the production areas of *Rosa* spp., *Sambucus nigra, Prunus spinosa, Crataegus* spp., etc., buying arrangements and centralized processing facilities were installed. Examples of centres of MAP production in the northern part of Hungary include Balassagyarmat, Pásztó, Veresegyháza and other towns and villages.

- **Specialization of cultivation areas**

In the development of the specialized cultivation areas for MAPs, beside traditional and accidental elements the ecological and economical considerations became ever more important.

- **Region 3**

The region for lavender cultivation (*Lavandula angustifolia* and *L. intermedia*) was created in the 1930s mostly based on ecological considerations. Suitable ecological conditions had to be chosen for the cultivation of species of Mediterranean origin (Németh 1996). Plantations were established on the southern slopes of the Tihany Peninsula and in the neighbouring territories (Balatonakali, Darányipuszta, etc.) where the ecological conditions show some similarity to the Mediterranean. The importance of lavender plantations has decreased in recent decades; however a relict of the first plantation still exists in the Tihany Peninsula.

- **Region 4**

Both ecological and economical considerations led to specialization in marjoram (*Origanum majoranna*) and basil (*Ocimum basilicum*) cultivation in the region. The climate of Region 4, situated in the southern part of Hungary, is relatively warm and receives appropriate sunshine. Its climate meets the ecological requirements of species of Mediterranean and subtropical origin. From an economical point of view, this area is also known as a major cultivation area for red pepper. The drying and postharvest processing of red pepper and medicinal and aromatic plant/spices are very similar. This means that the available technology and processing facilities located in the region can be utilized for both plant groups.

- **Region 5**

The introduction of the cultivation of ergot (*Claviceps purpurea*) in this area was motivated by both ecological and economical considerations. From the wide-ranging cultivation areas of the host plant (*Secale cereale*), a special area possessing suitable ecological conditions for the development of fungi had to be chosen. As a result of compromise the southwestern slopes of Bakony Mountain were selected for the purpose. The cultivation area has since been equipped with valuable cultivation and postharvest technological tools.

- **Region 6**

The cultivation of poppy (*Papaver somniferum*) has a great tradition in Hungary owing to its wide-scale use as both an important food plant and a source of oil and industrial raw material. Its large-scale cultivation is organized and controlled by the pharmaceutical factory "Alkaloida". The poppy cultivation region can be divided into two sub-regions. The cultivation of spring-sown varieties is concentrated in the Great Plain of Hungary and some parts of the northwestern regions of the country (Region 6a), while autumn-sown varieties, because of their susceptibility to winter damage, are situated in the western part of Hungary (Region 6b) where winters are usually much milder and snow covers the fields more frequently.

- **Species cultivated and collected country-wide**

- **Region 7**

Several MAP species can be cultivated and/or collected in Hungary without much restriction. Such cultivated species include members of the Apiaceae family (*Foeniculum vulgare, Carum carvi, Anethum graveolens, Coriandrum sativum, Pimpinella anisum,* etc.), mustard (*Sinapis* and *Brassica* spp.), *Silybum marianum, Cucurbita* spp., and among collected plants, examples include *Sambucus nigra*, *Rosa canina*, *Equisetum arvense*, *Crataegus* spp., etc.

### Preservation of Hungarian MAP production potential and biodiversity

Even today, a considerable proportion of the medicinal and aromatic plant drugs produced and sold by the Hungarian MAP sector comes from indigenous sources. This exploitation of natural plant populations without scientific analysis and control may reduce the productivity of natural plant populations and the biodiversity of the Hungarian flora. The analysis of the Hungarian situation shows that the ratio of MAPs is high among the 400-plus species that are legally protected in Hungary. The reduction of the local medicinal plant spectrum is shown, however, by the fact that 17 species which were recommended for collecting on a large scale and used as herbs and extracts in 1948 and in 1961 had to be removed from the production list. The danger is more obvious if we take into consideration the list of protected species which can be used as MAPs according to various literature references (Box 2).

**Box 2.** List of the Hungarian protected species which are used as medicinal and aromatic plants according to literature references

| | |
|---|---|
| *Achillea crithmifolia* | *Alchemilla crinita* |
| *Achillea ptarmica* | *Alkanna tinctoria* |
| *Acorus calamus* | *Armoracia macrocarpa* |
| *Adonis vernalis* | *Arnica montana* |
| *Betula pubescens* | *Carlina acaulis* |
| *Colchicum arenarium* | *Gentiana asclepiadea* |
| *Dictamnus albus* | *Gentiana cruciata* |
| *Equisetum hyemale* | *Gentiana pneumonanthe* |
| *Helichrysum arenarium* | *Globularia cordifolia* |
| *Helleborus purpurascens* | *Hippophae rhamnoides* |
| *Hepatica nobilis* | *Hypericum elegans* |
| *Inula helenium* | *Hypericum maculatum* |
| *Iris pumila* | *Isatis tinctoria* |
| *Lycopodium clavatum* | *Petasites albus* |
| *Menyanthes trifoliate* | *Peucedanum officinale* |
| *Nigricans* | *Phyllitis scolopendrium* |
| *Orchis morio* | *Plantago maxima* |
| *Polygonum bistorta* | *Polygala major* |
| *Primula elatior* | *Ruscus hypoglossum* |
| *Primula vulgaris* | *Scopolia carniolica* |
| *Quercus farnetto* | *Sempervivum marmoreum* |
| *Ribes nigrum* | *Sempervivum tectorum* |
| *Rosa pendulina* | *Tamus communis* |
| *Ruscus aculeatus* | *Taxus baccata* |
| *Vaccinium vitis-idaea* | *Urtica kioviensis* |
| *Valeriana sambucifolia* | *Vitis sylvestris* |

- **Factors playing a role in the decrease of the Hungarian MAP production potential and biodiversity**

The scientific study of the Hungarian indigenous flora has a long tradition. However the detailed study of populations utilized as MAPs (coenological, chemical and yield characters was undertaken only recently, in the early 1960s. This seems to be a very important phenomenon, especially in the case of medicinal plants which became protected or endangered. On the basis of recent investigations, the factors having an adverse effect on MAP productivity can be divided into two main groups:

**1. Direct factors limiting the spectrum and productivity of indigenous MAP populations**

- Overexploitation of MAP populations by intensive and inconsiderate collecting may result in irreversible damage. Owing to profit seeking and lack of scientific control, more and more species have to be included in the list of endangered species. Examples include *Adonis vernalis*, *Primula* spp. and *Dictamnus albus,* which became protected due to their unregulated exploitation from the wild.
- The overall reduction of forest areas resulting from progressive industrialization limits both the spectrum and productivity of MAP populations. The decrease in the populations of *Dryopteris filix-mas*, *Crataegus nigra*, *Primula vulgaris, Veronica officinalis,* etc., can be explained by this phenomenon.
- Drainage of marshy-moist ecosystems as a result of the political decision of the former socialist administration (especially in the 1950s) has had an adverse effect on the medicinal plant populations after many years. The most valuable species in this respect, which have had to be removed from the list of utilized plants, are *Acorus calamus* and *Menianthes trifoliata*.
- There are huge losses to former ruderal MAP populations, gathered from cultivated field headlands, meadows, farmyards, etc. The biodiversity of these systems has been reduced drastically. Species which were collected from these habitats on a large scale, such as *Marrubium vulgare* and *Fumaria schleicheri,* have become very rare and botanists consider there is a need for their protection.
- The natural distribution of MAPs is limited by pollution. Because of the increasing quality requirements, the production area has to be farther from the main roads, industrial facilities, etc. This means that more and more territories are withdrawn from the production of medicinal and aromatic plants. Some species, especially *Arnica montana* and *Vaccinium* spp., are very susceptible to pollution.

**2. Indirect factors limiting the spectrum and productivity of indigenous MAP populations**

The overall reduction in the species range in natural plant communities, which is a general phenomenon throughout Europe (plants becoming endangered, rare or extinct) is also observable in Hungary. Medicinal and aromatic plants, as constituents of the different ecosystems, are affected by any factor that damages plants generally. The severe reduction of biodiversity affects all Europe and is reflected in the large number of species now extinct (Bernáth 1988). This reduction also means the extinction of some chemotaxa existing in given natural systems, which may have had potential therapeutic use but are lost forever.

Several indirect factors such as industrial pollution, increasing amount of agrochemicals, harmful human influences, etc., damage the natural ecosystems continuously, including plant species utilized as MAPs.

- **Efforts made to preserve the MAP spectrum in the country and sustain their production capacity (*ex situ* and *in situ*)**

The importance of MAP preservation was recognized by the scientists, representatives of Hungarian authorities and drug producers, and concrete steps were taken recently to increase the effectiveness of protection. To harmonize and distribute the government funds allocated to MAP conservation in Hungary a Central Medicinal Conservation Board was established within the Ministry of Agriculture and Rural Development. The experts of the Department of Medicinal and Aromatic Plants of SZI University in Budapest head the Board.

**1. *In situ* conservation of MAPs**

- **Complex protection**

The complex protection of particular geographical regions or areas regulated by administrative measures is an indirect form of MAP conservation. The Hungarian protected areas that are important from the point of view of MAPs are listed in Table 1. However, the MAPs grown in these areas (with some exceptions) cannot be used as a source of raw material. Their plant spectrum and chemical diversity constitutes a source for scientific studies and further development.

**Table 1.** List of Hungarian conservation areas with potential importance for MAPs

| Name | Type[1] |
|---|---|
| Adony | PA |
| Aggtelek* | NP |
| Ágotapuszta | PA |
| Alcsút - arboretum | PA |
| Ásotthalom - marshland | PA |
| Aszófõ - forest | PA |
| Badacsony | PR |
| Baktalorándháza - forest | PA |
| Baláta-tó, Szente | PA |
| Balatonfüred - forest | PA |
| Barcs | PR |
| Bátorliget - pasture | PA |
| Bátorliget - marshland | PA |
| Béda-karapancsa | PR |
| Bélmegyer - forest-desert | PA |
| Bihar - pasture | PA |
| Biharugra | PR |
| Bikács, Ökör - hill | PA |
| Bodrogszeg, castle - hill | PA |
| Boronka region | PR |
| Borsod - fields | PR |
| Börzsöny - hill* | PR |
| Buda, Sas - hill | PA |
| Budapest - botanical garden ** | PA |
| Bükk* | NP |
| Cegléd - meadow | PA |
| Császártöltés - red marshland | PA |
| Csévharaszt - *Juniperus* pop. ** | PA |
| Csokonyavisonta - woody pasture | PA |
| Dabas | PA |
| Dámányadacs - meadow | PA |
| Darvas Lake (Sümeg) | PA |
| Dévaványa | PR |
| Devecser, Szék - forest | PA |
| Dinnyés, Fertô | PA |
| Lankóc - forest | PA |
| Láz-Horst | PR |
| Lesencetomaj | PA |
| Magas-Bakony | PR |
| Makó-landor - forest | PA |
| Mártély | PR |
| Mátra* | PR |

| Name | Type[1] |
|---|---|
| Csorvás - *Adonis* pop. | PA |
| Erdôbényei - woody pasture | PA |
| Erdôtelek – arboretum | PA |
| Fényi – forest | PA |
| Fertô - lake | NP |
| Fóti Somlyó** | PA |
| Füzérradvány – garden | PA |
| Gemenc | PR |
| Gerencse | PR |
| Gödöllô - ridge of hills | PR |
| Gyöngyös, Sár – hill | PA |
| Hajdúság - woodydeserts | PR |
| Hajós | PA |
| Hanság | PR |
| Hencida - *Quercus* forest | PA |
| Hortobágy* | NP |
| Jakab hegy, Kôvágószôlôs | PA |
| Jeli - arboretum** | PA |
| Káli - basin | PR |
| Kámon - arboretum | PA |
| Karancs-Medves | PR |
| Kardoskút - "white" lake | PA |
| Kecsker - pasture | PA |
| Kelemér, Mohos - lakes | PA |
| Kéleshalom - sand hills | PA |
| Kelet-Cserhát | PR |
| Kelet-Mecsek* | PR |
| Kerecsend - forest | PA |
| Kesznyéteni | PR |
| Keszthely | PR |
| Kis-Balaton | PR |
| Kiskunság* | NP |
| Kistápé - marshland | PA |
| Kôszeg | PR |
| Közép-Tisza* | PR |
| Szársomlyó, Nagyharsány | PA |
| Szarvas - arboretum** | PA |
| Szatmár-Bereg | PR |
| Szedres - *Crocus* pop. ** | PA |
| Szeleste - arboretum | PA |
| Szendrôlád - marshland | PA |
| Szentgál - *Taxus* pop. ** | PA |

**Table 1 (cont.).** List of Hungarian conservation areas with potential importance for MAPs

| Name | Type[1] |
|---|---|
| Melegmány völgy, Mánfa | PA |
| Nagycenk - *Tilia* pop. | PA |
| Nagyerdö, Debrecen - *Quercus* pop. | PA |
| Némethkér, Látó - hill | PA |
| Ócsa | PR |
| Ohat - forest | PA |
| Ôrség | PR |
| Ôrtilos, Szent Mihály - hill | PA |
| Pannonhalma - arboretum | PA |
| Pilis | PR |
| Pitvaros - pastures | PR |
| Pusztakócs - marshland | PA |
| Rinyaszentkirály - forest | PA |
| Ság-hegy | PR |
| Sárrét | PR |
| Sárvár - arboretum | PA |
| Somogyvár, Kapuvár - hill | PA |
| Sopron - Botanical Garden | PA |
| Sopron | PR |
| Sümeg, Mogyorós-Domb | PA |
| Szabadkígyós | PR |
| Szakadát, pasture | PA |
| Szaporca, Ó-Dráva - river bed | PA |

| Name | Type[1] |
|---|---|
| Szentgyörgyvölgy | PR |
| Szigetköz | PR |
| Szöllôskert - forest | PA |
| Tállya, Patócs - hill | PA |
| Tapolcafô - marshland | PA |
| Tatársánc - grassland, Pusztaföldvár | PA |
| Tátika - *Fagus* pop. | PA |
| Tihany ** | PR |
| Tiszadorogma, Göbe - forest | PA |
| Tiszaigar, arboretum | PA |
| Tiszavasvári, white-saline | PA |
| Tokaj-Bodrogzug | PR |
| Tura - pasture | PA |
| Újszentmargita - forest | PA |
| Úrkút - karst | PA |
| Uzsai - *Calluna* pop. | PA |
| Vácrátót - botanical garden ** | PA |
| Vajdalapos - forest | PA |
| Vértes | PR |
| Zákány, *Quercus* pop. | PA |
| Zemplén* | PR |
| Zirc - arboretum ** | PA |
| Zselic | PR |

[1] Type: NP = National Park, PR = Protected Region, PA = Protected Area
* = Conservation areas of main importance from the point of view of production
**= Conservation areas of main importance from the point of view of conservation

- **Controlled utilization of MAP species**

There are possibilities for the controlled utilization of MAP species grown in special protected areas. Model examples are the collections of *Juniperus communis* in the Kiskunság National Park and the collection of *Matricaria recutita* in Hortobágy National Park. These species are very abundant in these regions. The well organized collecting of the plants controlled by scientists and experts results in no harm to the plant populations and even contribute to a better knowledge of these *Juniperus* and *Matricaria* populations, which is the basis for further development and increased production. Another advantage of this method is that the plant drugs coming from such areas are very likely to be free of pollutants. This type of collection seems to be profitable from both biological and economic points of view and ought to be extended to other MAP species.

- **Protection of individual plants**

Protection of individual plants is used in the *in situ* conservation of MAP species in Hungary. However its effectiveness greatly depends on the actual biodiversity of the species existing at the time of protection. The plants can be divided into three groups according to their natural biodiversity status:

a. Individual protection of MAPs which have become extinct or extremely rare is only of limited practical importance. Some representatives of this group include *Arnica montana, Digitalis lanata, Digitalis ferruginea,* etc. The chemical, morphological and biological product diversity of these species is probably lost. There is little hope for their utilization as a genetic source for further development of the MAP sector.
b. A number of protected MAPs are widely studied for their chemical and biological characters in Hungary. The main representatives of this group include *Achillea* spp., *Adonis vernalis, Dictamnus albus, Primula* spp. and *Pulsatilla* spp. Many of these species show high chemical and biological diversity which can be utilized for the development of artificial production models or later on for cultivation. The simple protection of these

species hardly meets the theoretical and practical expectations. The different bio- and chemotypes must be preserved by other methods of plant conservation.

c. Only very limited information is available for the majority of MAP species under protection. Thus, for this group, protection is only an administrative measure. Again, this simple protection hardly meets the requirements of real plant conservation. The intensification of scientific activities in this respect appears necessary. The biological and chemical diversity of the species listed in this group could be utilized through international cooperation projects.

- **Monitoring of the abundance or extinction of MAPs**

Monitoring of the abundance or extinction of MAPs or their populations appears as a very important field of scientific activity related to MAP conservation. The changes occurring in the MAP populations must be monitored and predicted. The restriction of the collection of plant material from local sites should not be deferred until the populations become endangered. The stability of the natural populations should be guaranteed on the basis of scientific data. Model investigations in this field are carried out very efficiently in the following Hungarian research institutes:

- SZI University, Department of Medicinal and Aromatic Plants, Villányi str. 29-45, 1118 Budapest
- Research Institute for Medicinal Plants, Lupaszigeti str. 4, 2011 Budakalász
- Ecological and Botanical Research Institute of the Hungarian Academy of Science, Alkotmány str. 2-6, 2163 Vácrátot

Scientific activities in this area require greater international cooperation and funds allocated from international sources.

**2. *Ex situ* conservation of MAPs**

In addition to the protection of genetic resources, *ex situ* conservation of MAPs may contribute to the construction of a valuable genepool which serves as a reserve for the development of highly productive cultivars or plants possessing diverse chemical characters. The distinction of well defined chemical types seems to have both theoretical and practical importance. It is well known that the need for chemical compounds varies with time. In the case of poppy (*Papaver somniferum*) the demand for morphine, codeine or thebaine changes unexpectedly. The need for different chemotypes of *Matricaria recutita* also varies from time to time. In some cases the most recent pharmacological studies generate new requirements, such as requirement for low beta-thujone taxa of *Salvia officinalis*, or pyrrolisidine-free populations of Scrophulariaceae and Asteraceae species.

- **Chemotaxonomical gardens**

The simplest way of *ex situ* conservation of medicinal and aromatic plants is the establishment and management of chemotaxonomical gardens. There are two of these gardens specialized in MAPs in Hungary:

- Chemotaxonomical Garden of the Research Institute of Medicinal Plants, Budakalász (address as above) - based on the Dahlgren system (Dahlgren et al. 1981)
- Medicinal and Aromatic Plant Garden of the Department of Medicinal and Aromatic Plants, Soroksár (address as above)

- **MAP genebanks**

In Hungary, the conservation of seed material of MAP species started at the beginning of the 1980s (Tóth and Németh 1996). The following two genebanks are specialized in MAPs:

- Genebank of the Research Institute of Medicinal Plants, Budakalász (address as above)
- Genebank of the Department of Medicinal and Aromatic Plants, Soroksár (address as above)

Genetic conservation of MAPs is a difficult task compared to that of the agricultural crops cultivated on a large scale throughout the world. Many difficulties arise from the large number of species, varieties and chemotypes which are utilized in practice. The main factors making gene conservation work difficult can be summarized as follows:

- The number of MAP species in Hungary is large (ca. 200), while the number of chemotaxa with potential theoretical and practical importance is about ten times greater.
- The majority of medicinal plants are found at a very low level of domestication. Knowledge regarding their biological and chemical properties is lacking.
- There is no reliable information on MAP genetic conservation methods.
- The genetic conservation of medicinal plants needs to be accompanied by expensive chemical analyses.

As a result of the efforts of the above-mentioned two institutions, 1500 accessions of MAP seed are available in Hungary, including seeds of wild populations, chemotaxa, cultivated populations and registered cultivars.

- **Biotechnology**

Advanced biotechnological methods are applied for the conservation of MAP species and practised in both above-mentioned institutions. This is of great importance in the case of MAPs which cannot be propagated from seed or in the case of species whose chemical characteristics can be preserved only by clonal propagation. Microclonal propagation and biotechnological conservation methods have been developed for the following species: *Artemisia dracunculus, Lavandula intermedia, Melissa officinalis, Mentha piperita, Salvia officinalis, Sempervivum tectorum* and *Tanacetum parthenium.*

- **Introduction of protected and endangered MAPs into cultivation**

From a practical point of view, this seems to be a most effective *ex situ* conservation method (Bernáth 1992, 1993). It involves complex biological and chemical studies aimed at determining the ecological requirements, biological and chemical diversity as well as effective propagation methods of the given species. Achievements from Hungarian experts in this field are summarized in Table 2.

**Table 2.** List of protected and endangered MAPs introduced or being introduced into cultivation

| Species | Reason for conservation | Cultivation status |
|---|---|---|
| *Adonis vernalis* | protected | in progress |
| *Achillea crithmifolia* | protected | in progress |
| *Alkanna tinctoria* | protected | cultivated |
| *Arnica montana* | protected | cultivated |
| *Dictamnus albus* | protected | in progress |
| *Digitalis ferruginea* | protected to an increased degree | in progress |
| *Digitalis lanata* | protected to an increased degree | cultivated |
| *Equisetum arvense* | alkaloid-free populations | in progress |
| *Glycyrrhiza glabra* | endangered | cultivated |
| *Inula helenium* | protected | cultivated |
| *Primula vulgaris* | protected | in progress |
| *Isatis tinctoria* | protected | in progress |
| *Marrubium vulgare* | endangered | cultivated |
| *Sempervivum tectorum* | protected | cultivated |
| *Solidago virgaurea* | endangered | in progress |
| *Taxus baccata* | protected | Cultivated |
| *Valeriana sambucifolia* | protected | Cultivated |

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## *Medicinal and aromatic plants in the Israeli Gene Bank (IGB)*

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### Introduction

Israel is located at the meeting point of three phytogeographical regions: Mediterranean, Irano-Turanian and Saharo-Arabian. Thus it represents all three regions, with the consequence of profuse richness of plant species and varieties. Moreover, Israel is part of the Fertile Crescent and is located on the border between desert and temperate regions, where domestication of Old World crop plants began. Therefore, quite a number of its native species are wild relatives, feral derivatives or even direct ancestors of cultivated plants. In addition, other local species may have an untapped economic potential.

The Israeli Gene Bank (IGB) was established in 1979 and affiliated to two Ministries: the Ministry of Science (MOS) and the Ministry of Agriculture (MOA). The structural functions of the IGB are shown in Fig. 1. The headquarters are at Bet-Dagan (Volcani Center).

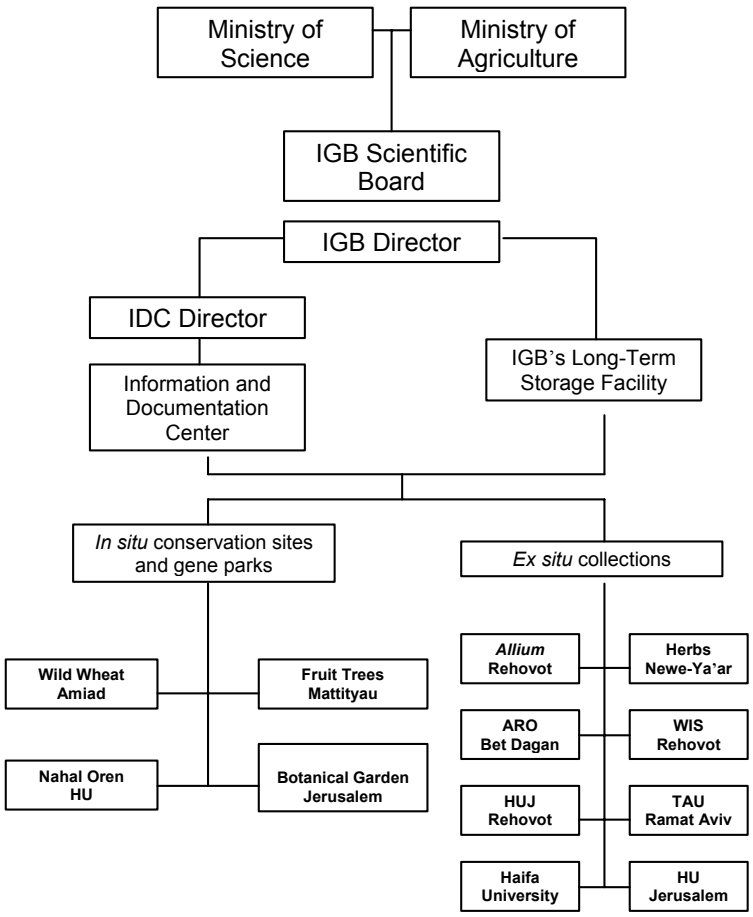



**Fig. 1.** Structure of the Israeli Gene Bank (IBG) for Agricultural Crops.

Today we face various types of immediate threats to the Israeli genepool:
- rapid increase in the human population and with it increased urbanization;
- road construction, industrial parks and military installations;
- pollution resulting from industry, traffic, sewage, agricultural fertilizers, etc.;
- farming activities such as general overgrazing, goat grazing, stubble burning, etc.;
- forestry activities and fire;
- collecting of plants from nature;
- tourism;
- gene contamination through introduction of new cultivars.

The IGB is responsible for the collection, preservation, documentation and practical evaluation of genetic resources of crop plants and their relatives across Israel; it holds in its central storage facilities some 16 000 accessions of indigenous species, landraces, agricultural and horticultural crops and their germplasm.

The Information and Documentation Center (IDC) at the IGB is responsible for the documentation of genetic resource holdings in all genebank collections throughout Israel, and for maintenance of the database and the information network.

Other activities of the IGB include: national and international exchange of plant material; promotion of national and international collaboration and coordination; organization of and participation in workshops, conferences and training activities; and support and guidance of research on genetic resources.

Today, the IGB also focuses on the collection and preservation of the genetic diversity of plants that are direct ancestors, wild relatives or landraces of domesticated plants which are endemic to our region. The establishment of a central modern seed bank as a long-term storage facility is in process (investment of more than US$ 2.5 million), and it will start to function in late 2004.

Until the IGB's new facility is finished, each specialist will be responsible for his own collection. Medicinal and aromatic plants (MAPs) in Israel are handled by a group of specialists located at the Newe-Ya'ar Research Center, where seeds and living plants are preserved (*ex situ*) and include all aromatic and some medicinal plants (Fig. 1).

## MAPs in Israel

All the threats that endanger any wild plants in Israel also endanger MAPs, especially the threat of collecting for dietary purposes. In Arabic culture, plants like za'atar (*Origanum syricum*) or three-leaved sage (*Salvia fruticosa*) are part of the diet. Therefore in 1956 a law was adopted (*The Plant Protection Law*) which made the gathering of certain protected plants illegal. Besides this law, some areas were designated as "National Park" and/or "Reserve Area" in which no grazing or cutting is permitted.

Special permission allows collecting of plants and seeds from all the wild MAP populations in Israel. Throughout the last 20 years or so, *ex situ* collections of some MAP species have been established. Table 1 lists part of these collections (more than 15 different species including 584 accessions), all growing at the Newe-Ya'ar Research Center, in the north of Israel. Besides these living plants we hold a large collection of MAP seeds, collected from wild populations in Israel and abroad, and seeds that are collected regularly from the *ex situ* plants at Newe-Ya'ar.

**Table 1.**Some examples of the MAP collection at Newe-Ya'ar and *in situ* in Israel

| | No. of accessions | | |
|---|---|---|---|
| **Species** | ***In situ*** | ***Ex situ*** | **Seed collection** |
| *Coridothymus capitatus* | 14 | 21 | 35 |
| *Foeniculum vulgare* | 17 | 5 | 25 |
| *Melissa officinalis* | 19 | 11 | 28 |
| *Mentha* sp. | 25 | 17 | 9 |
| *Micromeria fruticosa* | 17 | 17 | 22 |
| *Origanum* sp. | 18 | 97 | 115 |
| *Marjoram hortensis* | - | 15 | 35 |
| *Rosmarinus officinalis* | - | 48 | 10 |
| *Salvia officinalis* | - | 78 | 96 |
| *Salvia fruticosa* | 15 | 6 | 32 |
| *Satureja thymbra* | 12 | 28 | 37 |
| *Thymus vulgaris* | - | 141 | 75 |
| *Achillea fragrantissima* | 12 | 8 | 15 |
| *Achillea* sp. | 14 | 15 | 12 |
| *Artemisia* sp. | 34 | 77 | 30 |
| **Total** | **197** | **584** | **576** |

National competitive budgets received annually from the MOS and the MOA are applied to specific species by the IGB. For example national surveys have been conducted, covering *Foeniculum vulgare*, *Micromeria futicosa*, *Satureja thymbra* and other wild species.

For each species, after herbarium specimens had been studied and documentation prepared, physical searches were conducted in order to locate the largest wild populations covering the full range of variation, with primary attention to geographic and climatic variation (ecotypes). Then, from each of the populations that were selected, all morphological, phenological and chemical information was collected. Some of the plants were transferred to Newe-Ya'ar, mainly as cuttings but sometimes by removing the whole plant. The *ex situ* plants are compared with the *in situ* plants that grow in the wild, with respect to all the information mentioned above. Seeds were collected from the *in situ* and from the *ex situ* plants for at least two successive years.

Thus, from the living collections, as well as from *in situ* sites, we have obtained and hold data on morphological characters, flowering behaviour, harvest recovery and essential oil content and composition. This information is unique and helps us in our selection programme, in cooperation with other scientists, and as a link to the industry. The reports that are submitted to the IGB are in the public domain.

## MAP production

Israel produces MAPs on a commercial scale, including the various products listed in Table 2. Each group of products involves a great number of species and varieties. For example, we grow more than 42 species for fresh herb production (about 100 varieties) and more than 30 species for ornamental purposes (more than 80 varieties). Some of the varieties that are used commercially are cultivars but some are unregistered varieties that were brought, by us, directly from the wild.

Collection from wild populations (from more than 200 acres) includes a great number of species. The specific species that are collected depend on the origin and the diet of the collectors.

There is a small industry in Israel that distils or extracts secondary metabolites from MAPs growing in the country, but most of their raw material is imported.

If we exclude herbs (plants containing essential oils) that are used also for medicinal purposes, the "true" medicinal plants that are grown commercially are very limited, grown by a few farmers for local use.

**Table 2.** Production area (in acres) of MAPs in Israel

| Product | Green/net houses | Open field | Wild collection |
|---|---|---|---|
| Fresh herbs | 600 | 1450 | 50 |
| Dry herbs | 25 | 1650 | 100 |
| Seeds used as herbs | - | 1200 | 20 |
| Seeds for propagation | 5 | 25 | 5 |
| Ornamental herbs | 18 | 3 | -- |
| Distillation/extraction | - | 20 | -- |
| Medicinal plants | - | 30 | 10 |
| **Total** | **648** | **4378** | **285** |

## Wild MAPs in Israel

Some years ago a very extensive survey was carried out in order to identify potential wild MAPs in Israel that could be used in the future. The list is given in Table 3 for 74 species with their distribution area, shown in Fig. 2. This list is the basic national inventory of MAP natural resources of Israel. Normally from each population, depending on its size, the IGB stores 50 seeds or more, together with information on the name of the collector, the time of collection, and details of the site and its location.

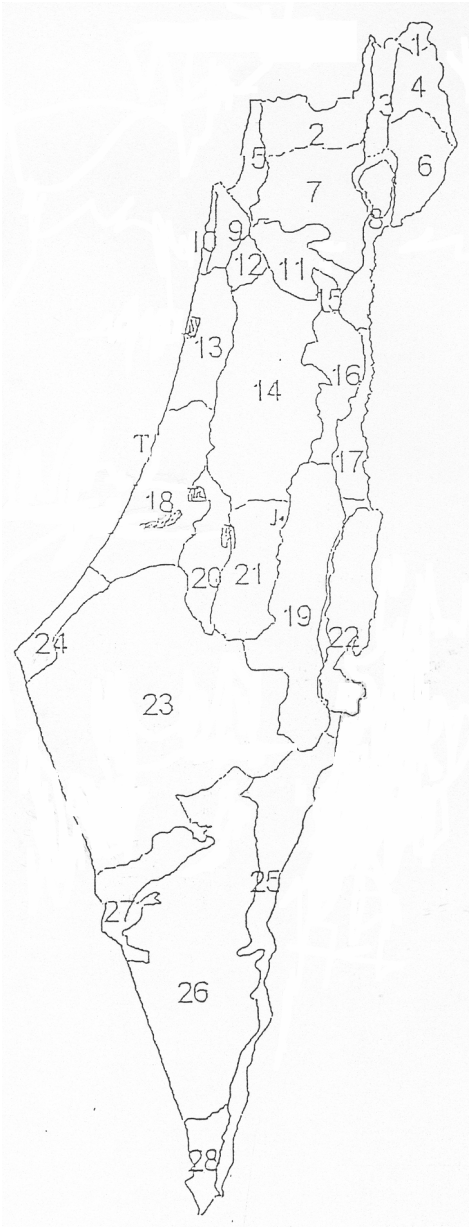
**Table 3**. Wild Israeli MAP species (74) with an economic potential

| Species | Family | Common name | Distribution area (*) | Abundance (**) |
|---|---|---|---|---|
| *Achillea biebersteinli* | Compositae | Yellow milfoil | 1 2 4 5 9 13 14 21 23 27 28 | R |
| *Achillea fragrantissima* | Compositae | Lavender cotton | 1 14 17 18 19 21 22 23 25 26 27 28 | CC |
| *Agrimonia eupatoria* | Rosaceae | Common agrimony | 12 | X |
| *Alhagi maurorum* | Papilionaceae | Cameltorn | 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 24 25 26 27 28 | C |
| *Ammi majus* | Umbelliferae | Common bishop's weed | 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 17 18 20 21 22 23 | CC |
| *Ammi Visnaga* | Umbelliferae | Toothpick | 2 3 4 5 6 7 8 9 10 11 12 13 14 16 18 20 21 23 | CC |
| *Anethum graveolens* | Umbelliferae | Dill | 3 7 14 18 2023 | RR |
| *Artemisia arborescens* | Compositae | Shrubby wormwood | 2 5 7 9 13 | RR |
| *Artemisia nerbaalba* | Compositae | White wormwood | 8 14 17 19 21 22 23 25 26 27 28 | CC |
| *Asphodelus aestivus* | Lillaceae | Tall asphodel | 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 | CC |
| *Asteriscus graveolens* | Compositae | Fragrant oxeye | 10 16 17 19 20 22 23 25 26 27 28 | C |
| *Atriplex halimus* | Chenopodiacea | Shrubby orache | 3 4 5 6 7 8 10 13 14 16 17 18 19 20 21 22 23 25 26 27 28 | C |
| *Balanites aegyptiaca* | Zygophyllaceae | Jencho balsam | 8 16 17 19 22 23 25 | F |
| *Brassica nigra* | Cruciferae | Black mustard | 1 2 3 4 5 6 7 8 9 10 11 13 18 | C |
| *Brassica tournefortli* | Cruciferae | Tournefort's mustard | 2 5 6 8 9 10 13 14 18 20 23 24 25 26 | C |
| *Calamintha incana* | Labiatae | Gray calamint | 23 7 9 12 14 19 20 21 | R |
| *Capparis spinosa* | Capparaceae | Thorny caper | 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 24 25 26 27 28 | CC |
| *Chenopodium ambrosioides* | Chenopodiacea | Common crosswort | 3 5 6 8 9 10 11 13 14 17 18 19 21 23 24 | F |
| *Cichorium pumilum* | Compositae | Dwarf chicory | 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 26 27 | CC |
| *Conium maculatum* | Umbelliferae | Mother die | 1 2 3 4 6 7 9 11 12 13 14 1518 21 | F |
| *Coriandrum sativum* | Umbelliferae | Coriander | 1 2 3 4 5 6 7 8 9 11 14 15 18 19 20 21 | R |
| *Coridothymus capitatus* | Labiatae | Capitate thyme | 1 2 4 5 6 7 9 10 13 14 16 17 18 19 20 21 22 23 26 | CC |
| *Crataegus aronia* | Rosaceae | Spiny hawthorn | 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 18 19 20 21 22 23 | C |
| *Crataegus azarolus* | Rosaceae | Mediterranean hawthorn | 2 4 9 21 | RR |
| *Crocus hyemalis* | Tridaceae | Winter crocus | 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 18 19 20 21 | CC |
| *Crocus pallasil* | Tridaceae | Pallas crocus | 1 2 4 6 10 14 21 | F |
| *Deverra tortuosus* | Umbelliferae | | 18 19 20 21 22 23 25 26 27 28 | C |
| *Ecballium elaterium* | Cucurbitaceae | Squirting cucumber | 1 2 3 4 5 6 7 8 9 10 11 12 13 14 16 18 19 20 21 23 | C |
| *Foeniculum vulgare* | Umbelliferae | Common fennel | 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 17 18 19 20 21 22 23 24 | CC |
| *Galium aparine* | Rubiaceae | Goose-grass | 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 17 18 19 20 21 22 23 | C |
| *Glaucium flavum* | Papaverceae | Yellow horned poppy | 2 5 9 10 13 18 21 | F |
| *Haptophyllum tuberculatum* | Rutaceae | Warty rue | 2 5 6 7 8 9 10 16 17 18 19 21 22 23 25 26 27 28 | C |
| *Hyoscyamus aureus* | Sotanaceae | Golden henbane | 1 2 3 4 5 6 7 8 9 10 12 13 14 15 16 17 18 19 20 21 22 23 25 26 27 | CC |

**Table 3 (cont.).** Wild Israeli MAP species (74) with an economic potential

| Species | Family | Common name | Distribution area (*) | Abundance (**) |
|---|---|---|---|---|
| *Inula viscose* | Compositae | Lesser eiecampane | 1 2 3 4 5 6 7 8 9 10 11 12 13 14 16 17 18 19 20 21 22 23 | CC |
| *Laurus nobilis* | Lauraceae | Laurel | 1 2 3 4 7 9 10 12 14 21 | C |
| *Lavandula stoechas* | Labiatae | French lavender | 1 3 5 9 13 18 19 20 21 22 | RR |
| *Mandragora autumnalis* | Solanaceae | Autumn mandrake | 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 23 | C |
| *Matricaria aurea* | Compositae | Wild chamomile | 2 5 6 7 8 9 11 13 14 17 18 19 20 21 22 23 26 27 | F |
| *Matricaria chamomilla* | Compositae | Wild chamomile | 1 2 3 5 6 7 9 13 20 21 23 | F |
| *Melissa officinalis* | Labiatae | Common balm | 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 21 | F |
| *Mentha longifolia* | Labiatae | Horse mint | 1 2 3 4 5 6 7 8 9 10 11 12 13 14 16 17 18 19 20 21 22 28 | C |
| *Micromeria fruticosa* | Labiatae | White savory | 2 4 5 6 7 8 9 10 11 12 13 14 15 19 20 21 22 | F |
| *Nasturtium officinale* | Cruciferae | True water-cress | 1 2 3 4 5 6 7 8 9 11 12 13 14 16 17 18 19 21 23 | C |
| *Origanum dayi* | Labiatae | | 14 19 21 23 25 26 27 | F |
| *Origanum syriacum* | Labiatae | Syrian marjoram | 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 | CC |
| *Papaver subpiriforme* | Papaveraceae | Corn poppy | 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 24 | CC |
| *Peganum harmala* | Zygophylaceae | Wild rue | 6 7 8 10 14 17 18 19 20 21 22 23 25 26 27 28 | F |
| *Plantago lanceolata* | Plantaginaceae | Ribwort plantain | 1 2 3 4 5 6 7 8 9 10 11 12 13 14 18 20 21 23 28 | C |
| *Plantago major* | Plantaginaceae | Great plantain | 12 3 4 5 6 7 8 9 10 11 13 14 16 18 19 | R |
| *Plantago ovata* | Plantaginaceae | Ovate plantain | 1 2 6 7 8 9 13 14 15 16 17 19 20 21 22 23 25 26 27 28 | CC |
| *Rhus coriaria* | Anacardiaceae | Sumach | 1 2 7 9 13 14 18 19 20 21 22 | F |
| *Ricinus communis* | Euphorbiaceae | Castoroil plant | 1 2 3 5 6 7 8 10 11 12 13 14 16 18 19 21 23 24 | F |
| *Ridolfia segetum* | Umbelliferae | Bishop's weed | 1 2 3 4 5 6 7 8 9 11 12 13 14 16 17 18 19 20 21 23 | CC |
| *Ruta chalepensis* | Rutaceae | Common rue | 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 18 20 21 23 | F |
| *Salix acmophylla* | Salicaceae | Willow | 1 2 3 4 5 6 7 8 9 10 11 12 13 14 16 17 18 19 21 22 | C |
| *Salix alba* | Salicaceae | White willow | 3 4 7 8 11 18 | F |
| *Salsola Kali* | Chenopodiacea | Prickly saltwort | 2 5 7 10 11 13 18 20 21 22 23 24 | C |
| *Salsola soda* | Chenopodiacea | French saltwort | 5 9 13 | O |
| *Salvadora persica* | Saivadoraceae | Toothbrush tree | 17 19 22 25 26 | RR |
| *Salvia dominica* | Labiatae | Dominica sage | 2 3 6 7 8 9 10 11 12 14 15 16 17 18 19 20 21 22 23 27 | C |
| *Salvia fruticosa* | Labiatae | Three-leaved sage | 1 2 3 4 5 6 7 9 10 12 13 14 15 19 20 21 22 23 | CC |
| *Salvia indica* | Labiatae | Blue sage | 1 2 4 6 7 14 18 19 | |
| *Silybum marianum* | Compositae | Holy thistle | 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 | CC |
| *Sinapis alba* | Cruciferae | White mustard | 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 | CC |
| *Styrax officinalis* | Styracaceae | Officinal storax | 1 2 3 4 5 6 7 8 9 11 12 13 14 15 16 20 21 | C |
| *Teucrium polium* | Labiatae | Mountain germander | 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 25 26 27 28 | CC |
| *Thymbra spicata* | Labiatae | Spiked thymbra | 1 2 3 5 7 9 12 13 14 19 20 21 | F |
| *Trigonella berythea* | Papilionaceae | Beirut fenugreek | 1 2 3 4 5 6 7 8 9 10 11 12 13 14 18 20 21 | F |
| *Urginea maritime* | Liliaceae | Maritime squill | 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 25 26 27 | CC |
| *Urtica pilulifera* | Urticaceae | Roman nettle | 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 23 | F |
| *Varthemia iphionoides* | Compositae | Goldilocks | 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 25 26 27 28 | CC |
| *Verbascum eremobium* | Scrophulariaceae | Round-leaved mullein | 19 20 23 25 26 27 28 | -1 |
| *Verbena officinalis* | Verbenaceae | Common vervain | 1 2 3 4 5 6 7 8 9 10 11 12 13 14 16 17 18 19 20 21 22 23 | C |
| *Withania somnifera* | Solanaceae | Common winter-cherry | 2 3 4 5 6 7 8 9 10 11 12 13 14 16 17 18 19 20 21 22 23 24 | C |
| *Ziziphus spinachristi* | Rhamnaceae | Christ-thorn jujube | 1 2 3 4 5 7 8 9 10 11 1314 16 17 18 19 20 21 22 23 24 25 26 28 | CC |

(*) see Fig. 2
(**) Abundance: CC = Very common; C = Common; F = Frequent (between C and R); R = Rare; RR = Very rare (3 to 30 sites); O = 1 to 3 sites; X = Not observed since 1960




1. Mount Hermon
2. Upper Galilee
3. Hula and Dan Valleys
4. North Golan
5. Akko Plain
6. South Golan
7. Lover Galilee
8. Upper Jordan Valley
9. Mount Carmel
10. Carmel Coast
11. Esdraelon Plain
12. Menashe Hills
13. Sharon Plain
14. Samaria Mountains
15. Mount Gilboa
16. Samaria Desert
17. Lower Jordan Valley
18. Philistean Plain
19. Judean Desert
20. Shefela
21. Judean Mountains
22. Dead Sea Area
23. Northern and Western Negev
24. Gaza Strip
25. Arava Valley
26. Southern and Central Negev
27. Negev Mountains
28. Elat

**Fig. 2.** Districts in which target species are distributed.

## *Conservation of medicinal and aromatic plants in Italy*

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### Legal protection of medicinal and aromatic plant species and their natural habitats

The fundamental text of Italian legislation concerning medicinal and aromatic plants (MAPs) is still the Royal Decree (R.D.) no. 772 of 1932, in which a list of MAPs is reported and where the maximum quantity allowed to be harvested, upon authorization, is indicated (Table 1).

**Table 1.** Italy: list of the MAPs cited in the Royal Decree n° 772 of 1932

| No. | Species | Quantity allowed to be harvested |
|---|---|---|
| 1 | *Achillea erba-rotta* All. | (aerial parts) 1 kg |
| 2 | *Achillea moschata* Wulfen | (aerial parts) 1 kg |
| 3 | *Aconitum* sp. | (leaves and roots) 0 kg |
| 4 | *Acorus calamus* L. | (roots) 2 kg |
| 5 | *Adonis* sp. | (full plants) 0 kg |
| 6 | *Angelica archangelica* L. | (seeds and roots) 2 kg |
| 7 | *Arctium lappa* L. | (roots) 5 kg |
| 8 | *Arnica montana* L. | (flowers and roots) 5 kg |
| 9 | *Artemisia absinthium* L. | (aerial parts) 1 kg |
| 10 | *Artemisia campestris* L. subsp. *borealis* (Pall.) | (aerial parts) 1 kg |
| 11 | *Artemisia genipi* Weber | (aerial parts) 1 kg |
| 12 | *Artemisia glacialis* L. | (aerial parts) 1 kg |
| 13 | *Artemisia pontica* L. | (aerial parts) 1 kg |
| 14 | *Artemisia umbelliformis* Lam. | (aerial parts) 1kg |
| 15 | *Artemisia vallesiaca* All. | (aerial parts) 1 kg |
| 16 | *Artemisia vulgaris* L. | (flowers, leaves, roots) 2 kg |
| 17 | *Atropa bella-donna* L. | (leaves) 0 kg |
| 18 | *Bryonia dioica* Jacq. | (leaves) 0 kg |
| 19 | *Centaurium erythraea* Rafn | (flowered plants) 5 kg |
| 20 | *Urginea maritima* (L.) Baker | (bulbs) 0 kg |
| 21 | *Citrullus colocynthis* (L.) Schrad. | (fruits) 0.5 kg |
| 22 | *Cnicus benedictus* L. | (aerial parts) 2 kg |
| 23 | *Colchicum autumnale* L. | (bulbs and seeds) 0 kg |
| 24 | *Conium maculatum* L. | (leaves) 0 kg |
| 25 | *Datura stramonium* L. | (leaves) 0 kg |
| 26 | *Delphinium staphisagria* L. | (seeds) 1 kg |
| 27 | *Dictamnus albus* L. | (flowered plants) 2 kg |
| 28 | *Digitalis purpurea* L. | (leaves) 0 kg |
| 29 | *Frangula alnus* Mill. | (bark) 0.5 kg |
| 30 | *Fraxinus* sp. L. | (manna) 0.5 kg |
| 31 | *Gentiana lutea* L. | (roots) 10 kg |
| 32 | *Glycyrrhiza glabra* L. | (roots) 10 kg |
| 33 | *Hyoscyamus niger L.* | (leaves) 0 kg |
| 34 | *Hyssopus officinalis* L. | (sprigs) 2 kg |
| 35 | *Peucedanum ostruthium* (L.) W.D.J.Koch | (roots) 2 kg |
| 36 | *Inula helenium* L. | (roots) 2 kg |
| 37 | *Juniperus sabina* L. | (sprigs) 0 kg |
| 38 | *Lavandula angustifolia* Mill. | (flowered tips) 10 kg |
| 39 | *Lavandula latifolia* Medik. | (flowered tips) 10 kg |
| 40 | *Lycopodium clavatum* L. | (spore) 0.5 kg |
| 41 | *Matricaria recutita* L. | (flowers) 10 kg |
| 42 | *Melissa officinalis* L. | (flowered tips, leaves) 5 kg |
| 43 | *Oenanthe aquatica* (L.) Poir. | (seeds) 0.5 kg |
| 44 | *Pinus mugo* Turra | (sprigs) 10 kg |
| 45 | *Psyllium afrum* (L.) Mirb. | (seeds) 0.5 kg |
| 46 | *Rhamnus cathartica* L. | (fruits) 0.5 kg |
| 47 | *Saponaria officinalis* L. | (leaves and roots) 10 kg |
| 48 | *Solanum dulcamara* L. | (stems) |
| 49 | *Tanacetum vulgare* L. | (flowers) 5 kg |
| 50 | *Taraxacum officinale* Weber | (roots, 5 kg.) |
| 51 | *Teucrium montanum* L. | (aerial parts) 2 kg |
| 52 | *Thymus vulgaris* L. | (flowered plants) 10 kg |
| 53 | *Tilia* sp. | (flowers) 10 kg |
| 54 | *Tussilago farfara* L. | (flowers) 5 kg |
| 55 | *Valeriana* sp. | (roots) 2 kg |
| 56 | *Veratrum lobelianum* Bernh. | (roots) 0 kg |

The attempts to substitute this national law with another one, more updated and opportune, have been as numerous as unsuccessful and still some months ago the umpteenth parliamentary bill for the regulation of the MAP sector was presented.

Nevertheless since the 1970s most regions and the two autonomous provinces of Trento and Bolzano have passed bills for flora and/or natural habitat protection (www.camera.mac.ancitel.it). In these laws a list is given of species whose harvesting is forbidden or restricted, but few regional laws contain specific references to MAPs (Table 2).

**Table 2.** Legal protection of plant species and MAP collecting regulation in various Italian regions

| Regions | Regional law (year/no.) | MAP collecting regulation | No. of protected species whose harvest is: | | Other species |
|---|---|---|---|---|---|
| | | | forbidden | restricted | No. of floral axes allowed [1] |
| Val d'Aosta | 1977/17 | specific | 40 | 22 | 20 (80) |
| Piedmont | 1982/32 | R.D. no. 772 | 174 | - | 5 |
| Liguria | 1984/07 | R.D. no. 772 | 32 | 20 | free harvest [4] |
| Lombardy | 1977/33 | R.D. no. 772 | - | 51 | free harvest [4] |
| Veneto | 1974/53 | - | 35 | - | 6 |
| Trentino | 1973/17 | derogations | 19 | - | 5 |
| Alto Adige | 1972/13 | derogations | 23 | - | 6 |
| Friuli V.G. | 1981/34 | specific | 20 | - | 10 |
| Emilia Romagna | 1977/02 | derogations | 46 | - | free harvest [4] |
| Tuscany | 2000/56 | free harvest [4] | 57 | 9 | free harvest [4] |
| Marche | 1985/7 | free harvest [4] | 15 [2] | - | free harvest.[4] |
| Umbria | 1987/49 | derogations | 48 | - | free harvest [4] |
| Lazio | 1974/61 | free harvest [4] | 72 | 2 | free harvest [4] |
| Campania | 1994/40 | R.D. n° 772 | 20 | 39 | free harvest [4] |
| Abruzzo | 1997/35 | free harvest [4] | 36 | - | free harvest [4] |
| Basilicata | 1994/28 | free harvest [4] | [3] | - | free harvest [4] |
| Puglia | 1997/19 | free harvest [4] | - | - | free harvest [4] |
| Calabria | 2001/30 | free harvest [4] | >30 | - | free harvest [4] |
| Sicily | 1981/98 | free harvest [4] | [3] | - | free harvest [4] |
| Sardinia | 1989/31 | free harvest [4] | [3] | - | free harvest [4] |

[1] Per person/day and (groups)
[2] Arborescent species
[3] Species present in the protected habitats
[4] Outside the protected habitats

Among these, most regions (Piedmont, Liguria, Lombardy and Campania) limit themselves to referring to species and quantities mentioned in the R.D., while only Friuli-Venezia Giulia and Val d'Aosta provide regional lists of MAPs and fix quantities which may be harvested. In Friuli for instance, it is allowed to collect the edible fresh parts of 26 species (maximum quantity/day = 1 kg) (Table 3), while for the species not included in the list, derogations are provided only for scientific, pharmaceutical, medicinal or educational purposes. The list of wild protected MAPs and corresponding quantities allowed in Val d'Aosta are almost the same as that of the R.D.

**Table 3.** Friuli Venezia Giulia: list of MAPs whose harvest is regulated [1]

| No. | Species | No. | Species |
|---|---|---|---|
| 1 | *Allium schoenoprasum* L. | 14 | *Rosa canina* L. |
| 2 | *Arnica montana* L. | 15 | *Rubus fruticosus* L. |
| 3 | *Aruncus vulgaris* Rafin | 16 | *Rubus idaeus* L. |
| 4 | *Asperula odorata* L. | 17 | *Ruscus aculeatus* L. |
| 5 | *Cardamine pratensis* L. | 18 | *Ruta graveolens* L. |
| 6 | *Chenopodium* sp. | 19 | *Silene cucubalus* Wibel |
| 7 | *Fragaria vesca L.* | 20 | *Symphytum officinale* L. |
| 8 | *Galium mollugo* L. | 21 | *Taraxacum officinale* Weber |
| 9 | *Humulus lupulus* L. | 22 | *Tragopogon pratensis* L. |
| 10 | *Melissa officinalis* L. | 23 | *Urtica dioica* L. |
| 11 | *Mentha* sp. | 24 | *Vaccinum myrtillus* L. |
| 12 | *Origanum vulgare* L. | 25 | *Vaccinum vitis-idaea* L. |
| 13 | *Papaver rhoeas* L. | 26 | *Valerianella olitoria* L. Poll. |

[1] Maximum quantity allowed =1 kg/person/day of fresh aerial parts

The regional laws to protect flora are more or less severe according to regions. In most regions the harvesting of unprotected species and of plants growing outside protected areas is unrestricted. On the contrary in other regions, in addition to a list of protected plants whose harvest is forbidden, severe limitations in the collection of other species are also provided and only a few specimens are allowed. Derogations to these restrictions are provided only for scientific, pharmaceutical, medicinal or educational purposes, but in any case harvesting needs an authorization. The regional law of Veneto does not provide derogations for "medicinal" purposes and as a consequence, in this region, MAP collecting is forbidden. The only exception is wild asparagus, of which you may harvest 1 kg/day.

In the regional lists of protected plants you may also find some "classic" MAPs (Table 4) among which *Dictamus albus* L., *Gentiana lutea* L. and two species of *Ruscus* (*R. aculeatus* L. and *R. ipoglossum* L.) are the most frequently recurring.

**Table 4.** MAPs present in the lists of protected plants in different Italian regions

| Genus | Species | Val d'Aosta | Piedmont | Liguria | Lombardy | Veneto | Trentino | Alto Adige | Friuli V.G. | Emilia Rom. | Tuscany | Umbria | Marche | Lazio | Campania | Abruzzo | Calabria |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| *Achillea* | *erba-rotta* All. | | | | | | | | | | | | | | | | X |
| *Aconitum* | sp. | X | X | | | | | | | | | | | | | | |
| *Arnica* | *montana* L. | X[1] | | X | | | | | X[2] | X | | | | | | | |
| *Artemisia* | *laxa* F. | | | | | X | | | | | | | | | | | |
| *Artemisia* | *umbelliformis* Lam | | | | | | | X | | | | | | | | | |
| *Atropa* | *bella-donna* L. | | X | | | | | | | | | | | X[4] | | X | |
| *Crocus* | sp. | | | | | | | | | X | X | | | X | | | |
| *Dictamus* | *albus* L. | X | X | X | | X | X | X | | X | | | | | X | X | X |
| *Gentiana* | sp. | X[1] | X | X | X | X | | X | | X | | X | | | X | X | X |
| *Glycyrrhiza* | *glabra* L. | | | | | | | | | | | | | | | X | |
| *Hyssopus* | *officinalis* L. | | X | | | | | | | | | X | | X | | | |
| *Iris* | sp. | | X | X | X | | | | X | | | | | X | | | |
| *Juniperus* | sp. | | X | | | | | | | | | X | | | | | |
| *Mandragora* | *autumnalis* Bert. | | | | | | | | | | | | | X | | | |
| *Myrtus* | *communis* L. | | | | | | | | | | | | | | | X | |
| *Paeonia* | *officinalis* L. | X | X | X | X | X | | | X | | X | X | | | | | X |
| *Pistacia* | *lentiscus* L. | | | | | | | | | | | X | | | | | |
| *Ruscus* | sp. | | X | X | X | | X | X | X[2] | | X[3] | X | | | X | X | |
| *Solidago* | *virgaurea* subsp. *littoralis* (Savi) Burnat | | | | | | | | | | X | | | | | | |
| *Taxus* | *baccata* L. | | | | | | | | | X | | X | X | X | | X | X |
| *Thymus* | *vulgaris* L. | | | | | | | | | | | | | X | | | |
| *Urginea* | *maritima* Bak. | | | | | | | | | | | | | X[4] | | | |
| *Viscum* | *album* L. | | | | | | | | | | X | | | | | | |

[1] It is allowed to collect 6 flowering stems per day/person and 24 flowering stems per groups >3 people
[2] It is allowed to collect daily 1 kg of edible, fresh parts per person
[3] It is allowed to collect daily 10 flowering stems/person
[4] A collecting authorization is needed

Instead of referring to "protected species", in some regions of southern Italy (Basilicata and Puglia) and in the islands (Sicily and Sardinia), the lawmakers have designated "protected areas" in which every kind of capture (animals) or collecting (plants, lichens and mosses) is forbidden. In the Basilicata region there are 9 natural protected areas, in Puglia there are 33 in total (7 in each of the Bari and Lecce provinces, 11 in the province of Taranto and 4 in each of the Brindisi and Foggia provinces); in Sardinia 9 natural parks are present, covering more than 300 000 ha.

These laws have also undoubtedly contributed to preserving MAP genetic resources; nevertheless, in the meantime the reasons for the increasing rarity and/or extinction of these

plants have changed: apart from oregano (*Origanum vulgare* L. subsp. *hirtum* (Link) Ietswaart), still abundantly collected in central and southern Italy, and myrtle (*Myrtus communis* L.), collected in Sardinia to make liqueurs, wild collecting has drastically decreased because very few people still take part in this activity.

### Conservation *ex situ*

It is well known that the earliest attempts to conserve MAPs *ex situ* date back to the Middle Ages when the first "*Horti sanitatis*" were established in the monasteries. Afterwards, during the Renaissance the first botanical gardens called "gardens of simples" were laid down according to similar patterns (Del Prete *et al*. 2001). The earliest botanical garden in the world was established in Pisa in 1543-1544, while a garden connected with the Medicean court, in which MAPs were grown, already existed in Florence in 1544. While no traces of either of these gardens remain, the botanical garden of Padua, founded in 1545, still exists and is tended. Furthermore numerous other universities endowed themselves with similar institutions and there are currently 38 botanical gardens in Italian universities. In addition there are 31 alpine gardens, including 16 located in the actual Alpine areas, 15 others in the Apennines (Sicily included) and 12 thematic gardens of which one is dedicated to MAPs, the "Giardino delle erbe" of Casola Valsenio. Nevertheless, this large number of institutions does not mean that MAP genetic resources are abundant and properly conserved, as in most cases these institutions have only an educational function.

### Activities at a national level to record MAP natural resources

Because of the huge environmental variation in Italy and the restricted economic importance of MAPs in our country, it is not feasible to record MAP natural resources at the national level. In the recent past some studies have been carried out on wild populations of oregano (De Mastro 1996; Leto 1996). More recently, research on the characterization of different wild populations of *Chamomilla recutita* Rausch. collected in northern (Aiello *et al.* 2001) and central Italy (Taviani *et al.* 2002) and numerous populations of myrtle and *Rosmarinus officinalis* L. collected in Sardinia (Mulas 2001) is still underway within the National Project IPPO ("Increase of medicinal and aromatic plants production").

According to research carried out in Trentino (Prosser 2000), involving 10 years of plant mapping, 30.6% of the species (723 out of 2359) are considered as threatened, including some MAP species. For instance, according to the categories adopted by IUCN (International Union for the Conservation of Nature): *Centaurea cyanus* L. and *Marrubium vulgare* L. are considered critically endangered (CR); *Lactuca virosa* L. and *Rosa gallica* L., endangered (EN); *Gratiola officinalis* L., *Hyoscyamus niger* L. and *Hyssopus officinalis* L., vulnerable (VU); *Althaea officinalis* L., *Equisetum pratense* Ehrh., *Leonorus cardiaca* L. and *Lithospermum officinale* L., at low risk (LR). Also according to this research, the main factor threatening the conservation of plant genetic resources (PGR) seems to be the change in land use in the agricultural and pastoral sectors: land abandonment and high-input agricultural practices together account for almost 3/4 (73%) of all the threatening factors identified.

As regards the inventory of PGR *ex situ,* within the project "Cultural Assets", the group "Botanical and historical garden" has been working for about 4 years to draw up a complete list of the gardens being included in this project and of their corresponding collections (Del Prete *et al*. 2001). On a smaller scale, taking the opportunity of this meeting, the authors of this paper contacted numerous botanical gardens and recorded the list of MAPs held by 15 of these (Table 5).

**Table 5.** Information recorded about the Italian Botanical Gardens

| Botanical gardens | No. | (%) |
|---|---|---|
| Total | 58 | |
| With a specific collection for maps | 25 | 43 |
| Having their own Web site | 26 | 45 |
| Having a general list of plants on the Web site | 6 [(1)] | 10 |
| Having a specific list for MAPs on the Web site | 3 [(2)] | 5 |
| Having an *Index seminum* on the Web site | 4 [(3)] | 7 |
| Botanical gardens recorded | 15 [(4)] | 26 |
| **Total number of MAP species in conservation** | **1247** | **-** |
| Spontaneous in Italy | 731 | 59 |
| Endemic in Italy | 30 | 2.4 |
| Exotic | 407 | 33 |
| Cultivated | 467 | 37 |

[(1)] Casola val Senio (RA), Camerino, Majella, Lecce, Bari, Catania.
[(2)] Casola val Senio (RA), Camerino, Majella.
[(3)] Trieste, Modena, Siena, Majella.
[(4)] Torino, Bergamo, Brescia, Padova, Trieste, Bologna, Ferrara, Casola val Senio (RA), Firenze, Camerino, Majella, Siena, Bari, Lecce, Catania.

## Review of the acreage of MAPs under cultivation

In 1999-2000, ISAFA carried out a survey of the acreage and the characteristics of MAPs under cultivation in Italy (Vender 2001). According to this survey, the total area occupied by MAPs was 3342 ha and the number of species grown in Italy was over a hundred. However, slightly more than 30 species occupy over 90% of the total area. Beside *Citrus bergamia* Risso (bergamot), the only species with an area greater than 1000 ha, the other main species are: *Mentha* x *piperita* L. (peppermint), *Fraxinus* sp. (manna ash), *Chamomilla recutita* Rausch. (chamomile), *Glycyrrhiza glabra* L. (liquorice), *Lavandula* sp. (lavender and lavandin), *Hypericum perforatum* L. (St John's wort) and *Linum usitatissimum* L. (linseed). The main species cultivated, assembled into six groups according to the area occupied, are listed in Table 6. Compared with a similar survey carried out 10 years earlier (ISMEA 1989) the list of species has changed: tarragon, orris, roman wormwood, summer savory and jasmine have declined; peppermint, manna ash and saffron have fallen in importance, while chamomile, St John's wort, liquorice, lavenders, linseed, rosemary, fennel, sage, lemon balm and coneflower have greatly increased.

As to distribution, more than 50% of the total area cultivated in MAPs is located in Calabria where, besides bergamot, liquorice and cedar are also grown. Other regions where MAP cultivation has some importance are Piedmont, where excellent peppermint oil is obtained, and Tuscany, where numerous herbs are grown on a huge farm.

Excluding bergamot, from which we obtain the famous essence, about 70% of the total area cultivated in MAPs is used for the production of dried herbs, 20% for essential oil extraction and less than 10% for fresh consumption (Vender 2002).

As regards farm size, there are huge individual and regional differences, but in general, only a small part of the arable area (a.a.) is reserved for MAPs. Farms specialized in MAP production (>80% of the a.a. planted to MAPs) represent only 17% of the total and most of them are located in the north. Nevertheless there are some exceptions: in Piedmont numerous large farms specializing in MAP cultivation are present, and in Tuscany the Aboca farm dedicates more than 300 ha to the cultivation of many species.

Concerning field management, most farms adopt organic techniques (61%), the largest proportion of organic farms being in the centre (75%) and the lowest in the south (43%); nevertheless we may note that only bergamot, peppermint and St John's wort are handled as industrial crops, sustainable practices being used for other species.

Taking into consideration different parameters (proportion of specialized farms, postharvest machinery availability, trade association importance, etc.) we may conclude that in the last 10 years there has been no widespread development of the sector or of associated

facilities. This sector could have useful economic potential in our country, but systems and organizations aimed to match supply and demand are lacking, and besides the supply is widely scattered.

**Table 6.** Medicinal and aromatic plants grouped according to the cultivated area, 1999 (source: Vender 2002)

| First group: >1000 ha | Surface (ha) |
|---|---|
| Bergamot | 1500 |

| Second group: 250-100 ha | Surface (ha) |
|---|---|
| Chamomile | 171 |
| Lavender and lavandin | 133 |
| Liquorice | 146 |
| Manna ash | 200 |
| Peppermint | 239 |
| St John's wort | 156 |
| **Total** | **1045** |

| Third group: 100-50 ha | Surface (ha) |
|---|---|
| Linseed | 81 |

| Fourth group: 50-10 ha | | | Surface (ha) |
|---|---|---|---|
| Anise | Flea-wort | Passion flower | |
| Blue gum tree | Ginkgo | Roman chamomile | |
| Burdock | Hash | Roman wormwood | |
| Citron | Hawthorn | Rosemary | |
| Clary sage | Hyssop | Saffron | 593 |
| Coneflower | Immortelle | Sage | (in total) |
| Dandelion | Lemon balm | Summer savory | |
| Dog rose | Mauve | Sweet clover | |
| Fennel | Oregano | Thyme | |

| Fifth group: 10-1 ha | | | Surface (ha) |
|---|---|---|---|
| Alder | Fenugreek | Myrtle | |
| Artichocke | Genepi | Orange (flower) | |
| Black currant | Grindelia | Orris (roots) | |
| Californian poppy | Linden | Tarragon | 108 |
| Caraway | Marigold | Witch hazel | (in total) |
| Chives | Marjoram | Wormwood | |
| Coriander | Marshmallow | Yarrow | |
| Dill | Meadow-sweet | Yew | |

| Sixth group: <1ha | Surface (ha) |
|---|---|
| About 50 different species | 1 |

## Use of MAPs by processing industries

About 1500 plant species are regarded as MAPs, most of which come from tropical areas and are used in different sectors. Thus it is clear that we face a global market in MAPs.

Following an increasing demand for a wide range of products of natural origin, the national manufacturing industries' demand for MAPs has increased and firms processing herbs for different outlets are numerous (Table 7). However, apart from those of a few firms, most herbs, spices and essential oils come from abroad. Such products are bought directly or by a few big wholesalers belonging to the Assoerbe organization, but some large firms draw up cultivation contracts. For instance the Italian firm Indena, a world leader in pharmaceutical plant products, to assure itself high quality standards and to limit its costs, makes cultivation contracts with growers throughout the world, apart from Italy (R. Iguera, personal communication, 2002).

**Table 7.** Italy: number and kind of industries operating in the herb sector (source: Anonymous 2000)

| Productive typology | No. of industries |
|---|---|
| Raw materials and botanical extracts (fresh and dried) | 77 |
| Dietary supplement, special nutrition and health food | 62 |
| Natural cosmetics | 110 |
| Other products of natural origin | 13 |
| Manufacturing services | 45 |
| Multi-tasks firms | 248 |
| **Total** | **555** |

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## *Medicinal and aromatic plants in Latvia*

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### Legal protection of medicinal and aromatic plant species and their natural habitats

The central executive institution for nature protection in Latvia is the Ministry of Environmental Protection and Regional Development. With the help of the institutions under its supervision, the Ministry is responsible for: preparing and implementing a national policy for nature protection, preservation and rational use of natural resources; drafting legal acts within its jurisdiction; harmonizing government mission statements with the requirements of the European agreements and Commission; and ensuring their implementation.

The National Environmental Policy Plan for Latvia included tasks to cooperate with international organizations for nature conservation to ensure consideration of Latvian interests and to draw financial investments into the adoption of the National Programme for Biological Diversity.

The Nature Protection Department of the Ministry of Environmental Protection and Regional Development is regulated by the laws of Latvia: *Law on Protected Territories* and *Law on Conservation of Species and Habitats* (National Programme of Biodiversity 2000). Medicinal and aromatic plants (MAPs) are included together with other species.

The Department prepares the National Programme on Biological Diversity. Its main tasks are *in situ* conservation and prevention of the decline in numbers and distribution of local wild species. The programme includes:

- Survey of medicinal plant resources;
- Development of nature reserves for the most important sites of medicinal herbs, where priority is given to resource maintenance and sustainable use;
- Development and implementation of commercial regulations for harvesting and trade in medicinal plants, including requirements for protection of herbs; and
- Popularization and promotion of herb cultivation.

### Inventory of natural resources of MAPs

The inventory data of MAP natural resources are not suitable for today's requirements, since it was obtained and published 20-30 years ago. Since then, the status of many species has changed, and now information on many species including MAPs is quite problematic. Additionally much useful information can be found on birds and plants in the Red Book of Latvia (Aigare *et al.* 1985).

The last inventory project was the CORINE Biotopes Project[14] conducted in Latvia 1994-1997 (Opermanis *et al.* 1997). At the end of the project, 251 "Biotopes sites" for a total area of 598 106 ha were designated in Latvia. The total terrestrial plus inland water Biotopes sites area covers 504 000 ha, or 7.8% of the area of Latvia. About 6% of the total number of Biotopes sites contained areas free from human influence. About 174 750 ha (34.6%) of the total Biotopes sites coverage is protected by the State.

[14] CORINE = CO-oRdination of INformation on the Environment. The CORINE Biotopes inventory aims at identifying the sites of major importance for nature conservation on the European level (Biotopes sites).

**Protected territories in Latvia with medicinal and aromatic plants**

- State Nature Reserves (5): Moricsalas (1912), Slitere (1921), Grini (1936), Krustkalni (1977), Teichi (1982)
- National Parks (2): Gauja National Parka and Kemeri National Park
- Nature Parks (21): the most popular are "Tervete" Nature Park and "Daugavas Loki" Nature Park
- Nature Reserves (211): these areas are natural complexes unaltered by human activity, areas where rare and endangered species can be found.

### *Ex situ* conservation of MAPs

The collections of medicinal and aromatic plants are held in five institutes:

- Botanical garden of Latvia at Salaspils (largest MAP collection)
- Botanical garden of the Latvian University in Rîga
- Latvia University of Agriculture in Jelgava
- State Selection Station at Skriveri
- Bulduri Horticultural College.

### Collecting of wild medicinal plants

The following medicinal plants are regularly collected from the wild only: *Filipendula ulmaria, Tanacetum vulgare, Taraxacum officinale* and *Tussilago farfara.*

Most of the marketed raw materials from the following species are collected from the wild: *Achillea millefolium* (~70%), *Bidens tripartita* (~70%), *Thymus serpyllum* (~95%) and *Hypericum perforatum* (~10%).

### MAP cultivation in Latvia

Exact data on cultivation are not officially obtainable, since growers seem to be afraid to give accurate information owing to high taxes. The Medicinal Plant Association in Latvia, "Metra", consists of 95 members. Most growers with large cultivated areas are situated in the districts of Limbazi, Valmiera, Cesis and Aizkraukle.

According to the statistical information of Metra, the area cultivated under MAPs in 2001 was about 200 ha, but according to the statistical information of the Central Statistical Bureau of Latvia, the total area cultivated with MAPs is estimated to be about 300 ha (Table 1).

**Table 1.** Estimated area of cultivated medicinal and aromatic plants in Latvia

| Species | Total area (ha) | Note |
|---|---|---|
| *Valeriana officinalis* | 50 | one farm has about 20 ha |
| *Chamomilla recutita* | 60 | one farm has about 30 ha |
| *Hypericum perforatum* | 35 | |
| *Mentha* x *piperita* | 30 | one farm has about 10 ha |
| *Calendula officinalis* | 20 | |
| *Carum carvi* | 50 | |
| *Origanum vulgare* | 20 | |
| *Viola arvensis* and *V. tricolor* | 3 | |
| *Artemisia absinthium* | 3 | |
| *Salvia officinalis* | 2 | |
| *Bidens tripartita* | 2 | |
| *Helichrysum arenarium* | 2 | |
| *Leonorus cardiaca* | 2 | |
| Other MAP species (*Hyssopus officinalis, Majorana hortensis, Lavandula angustifolia, Thymus serpyllum*, etc.) | 10 | |
| **Total** | **289** | |

### Use of MAPs by the processing industry

No precise data are available. About one third of the production is processed by the Riga Pharmaceutical Factory. The growers sell about one third of the production directly to drugstores (chemist shops) for teas and homeopathic drug production. The last third is exported to Russia and to other former Soviet Union republics.

### Experience with sustainable use of MAPs

The first experiences in different projects are connected with organic (ecological) agriculture and country tourism.

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## *Conservation of medicinal and aromatic plants in Lithuania*

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### Introduction

The use of wild medicinal and aromatic plants (MAPs) is widespread and has old traditions in Lithuania, especially in forested areas in the southern and southeastern parts of the country. About 100 taxa are still harvested from the wild for medicinal purposes, representing a significant part of the Lithuanian natural biodiversity which comprises 1323 vascular plant taxa. The changes in political and economic life (Lithuania regained its independence in 1991) have increased the demand for MAPs and therefore determinate the importance of germplasm conservation. Active conservation of MAPs is pursued within the context of the National State Plant Genetic Resources Programme (1998-2002).

The Nordic Gene Bank has made a substantial contribution in starting and developing plant genetic resources (PGR) conservation activity in Baltic countries. The Nordic-Baltic collaborative project on PGR for food and agriculture was initiated in 1994 and is still ongoing (Weibull 2001). The MAP working group is one of the crop-specific working groups within this project. The new project SPIMED (*Spice and medicinal plants in the Nordic and Baltic countries. Strategies for conservation of genetic resources*) started in 2002.

This paper provides a brief review of the current research and activities dealing with MAP germplasm conservation in Lithuania.

### Material and methods

The analysis of trade figures is based on data collected by the governmental Department of Statistics and the Division of Plant Resources of the Ministry of Environment in 1996-2001.

Target species for conservation were selected on the basis of their current economic value, usefulness for breeding and conservation status. The evaluation of accessions was carried out according to the morphometric analysis of the phenotype and the quality and quantity of secondary metabolites. Analytical methods were employed for chemical evaluation of the raw material of target species. Spectrophotometry was used for the determination of total flavonoids. The quantitative analysis of phenolic compounds was carried out by a modified HPLC gradient elution method (Hölzl and Ostrowski 1987). Essential oils were isolated by hydro-distillation. GC and GC–MS analyses were performed for qualitative and quantitative evaluation of the essential oils.

The species distribution maps were prepared on the basis of herbarium data from the Institute of Botany and Vilnius University and our observations on MAP sources.

### Results and discussion

- **The trade in MAPs**

The average annual volume of plant material coming from the wild represents about 29% of the total volume used in pharmaceutics. This means that about 85 t dry raw material is extracted from the wild populations yearly (Fig. 1). The most frequently used native species in herbal medicine are *Crataegus* sp., *Arctostaphylos uva-ursi, Menyanthes trifoliata, Hypericum perforatum, Thymus serpyllum*, *Tussilago farfara*, *Polygonum aviculare, Urtica dioica* and *Frangula alnus.*

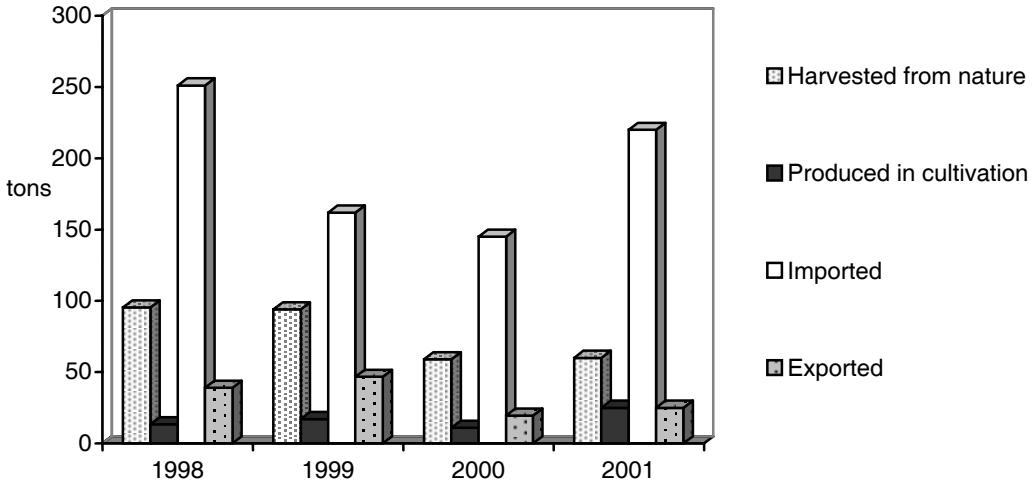


**Fig.1.** Total volumes of raw material of medicinal and aromatic plants produced in natural habitats, cultivated, imported and exported in 1998-2001 in Lithuania.

The increased MAP trade has been greatly influenced by the liberalization of the export-import activity. According to import-export data Lithuania can be characterized as a country mainly importing the raw material in contrast to other eastern and central European countries (Bernáth 1999) (Fig. 2). The average annual volume of imported MAP raw material is about 65% of all material used in the pharmaceutical industry. Exports make up a small part of the trade and are mainly directed to the countries of the former Soviet Union.

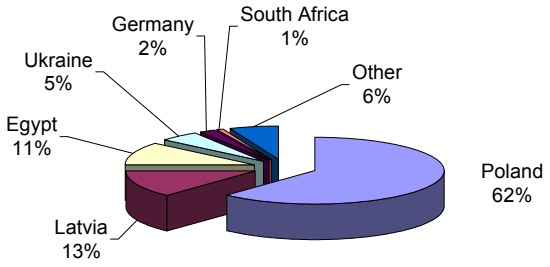


**Fig. 2**. Structure of MAP imports in Lithuania (1996-2001).

Cultivation is one of the solutions to the problem of over-exploitation of wild species. During recent years an ever-increasing number of the rural population became involved in medicinal plant cultivation. The cultivated production covers only 4-6% of the demand for MAP raw material. The main plants under more intensive cultivation are caraway, valerian, marigold, chamomile, peppermint, and lemon thyme.

- **Legislation**

The harvest and trade of MAPs are regulated by the *Law on Protected Areas* (1993, 2001), *Law on Endangered Wildlife* (1996), *Law on Wild Vegetation* (1999) and *Law on Plant Genetic Resources Conservation* (2001). Recently, a draft law on the ratification of the *Convention on International Trade in Endangered Species of Wild Fauna and Flora* was prepared. The Ministry of Environment regulates the gathering of wild MAPs and revises the list of species whose collecting is prohibited on the basis of research carried out by specialists from the Institute of Botany. The State Register (2000) of the *Law on Wild Vegetation* lists MAP species for which gathering from the wild indicated existing or potential conservation problems and whose collecting is therefore limited. Some medicinal plants are included in the National Red Book and are subjected to national legislation (Box 1).

**Box 1.** List of MAP species regulated by law in Lithuania

| **Threatened MAP species whose collecting from the wild is regulated by law** | **MAP species included in the National Red Book** |
|---|---|
| *Acorus calamus* | *Arnica mntana* |
| *Arctostaphylos uva-ursi* | *Allium angulosum* |
| *Angelica archangelica* | *Allium vineale* |
| *Centhaurium erythraea* | *Allium scorodoprasum* |
| *Centhaurium pulchellum* | *Allium ursinum* |
| *Cetraria islandica* | *Arctium nemorosum* |
| *Chimophila umbellata* | *Gentiana cruciata* |
| *Digitalis grandiflora* | *Hedera helix* |
| *Gentiana cruciata* | *Hypericum montanum* |
| *Hierochloe australis* | *Hypericum hirsutum* |
| *Hierochloe odorata* | *Mentha longifolia* |
| *Helichrysum arenarium* | *Polemonium coeruleum* |
| *Lycopodium clavatum* | *Pulmonaria angustifolia* |
| *Origanum vulgare* | *Salvia pratensis* |
| *Potentilla erecta* | |
| *Poleminium caeruleum* | |
| *Primula veris* | |
| *Plantago arenaria* | |
| *Sanguisorba officinalis* | |
| *Viscum album* | |
| *Viola tricolor* | |

- **Conservation methods**

Different methods are used to avoid potential danger and existing threats to MAP species, i.e. *in situ* and *ex situ* conservation. The biological peculiarities of species and their sources of raw material determine the conservation method. MAP species have been divided into three groups according to the source of raw material: sufficient, limited and insufficient (Table 1):

1. For species with sufficient sources of raw material, the *Law on Wild Vegetation* regulates the exploitation of common species.
2. Widespread species characterized by a large phenotypic diversity but with low sources of raw material can be or are cultivated. Wild populations of these species are not endangered in their natural habitats; however the sampling of their diversity has been initiated to ensure the conservation of their germplasm and further use in breeding.
3. Species with low sources of raw material and narrow ecological adaptation, which are difficult to introduce into cultivation, are in the most critical situation. This group of species includes rare and endangered plants. The main factor causing the decrease in populations of these species is the changing environmental conditions.

**Table 1.** Medicinal and aromatic plants of commercial interest grouped according to the sources of raw material

| Sources of raw material | Species |
|---|---|
| 1. Sufficient | *Achillea millefolium, Artemisia vulgare, Artemisia absinthium, Calluna vulgaris, Crataegus* sp., *Epilobium angustifolium, Equisetum arvense, Filipendula ulmaria, Frangula alnus, Fragaria vesca, Glechoma hederacea, Hypericum perforatum, Humulus lupulus, Ledum palustre, Menyanthes trifoliata, Pulmonaria officinalis, Rubus idaeus, Tilia cordata, Tussilago farfara, Taraxacum officinale, Urtica dioica, Vaccinium vitis-idaea, Vaccinium myrtillus* |
| 2. Limited | *Arctostaphylos uva-ursi, Arctium lapa, Arctium tomentosum, Agrimonia eupatoria, Angelica archangelica, Bidens tripartite, Chamomilla recutita, Chelidonium majus, Cichorium intybus, Convallaria majalis, Helichrysum arenarium, Inula helenium, Leonurus cardiaca, Malva sylvestris, Oenothera biennis, Origanum vulgare, Plantago arenaria, Primula veris, Plantago major, Polygonum bistorta, Potentlla erecta, Sambucus nigra, Symphytum officinale, Tanacetum vulgare, Viburnum opulus, Verbascum nigrum, Valeriana officinalis, Viola arvensis, Thymus serpyllum* |
| 3. Deficient | *Acorus calamus, Arnica montana, Centhaurium erythraea, Cetraria isandica, Drosera anglica, Gentiana cruciata, Gnaphalium uliginosum, Herniaria glabra, Herniaria hirsuta, Lycopodium clavatum, Mentha longifolia, Petasites officinalis, Polygonum aviculare, Polemonium ceruleum, Viola tricolor, Viscum album* |

### *In situ* conservation

This method of conservation is used mainly for those species which are most endangered or for which conservation problems were identified. *In situ* conservation can be implemented using the existing protected areas that contain a broad range of MAP germplasm material. Changes in the distribution and survival of natural populations have been observed in recent years. The large areas of wasteland previously cultivated or owned by the military are currently used for gathering of raw material.

The monitoring of the vitality of populations, coenodiversity changes and regeneration has been carried out on *Allium* L. spp., *Arctostaphylos uva-ursi* L., *Arnica montana* L., *Centhaurium erythraea* Rafn., *Origanum vulgare* L. and *Menyanthes trifoliata* L.

- The distribution area of ***Arnica montana*** originates in the European mountain province and the northern border of its distribution area crosses the Lithuanian territory. In Lithuania it grows only in the southern and southeastern parts of the country (Fig. 3). It was observed that populations of *Arnica montana* growing in optimum lichen-type pine stands have not decreased and are potential sources for their exploitation (Radušienė *et al.* 1997).

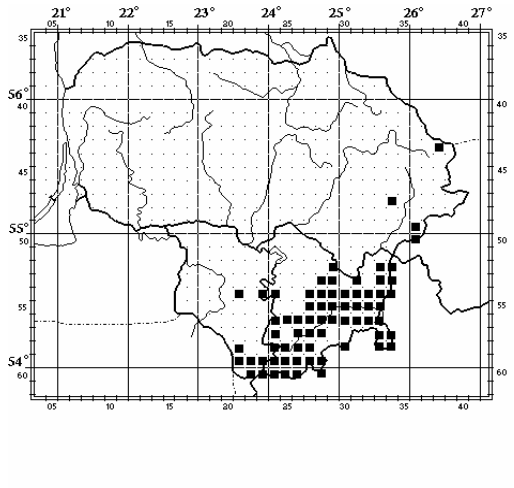


**Fig. 3.** Distribution of *Arnica montana* L. indigenous populations in Lithuania.

- The collecting of ***Arctostaphylos uva-ursi*** raw material increased in former military areas where populations of high vitality have been observed. In other locations the collecting of raw material seriously damages the stands of this plant.

- ***Centaurium erythraea*** is native to oceanic Europe and the Mediterranean, western Asia and northern Africa, and naturalized in North America. It is one of the traditional panaceas. Wild populations of centaury fluctuate greatly in size and are short-lived. They are greatly endangered by collecting and destruction of natural habitats. The fluctuations are due to the limited competitive ability of the plant and depend highly upon the environment (Radušienė 1995). Several attempts have been made to cultivate centaury, but the results were unsatisfactory.

- It was revealed recently that sources of the widespread species ***Menyanthes trifoliata*** have decreased significantly of late. The vitality of this plant in most stands is moderate or low. This fact indicates changes in moist habitat conditions that threaten the populations of *Menyanthes trifoliata.*

- ***Origanum vulgare***, in spite of its economic importance, is often referred to as an underutilized taxon, in the sense that its genetic resources and variability have not yet been fully explored (Padulosi 1997). The research on germplasm conservation is very limited outside the Mediterranean region where most resources of oregano are concentrated. The whole genepool of oregano should be conserved to have a representation of the diversity of this species. The populations of oregano in Lithuania are characterized by a limited distribution and low sources of raw material. The majority of populations are concentrated in southeast and east Lithuania (Fig. 4). The initial step of conservation of oregano consisted in identification of growing sites, gathering of wild accessions, their characterization and introduction into the field, further evaluation and multiplication of the selected material.

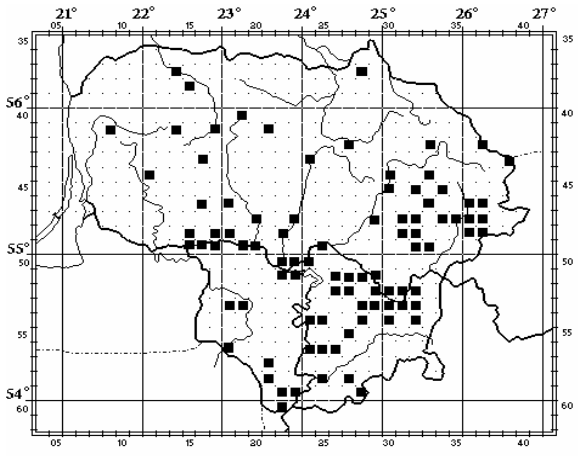


**Fig. 4.** Distribution of *Origanum vulgare* L. indigenous populations in Lithuania.

- All native species of ***Allium*** are subject to germplasm conservation in Europe. There are seven spontaneous species of *Allium* in Lithuania. Four of these, **A. *angulosum* L., *A. scorodoprasum* L., *A. ursinum* L. and *A. vineale* L**. are included in the National Red Data Book. In Lithuania *A. angulosum* stretches over the northern border of its distribution area. Only two sites of this species have been found in water meadows. *A. scorodoprasum* is under imminent threat. It is found in over 20 localities, mostly situated in protected areas in the western part of Lithuania. Only a few sites of A. *vineale* are known. The first three species are under imminent threat, while *A. ursinum* has been found in 50 localities all over the country. *A. ursinum* covers large areas (up to 10 ha) as pure stands in broadleaved forests or as isolated plants (Fig. 5).

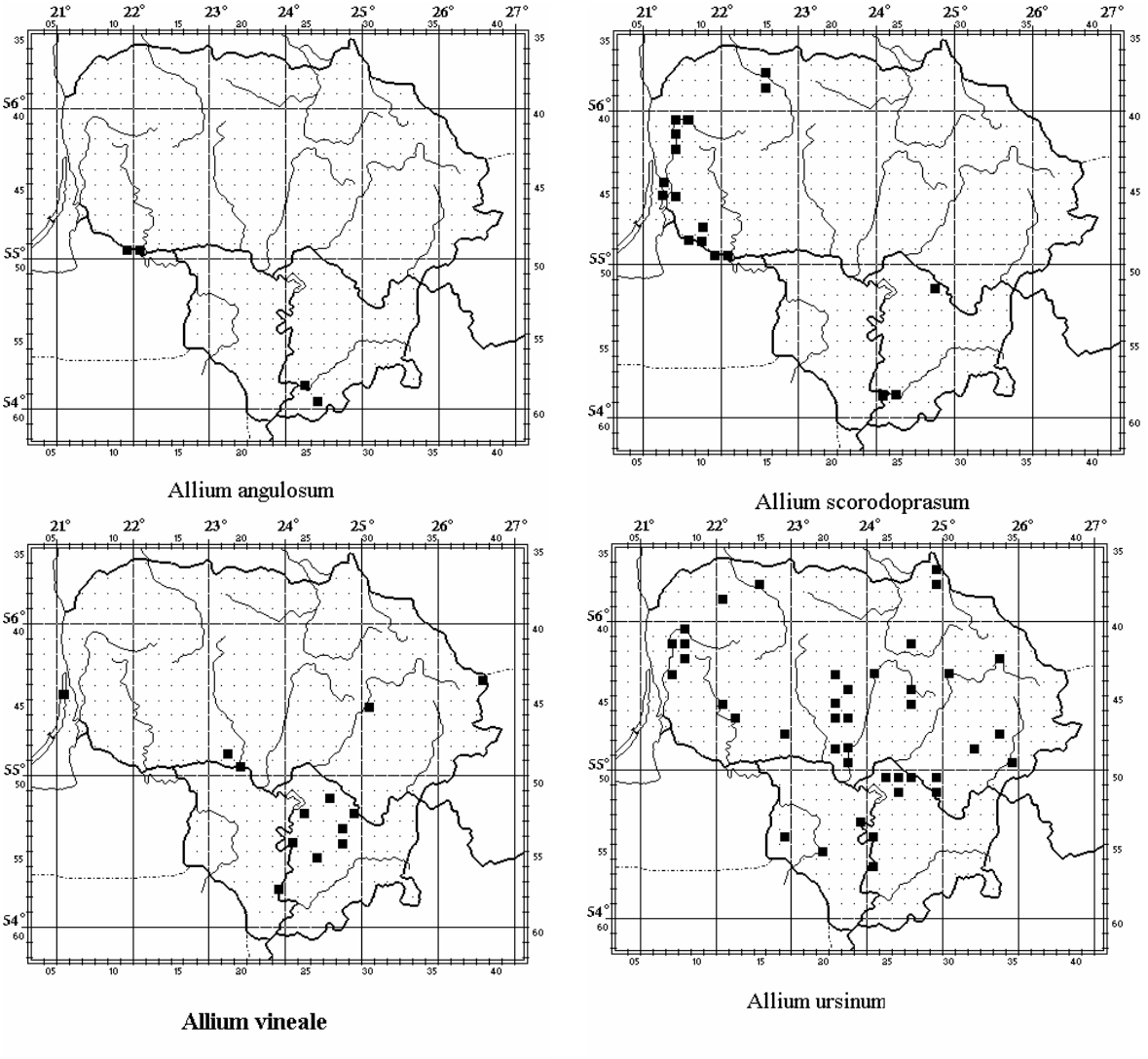


**Fig. 5.** Distribution of threatened *Allium* L. species in Lithuania.

***Ex situ* conservation**

Conservation of MAPs in field collections is carried out at the Institute of Botany, at the Kaunas Botanical Garden of Vytautas Magnus University, and at the University of Agriculture.

Target species for *ex situ* germplasm conservation are the following: *Achillea millefolium* L., *Allium oleraceum* L., *Carum carvi* L., *Helichrysum arenarium* L., *Hypericum perforatum* L., *H. maculatum* Cranz, *Humulus lupulus* L., *Origanum vulgare* L., *Thymus pulegioides* L., *T. serpyllum* L. and *Viola tricolor* L.

- ***Helichrysum arenarium*** is a perennial herbaceous plant native to Europe and West Asia. It is one of the most frequently used plants in Lithuanian pharmaceutics. As gathering from the wild has threatened the populations of this species and does not satisfy the market demand, there is a need for its conservation and cultivation *ex situ*. *H. arenarium* belongs to the species of rather easy introduction and acceptable conservation in field collections. The field collection holds 10 accessions of this species. The plants of the same population showed a wide range of variation of yield-related characters such as mass of inflorescences, number and colour of flowers and plant height (Radušienė 2002). The high phenotypic variation of *H. arenarium* may allow selection of the most valuable accessions.

- Investigations on ***Hypericum perforatum*** and ***H. maculatum*** described the variation of morphological and chemical characters in wild populations and field accessions (Bagdonaitė *et al.* 2000; Radušienė and Bagdonaitė 2001; Radušienė 2002). The expression of morphological diversity in wild populations and field accessions of *Hypericum* is influenced by both environmental and genetic factors, while chemical variability is based mainly on the genetic background. The content of flavonoids and hypericin varies greatly between species, different accessions, and parts of the plant. Significant differences were detected in the mean concentrations of quercetin, hypericin, rutin and hyperoside+isoquercetrin in flowers and leaves of both species. The flavonoid content appeared to be the most constant character in both species. The poorest growth and lowest mean concentrations of secondary metabolites were found in *H. maculatum.*

- The oregano collection was established in the MAP field collection at the Institute of Botany. It holds 15 accessions gathered from indigenous populations and 4 from local cultivation. The results of primary evaluation on ***Origanum vulgare*** indicated a high heterogeneity within the material studied. The accessions of oregano significantly differed in all measured morphological and yield-related characters (Radušienė *et al.* 2002). The results of essential oil analysis show a rich composition and high variation of its compounds. Furthermore, comparison showed that the values of oil yield obtained from wild populations and field accessions were not significantly different.

- The studied morphological variation of thyme resulted in the identification of 8 varieties of ***Thymus pulegiodes*** and 4 varieties of ***T. serpyllum*** in the wild (Ložienė and Vaičiunienė 1999). Introduction of 26 wild populations of thyme in the field demonstrated that the morphological differences were not stable with the change of conditions. Chemical analysis revealed a wide variability in the content and composition of essential oil of thyme. Different populations of *T. serpyllum* and *T. pulegioides* yielded from 0.25-0.11% to 0.78–1.20% of essential oil respectively (in absolute dry matter of the leaves+flowers fraction). A total of 80 constituents were identified in *T. pulegioides* and 75 in *T. serpyllum* (Ložienė *et al.* 2002). The chemical variability among thyme accessions was

characterized by a significant presence of 5 chemotypes in *T. pulegioides* (thymol, carvacrol, geraniol, linalool and α-terpenyl acetate) and 4 chemotypes in *T. serpyllum* (1,8-cineole, dihydrocarvone, germacrene and (Z)-β-ocimene). The chemical variability of thyme remained the same in the field and could be predetermined genetically.

- In selecting ***Achillea millefolium*** accessions we tried to sample a wide range of environmental factors to obtain as many different genotypes as possible. The variation in total essential oil content in *A. millefolium* field accessions was high: 0.08-0.78% of dry mass in inflorescences and 0.05-0.13% in leaves. Wide variation was found in the content of proazulenes among *A. millefolium* populations. Some of them contain no proazulenes while others vary widely in their content of these substances. Proazulene-free and proazulene-containing individuals occurred at the same site. The quantitative variation of proazulenes in natural populations of the *A. millefolium* group and its relationship with abiotic factors and phytosociological releves has been examined.

- Anthropogenic and technological factors have caused a decrease in ***Carum carvi*** habitats (natural meadows) and a reduction in genetic resources of this species. The field collection of caraway at the University of Agriculture holds 130 accessions, which vary in blossoming time, colour of inflorescence and amount of volatile oil. Two different forms – pasture and meadow – have been distinguished according to morphological characters (Petraitytė *et al.* 2001).

## Conclusion

It seems obvious that many of the indigenous species will continue to be collected from the wild in the future, as their cultivation is often difficult due to their biological peculiarities. Different possibilities for avoiding potential dangers and existing threats to MAP species include legislation and conservation projects. MAP germplasm conservation ought to be carried out jointly with the research and plant breeding sectors. The material appeared to be phenotypically quite variable and therefore needs to be further studied, using more sophisticated methods. The germplasm of evaluated species may be a potential source of genetic variation to allow selecting valuable material for breeding.

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## *Status of medicinal and aromatic plants in the Republic of Macedonia F.Y.R.*

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The Republic of Macedonia is a small, landlocked Balkan country in southeastern Europe, bordered by Yugoslavia, Bulgaria, Greece and Albania. Largely surrounded by high mountains, much of the country is a plateau more than 1000 m above sea level. The western part of the country, with its alpine climate, and the eastern part, with its continental climate, are divided by the largest river, the Vardar, rising in the north and flowing south to Greece and into the Mediterranean Sea, drawing the influence of a sub-Mediterranean climate deep into the continental territory. This predominantly mountainous country is made up of valleys, gorges, plateaux and highlands. The territory of Macedonia includes:

| | | |
|---|---|---|
| - | Pastureland | 25% |
| - | Grassland and meadows | 2% |
| - | Arable land, vineyards, orchards | 23% |
| - | Barren land | 8% |
| - | Forest land | 37% |
| - | Lakes | 2% |
| - | Urban or industrial land | 3% |

Four phytogeographic regions can be distinguished, characterized by their different climates and altitudes and consequently by their different flora:
- sub-Mediterranean region (40% of the territory): Mediterranean plants dominate, growing mainly up to 600 m;
- sub-continental region (37% of the territory): it lies mainly between 600 and 1200 m and is subject to some Mediterranean influence and is thus not completely continental;
- sub-humid region (22% of the territory): between 900 and 1700 m with mainly forest vegetation;
- sub-alpine region (1% of the territory): may be characterized as a real alpine region (above 2200 m).

In spite of its small territory (25 333 km$^2$), Macedonia is very rich in plant diversity, having approximately 3500 vascular plant species, and is at the top of the list of countries called "European hotspots". According to data from local and neighbouring countries' folk medicine there are more than 700 plants with medicinal properties, but only about 150 species are used frequently. All these approximations are due to the fact that the inventory of the flora of Macedonia is still not complete; therefore an exhaustive inventory of the medicinal and aromatic plants (MAPs) is not yet available. The Department of Pharmacognosy, Phytochemistry and Pharmaceutical Botany of the Faculty of Pharmacy in Skopje, through its scientific research and educational activities in this field, has gathered a significant amount of data.

For better consideration of the overall situation of MAPs in Macedonia, it has to be mentioned that Macedonia was the last of all the former Yugoslavian republics to acquire its independence, withdrawing from Yugoslavia in 1991. This is not a very long period for establishing relevant legislation. Some laws are still those that were in force in the former Yugoslav legislation, most of them dating from the early 1970s. There is no law for MAPs or specific regulation contained in any other law. The only regulation is found in one half-page

article (art. 42 of the *Law of the Forests,* 1997), classifying MAPs as "other forest products", regulating almost nothing. Therefore Macedonia is missing a fully comprehensive legislation regulating collecting, trade, cultivation, manufacturing, quality control, maintenance, preservation, conservation and evaluation of indigenous and introduced MAPs.

Research studies show that more than 20% of the territory is of special interest regarding natural heritage, but only 6% is protected by the *Law for protection of natural rarities*.

*In situ* conservation of medicinal and aromatic plants, not exactly specified as MAPs, but as part of the whole flora, can be found in the three national parks, with a total area of 108 000ha; they are regulated by the following laws:

- Law for National Park "Pelister" (1948)
- Law for National Park "Mavrovo" (1949)
- Law for National Park "Galichica" (1958)

These laws date from the Yugoslavian regime and are not in accordance with IUCN categories for protected areas but there are initiatives and efforts dedicated to changing this situation.

The Macedonian legislative body has ratified the following international regulations/conventions:

- Convention on Biological Diversity (CBD), Agenda 21 (Rio de Janeiro, 1992) ratified in 1998. So far, a National Coordinator (focal point) has been nominated and a National Committee for Biodiversity has been formed and is functioning;
- Convention on the Conservation of European Wildlife and Natural Habitats (CCEWNH or Bern Convention, 1982) ratified in 1999. A National Coordinator (focal point) operates and a National Committee for the Bern Convention is being formed.
- Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES) (Washington, 1973) ratified in 2000; a National Coordinator (focal point) has been designated.

As a permanent member of the CBD, Macedonia is liable to prepare a national biological and landscape diversity conservation strategy and an appropriate legislative background. These conventions being ratified, they are considered as laws in Macedonia. Beside these, no strategy has yet been prepared (the first phase, dealing with data evaluation, is currently underway), nor any legislative background.

In the Republic of Macedonia, there is no *ex situ* conservation of medicinal plants, because of the lack of a national strategy in this field. Yet there are facilities at the Institute for Southern Crops in Strumica, which could easily be adapted and used for this purpose.

The most common way of providing herbal raw materials is traditional collecting of medicinal plants from the wild. Organized collecting from natural resources has existed for a long time but the people involved do not always have the necessary skills. The collectors are mainly private individuals, especially villagers in the vicinity of collecting areas. In most cases collecting is done to meet a demand for certain species. Only a few companies in the Republic of Macedonia can be regarded as serious and important in this business. The most important and by far the largest pharmaceutical company which buys aromatic plants in Macedonia, and the only one to have a permanent list of required medicinal plants and eight buying stations throughout Macedonia, is "Alkaloid". In addition, 4-5 smaller companies are involved in business with MAPs, mainly packing tea-bags. From 90 to 95% of the total plant material used in the pharmaceutical industry is of wild origin, 80% being of native origin and 20% imported.

One of the greatest threats for wild medicinal flora is due to the large requirements for certain species from abroad, guided by very small companies with incompetent personnel. Likewise, the presence of *Gentiana* spp., of all Orchidaceae bulbs (which can be used in the production of salep), of *Sideritis scardica, Sideritis syriaca, Arctostaphylos uva-ursi* and others, on the buying list with the most tempting prices, endanger these species. Recently, the Ministry of the Environment has regulated the export of 50 plant species from Macedonia, including many medicinal plants.

Despite the very good climate and soil for cultivation, today in Macedonia the cultivation of medicinal plants on a large scale has been abandoned. Some smaller companies are trying to organize cooperative contracts with individual farmers, supplying them with raw material and seeds with an obligation to buy their herbal material. This kind of cultivation has appeared only recently; it receives no support from the government and the acreage under medicinal plants at the moment can be estimated at approximately 50 ha.

## *Status of medicinal and aromatic plants in Malta*

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### Introduction

Although the Maltese Archipelago has a restricted environment due to its small size, the diversity in biological species is vast. The variation in biodiversity is mainly linked to the connection of the warm North Africa to the cold southern Europe. The bioclimatic conditions of Malta are also found in coastal regions of other Mediterranean countries. In fact, about 66% of the flora of Malta is common to other Mediterranean regions. Common medicinal flora of the Mediterranean region and Malta include conifers (*Pinus halepensis* and *Cupressus sempervirens*), broadleaved trees (*Laurus nobilis, Morus nigra, Tamarix gallica* and *Rhus coriaria*), fruit trees (*Ceratonia siliqua, Ficus carica, Punica granatum, Cercis siliquastrum, Nerium oleander, Citrus aurantium* and *Olea europea*) and others (*Allium sativum, Aloe ferox, Arbutus unedo, Capparis spinosa, Myrtus communis, Opuntia ficus-indica, Origanum vulgare, Papaver somniferum, Phytolacca decandra* and *Pistacia lentiscus*). The other 34% of the flora is adapted to cold European conditions (*Populus alba, Salix* spp. and *Crataegus monogyna*) and subtropical conditions (*Ceratonia siliqua, Ficus carica, Myrtus communis* and *Cynomorium coccineum*).

There are about 1264 vascular plant species, including casual or naturalized aliens. Of these, 458 species possess medicinal value and have been used in traditional medicine for several ailments (Lanfranco 1975, 1993) particularly related to the Mediterranean region. These refer to the nervous system (15%), gastrointestinal (24%), cardiovascular (14%), dermatological (13%) and antimicrobial (4%). Predominating medicinal plant families include Compositae (15%), Labiatae (7%), Leguminosae (6%), Rosaceae (4%), Umbelliferae (4%), Liliaceae (3%), Solanaceae (3%), Cruciferae (3%), Caryophyllaceae (2%), Ranunculaceae (2%) and Papaveraceae (1%). These aromatic and medicinal herbs are found in small patches throughout the country (Haslam *et al.* 1977).

### Medicinal and aromatic plant status

A considerable number of medicinal plants are under threat, mainly not due to overuse but to degradation by human activities including building and agricultural practices. Table 1 shows the status of the threatened population of medicinal plants in relation to the local threatened plant species (Lanfranco 1989).

**Table 1.** Medicinal plants under threat in Malta (adapted from Lanfranco 1989)

| | Extinct | Endangered | Vulnerable | Rare | Indeterminate | Total |
|---|---|---|---|---|---|---|
| Medicinal plants | 9 | 9 | 5 | 20 | 1 | 44 |
| Total species | 109 | 74 | 44 | 137 | 34 | 398 |

### *Ex situ* conservation

In Malta, few *ex situ* conservation measures are undertaken for medicinal plants. However, the Institute of Agriculture, University of Malta in collaboration with the Micropropagation Centre, Department of Plant Health, has looked into conservation of medicinal plants through micropropagation. Another area of interest for medicinal plants is their yield of secondary metabolites under *in vitro* and *in situ* conditions (Cassar 1998; Attard 2001). In the near future, the threatened species will be considered on an individual basis in order to regenerate their populations. The University Botanic Gardens and the Government's nursery

at Wied Incita have propagated a large variety of plants. In fact threatened medicinal plants are found both at the Botanic Gardens and on the University Campus, and in public gardens or public areas. Amongst these are included plants such as *Ephedra fragilis*, *Paleocyanus crassifolia*, *Rosmarinus officinalis*, *Vitex agnus-castus*, *Quercus ilex*, *Coronilla emerus*, *Viola odorata*, *Aloe vera*, *Pancratium maritimum*, *Salvia triloba*, *Spartium junceum*, *Centranthus ruber*, *Laurus nobilis*, *Ruscus hyphophyllum*, *Tamarix africana* and *Orchis morio.*

### Inventory of MAP natural resources

A Medicinal and Aromatic Plant Database was initiated in 1995 with the emergence of a MAP project at the University of Malta consisting of a multidisciplinary research group. Throughout the years collection of data on the local medicinal flora has led to the creation of another more powerful and user-friendly database at the Institute of Agriculture, University of Malta. This database is multidisciplinary with the main focus on:

- plant nomenclature
- botanical description
- local and foreign ethnobotanic uses
- description of the natural habitat and cultivation requirements
- plant pests and diseases
- local, Mediterranean and worldwide distribution
- plant toxicity to humans and animals
- chemical constituents and pharmacological uses.

To complement this, a photo gallery is being compiled alongside the database.

### Review of the acreage of MAPs under cultivation and use of MAPs by processing industries

In Malta, commercial natural products, found in pharmacies, are mainly obtained from European countries. Due to the lack of space available for agricultural practices, to date no medicinal or aromatic plants are being cultivated for purposes other than for culinary uses. Considering the entry of MAP-related produce into the national marketplace for the year 2001 (Table 2), it can be noted that the local cash crop, the potato tuber, is the most cultivated crop. Besides, the most prominent export for the Maltese Islands is the potato export to the Dutch markets, with a total of 6069 tonnes for the year 2001 (PQS 2002).

The local medicinal and aromatic plant industry is still very young. Attempts have been made to set up an industry but due to marketing reasons, commercial problems outweigh the feasibility of such projects.

**Table 2.** Entry of MAP-related produce for the year 2001 into the national marketplace (Anonymous 2002)

| Produce | Weight (kg) |
|---|---|
| *Allium cepa* | 1319637 |
| *Allium sativum* | 124941 |
| *Anethum graveolens* | 1916 |
| *Capparis spinosa* | 91 |
| *Capsicum* spp. | 36436 |
| *Ficus carica* | 16641 |
| *Foeniculum vulgare* | 838 |
| *Morus alba* | 39 |
| *Morus nigra* | 1725 |
| *Ocimum basilicum* | 4857 |
| *Passiflora caerulea* | 220 |
| *Petroselinum crispum* | 332 |
| Potato tuber | 2625209 |
| *Thymus vulgaris* | 85 |
| Other herbs | 19308 |

## Conclusion

Although the Maltese Islands have a high level of biodiversity, the limited land area lowers the potential for large-scale cultivation of medicinal and aromatic plants. Therefore the main aim, as regards medicinal and aromatic plants, is to sustain and conserve the genetic stock by *in situ* and *ex situ* methods. This includes afforestation projects involving, for example, *Coronilla emerus* and *Spartium junceum*. Also efforts are made in order to promote alternative crops (MAPs) that might be the starting material for processing industries.

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## *Medicinal and aromatic plants in the Nordic Countries*

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### MAPs at the Nordic Gene Bank

Within the Nordic countries the production and the collection of herbs and medicinal plants is quite low. During recent years the interest has increased.

In 1985, a working group for spices and medicinal plants was established at the Nordic Gene Bank. Among other working group activities, the group collected information in the area of medicinal and aromatic plants (MAPs). Only a few accessions of material where collected and conserved at the Nordic Gene Bank. Unfortunately, the group was closed down in 1993 due to lack of resources. Before it was closed down, the working group produced a study on Nordic medicinal plants, covering information such as content of chemical substances in the plants, distribution, uses and recommendations for conservation. When the working group was closed down the material and the responsibility were carried over to the working group for vegetables. Table 1 lists the MAP species conserved at the Nordic Gene Bank.

**Table 1.** MAP material stored at the Nordic Gene Bank (ACC = accepted; TEM = temporary)

| Species | | Denmark | | Finland | | Iceland | | Norway | | Sweden | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | ACC | TEM | ACC | TEM | ACC | TEM | ACC | TEM | ACC | TEM |
| *Anthriscus cereifolium* | Chervil | 2 | | | | | | | | | 1 |
| *Carum carvi* | Caraway | 1 | | | | | | | 3+3 | | |
| *Angelica archangelica* subsp. *archangelica* | Angelica | | | | | | | | 1 | | 1 |
| *Levisticum officinale* | Garden lovage | | | | | | | | | | 1 |
| *Solanum nigrum* | Garden nightshade | | | | | | | | | | 1 |
| *Petroselinum crispum* var. *crispum* | Parsley | 11 | 7 | | | | | | | 2 | |
| *Anethum graveolens* | Dill | 4 | 3 | | | | | | | 5 | |
| *Humulus lupulus* | Hops | 19 | | 10 | | | | 31 | | 32 | |

### The SPIMED project

However, during the late 1990s interest in this material increased and in 2000 the working group for vegetables initiated a project called SPIMED (SPIce and MEDicinal plants). It will run for five years (2001-2005) and is supported by the Nordic Gene Bank. The full title of the project is *"Spice and medicinal plants in the Nordic and Baltic countries. Strategies for conservation of genetic resources in minor crops"*. Its aim is to develop strategies for conserving cultivated spice and medicinal plants *in situ* or *ex situ* in the Nordic and Baltic countries. The three Baltic countries (Estonia, Latvia and Lithuania) are participating in the project as well as Norway, Denmark, Finland and Sweden; in 2003, the project will expand to include Iceland.

Planned activities in the project are:

1. To establish a priority list of species to be included in the project; list existing *in situ* populations of these species;
2. Consider if these populations are safe in terms of plant genetic resources conservation;
3. Collect threatened species/populations for *ex situ* conservation;
4. Develop descriptors for characterization of the species on the priority list;
5. Characterize the collected material according to these descriptors;
6. Develop systems for documentation of *in situ* conservation;
7. Map genetic variation in the species;
8. Develop strategies for conservation, *in situ* or *ex situ*.

The project is divided into two parts, one dealing with the establishment of collections and characterization of the material, the other with conservation strategies.

Table 2 shows the species on the A-list, selected according to the results from the different country reports, with priority given to these species.

**Table 2.** Species on the A-list and the different countries' selection of priority plants

| Species | Estonia | Denmark | Latvia | Finland | Lithuania | Sweden | Norway | Iceland |
|---|---|---|---|---|---|---|---|---|
| *Acorus calamus* | | | | X | | | | |
| *Arnica* sp. | | | | X | | X | | |
| *Helichrysum arenarium* | X | | | | X | X | | |
| *Hypericum* sp. | | X | | | X | | | |
| *Origanum vulgare* | X | X | X | | X | | X | |
| *Rhodiola rosea* | | | | X | X | X | X | X |
| *Thymus* sp. | | X | X | | X | | | X |
| *Valeriana officinalis* | | X | | | | X | | |

The purpose of this work is to describe genetic diversity within species with value as spice and medicinal plants in our region, mainly based on morphological characterizations in the collected material.

These species cover differences in:

1. geographical distribution (southern/northern, montane/widely distributed)
2. Red List status (rare/common)
3. origin (indigenous wild/cultivated/subspontaneous)
4. use (traditional use/actual commercial production/potential for use)
5. purpose (medicinal/spice)
6. need for conservation activities

This year [2002] will be dedicated to the establishment of collections. The plants will be collected both as seeds and living plants from different regions and if possible different kinds of habitats.

The delegates from different countries will be responsible for developing descriptor lists for the species. The second part of the project is to investigate which species are threatened and consider needs for action and strategies for conserving such species.

### Status report for Denmark, Norway and Sweden

A short summary of the status reports from Denmark, Norway and Sweden is given below. The results from this year's work (2002) are not yet ready. This part only covers the above-mentioned countries.[15]

- **Denmark**

During the summer, Denmark has collected *Origanum, Hypericum, Thymus* and *Valeriana* in order to establish collections.

In Denmark there is a production of freeze-dried herbs and spices for the processing industry, e.g. *Anethum graveolens, Petroselinum crispum* var. *crispum, Thymus* sp., *Ocimum basilicum* and *Satureja montana*. There is also an increased production of small pot-plants as a fresh supply of spices and herbs. The most common are *Petroselinum crispum* var. *crispum, Anethum graveolens, Ocimum basilicum, Thymus* sp., *Origanum vulgare, Melissa officinalis, Artemisia dracunculus, Rosmarinus officinalis* and *Mentha* sp.

[15] For Finland, see detailed report by B. Galambosi, following paper.

- **Norway**

A collection of *Origanum vulgare* has been started at the Norwegian Crop Research Institute. Norway will also work further on the recently established *Rhodiola* collection, which consists of 90 clones. In 2000 material from *Chrysanthemum vulgare* from 40 different places in Norway was collected as well as 10 clones of *Mentha* x *piperita*.

- **Sweden**

The production of medicinal and aromatic plants is quite small in Sweden but it has increased during recent years.

In 2000, the production of herbs grown in the field covered an area of 236 ha (154 ha of *Anethum graveolens*, 43 ha of *Petroselinum crispum*, 18 ha of *Amoracia rusticana* and 21 ha of different other kinds of herbs) (Anonymous 2000).

The greenhouse production of herbs more than doubled between 1999 and 2000, from 11 000 plants to 26 200 (Anonymous 2001).

Within the SPIMED project, material from the following species was collected during the summer: *Arnica* sp., *Helichrysum arenarium, Rhodiola rosea* and *Valeriana officinalis.*

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## *Medicinal and aromatic plants in Finland*

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Despite the severity of the environmental conditions in Finland, more than 100 medicinal and culinary herbs grow wild over the entire country during the relatively short but intensive photoassimilation period (Hälvä 1988). Knowledge of the utilization of these plants varies with the species and part of the country. Quite a limited variety of natural, wild-growing herb plants have been used since the early days of Finland.

Historically, collecting and cultivation of herbs have been occasionally promoted and many new species have been introduced in Finland. During the last 15 years intensive research and education programmes have been carried out to promote the production of herbs as alternative crops (Galambosi 2000). At the same time, attention has been focused on the protective management of those threatened plant species that have some practical value as medicinal plants.

### Legal protection of medicinal and aromatic plants (MAPs)

According to the present Finnish law, the following plant species have been assigned endangered or protected status in Finland (Table 1). The practical and commercial importance of these species as medicinal plants is variable.

**Table 1.** Endangered and protected plant species in Finland

| Category | Species |
|---|---|
| Critically endangered plants (CR) | *Pimpinella major, Rosa canina* |
| Endangered species (EN) | *Agrimonia pilosa, Arctium nemorosum* |
| Vulnerable (VU) | *Arnica angustifolia*, *Asarum europaeum*, *Carlina vulgaris, Crataegus monogyna, Galium verum*, *Ononis arvensis* |
| Near-threatened (NT) | *Allium ursinum*, *Anchusa officinalis*, *Cinna latifolia, Galium odoratum, Dryopteris fragrans*, *Drosera intermedia*, *Allium schoenoprasum* var. *sibiricum* |
| Regionally threatened species | |
| - regionally threatened | *Agrimonia eupatoria, Arctium lappa, Convallaria majalis* |
| - regionally protected | *Angelica archangelica* subsp. *archangelica* |
| Restricted collecting | *Daphne mezereum*, *Hepatica nobilis*, *Hippophae rhamnoides*, *Juniperus communis, Primula veris* |

### Inventory of MAP natural resources at national level

Medicinal plants have not been inventoried officially in Finland. According to personal communications from different experts (P. Alanko, University of Helsinki, 2001; A. Alanen, Finnish Environmental Centre, Helsinki, 2002) more attention should be paid to the following species, because their populations are decreasing in Finland: *Acorus calamus*, *Antennaria dioica, Artemisia abrotanum*, *Hierochloa odorata* subsp. *odorata*, *H. odorata* subsp. *baltica, Pimpinella saxifraga*, *Isatis tinctoria, Leonorus cardiaca* subsp. *cardiaca, L. cardiaca* subsp. *villosa.*

### *Ex situ* conservation

In Finland there is no special *ex situ* conservation for medicinal herbs. During 1990-1994 the Nordic Gene Bank set up a specialist group to check the situation of MAPs in the Scandinavian countries. The group collected basic information about the most important medicinal plants used in the Nordic countries and the results have been published (Wahlin and Blixt 1994). No official activity was proposed for the protection and inventory of MAPs

at that time, but presently a project named SPIMED (SPIce and MEDicinal plants) has been started aiming to analyze more accurately the threatened status of MAPs in the Baltic and Scandinavian countries (Asdal 2001).

**Herb gardens and herb collections in Finland**

Historically herb gardens have had an important role in supplying medicinal plants and medical raw materials to pharmacies and doctors (Peldan 1967). Presently the function of herb gardens has changed: they are a tool for education and research, or serve for touristic purposes and special collections. These gardens may provide good possibilities for the maintenance and conservation of threatened or endangered medicinal plant species (Väre and Siuruainen 1994). At the moment the following herb collections exist in Finland:

- **Botanical gardens**

| | |
|---|---|
| - Botanical garden, University of Turku | 20100 Turku |
| - Botanical garden, University of Helsinki | 00014 Helsinki |
| - Botanical garden, University of Oulu | 90570 Oulu |

- **Agricultural schools and private farms**

| | |
|---|---|
| - Herb Garden of Lönnrot | 09220 Sammatti |
| - Kenkävero, Herb Garden | 50600 Mikkeli |
| - Tertin Kartano, Herb Garden | 50600 Mikkeli |
| - Lehtiniemen Kartano, Herb Garden | 57600 Savonlinna |
| - Luonnon Aromit, Herb farm | 88670 Sotkamo |
| - Rohto-ja yrttipuisto, Herb Collection and Garden | 42100 Jämsä |
| - Frantsila Herb Garden, Herb Collection and Garden | 39100 Hämeenkyrö |
| - Wetersin puutarha, Herb Farm, Collection | 25700 Kemiö, Kila |
| - Tuorlan koulutila, Herb Garden | 1500 Piikkiö |
| - Pukkilan kartano, Historical Herb Garden | 21500 Piikkiö |

- **Research institutes**

| | |
|---|---|
| - Agrifood Research Centre, Ecological production | 50600 Mikkeli |

**Collection of wildflower medicinal plants**

In Finland there are about 50 wildflower medicinal plants of commercial importance (Hälvä 1988). Accurate data on the quantities of collected wildflower medicinal plants is quite difficult to obtain. Many of these plants are collected for personal consumption by thousands of families. Numerous small local herb farms produce dry tea mixtures or other products. Finally, some producers of health food products are using larger quantities for further processing/extraction.

According to a recent review, only a few firms deal with commercial collection and processing of these raw materials. On the basis of a telephone interview and personal discussions, the quantity of dry wildflower medicinal plants can be estimated at about 4000-5000 kg/year (Galambosi 1996).

The most important wild medicinal plants are stinging nettle (*Urtica dioica*) and birch leaves (*Folium betulae*) with 1000-5000 kg collected each year. The quantity of dry plant material of *Calluna vulgaris, Juniperus* berries and *Achillea millefolium* flowers varies from 500 to 1000 kg/year, while 100-500 kg of dried raw material of *Solidago, Filipendula, Taraxacum, Epilobium* and *Vaccinium myrtillus* leaves are collected each year. About 15 other species are collected (less than 100 kg/year per species).

In addition to these species, there is a special product collected by the Oulu District of the Finnish 4H Association. During the last 10 years, 500-2200 kg of fresh sundew *(Drosera rotundifolia)* was collected from peatlands and marketed mainly to Switzerland, and partly domestically.

### Cultivated herbs

During the last 15 years, interest in and cultivation of herbs has increased significantly. The acreage of herbs in 1985 was 100 ha and in 2001 it reached 3600 ha. At present about 30 different herbs and medicinal plants are cultivated in Finland to various extents.

The most important herb is biannual caraway (*Carum carvi*), since its cultivation can be easily mechanized. The area grown in l994 was 1560 ha. In 2001 it had reached 3600 ha, spread over about 350 farms.

The second important spice seed is mustard (*Sinapis alba*). At its largest, in l991, the growing area was about 400 ha, but due to price and quality problems its cultivation has now nearly ceased. Presently the area of mustard is barely 50 ha.

The third important herb is the popular leaf dill (*Anethum graveolens*). In 1999 it was cultivated on 162 ha (323 farms) outdoors and on 8.2 ha indoors. Nearly all dill is used in fresh form for direct sale or in the processed food industry. Only a few tonnes are dried in Finland.

Parsley (*Petroselinum hortense*) is the fourth important herb. It is cultivated outdoors and indoors on a total of 20-28 ha, mainly for fresh consumption.

During the last 4-5 years, the cultivation of garlic (*Allium sativum*) has increased to 24 ha. Production is widely distributed and is carried out on small-scale plots. Nearly 170 farms are engaged in garlic cultivation.

About 10 different herbs, including some medicinal plants (e.g. *Coriander, Angelica, Oreganum*, *Mentha* sp., *Echinacea purpurea, Urtica* and *Artemisia dracunculus*), are cultivated on an area of between 1 and 5 ha/each. These plants are produced for further processing, for the health food industry or for the growers' own products. The harvested raw materials are dried, frozen or extracted.

About 20 different herb and medicinal plant species (*Hypericum, Hyssopus, Matricaria*, *Agastache, Calendula,* etc.) are cultivated on less than 0.5 ha/species.

### Use of medicinal plants by processing industries

Due to the small quantities and lack of information, only limited data are available about the industrial use of herbs and medicinal plants. The data presented here were obtained through personal contacts.

Nearly the whole caraway yield is exported from the country. Only 100-120 t of seeds are used as a spice by the domestic food industry companies yearly.

The domestic food industry uses dill mainly in fresh forms (leaf, flowering shoot) and only a few farms produce dried dill leaf packed for retail marketing (ca. 5 ha). Experimental distillation of caraway seed oil and dill plant oil has been carried out for industrial use.

Some mint species (*Mentha* x *piperita, M.* x *dalmatica*) are cultivated, mainly for dry leaf production and some experiments have been started for distillation of mint oils, mainly for aromatization of local herb products.

Fresh leaf production of stinging nettle has been started from both collecting in the wild and field cultivation for the production of special frozen food products.

The wild-collected sundew (*Drosera rotundifolia*) is exported in fresh form for the use of medicinal industry, mainly to Switzerland, but it is also used to some extent in Finland.

Bearberry leaves (*Arctostaphylos uva-ursi*) are collected from the wild for further industrial processing and the extract is used in the perfume industry abroad and in Finland.

Cultivation of several medicinal plants has been started by the requirements of the domestic medical industry. Alcoholic extracts, tablets and other special products are produced from *Echinacea purpurea, Solidago virgaurea, Rhodiola rosea, Plantago lanceolata,* etc. The quantities of these plants together are variable (ca. 15-30 t/year).

## Experiences in sustainable use of MAPs

- **Organic production**

Organic production is expanding continuously in Finland. In 1998 the total organic area controlled by the local authorities was 126 000 ha. Organic herb production is increasing, but is still very small: the organic and converting area was 71 ha in 1995, 121 ha in 1997 and 228 ha in 2001.

Since the health drug stores require organically grown raw materials, all medicinal plants are grown organically. Finnish consumers prefer herbicide-free products, therefore the local producers grow herbs using organic cultivation methods. Since weed control is not mechanized in organic cultivation, the production units and quantities produced remain small.

The areas of organically grown mustard, caraway and coriander are between 3-12 ha, dill and mint about 5 ha, and those of nearly 20 other herbs about 0.5-3 ha.

- **Sustainable collecting of herbs**

In the guide for wild herb collecting (Mäkinen *et al.* 1994) general instructions are given for sustainable collecting methods. These instructions were given in more detail in some cases, e.g. *Drosera rotundifolia*, which is collected in quite large quantities regularly.

During 1993-1999 the effect of regular collecting on the regeneration of natural populations of *Drosera rotundifolia* were studied (Galambosi *et al.* 2000). The results showed that after two years of collecting, the density of the plant populations and the available fresh yields had decreased by 50%. On the basis of these results, a new environmentally sustainable collecting method was developed and applied by the collectors.

As shown by the results of the experiment, sundew populations seem to regenerate by seed. Therefore the Finnish 4H Association immediately elaborated a new, environmentally sustainable collecting programme for young collectors to minimize the harmful effects of collecting and ensure safe regeneration of natural populations. According to the new guidelines, the collector must leave 5-10 flowering plants per square metre on the peatland. In theory, this leaves about 400-900 seeds/$m^2$. Additionally, sundew may be collected only once in the summer from each site. According to our results, 35% of the whole population flowered after the first collecting. Following the new guidelines, also later-flowering plants can produce seeds safely, disperse them and ensure the natural regeneration of sundew populations. These instructions are emphasized in collectors' training programmes.

- **Production-oriented dynamic preservation of threatened medicinal plants**

According to a recent study of the commercial importance and threatened status of medicinal plants in Europe, about 150 species were reported to be threatened in at least one European country as a result of over-collecting from the wild (Lange 1998). Some of the threatened medicinal plants are cold-tolerant or belong to the original Finnish flora.

A series of 3-year cultivation experiments were carried out in Finland to study the suitability of several cold-tolerant medicinal plants for cultivation and possible raw material production (Bernáth 1988). Reasonable yields were obtained in southern Finland from the following species in organic conditions: *Achillea ptarmica, Acorus calamus, Alchemilla alpina, A. xanthochlora, Arnica montana, Gentiana lutea, Herniaria glabra, Leontopodium alpinum, Primula veris* and *Rhodiola rosea*. After 3-year growing observations, *Antennaria dioica, Asarum*

*europaeum*, *Allium ursinum* and *Menyanthes trifoliata* showed a slow growth rate and insufficient yield level.

By producing raw material from controlled farming systems, we hope to decrease the economic pressure imposed on some threatened plant species and we also wish to provide additional income to the growers specialized in herb production. The results of plot experiments have to be checked in semi-large scale experimental conditions before educating the growers for production of these new medicinal plants.

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### Web site

Finnish Ministry of Environment: http://www.environment.fi

## *The status of medicinal and aromatic plants in Poland*

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### Introduction

Poland is an important producer of medicinal and aromatic plant (MAP) raw materials. About 200 MAP species are collected in larger or smaller amounts from natural sites, including 80 for commercial purposes. About 60 species are cultivated. The average annual harvest of raw materials from natural sites is estimated at 8000-10 000 t and more than half is exported. About 20 000 t of MAP raw materials originate from cultivation (Jambor 1999).

Over 10 000 people in Poland are directly or indirectly involved in herb collecting, cultivation, processing and trading.

The distribution of medicinal plants grown wild in Poland is irregular. Most of them, taking into consideration both the number of species and the area of natural sites, occur in the east of Poland. It is a special region representing the transitional area between continental and Atlantic climates. It is one of the ecologically cleanest parts of Poland, without heavy industry, large urban agglomerations or large agricultural farms.

Wild-growing MAP species are endangered by two main factors: the first, not specific to MAPs, is connected with irreversible abiotic changes of the environment (e.g. lowering of ground water level, pollution of the environment and eutrophication of natural water reservoirs); the second is specific to MAPs, i.e. uncontrolled and excessive harvesting.

### Legal protection of MAPs

The legal protection of MAPs in Poland is based both on:

- international conventions ratified by Poland in 1995: Convention on the Conservation of European Wildlife and Natural Habitats, Bern Convention 1982; Convention on Biological Diversity, Rio de Janeiro 1992; and
- national regulations: Preservation of Nature Act (16.10.1991); Decree of the Minister of Environment Protection, Natural Resources and Forestry on the protection of plant species (06.04.1995); Decree of The Minister of Environment Protection, Natural Resources and Forestry in consultation with the Minister of Health and Social Welfare and the Minister of Agriculture and Food Management, concerning the rules of harvesting of wild-growing medicinal plants or those important for pharmaceutical industry, as well as the management of the plantations of those plants (in preparation); and the Polish Red Book of Plants.

The official control organizations involved in the preservation of MAPs in Poland are the National Conservatory of Nature (attached to the Ministry of Environment), Provincial Conservatories of Nature, and Nature Preservation League (inspectors). Other institutions are also interested in this issue such as the Ministry of Agriculture and Rural Development, the Scientific Council for the Conservation of Plant Genetic Resources affiliated to the above Ministry, universities and research institutes.

According to the law, 111 species of Polish flora are strictly protected, including 20 species of herbaceous medicinal plants (*Lycopodium* sp., *Gypsophila paniculata*, *Cimicifuga europaea*, *Aconitum* sp., *Adonis vernalis*, *Nymphaea alba*, *Nuphar luteum*, *Drosera* sp., *Archangelica officinalis*, *Polemonium coeruleum*, *Echium rubrum*, *Atropa belladonna*, *Digitalis purpurea*, *Gentiana* sp., *Arnica montana*, *Veratrum* sp., *Colchicum autumnale*, *Leucoium vernum*, *Galanthus nivalis* and *Hierochloe odorata*).

Sixteen other MAP species are partly protected (*Cetraria islandica*, *Ribes nigrum*, *Ononis spinosa*, *Frangula alnus*, *Ledum palustre*, *Arctostaphylos uva-ursi*, *Viburnum opulus*, *Polypodium vulgare*, *Asarum europaeum*, *Primula elatior* and *P. officinalis*, *Asperula odorata*, *Centaurium umbellatum*, *Gentiana asclepiadea*, *Helichrysum arenarium*, *Convallaria maialis,* and *Hierochloe australis*). Their harvesting is possible in specified amounts and time limits, with the permission of the Provincial Conservatories of Nature.

## Wild-growing MAPs in Poland

There is no detailed documentation so far concerning the distribution of wild-growing MAP species in Poland. The investigations on natural habitats have been made only for some species (e.g. *Achillea* sp., *Centaurium umbellatum*, *Hypericum* sp., *Euphrasia* sp., *Adonis vernalis*, *Atropa belladonna*, *Arctostaphylos uva-ursi*) and for chosen areas. More thorough studies are currently underway in the Research Institute of Medicinal Plants in Poznań and in the Department of Vegetable and Medicinal Plants, Warsaw Agricultural University (SGGW) (Table 1). They concern geographic location, number of sites, population size, morphological, developmental and chemical traits. For chosen species molecular research is also carried out.

**Table 1.** MAP species studied *in situ*

| Species | No. of *in situ* sites studied | Frequency of population size (no. of sites x most frequent population size*) | No. of accessions transferred to the Polish Gene Bank |
|---|---|---|---|
| **Under strict or partial protection** | | | |
| *Adonis vernalis* | 29 | 2 x b, 10 x c, 14 x d, 3 x e | 26 |
| *Archangelica officinalis* | 1 | 1 x c | 6 |
| *Arctostaphylos uva-ursi* | 26 | 4 x a, 8 x b, 10 x c, 3 x d, 1 x e | 12 |
| *Arnica montana* | 15 | 7 x b, 6 x c, 2 x d | 21 |
| *Asarum europaeum* | 21 | 2 x b, 8 x c, 9 x d, 2 x e | 13 |
| *Centaurium umbellatum* | 13 | 1 x a, 4 x b, 7 x c, 1 x d | 15 |
| *Colchicum autumnale* | 23 | 2 x a, 1 x b, 6 x c, 11 x d, 3 x e | 19 |
| *Convallaria majalis* | 26 | 1 x b, 6 x c, 11 x d, 8 x e | 13 |
| *Hierochloe australis* | 3 | 1 x a, 2 x b | 3 |
| **Others** | | | |
| *Hypericum maculatum* | 4 | 3 x b, 1 x c | 2 |
| *Hypericum perforatum* | 51 | 6 x a, 19 x b, 11 x c, 9 x d, 6 x e | 20 |
| *Thymus pulegioides* | 2 | 1 x a, 1 x b | |
| *Thymus serpyllum* | 20 | 8 x a, 10 x b, 2 x c | 9 |

* Population size:
a = one to several individuals
b = a dozen or so individuals
c = dozens of individuals
d = hundreds of individuals
e = thousands of individuals

Figures 1-4 illustrate the chemical variability within two investigated species (Osińska and Węglarz 2000; Osińska *et al.* 2002).

In the last few years more than 100 samples of seeds of investigated accessions were transferred to the Polish Gene Bank in Radzików.

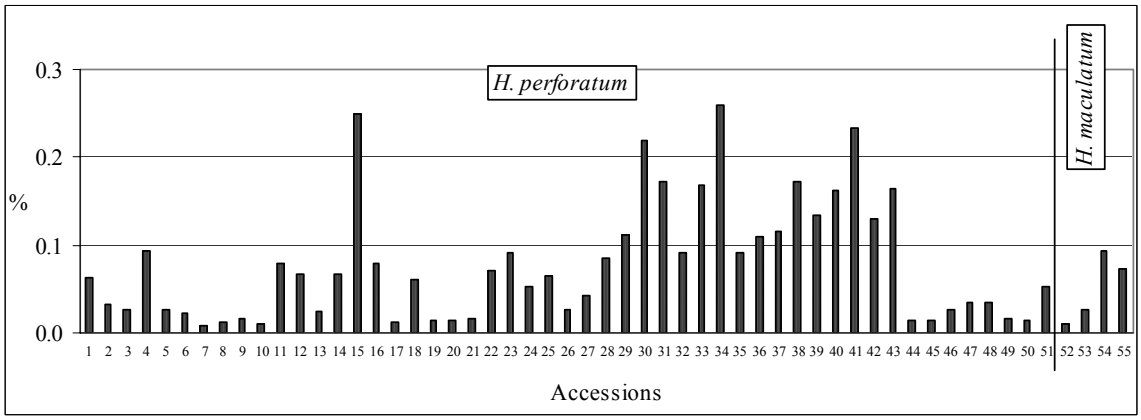


**Fig. 1.** Hypericin content in the herb of *Hypericum* sp. accessions studied *in situ*.

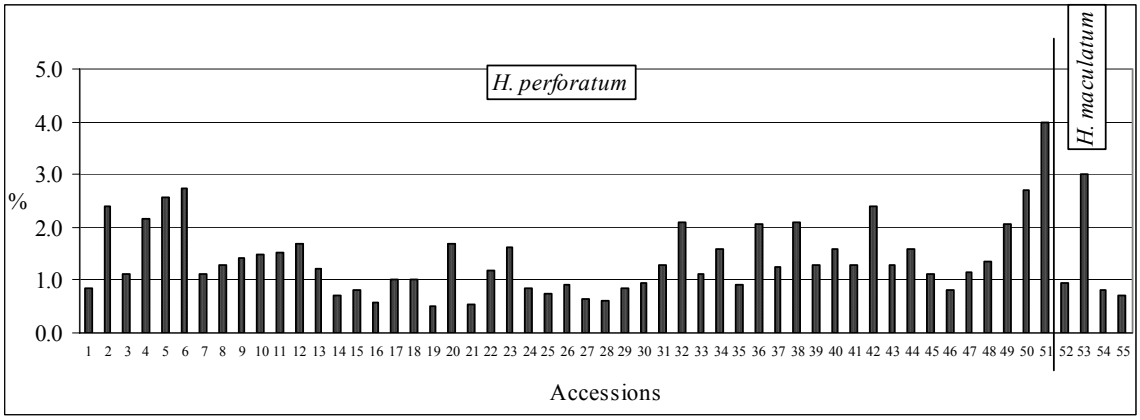


**Fig. 2.** Flavonoid content in the herb of *Hypericum* sp. accessions studied *in situ.*

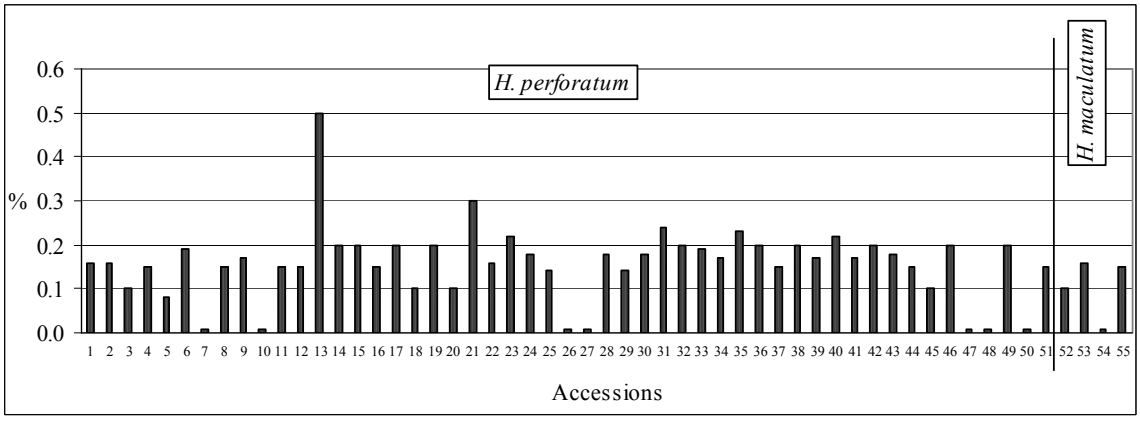


**Fig. 3.** Essential oil content in the herb of *Hypericum* sp. accessions studied *in situ.*

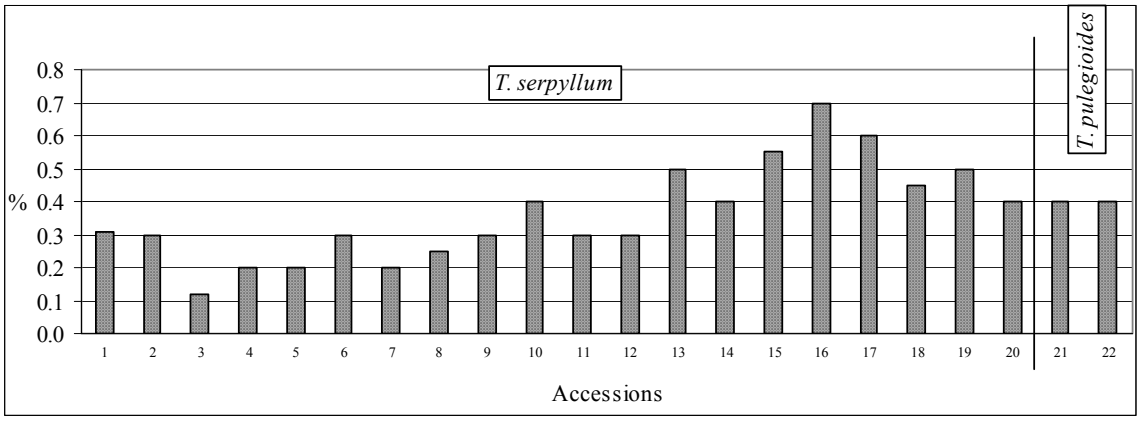


**Fig. 4.** Essential oil content in the herb of *Thymus* sp. accessions studied *in situ*.

## *Ex situ* MAP collections

- **Botanical gardens**

Collections of both wild and cultivated MAP species are maintained in botanical gardens. Six of the numerous botanical gardens in Poland maintain large MAP collections and offer the seeds of those plants according to the *Index Seminum*. These are mainly the seeds of plants cultivated in those gardens (1765 accessions from 110 families). However some botanical gardens (e.g. Botanical Garden of Medicinal Plants, University of Medicine, Wrocław) also possess seeds of wild medicinal plants collected from natural sites (208 accessions) (Table 2).

- **Warsaw Agricultural University**

The Department of Vegetable and Medicinal Plants, Warsaw Agricultural University also maintains *ex situ* collections of some MAP species (Table 3). The collected accessions differ both in quantitative and qualitative traits, e.g. content and composition of biologically active compounds (Osińska 2000; Węglarz *et al.* 2002) (Tables 3 and 4).

**Table 2.** MAP seed collections in Polish botanical gardens (number of accessions from the garden + from *in situ*)

| Family | Botanical garden (*) | | | | | |
|---|---|---|---|---|---|---|
| | A | B | C | D | E | F |
| Acanthaceae | | 6 | 2 | | | |
| Aceraceae | 3 | 2 | | | 2 | |
| Agavaceae | | 4 | | | | |
| Alliaceae | 9 | 2 | | | 16 | 3 |
| Amaranthaceae | 6 | 1+2 | 4 | | 1 | 5 |
| Amarylidaceae | 2 | 1 | | 1 | 2 | |
| Anacardiaceae | | 1 | 1 | | | 1 |
| Apiaceae | 27 | 13+3 | 13+1 | 14 | 13 | 8+2 |
| Apocynaceae | 1 | 2 | 5 | 1 | | |
| Aquifoliaceae | | 1 | | | | |
| Araceae | | 1 | | 1 | 1 | |
| Araliaceae | 1 | 1 | | | 1 | |
| Aristolochiaceae | 2 | | | 1 | 1 | |
| Asclepiadaceae | 5 | | 3 | | 1 | 1 |
| Asparagaceae | | 2+2 | | | | |
| Asphodelaceae | | 3 | | | | |
| Asteraceae | 78 | 40+40 | 102+3 | 30 | 37 | 27+3 |
| Balsaminaceae | | 0+2 | | | | |
| Berberidaceae | 5 | 1+1 | 2 | | 3 | |
| Bignoniaceae | | | 1 | | | |
| Betulaceae | 1 | 0+2 | | | | |
| Boraginaceae | 10 | 4+2 | 4 | 3 | 6 | 4 |
| Brassicaceae | 23 | 13+6 | 3 | 5 | 7 | 3 |
| Bromeliaceae | | 1 | | | 1 | |
| Buxaceae | | | | 1 | | |
| Campanulaceae | 4 | 5+2 | 1 | | 3 | 1 |
| Cannabaceae | 2 | | | 1 | 1 | |
| Capparaceae | | 1 | | | | 1 |
| Caprifoliaceae | 9 | 2+8 | 2+1 | | 2 | |
| Caryophyllaceae | 23 | 13+2 | 6 | 4 | 15 | 5+1 |
| Celastraceae | 1 | 2+1 | 1 | | 1 | |
| Chenopodiaceae | 6 | 4+1 | | 2 | 3 | |
| Cistaceae | | 4 | | | | |
| Clusiaceae | 3 | 2+2 | 2 | 1 | 2 | 1+1 |
| Convolvulaceae | 3 | 0+1 | | | | |
| Cornaceae | 1 | 1+1 | | | 1 | |
| Crassulaceae | 4 | 0+1 | 2 | 1 | 1 | 3 |
| Cucurbitaceae | 1 | 1+1 | | 1 | | 1 |
| Cupressaceae | | 1 | 1 | | 1 | |
| Cuscutaceae | 1 | | | | | |
| Cyperaceae | 2 | 5 | | | | |
| Dioscoreaceae | | 1 | | | 3 | |
| Dipsacaceae | 1 | 8+3 | 1 | | | |
| Equisetaceae | 1 | | | 1 | | |
| Ericaceae | | 0+1 | | 2 | 2 | |
| Euphorbiaceae | 1 | 2+1 | 2 | 1 | 2 | |
| Fabaceae | 26 | 15+23 | 12 | 16 | | 16+4 |
| Fagaceae | | 0+2 | | | 10 | |
| Gentianaceae | 4 | | 1 | | 2 | |
| Geraniaceae | 6 | 1+2 | | | | |
| Globulariaceae | | 1 | | | | |
| Grossulariaceae | | 1 | | | | |
| Hemerocallidaceae | | 1 | | | | |
| Hippocastanaceae | 2 | 0+1 | | | 1 | |
| Hydrophyllaceae | | 1 | | | | |
| Iridaceae | 7 | 2 | | | 1 | |
| Juglandacae | 2 | | | | | |
| Lamiaceae | 52 | 43+8 | 34 | 32 | 23 | 23+1 |
| Liliaceae | 11 | 2 | 9 | 10 | 12 | |
| Linaceae | 2 | 2 | 1 | 1 | 1 | 7 |
| Lobeliaceae | 1 | 1 | 1 | | | |
| Loganiaceae | | | 1 | | 1 | |
| Loranthaceae | | 0+2 | | | | |
| Lythraceae | 1 | | 2 | | 1 | |
| Malvaceae | 7 | 7+1 | 4 | 3 | 4 | 2 |

**Table 2 (cont.).** MAP seed collections in Polish botanical gardens (number of accessions from the garden + from *in situ*)

| Family | Botanical garden (*) | | | | | |
|---|---|---|---|---|---|---|
| | A | B | C | D | E | F |
| Moraceae | 1 | 2+2 | 1 | | | |
| Myrtaceae | | 1 | | | | |
| Nyctaginaceae | | 1 | | | | |
| Oenotheraceae | 2 | 0+4 | 5 | 1 | 3 | 3+1 |
| Oleaceae | 1 | 0+1 | 1 | | 1 | |
| Osmundaceae | | 0+1 | | | | |
| Oxalidaceae | | | | 1 | | |
| Papaveraceae | 9 | 6 | 1 | 5 | 6 | 12 |
| Passifloraceae | | 2 | | | | |
| Phytolaccaceae | 1 | 2 | 3 | | 1 | |
| Pinaceae | 4 | 0+1 | 1 | | 2 | |
| Plantaginaceae | 5 | 2+3 | 3 | 3 | 2 | 2+1 |
| Plumbaginaceae | 2 | 2+3 | 1 | | 1 | |
| Poaceae | 12 | 4+9 | 5 | 3 | 10 | 7 |
| Polemoniaceae | 1 | 3 | 1 | | 1 | 1 |
| Polygonaceae | 14 | 3+5 | | 5 | 12 | 2 |
| Polypodiaceae | 2 | | | | | |
| Portulaccaceae | | | 1 | | | |
| Primulaceae | 4 | 2 | | 4 | 2 | 1 |
| Ranunculaceae | 34 | 16+1 | 6 | 8 | 6 | 5 |
| Resedaceae | 3 | | | | | |
| Rhamnaceae | 2 | 1+1 | | | 2 | |
| Rosaceae | 20 | 20+17 | 9 | 5 | 12 | 5+3 |
| Rubiaceae | 3 | 1 | | 1 | | |
| Rutaceae | 3 | 4 | 3 | 2 | 2 | 2 |
| Sapindaceae | | 1 | 2 | | 1 | |
| Saxifragaceae | 2 | 2+1 | 1 | 2 | 1 | 1 |
| Schisandraceae | | 1 | | | | |
| Scrophulariaceae | 8 | 9+2 | 10 | 5 | 6 | 4+1 |
| Simarubaceae | | | 1 | | 1 | |
| Solanaceae | 8 | 6+2 | 8 | 4 | 7 | 3+1 |
| Staphyleaceae | | 1 | | | | |
| Taxaceae | 1 | 1+1 | | | 1 | |
| Thymelaeaceae | | 4 | | | | |
| Tiliaceae | 1 | | 0+1 | | 2 | |
| Tropaeolaceae | 1 | 1 | 1 | 1 | | |
| Ulmaceae | | 1 | | | | |
| Urticaceae | 2 | 2+2 | | 1 | 3 | |
| Valerianaceae | 4 | 1 | 3 | 1 | 1 | 1+1 |
| Verbenaceae | 1 | 2 | 3 | | 1 | |
| Violaceae | 3 | 2+1 | | 3 | | 2 |
| Vitaceae | | 1+1 | | | | |
| Woodsiaceae | | 1 | | | | |
| Zingiberaceae | | 1 | | | | |
| Zygophyllaceae | | 1 | | | | |

(*) Botanical gardens:

A = Research Institute of Medicinal Plants, Poznań

B = Botanical Garden of Medicinal Plants, University of Medicine, Wrocław

C = Polish Academy of Sciences, Institute of Pharmacology, Kraków

D = Garden of Medicinal Plants, Faculty of Pharmacy, Jagiellonian University, Kraków

E = Botanical Garden of the Jagiellonian University, Kraków

F = Botanical Garden of Plant Breeding and Acclimatization Institute, Bydgoszcz

**Table 3.** *Ex situ* collections of MAP species (Dept. of Vegetable and Medicinal Plants, Warsaw Agricultural University – SGGW)

| Species | No. of accessions | Variability range | | | No. of seed samples transferred to the Polish Gene Bank |
|---|---|---|---|---|---|
| | | Dry mass of herb (g/plant) | Essential oil content (%) | | |
| *Hyssopus* sp. | 17 | 60.1-245.6 | 0.85-2.20 | | 10 |
| *Satureja* sp. | 11 | 69.2-169.2 | 0.75-2.30 | | 4 |
| *Thymus serpyllum* | 10 | 28.2-123.0 | 0.20-0.40 | | 4 |
| *Origanum vulgare* | 14 | 12.1-276.0 | 0.30-1.30 | | 6 |
| | | | Coumarin content (%) | | |
| *Hierochloe australis* | 38 | 2.5-28.0 | 0.22-0.60 | | 12 |
| | | | Hypericin content (%) | Flavonoid content (%) | |
| *Hypericum* sp. | 15 | 61.0-333.5 | 0.01-0.16 | 0.55-2.58 | 5 |

**Table 4.** Content of major constituents of essential oils of MAP species studied *ex situ*

| Compound | % essential oil | | |
|---|---|---|---|
| | ***Hyssopus* sp.** | ***Satureja* sp.** | ***Origanum vulgare*** |
| α-thujene | 0.10-0.74 | 0.03-1.16 | 0.22-2.52 |
| α-pinene | 0.05-0.32 | 0.38-2.03 | 0.13-1.25 |
| β-pinene | 1.51-15.07 | 0.08-1.40 | 0.72-18.03 |
| sabinene | 1.00-7.56 | 0.02-0.63 | 5.46-35.52 |
| myrcene | 1.15-9.46 | 0.32-2.46 | 1.10-7.02 |
| limonene | 0.60-2.71 | 0.17-4.22 | 0.50-4.61 |
| cineol | 1.49-46.98 | 0.29-6.08 | 0.75-44.50 |
| α-terpinene | - | 0.67-2.18 | 0.00-6.14 |
| γ-terpinene | 0.06-4.30 | 1.79-16.35 | 0.00-17.12 |
| p-cymene | 0.03-0.51 | 2.97-9.76 | 0.00-9.14 |
| menthone | 0.04-0.18 | 0.92-1.62 | - |
| pinocamphone | 0.21-58.85 | - | - |
| izopinocamphone | 2.78-70.00 | - | - |
| linalool | 0.15-3.44 | 0.20-13.04 | 0.00-15.60 |
| β-caryophyllene | 0.70-3.48 | 1.04-4.28 | 0.39-12.28 |
| terpinene-4-ol | 0.05-0.55 | 0.08-3.94 | 0.59-18.48 |
| α-terpineol | - | 0.14-2.60 | 0.00-4.35 |
| borneol | 0.03-5.69 | 0.11-1.92 | - |
| caryophyllene oxide | 0.11-0.81 | 0.06-0.82 | 0.13-15.03 |
| thymol | - | 0.38-8.36 | 0.21-7.35 |
| carwacrol | - | 27.26-70.53 | 0.00-0.40 |

Transfer of wild plants into cultivation may affect the chemical composition of raw materials. For example, accessions of *Hypericum perforatum* studied in *ex situ* conditions were characterized by higher content of hypericin and lower content of flavonoids and essential oil in comparison with raw materials collected from natural sites (Figs. 5-7).

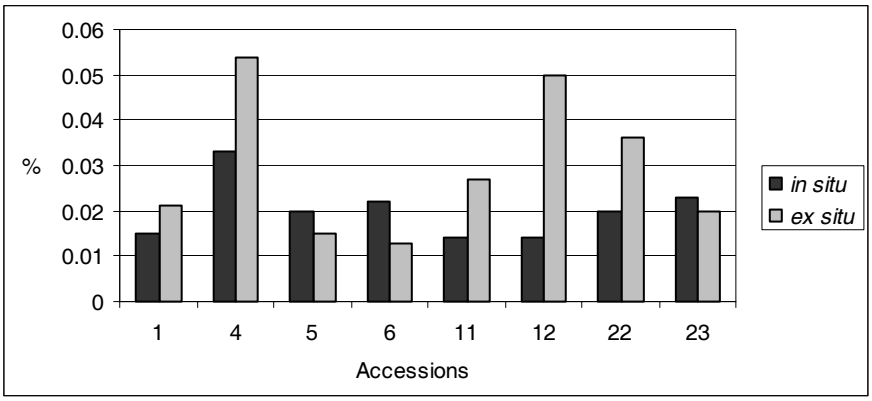


**Fig. 5.** Hypericin content in the herb of *Hypericum perforatum* in *in situ* and *ex situ* conditions.

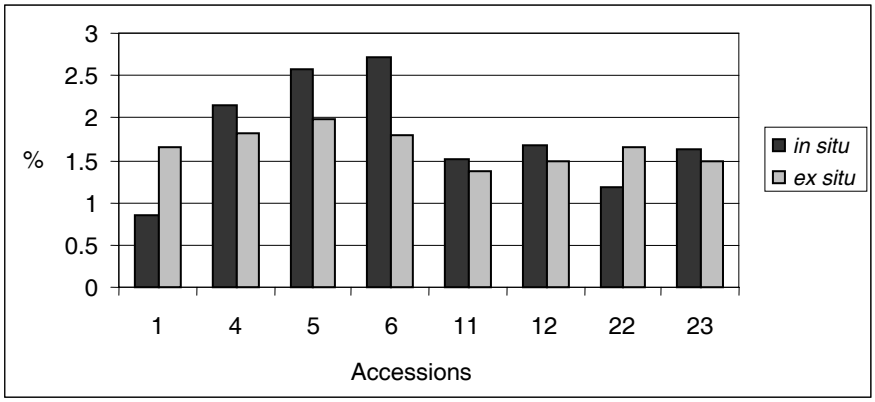


**Fig. 6.** Flavonoid content in the herb of *Hypericum perforatum* in *in situ* and *ex situ* conditions.

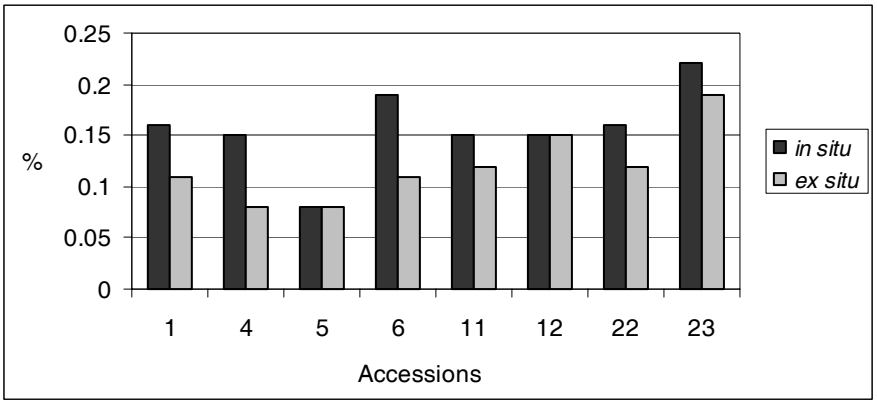


**Fig. 7.** Essential oil content in the herb of *Hypericum perforatum* in *in situ* and *ex situ* conditions.

## MAP cultivation and trade

It is difficult to present exact data on the acreage and production volume of MAPs in Poland owing to the increasing number of new Polish private enterprises involved in MAP production and the activity of many foreign firms on this market.

The most important MAP raw materials collected from natural sites are (in descending order of tonnage of purchased raw material): *Urtica dioica* (leaves) > *Hypericum perforatum* (herb) > *Tilia cordata* (inflorescences) > *Frangula alnus* (bark) > *Sambucus nigra* (fruits) > *Rosa canina* (fruits) > *Equisetum arvense* (herb) > *Betula verrucosa* (leaves) > *Solidago virgaurea* > *Quercus robur* (bark) > *Salix purpurea* (bark) > *Aesculus hippocastanum* (bark) > *Pinus sylvestris* (buds) > *Agropyron repens* (rhizomes) > *Taraxacum officinale* (roots) > *Potentilla erecta* (rhizomes) > *Juniperus communis* (fruits) > *Crataegus oxyacantha* (fruits) > *Sambucus nigra* (flowers) > *Helichrysum arenarium* (inflorescences).

Nowadays, MAPs in Poland are cultivated on an area of ca. 20 000 ha. The main cultivated species are (in descending order of acreage and tonnage of purchased raw material): *Oenothera* sp. (seeds) > *Carum carvi* (fruits) > *Silybum marianum* (fruits) > *Mentha piperita* (leaves) > *Chamomilla recutita* (inflorescences) > *Origanum majorana* (herb) > *Valeriana officinalis* (roots) > *Thymus vulgaris* (herb) > *Melissa officinalis* (herb) > *Coriandrum sativum* (fruits) > *Borago officinalis* (seeds) > *Cynara scolymus* (herb) > *Althaea officinalis* (roots) > *Archangelica officinalis* (roots) > *Levisticum officinale* (roots) > *Salvia officinalis* (leaves) > *Echinacea purpurea* (herb and roots) > *Ribes nigrum* (seeds) > *Ruta graveolens* (herb) > *Plantago lanceolata* (leaves).

The main MAP raw materials exported by Poland are: *Carum carvi* (fruits) > *Oenothera* sp. (seeds) > *Valeriana officinalis* (roots) > *Mentha piperita* (leaves) > *Thymus vulgaris* (herb) > *Chamomilla recutita* (inflorescences) > *Potentilla erecta* (rhizomes) > *Hypericum perforatum* (herb) > *Borago officinalis* (seeds) > *Tilia cordata* (inflorescences).

The cultivation of MAPs in "ecological farms" (practicing organic farming) in Poland started in the beginning of the 1990s. Unlike that of agricultural and animal organic production, the increase in organic MAPs' acreage is relatively slow. Organic MAP production is currently concentrated mainly in southeastern Poland. Seven major MAP species are cultivated in 80 organic farms (Table 4) and about 15 other MAP species are cultivated on smaller areas.

**Table 4.** Major MAP species cultivated in organic farms in Poland

| Species | Cultivated area (ha) |
|---|---|
| *Oenothera* sp. | 25 |
| *Borago officinalis* | 5 |
| *Valeriana officinalis* | 3 |
| *Thymus vulgaris* | 3 |
| *Nigella sativa* | 3 |
| *Mentha piperita* | 2 |
| *Ribes nigrum* (leaves) | 5 |

In 2001, 10 t of *Rubus idaeus* leaves and 6 t of *Urtica dioica* leaves were collected from natural sites as raw materials.

In a few major Polish cities (e.g. Warsaw, Kraków, Poznań and Gdańsk) about 20 shops specialized in organic food offer MAP products originating both from Poland and from abroad. Such products are also sold in chosen grocers' shops, under the special control of the Polish Ecological Club in the town of Gliwice.

There are about 2500 herbal medicines, estimated at 200 million US$ per year, on the Polish market. These include simple medicinal forms (e.g. dried herbs, herb mixtures, granulates), more processed forms (e.g. tablets, capsules, dragees) and half-finished products for pharmaceutical, cosmetic and food industry (extracts and essences).

For the production of extracts about 4000 t of raw materials of wild-growing plants and about 8000 t of cultivated plants are used, most frequently *Hypericum perforatum*, *Urtica dioica*, *Valeriana officinalis*, *Betula verrucosa*, *Cynara scolymus*, *Chamomilla recutita*, *Silybum marianum*, *Equisetum arvense* and *Thymus vulgaris*. Herbal medicines are used mostly against disorders of the digestive tract and as tonics and strengtheners. Among Polish plant preparations the most popular are Sylimarol, Raphacholin C and Tussipect (Lutomski 1999).

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## *Medicinal and aromatic plants in Portugal – a survey*

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### Introduction

A set of questions was submitted to 43 institutions about research and other activities concerning medicinal and aromatic plants (MAPs). The topics defined were: collecting, conservation, characterization, preliminary evaluation, further evaluation and utilization. Twenty-three institutions replied to the questionnaire. Some replies arrived too late to be included in this survey, but the work done at the institutions concerned will be taken into account in future publications.

### Conservation

- ***In situ* conservation**

Since 1999 the Institute for the Conservation of Nature (ICN) that oversees one National Park, ten Natural Parks, six Natural Reserves and two Protected Landscapes, started a 5-year integrated project based on four main objectives: survey of medicinal and aromatic plant species; ethnobotany survey; environmental education programmes and public awareness; correct management and promotion of sustainable utilization of MAPs.

Table 1 lists the research activities carried out for this project in the various protected areas.

**Table 1.** List of protected areas in Portugal and their activities

| Protected areas | Species survey | Ethnobotany survey | Environmental education programme | Sustainable management |
|---|---|---|---|---|
| Reserva Natural da Serra da Malcata | ■ | ■ | | ■ |
| Reserva Natural das Dunas de S. Jacinto | ■ | | | |
| Parque Natural do Douro Internacional | ■ | ■ | ■ | ■ |
| Parque Natural da Serra da Arrábida | ■ | ■ | | |
| Parque Natural do Alvão | ■ | ■ | | |
| Reserva Natural do Estuário do Tejo | ■ | | | |
| Parque Nacional da Peneda-Gerês | ■ | ■ | ■ | |
| Parque Natural da Serra da Estrela | ■ | ■ | ■ | ■ |
| Área de Paisagem Protegida da Serra do Açor | ■ | ■ | | |
| Parque Natural de Sintra Cascais | ■ | ■ | | |

- ***Ex situ* conservation**

In order to conserve, evaluate and use MAP species, different Portuguese institutions such as universities, polytechnic schools, the Ministry of Agriculture, Rural Development and Fisheries and foundations all over the country have been developing research in this area.

Since 1990, the Banco Português de Germoplasma Vegetal (BPGV) – Ministry of Agriculture, Rural Development and Fisheries, has been conducting, all over Portugal, a systematic collection of *Allium sativum* L., *Allium cepa* L., *Allium* spp., *Petroselinum crispum* (Mill.) Nym. ex AW Hill, *Coriandrum sativum* L., *Humulus lupulus* L., *Thymus* spp., *Origanum vulgare* L. and *Mentha* spp. This material is conserved in long-term storage conditions at -20ºC, in medium-term storage conditions (0-5ºC) and as a field collection.

The Direcção Regional de Agricultura de Entre Douro e Minho (DRAEDM) has done a systematic collection of seed and propagules of the following species: *Hypericum androsaemum* L., *Ruscus aculeatus* L., *Melissa officinalis* L., *Cynara cardunculus* L. and *Centaurium erythraea* Rafn.

Several other institutions such as polytechnic schools and delegations of the Ministry of Agriculture, Rural Development and Fisheries have been collecting material in order to create small gardens as a source for environmental education. An excellent example is the Jardim Algarve located at the Direcção Regional de Agricultura do Algarve.

The Serralves Foundation has organized a medicinal and aromatic garden at Oporto

As a commercial enterprise the Cantinho das Aromáticas–Viveiros develops the production and sale of MAPs using organic farming technology.

## Evaluation

The BPGV and the Escola Superior Agrária de Elvas have been evaluating morphologically and agronomically the accessions of local populations of garlic, onion, coriander, parsley and hops. Some garlic and coriander accessions are being subjected to molecular analysis (Table 2).

**Table 2.** Evaluation of MAP species

| Action | Carried out by | Genera / Species |
|---|---|---|
| Characterization and preliminary evaluation | DRAEDM | Coriander, garlic, onion, parsley, hops |
| Chemical characterization - essential oils | ESAPL | *Thymus mastichina, Rosmarinus officinalis* |
| Chemical characterization - essential oils | UC | *Thymus, Juniperus, Teucrium, Origanum, Mentha* |
| Ethnopharmacological survey and essential oils of aromatic plants from Africa (Popular Republic of S. Tomé and Principe and Mozambique) | UC | |
| Technological valorization of the essential oils | UC | |
| Antibacterial and antifungal activity of essential oils | UC | |

DRAEDM = Direcção Regional de Agricultura de Entre Douro e Minho
ESAPL = Escola Superior Agrária de Ponte de Lima
UC = Universidade de Coimbra

## Multiplication and utilization

The sustainable use of all genetic material collected, conserved and evaluated must be promoted. Several institutions are working on propagation and multiplication, at the same time carrying on with further evaluation (Table 3).

**Table 3.** Multiplication and utilization of MAP species

| Action | Carried out by | Genera / Species |
|---|---|---|
| Propagation (trials on seminal and vegetative propagation) | DRAEDM | *Hypericum androsaemum, Melissa officinalis, Cynara cardunculus, Centaurium erythrea* |
| Production trials | DRAEDM | *Hypericum androsaemum* L., *H. perforatum* L., *Vaccinium myrtillus* L., *Melittis melissophyllum* L., *Woodwardia radicans* L. |
| Selection and propagation | DRAEDM | *Origanum virens, Lavandula* spp., *Thymus* spp. and *Apium graveolens* |
| Variety study | ESAC | *Mentha* x *piperita, Origanum vulgare, Salvia officinalis* cv. "green", *S. officinalis* cv. "purpurescens", *Lavandula angustifolia* and *Lippia citriodora* |
| Selection and propagation | ESAPL | *Petroselinum crispum, Coriandrum sativum, Thymus vulgaris* |

DRAEDM = Direcção Regional de Agricultura de Entre Douro e Minho
ESAC = Escola Superior Agrária de Coimbra
ESAPL = Escola Superior Agrária de Ponte de Lima

**Ethnobotany**

A major 3-year National Programme entitled "Ethnobotany, the use and management of aromatic and medicinal plants and their sustainable utilization as contribution to improvement of rural areas" involves several institutions belonging to the areas of agriculture, education, and environmental protection. During this project we will be collecting, conserving, evaluating and using the following species:

*Centaurium erythraea* Rafn.
*Coriandrum sativum* L.
*Cynara cardunculus* L.
*Melittis melissophyllum* L.
*Mentha aquatica* L.
*Mentha cervina* L.
*Mentha pulegium* L.
*Mentha suaveolens* Elrh
*Origanum vulgare* L.
*Origanum virens* Hoffmanns & Link
*Thymus caespititius* Brot.
*Thymus pulegioides* L.
*Thymus zygis* L.
*Vaccinium myrtillus* L.

The Direcção Regional de Agricultura do Algarve (DRAAL) and the Escola Superior Agrária de Bragança (ESAB) are also developing two projects in this subject, circumscribed to the areas of Portel, Ponte de Sôr and Bragança.

## *Status of the Romanian medicinal and aromatic plant collection*

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[3] *Central Research Station for MAP Fundulea, Romania*

### Introduction

The relief of Romania is distributed harmoniously: the mountains form an arch in the central part of the country and represent 31% of the total area of the country. Hills and plateaus covering 33% of the country surround the Carpathian mountain chain, and the plains, situated to the south and to the west of the country, represent the remaining 36%.

The Romanian flora includes 3450 cormophyte species which represent 30% of the vascular European flora. Among these, 283 species have therapeutic effects and 180 species are studied, but only 52 species are cultivated (Mocanu 1999). *In situ* and *ex situ* preservation are achieved in natural reserves and on farms respectively, in the institutions and agricultural research stations that have breeding programmes and by the Suceava Genebank, a governmental institution.

### Legal protection of MAP species and their natural habitats

The decrease of biological diversity on a global level has led to the elaboration of a Red List which includes medicinal and aromatic plants. Table 1 lists the endangered medicinal plants from the spontaneous flora which are included in the Red List.

**Table 1.** Endangered medicinal plants from the spontaneous flora of Romania

| Plant organs harvested | Species |
|---|---|
| Flowers | *Arnica montana*<br>*Primula officinalis*<br>*Tussilago farfara* |
| Leaves | *Atropa belladonna*<br>*Allium ursinum*<br>*Althaea officinalis* |
| Stems, leaves | *Adonis vernalis*<br>*Galium verum*<br>*Lycopodium clavatum* |
| Roots, bulbs, tubers | *Gentiana lutea*<br>*Arnica montana*<br>*Helleborus purpurascens*<br>*Angelica archangelica* |

There are currently 450 reserves in Romania, including:
- 1 World Heritage Site - Danube Delta (591 200 ha)
- 2 biosphere reserves: Retezat (54 400 ha) and Rodna (56 700 ha)
- 12 national parks covering 287 084 ha
- 2 natural parks (10 150 ha)
- 161 ornamental plant collections (22 000 ha)
- 52 forestry reserves of over 113 668 ha

Thus, the total protected areas cover 1 072 498 ha, i.e. 4.59% of the whole country (Anonymous 1995).

Activities dealing with the preservation of plant genetic resources present in parks and natural reserves are coordinated at national level by the Romanian Academy, Commission for the Protection of Nature Monuments.

The following laws are currently active (Anonymous 1994, 2000, 2002):

- Law of the protected areas (5/2000), which includes rules for the protection of natural areas and for the conservation of natural habitats and of the wild flora and fauna of Romania;
- Law 69/1994, referring to the international trade agreement related to species of the Romanian wild flora and fauna threatened with extinction.

At the beginning of 2002, the law regulating the use of medicinal, aromatic, toxic and drug plants was under discussion in the Romanian Parliament.

### *Ex situ* conservation of MAP genetic resources

This is accomplished in Romania through field conservation and medium- and long-term conservation.

Field conservation is carried out in the following institutions:

- Central Research Station for MAP Fundulea (931 samples - 297 species)
- Agricultural University Iassy (80 samples - 67 species)
- Agricultural University Timisoara (74 samples – 66 species)

Regarding medium- and long-term conservation, two institutions hold MAP collections: the Suceava Genebank and the Agricultural University Iassy.

The Suceava Genebank holds 246 samples (174 species) in the active and base collections distributed as shown in Fig. 1.

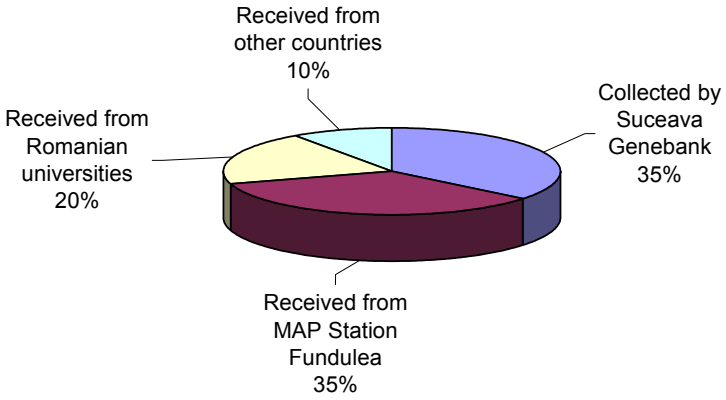


**Fig. 1.** Distribution of the MAP samples conserved in the Suceava Genebank.

The Agricultural University Iassy holds a collection of 1200 species kept under medium-term conditions. In 2001 a genetic centre for medicinal, aromatic and tinctorial plants collected from 52 Asian and European countries was established.

### National inventory of MAP genetic resources

In 2000 the Suceava Genebank, with IPGRI's support, published the National Catalogue of Plant Genetic Resources. One fascicle covers medicinal and aromatic plant collections. This catalogue includes only information on passport descriptors.

### MAP cultivation areas and yields in Romania

The dynamics of areas cultivated with MAPs over a six-year period (1995-2000) depended on many factors, among which the relief and climatic conditions are decisive. Social factors and local customs, economic factors and agricultural policy also had a great influence. Whilst in 1995 the area covered with MAPs was 26 500 ha (Anonymous 2000), by 2000 it had decreased by 4000 ha (Fig. 2).

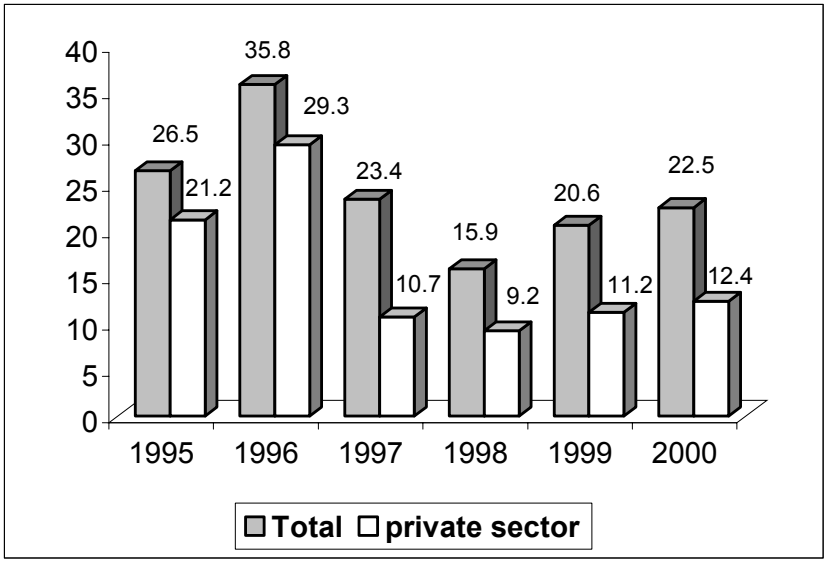


**Fig. 2.** MAP cultivation areas in Romania, 1995-2000 (x 1000 ha).

The same situation applies to MAP yields in Romania: Fig. 3 shows the fall in MAP yield during the same period (1995-2000).

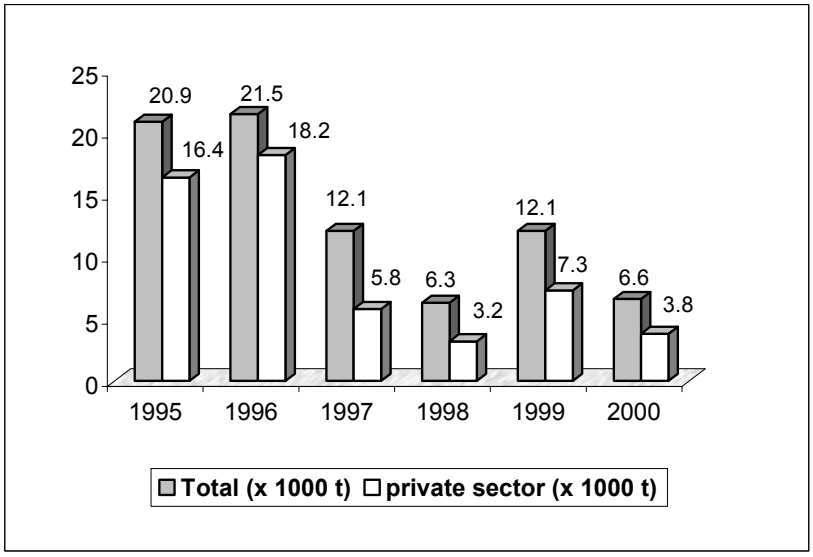


**Fig. 3.** Yields of cultivated MAPs in Romania, 1995-2000.

### Use of MAPs by processing industries

The MAP processing industry in Romania started in 1948 with the establishment of the National Society "Plafar" (Pharmaceutical Plants). The main objective of this institution is the sustainable exploitation of the medicinal, aromatic and drug plants from the wild flora of Romania. The National Society "Plafar" has nine inter-district branches (Botosani, Iassy, Bucarest, Cluj, Brasov, Timisoara, Constanta, Mures and Maramures). There are also five processing companies which process and produce medicine only from medicinal and aromatic plants: Vitasana, Brasov; Herborium, Cluj; Vorel, Piatra Neamt; Digitalis, Orastie; and Naturavit, Oradea.

The quantities of raw materials gathered in the wild and processed in 2001 are presented in Table 2.

**Table 2.** Quantities of raw materials gathered in the wild and processed in 2001

| Harvested organs | Quantity (tonnes) | No. of species gathered |
|---|---|---|
| Flowers | 1680 | 27 |
| Leaves | 3500 | 36 |
| Stems, leaves | 5000 | 63 |
| Roots, tubers, bulbs | 1000 | 18 |
| Seeds and fruits | 100 | 10 |

### Sustainable use of MAPs in Romania

This is accomplished in the following ways:

1. MAP breeding
2. Research on biology and cultivation of new MAP species
3. Foundation of a genetic centre of medicinal, aromatic and tinctorial plants and their use in herbal medicine.

1. MAP breeding is carried out in Romania out by the Central Research Station Fundulea. The species being bred include *Cynara scolymus, Digitalis lanata, Dracocephalum moldavica, Foeniculum officinale, Hyssopus officinalis, Coriandrum sativum, Lavandula angustifolia, Matricaria chamomilla, Mentha piperita, Ocinum basilicum, Papaver somniferum*, etc. The research activity during 1980-2002 resulted in 27 advanced cultivars of *Cynara scolymus, Ocimum basilicum, Thymus vulgaris, Coriandrum sativum, Digitalis lanata, Calendula officinalis, Datura innoxia, Lavandula angustifolia, Mentha piperita* and *Matricaria chamomila.* In addition, 31 local landraces were certified (*Pimpinella anisum, Capsicum annuum, Silybum marianum, Satureja hortensis, Tagetes patula, Digitalis purpurea, Foeniculum vulgare*, etc.).
2. Cultivation of new MAP species: during the period 1995-2000 the researchers from Central Research Station Fundulea introduced into cultivation five species (*Aconitum napellus, Artemisia abrotanum, Gentiana lutea, Leonurus cariaca* and *Lychnis coronaria*), taking into consideration their mode of use, pharmacodynamic action, chemical composition and environmental requirements (Paun 1998).
3. The foundation at the Agricultural University Iassy of a genetic centre for medicinal, aromatic and tinctorial plants and their use in herbal medicine was carried out through the following activities:
   - Inventory of MAPs from the wild and cultivated flora;
   - Establishment of a biological collection conserved in the Iassy Genebank;
   - Foundation of a herbal medicine library;
   - Development of some complex therapeutic prescriptions;
   - Testing of the efficacy of the elaborated products;
   - Organization of a network of collaborators interested in herbal medicine.

## Future strategies

The institutions specializing in breeding and processing of MAPs propose a number of measures for the near future, such as:

- Sustainable exploitation of wild medicinal plants; improvement of research through breeding and cultivation of new species;
- increase in the area cultivated with MAPs by the private sector through higher prices of dry products collected by the processing enterprises;
- use of advanced technology for raw material processing and of essential quantitative indices for the major medicinal plants cultivated in Romania.

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## *Conservation of medicinal and aromatic plant genetic resources in Slovenia*

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### Introduction

In 1996, the Slovenian Fund for nature conservation ratified the Resolution on conservation of biological diversity and permanent landscape development, including medicinal and aromatic plants (MAPs). In order to ensure genepools for future investigations, the genebank, a national genebank collection of medicinal and aromatic plants containing 650 indigenous or foreign/introduced MAP accessions, was established in 1994, officially recognized in 1995 and is annually supported by the Slovenian government. The programme on MAP conservation in Slovenia involves activities at different conservation levels (*ex situ*, *in situ* and on-farm) as well as morphological and chemical evaluation of MAP accessions held in the MAP genebank (*Salvia officinalis* L., *Origanum vulgare* L., *Gentiana lutea* L., *Thymus vulgaris* L., etc.) (Baričevič 1997a, 1997b; Baričevič *et al*. 1997; Rode *et al.* 1998).

### *Ex situ* conservation and evaluation

Maintenance of germplasm as seed, *in vitro* and *in vivo* collections and seed propagation for cultivation purposes (*Cynara scolymus* L., *Gentiana lutea* L., *Salvia officinalis* L., *Origanum vulgare* L., *Trigonella foenum-graecum* L., *Melissa officinalis* L., *Hyssopus officinalis* L., *Thymus vulgaris* L., *Satureja montana* L.) are the main activities of the Slovenian MAP genebank. Because of the high morphological and chemical variability in MAPs, *in vitro* culture techniques have been applied for practical purposes, so the screening for optimal *in vitro* conditions (considering multiplication rate, morphological uniformity and low-cost input) has been considered. The micropropagation of MAPs has been recognized as an essential tool in obtaining uniform descendants in cross-pollinating species, in species with poor or slow germination as well as in virus-infected plant material. The evaluation of MAP accessions, which regularly accompanies the routine maintenance work, can be divided into three research categories:

- Evaluation of morphological and chemotaxonomic characteristics of MAPs (*Origanum vulgare* L. subsp. *vulgare*, *Origanum* L. subsp. *vulgare hirtum, Gentiana lutea* L. subsp. *symphyandra*) (Zupančič and Baričevič 2002);
- Evaluation of MAP ecotypes for quantitative and qualitative differences in secondary metabolites with regard to growth and development and/or biological activity (Filipič and Baričevič 1997, 1998; Bolta *et al*. 2000; Baričevič *et al*. 2001a, 2001b; Zupančič *et al*. 2001; Zupančič and Baričevič 2002);
- Evaluation of susceptibility of descendants to environmental stress (drought, low temperature, depleted soils, etc.) in pot trials in a controlled environment (Baričevič 1995; Baričevič *et al*. 1999; Železnik *et al.* 2002; Železnik and Baričevič 2002).

### *In situ* conservation

Slovene botanists have made an inventory of the Slovenian flora (not exclusively MAPs) throughout the last decade. The inventory is intended to provide the basis for conservation of the remaining natural resources of rare and/or endangered species. The result of this intensive research was "The Red Data List of Threatened Vascular Plants in Slovenia" (Wraber and Skoberne 1989) upheld by the Institute for the Conservation of Natural and Cultural Heritage of Slovenia. This document is based on IUCN categories and results from

the classification of endangered plants according to the degree of danger. Amongst approximately 3200 plant species (400 of them MAPs) known to be indigenous or well adapted to the Slovenian climate, some 10% are considered to be endangered (34 have been harmed, 77 are vulnerable, 192 are rare). Slovenia has shown much concern in the preservation of natural resources such as medicinal plants.

In Slovenia, directives for the national programme of production, processing and quality control of MAPs have been developed. The strategy for the conservation of natural resources is perceived as of the utmost importance. The first stage of the programme consists in the monitoring and characterization of natural populations and *in situ* conservation of evaluated indigenous plant material. Data from different sources (e.g. Ljubljana Herbarium) and plant inventories are used to obtain data on wild MAP accessions in Slovenia.

Conservation of the natural heritage for future generations, followed by the importance of attractive landscapes, are among the principal aspects of the conservation strategy in the field of MAPs. This can be achieved by:

- collecting and estimation of the danger level;
- active conservation (conservation *in situ*);
- sensible/sustainable use of wild plants (for *ex situ* conservation);
- limitation of massive exploitation of wild plants by successive introduction into cultivation of known genotypes in suitable environments.

In Slovenia, *in situ* inventorying/monitoring and estimation of population density as well as quality control with the objective of defining particular/optimal market use of raw materials are planned to be carried out through the multi-user relational database MEDPLANT. Wild populations will be included in the national collection of medicinal and aromatic plants. Further activities related to future cultivation (such as propagation of plant materials and seed breeding) are planned.

### On-farm conservation

Principles and techniques of on-farm conservation of local ecotypes and landraces of cultivated plants and traditional cultivation techniques have been widely introduced in less developed parts of the world. In the last few years a need for similar actions was identified for Europe, where a high degree of genepool erosion was found in field crops.

In Slovenia, there is no special institution engaged in the programme of on-farm conservation, breeding or management. Some of the institutions that maintain genetic resources of different crops have contacts with individual farmers, who maintain old varieties on farm. Some NGOs do include activities similar to on-farm conservation but there is little coordination between different parties. There is almost no cooperation between farmers and genebanks in the field of breeding or genetic improvement of local ecotypes.

Regarding MAP on-farm conservation, some valuable species are found in Slovenia. The most interesting crop that is considered as a traditional oil and medicinal plant is *Camelina sativa* (L.) Crantz, still widespread in the Koroška region. It is predominantly used as a raw material in oil processing. Some incomplete data on its cultivation were collected by the extension service offices in Dravograd and Slovenj Gradec and need further survey. Now almost 20 seed samples are under investigation at the Agricultural Faculty of the University of Maribor and partly in the fields of the Institute for Hop Research and Brewing in Žalec, where some data on cultivation technology and oil processing machinery have been collected.

The second interesting potential medicinal crop is buckwheat, *Fagopyrum esculentum* Moench. Buckwheat was very popular in the old days and some knowledge about technology and landraces is still available. But since very good cultivars were developed in

Slovenia they have been preferred by the extension service and many old varieties are no longer used. At the Biotechnical Faculty of the University of Ljubljana the buckwheat genebank maintains a number of landraces from Slovenia. Its cultivation has decreased considerably in recent decades because of crop failures. Unfortunately the records on genetic resources have been lost because the farmers were not encouraged to maintain old varieties in the past. Besides being a valuable source of healthy food, indigenous accessions need re-evaluation as MAPs.

### Home gardens as a source of biodiversity

The idea of studying and conserving biodiversity in home gardens, proposed by the task force for on-farm conservation, developed from the on-farm practices. In many European countries much biodiversity present in the fields has been lost because of large-scale production of new crop cultivars. Home gardens could be considered as a refuge for many landraces of traditional crops. They are less susceptible to the pressure of commercial seed producers. But the diversity is rapidly decreasing, concomitantly with the ageing of the people (women) maintaining the gardens. Among species maintained in the home gardens MAPs can also be found. In Slovenia interesting species, traditionally kept in the home gardens, are: *Dracocephalum moldavicum* (south-west Slovenia), *Artemisia dracunculus* L., *Carthamus tinctorius* L. (Koroška), *Salvia officinalis* L., pelargoniums with scented leaves, *Ruta graveolens* L., *Artemisia abrotanum* L., *Artemisia absinthium* L., old landraces of tobaccco (Goričko, Premurje), *Oenothera biennis* L., *Tanacetum balsamita* L., *Tanacetum parthenium* (L.) Schultz-Bip. and others.

In Slovenia there is no special funding for MAP on-farm conservation activities. They are partly incorporated into the routine work of the MAP genebank. In 2001 the first serious attempts at cooperation with the municipality of Dravograd in the Koroška region were conducted for *Camelina sativa*, but the funds are still not available. There is a need to build awareness of on-farm conservation and MAP conservation in Slovenia among farmers, scientists, the general public, processing industry and decision-makers.

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## *Current status of medicinal and aromatic plants in Spain*

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### Legal protection of medicinal and aromatic plant species and their natural habitats

In Spain the legal protection of nature is under the responsibility of the Spanish Governement and of the "Autonomous Communities", each regulated by its own laws.

The general legislation at national level includes numerous laws and decrees. Reference documents include, *inter alia:*

- the catalogue of threatened species (*Catalogo de especies amenazadas*); in April 2000 the following categories were identified: threatened species (118), species sensitive to habitat alteration (5), vulnerable species (4) and species of special interest (133).
- the Red List of the Spanish vascular flora (Aizpuru *et al.* 2000).

The catalogue of threatened species provides local regulations for the communities of Catalunya, Aragón, Asturias, La Rioja, Navarra, Madrid, Euskadi, Valencia, Andalucia, Canarias, Murcia and Castilla-La Mancha.

Other by-laws exist for Catalunya, Aragón and Castilla-La Mancha.

### *Ex situ* conservation / Inventory of MAP natural resources at national level

Botanical gardens such as the Jardín Botánico de Córdoba, Jardí Botànic de Soller, Jardín Botánico de Madrid and some research institutes and universities are involved in the conservation of MAPs, mainly in germplasm banks. The biological material conserved in these banks usually comes from various projects carried out by these centres. There are currently four projects dealing with these subjects:

1. **Collection, conservation and characterization of germplasm from Spanish populations of different species of the genera *Rosmarinus* and *Origanum*** (Elena Gonzalez, EUETA, Dept. Biología, Ciudad Universitaria s/n, 28040 Madrid). The main aim of this project is the study of species populations (listed in Table 1) and their conservation in the germplasm bank of the Universidad Politécnica de Madrid.
2. **Screening of MAP genetic resources in Castilla-La Mancha and Extremadura** (Federico Varela, Instituto Nacional de Investigaciones Agrarias, Centro de Recursos Fitogenéticos, Apartado 1045, Alcala de Henares, Madrid). Since 2000 this team has been collecting samples as listed in Table 1.
3. **Screening of MAP genetic resources in Aragón** (Jesús Burillo, Servicio de Investigación Agroalimentaria, Apartado 727, 50071 Zaragoza). Seed samples studied in Aragón are listed in Table 1.
4. **Screening of some MAP species in Catalunya** (no list available).

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[18] Full details: see list of participants.

**Table 1.** Projects carried out in Spain on MAP species

| Project | Species studied | No. of populations studied |
|---|---|---|
| Collection, conservation and characterization of germplasm from Spanish populations of different species of *Rosmarinus* and *Origanum* | *Rosmarinus officinalis* L. | 19 |
| | *R. eryocalys* | 2 |
| | *Origanum vulgare* (subsp. *virens* and *vulgare*) | 16 |
| | *Arctostaphylos uva-ursi* | 21 |
| Screening of MAP genetic resources in Castilla-La Mancha and Extremadura | *Hypericum perforatum* | 47 |
| | *Lavandula latifolia* | 17 |
| | *Lavandula pedunculata* | 28 |
| | *Lavandula stoechas* | 22 |
| | *Satureja montana* | 5 |
| | *Thymus mastichina* | 58 |
| | *Thymus vulgaris* | 64 |
| | *Thymus zygis* | 56 |
| Screening of MAP genetic resources in Aragón | *Althaea officinalis* | 1 |
| | *Arctostaphylos uva-ursi* | 2 |
| | *Artemisia absinthium* | 2 |
| | *Evonymus europaeus* | 1 |
| | *Hypericum perforatum* | 3 |
| | *Lavandula latifolia* | 4 |
| | *Melilotus officinalis* | 1 |
| | *Plantago major* | 1 |
| | *Ricinus communis* | 1 |
| | *Santolina chamaecyparissus* | 1 |
| | *Tanacetum parthenium* | 1 |
| | *Thymus mastichina* | 19 |
| | *T. vulgaris* | 19 |
| | *T. zygis* | 1 |

### Review of the acreage of MAPs under cultivation

According to various information sources the main species cultivated commercially in Spain are lavandin (*Lavandula* x *intermedia*), mint (*Mentha piperita* and *M. pulegium*), sage (*Salvia officinalis* and *S. lavandulifolia*), fennel (*Foeniculum vulgare*), lemon balm (*Melissa officinalis*), chamomile (*Matricaria chamomilla*), oregano (*Origanum vulgare, O. virens*) and anise (*Pimpinella anisum*); there have also been recent experiences in the production of echinacea (*Echinacea purpurea*), lemon verbena (*Lippia citriodora*) and spike lavender (*Lavandula latifolia*). The main cultivation areas are Andalucia, Valencia, Murcia, Catalunya, Aragón and Mallorca, and, although there are no data for the current area cultivated, it can be estimated to be about 6000 ha in the whole of Spain. There are two kinds of farm: the small ones are more or less experimental, while the bigger ones are usually managed directly by foreign companies.

### Use of MAPs by processing industries

In Spain, 10-20% of the plant raw material used by the industry comes from Spain and 80-90% is imported, mainly (40-60%) from eastern Europe.

Every year about 100 000 ha are subjected to collecting from the wild in Spain. The main collecting areas are Granada, Almería and Murcia, all in southern Spain.

Some medicinal plants are collected in large amounts (more than 1 t/year). Most of them do not have major conservation problems (Box 1). However Blanco and Breaux (1997) reported that some wild plants collected in significant quantities in Spain have potential conservation problems and it is necessary to update the collecting regulations as soon as possible (Box 2).

**Box 1.** Collected species with no major conservation problems (van Ginkel 2002)

| | |
|---|---|
| *Equisetum ramosissimum* Desf. subsp. *ramosissimum* | *Lavandula* spp. |
| *E. telmateia* Ehr. | *Paronychia* spp. |
| *Lepidium draba* L. | *Jasonia glutinosa* De Cand. |
| *Juniperus communis* L. | *Centaurea aspera* L. |
| *Taraxacum officinale* Weber. | *Artemisia campestris* L. |
| *Arctostaphylos uva-ursi* Spreng. | *Spiraea ulmaria* L. |
| *Gentiana lutea* L. | *Capsella bursa-pastoris* Medik. |
| *Malva sylvestris* L. | *Santolina* spp. |
| *Viscum album* L. | *Sideritis* spp. |
| *Rosmarinus officinalis* L. | *Satureja* spp. |
| *Thymus* spp. | Others |
| *Salvia* spp. | |

**Box 2.** Species with potential conservation problems (Blanco and Breaux 1997)

| | | |
|---|---|---|
| *Arnica montana* | *Gentiana lutea* | *Petasites hybridus* |
| *Cetraria islandica* | *Jasonia glutinosa* | *Ruscus aculeatus* |
| *Drosera rotundifolia* | *Menyanthes trifoliata* | *Valeriana officinalis* |

## Experience in sustainable use of MAPs

Various organizations have carried out local projects in order to promote the sustainable use of MAPs and their cultivation. However in most cases the marketing of the final products has been too difficult or impossible, and most of these initiatives have been abandoned.

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# *Activities on medicinal and aromatic plants at the Aegean Agricultural Research Institute*

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The use of medicinal and aromatic plants (MAPs) has a long history in the world. According to the World Health Organization, 20 000 species are used for medicinal and aromatic purposes. Today, 4000 drugs are widely used and 10% of them are commercially exploited or produced. Around 500 plant species are used for medicinal and aromatic purposes in Turkey (Başer 1998). However, all these species are neglected or underutilized and only a few are cultivated or subjected to research. The Aegean Agricultural Research Institute (AARI) is one of the institutions working on and giving importance to MAPs to improve commercial varieties, find out new crops and assess agronomic requirements of related plant species.

Activities on MAPs at AARI were started in the 1970s. The first studies were on identification, collection and conservation of MAPs. The most economically important species were given research priorities. The MAPs studied in this institute and research results obtained are summarized below.

## Anise (*Pimpinella anisum* L.)

Studies on anise were started in 1979. Several seed lots were collected from farmers growing anise in Turkey. After six years, a cultivar called 'Gölhisar' was registered.

Another study focused on the changes in current and real production costs of anise between 1989 and 1993. In this study, data were obtained from 67 farmers using different farming systems. Labour and weed control costs were the highest share of the total production cost (Fig. 1). Anise prices did not repay production costs during the research period.

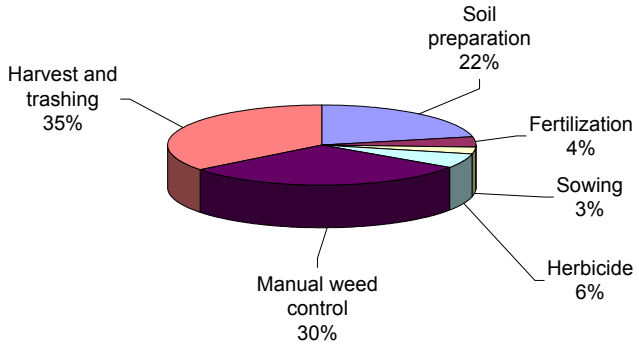


**Fig. 1.** Rates of production inputs in anise production.

Anise growers used to broadcast anise seed. Research on sowing methods and plant density was conducted for three years. Anise yield increased while plant density was increased from 20 plants/m² to 90 plants/m² (Fig. 2). After seeing the research results, farmers started to sow, harvest and thrash the anise mechanically. Production costs decreased and low rural incomes increased.

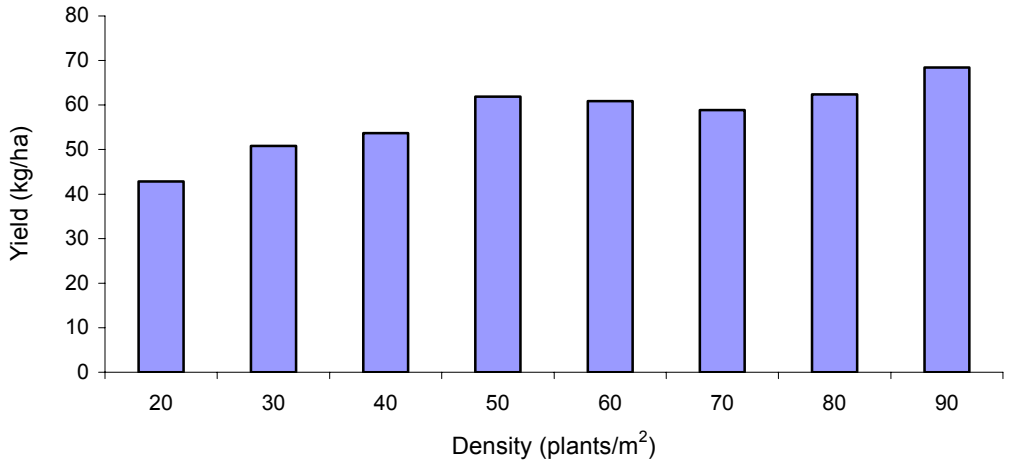


**Fig. 2.** Effect of plant density on anise (*Pimpinella anisum* L.) yield.

In another study, weed species and the effect of some herbicides were identified in anise fields in the Aegean region of Turkey. The most important species are as follows: *Chenopodium album, C. glaucum, Centaurea cyanus, Convolvulus arvensis, Chondrilla juncea, Chonringia orientalis, Coronilla scorpioides, Cynodon dactylon*, *Euphorbia exigua*, *Gallium tricorne, Geranium dissectum, Hypocum procumbens, Lathyrus aphaca, Lithospermum arvense, Polygonum aviculare, Polygonum* sp., *Ranunculus arvensis*, *Sinapis arvensis*, *Tragopogon* sp., *Vicia* sp. (Otan *et al.* 1993). The effects of herbicides used in this study varied with years and locations; however the highest yield was obtained with mechanical weed control methods.

### Caper (*Capparis spinosa* L.)

Experiments were conducted on caper (*Capparis spinosa* L.) during 1991-1993. Several experiments on germination and sowing methods were conducted to find out the best method to grow caper. Direct seed sowing in the field was not suitable. Instead, sowing non-stratified seeds in raised seed beds and transplanting seedlings were the best methods (Otan and Sarı 1994).

In another study made between 1997 and 1999, yield and morphological characteristics of a caper population were assessed. There were significant variations among caper plants for yield (ranging 0-87 g), number of primary branches (2-12), primary branch length (50-164 cm) and canopy width per plant (80-260 cm) (Table 1). In addition, there were significant positive correlations between the following characters: yield and number of primary branches, branch length and canopy width. The plants in the population had three growth types (prostrate, semi-erect and erect) in the proportions 51%, 29% and 20%, respectively (Sarı *et al.* 2002).

**Table 1.** Yield, number and length of primary branches and canopy width intervals and frequencies (F) of a caper (*Capparis spinosa* L.) population

| Yield (g) | F (%) | No. of primary branches | F (%) | Primary branch length (cm) | F (%) | Canopy width (cm) | F (%) |
|---|---|---|---|---|---|---|---|
| 0 | 8 | 5< | 4 | 50-80 | 24 | 100< | 1 |
| 11-30 | 71 | 5-10 | 87 | 80-110 | 40 | 100-150 | 20 |
| 31-50 | 11 | >10 | 9 | 111-150 | 32 | 151-200 | 45 |
| 51-60 | 5 | | | >150 | 4 | 200-250 | 32 |
| 61-80 | 4 | | | | | >250 | 2 |
| >80 | 1 | | | | | | |

The germination rate for untreated caper seeds is quite low. Some seed treatments were tested to increase germination. Freshly harvested seeds of the year 1999 germinated at 29% when immersed for 12 h in 2000 ppm $GA_3$. In control seed lots the germination rate was only 1% (Fig. 3).

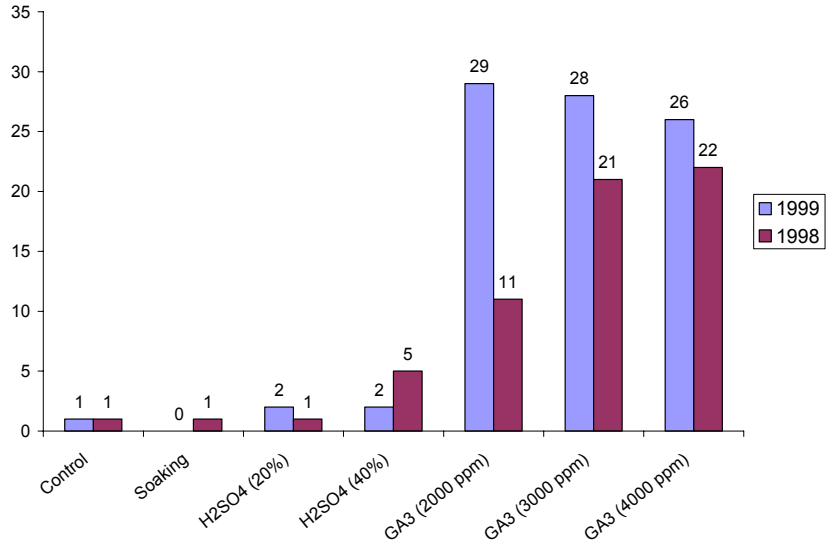


**Fig. 3.** Effect of different seed treatments on germination of caper (*Capparis spinosa* L.) seed lots.

## Oregano (*Origanum onites* L.)

The effects of three levels of nitrogen application (0, 60, 120 kg/ha) and four different planting patterns on *Origanum onites* L. were investigated for three years (between 1991 and 1993) in three locations. Green herb and drug leaf yield and essential oil percentage were measured. Yields and essential oil percentage were generally highest at 45 x 15 cm with an application of 60 kg/ha nitrogen (Table 2).

**Table 2.** The effects of four planting patterns and three nitrogen levels on green herb and drug leaf yields and essential oil percentage of oregano (*Origanum onites* L.)

| | Green herb yield (kg/ha) | Drug leaf yield (kg/ha) | Essential oil (%) |
|---|---|---|---|
| **Planting pattern** | | | |
| 45 x 15 cm | 26520 | 5210 | 2.93 |
| 70 x 10 cm | 23320 | 4580 | 2.96 |
| 22.5 x 30 cm | 25570 | 5170 | 2.91 |
| 25 x 20 cm | 25070 | 4880 | 2.89 |
| **N level** | | | |
| $N_0$ | 24060 | 4800 | 2.93 |
| $N_{60}$ | 25270 | 4990 | 2.97 |
| $N_{120}$ | 26030 | 5100 | 2.88 |

In another study, a sample of 52 *Origanum* spp. were collected from 12 provinces situated in the Aegean and West Mediterranean regions of Turkey. Samples of 10 species (*O. sipyleum* L., *O. onites* L., *O. majorana* L., *O. vulgare* L., *O. saccatum* P.H.Davis, *O. syriacum* L., *O. hypericifolium* O.Schwarz & P.H.Davis, *O. laevigatum* Boiss. and *O. bilgeri* P.H.Davis) and two hybrids (*O. sipyleum* x *O. vulgare* subsp. *hirtum*, *O. onites* x *O. vulgare* subsp. *hirtum)* belonging to *Anatolican, Chilocalyx, Majarona, Prolaticorolla, Amaracus* and *Origanum* sections were investigated for 40 characters (Kıtıkı *et al.* 1997). The chromosome numbers of *O. vulgare*, *O. sipyleum* and *O. onites* were found as 2n=30. The samples could not be distinguished by their cytological characters. Principal component analysis (PCA) was applied on morphological and anatomic characters. According to PCA results, the *Anatolican* section formed a distinct group from the main group which consists of other sections and hybrids.

A breeding programme has been continued on *O. onites* L. since 1992. Initiation material was constituted by 5000 single plants originating from different locations in western and southwestern Turkey. Today, two groups of selected lines consisting of four and three lines having similar morphological and agronomic characters, high yield and quality, are grown to develop two synthetic lines. There will be two commercial synthetic varieties in a few years.

### Sage (*Salvia officinalis* L.)

The effect of nitrogen application (0, 80 and 160 kg/ha) and six different cropping systems on *Salvia officinalis* L. (common sage) were investigated in three locations for three years, between 1991 and 1993. Green herb, drug herb, drug leaf, essential oil %, essential oil yield, dry matter, plant height, plant nutrient composition (Mg, N, Na, K, Ca and P) and composition of essential oil were examined. Only the results for drug leaf yield and essential oil % are presented here (Table 3).

**Table 3.** The effects of six planting patterns and three nitrogen levels on drug leaf yields and essential oil % of sage (*Salvia officinalis* L.)

| | | Row space | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | | 45 cm | 70 cm | 11.5 cm | 22.5 cm | 12.5 cm | 25 cm | Mean |
| **Drug leaf yield (kg/ha)** | $N_0$ | 9560 | 7650 | 7510 | 7560 | 7550 | 7690 | 7920 |
| | $N_{80}$ | 9350 | 7530 | 8860 | 8560 | 8680 | 8810 | 8630 |
| | $N_{160}$ | 9400 | 7570 | 7850 | 8130 | 8040 | 8800 | 8300 |
| | Mean | 9440 | 7580 | 8070 | 8080 | 8090 | 8430 | 8280 |
| **Essential oil (%)** | $N_0$ | 1.89 | 1.90 | 1.88 | 1.91 | 1.85 | 1.83 | 1.88 |
| | $N_{80}$ | 1.83 | 1.81 | 1.89 | 1.85 | 1.87 | 1.96 | 1.87 |
| | $N_{160}$ | 1.84 | 1.76 | 1.81 | 1.81 | 1.80 | 1.88 | 1.82 |
| | Mean | 1.85 | 1.82 | 1.86 | 1.86 | 1.84 | 1.89 | 1.85 |

### Lemon balm (*Melissa officinalis* L.)

Eleven lemon balm (*Melissa officinalis* L.) populations originating from different sources in Turkey and European countries were investigated for three years in two ecologically different locations, Menemen and Bozdağ, to find out the populations giving high quality and yield. There were significant variations between locations, years and populations for yield and quality characters. The ecology was highly suitable in Menemen to grow lemon balm successfully and almost all yield and quality characters at Menemen were significantly higher than those in Bozdağ (Table 4). The growth of populations in this study increased after the first year of trials in both locations; therefore all aspects of yield were significantly higher in the second and third years of trials compared with the first year. The populations

numbered 9 and 7, originating from Germany and Romania respectively, had high yield and quality and were the two most promising populations for lemon balm cultivation in these locations.

**Table 4.** Yield and quality characters of lemon balm (*Melissa officinalis* L.) in two locations (Sarı and Ceylan 2002)

| Location | Plant height (cm) | Green herb yield (kg/ha) | Drug leaf yield (kg/ha) | Essential oil (%) |
|---|---|---|---|---|
| Menemen | 47.58 | 28690 | 4969 | 0.067 |
| Bozdağ | 20.73 | 4160 | 900 | 0.036 |

The main component of the essential oil of the lemon balm was geranial in both locations and geranial concentration averaged over years and populations was 38.13% in Menemen and 53.68% in Bozdağ. Hence, Menemen ecological conditions were quite suited to growing lemon balm with a high yield and quality and the populations originating from Romania and the Ege University Agricultural Faculty (Germany) were especially recommended to growers.

### Mint (*Mentha* spp.)

A sample of 232 different mint (*Mentha* spp.) populations were collected from different regions of Turkey in collaboration with the Turkish Extension Service and conserved in a field genebank at AARI. Mint populations in the conservation field were characterized using morphological and yield parameters. Results were as follows: total green herb ranged 10 160-168 240 kg/ha, drug herb yield 3760-46 760 kg/ha, drug leaf yield 1000-28 000 kg/ha, and plant height 28-116.5 cm.

### *Sideritis* spp.

A sample of 68 *Sideritis* L. was collected from 12 provinces of Aegean and Mediterranean regions (Otan *et al.* 1994). Morphological, anatomical and cytological characters of 20 species belonging to *Empedoclia* and *Hesiodia* sections were examined. Chromosome numbers were found to be 2n=28 for species of the *Hesiodia* section and 2n=32 for those of the *Empedoclia* section. Submetacentric chromosomes were found on *S. leptoclada* and some samples of *S. perfoliata*. Hairy and glabrous samples of *S. perfoliata* were also different according to centromer position of their chromosomes. Chromosome 1B was observed for *S. tmolea.* There was intra- and interspecific diversity for stem, pollen and leaf characters. PCA was applied to morphological and anatomical characters. According to PCA the *Hesiodia* section formed a distinct group from the main group which consists of the *Empedoclia* section.

The interest and demand for *Sideritis* species is increasing nowadays because of their antioxidant properties. Hence a new research project was initiated in 2002 to determine optimum plant density and agronomic characters of *S. perfoliata*, one of the most promising species for cultivation.

### Dye plants

Synthetic dyes were invented in the 19th century and replaced natural dyes owing to their easy application, high fastness and vibrant colours. However, it is realized that synthetic dyes may harm the environment. Environmental concern and interest for environmental protection and conservation has increased since the 1980s. As a consequence, interest and demand for natural dyes and items coloured with them such as carpets, rugs and foods have also increased since that time. However, dye plants are not cultivated but collected from the wild, with damage to natural habitats.

A new research project was started to conserve natural habitats and obtain standard natural dye extracts. Agronomic and chemical characters and dying properties of three dye plants, woad (*Isatis tinctoria*), madder (*Rubia tinctorum*) and weld (*Reseda luteola*), providing three main colours (blue, red and yellow, respectively) will be investigated.

### Indigenous knowledge

The indigenous knowledge on usage of MAPs as folk remedies is getting lost owing to migration from rural to urban areas, industrialization and changes in life style. The Aegean region in Turkey is one of the areas most affected by industrialization. A new project was initiated to collect and record the information on MAPs used as folk remedies.

### Possible cultivation methods for orchid species

The use of salep (ground orchids bulbs) has a long history in Turkey. It is estimated that about 20 million orchid bulbs are collected annually in Turkey. This figure clearly shows that population density decreased dramatically due to annual harvesting from the wild. A new research project aims to find out cultivation methods for salep orchids.

### Genebank activities

Target areas and species are determined every year. Herbarium samples collected from those areas are submitted to the genebank. In addition, seed multiplication is carried out for seed lots having insufficient or weak germination rates. Characterization and evaluation studies for selected species are also conducted for the genebank.

### Seminars and training courses

Seminars and training courses on MAP cultivation for Extension Service experts and farmers are held at AARI every year. The number of trainees ranges between 10 and 50 from year to year.

### Conclusion

There are many plant species in the world with medicinal properties. However, almost all MAPs are neglected or underutilized and insufficiently documented. Cultivation of MAPs is limited and most are wild crafted, causing degradation of natural habitats. Therefore, a need for conservation is arising for many species. The number of research activities, scientists studying MAPs and species investigated should be increased to produce information and better understanding to conserve MAPs. An international network storing information on MAPs should be established for better use of MAPs and sharing related information.

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## *Medicinal and aromatic plant production in the United Kingdom*

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Natural solutions are increasingly being sought to agrochemical, pharmaceutical and food issues and the market for herb products is growing rapidly, putting an additional strain on resources as many herb species are wild harvested. A report by Lange (1998) claimed that 70-90% of herbal products imported into Germany are wild harvested. Current production of herbs in the UK occupies a relatively small area (ca. 4000 ha), mostly of culinary herbs. Herb companies still import large quanitites of herbs which could be grown and there is considerable potential for expansion to provide industrial raw materials. In order to realize this potential and thus meet the UK government's policy aim of encouraging the development of crops for industrial applications, a number of projects have been initiated. But evidence of the biological activity of these materials, the feasibility and economics of UK production of these crops needs to be obtained.

A number of factors have been shown to influence the yield and composition of essential oils and bioactives from herb crops. These include genetics (origin, variation), morphogenesis (leaf position and age, harvest, flowering), environment (temperature, day length and light intensity) and finally agricultural practices (nutrition, irrigation, propagation, harvesting and extraction). Selection of plant material and the optimization of agricultural practices is vital for the production of high quality herbs, essential oils and extracts. The need to know how the crop has been produced (traceability of raw materials) and concern over the quality of imports (pesticides, heavy metals) suggests there is scope to supply extracted natural products from herbs grown in the UK to provide high quality raw materials for industry. There is potential to develop the UK herb industry, which encompasses a number of small- and medium-sized companies, if they meet the stringent specifications, supply chain and quality requirements. It is possible to capitalize on this opportunity with a better knowledge of cultivar selection, how herbs can best be grown under UK conditions and by developing new techniques which may lead to safer and more efficient extraction of natural products.

Below are three projects in which the National Herb Centre is involved.

**The Rapfi project** (*Rosemary antioxidants for the pharmaceutical and food industries)* is part of the Competitive Industrial Materials from Non Food Crops (CIMNFC) programme and is a collaboration between the National Herb Centre (NHC), Department of Biology at the University of Reading, Department of Sonochemistry at the University of Coventry, J.K. Kings of Coggeshall (Seedsmen), Advanced Phytonics (Extraction), Langford Electronics (Ultrasonic equipment) and Checkmate (Audit trail). The project objectives are to select rosemary accessions for high antioxidant activity, enhance antioxidant activity through the use of elicitors, produce a predictive model relating antioxidant activity to UV radiation, to develop an affordable extraction process and produce an audit trail for the whole production process.

Accessions of *Rosmarinus officinalis* were collected from throughout Europe. In total 80 accessions have been trialed and analyzed at the National Herb Centre. Results have shown that antioxidant potential as measured by the free radical scavenging activity and chain propagation correlated with carnosic acid (r=0.82 and 0.53 respectively). Rosemary antioxidants also reduced production of "off flavours" such as hexanal. Rosemary accessions were therefore selected for antioxidant activity by screening for carnosic acid concentration. Field trials (30 accessions and 6 replicates) had been planted September 1999 in three

different areas of the UK. Analysis of samples showed that plants were reproducible at sites and between sites for carnosic acid. In addition there was a fourfold range of carnosic acid analyzed between low and high carnosic acid producers. Six high-yielding accessions were selected and propagated (1000 plants) to provide for larger scale field trails at the three sites, planting in spring 2001 for harvest in 2002.

Factors affecting the concentration of carnosic acid and antioxidant activity have been investigated using abiotic and biotic elicitors and also the effects of water stress. In addition the essential oil profiles and yields have been compared with the concentrations of antioxidants to determine whether there is a "trade off" in production.

**The Phytoderm project (***Optimized production and extraction techniques for consistent yield and quality of skin-protecting phytochemicals)* is also part of the CIMNFC programme and is a collaboration between ADAS, NHC, Molecular Nature Ltd., University of Leeds, Express Separations, Boots PLC, Cornwall College and Hydroponic Herbs. The project is investigating the effects of root polysaccharides of *Astragalus membranaceus* and *Althaea officinalis* on cell surface markers, their skin healing and anti-ageing properties. For both plants, the identity and structure of the active compounds is largely unknown, and will greatly affect the optimal extraction protocol. UV absorbers are important to reduce UV-mediated skin ageing, photoallergenic reactions and even skin cancer. Preliminary evidence suggests that both *Althaea* and *Astragalus* roots contain unusual UV-absorbing compounds.

Hydroponic or soil-less culture techniques are being investigated to enable greater control over production, significantly increasing biomass yield per unit time and content of active compounds and with less variability in a range of plants grown for phytochemicals.

**Antimicrobial and pesticidal properties of herbs:** this project is funded by the Department of Food and Rural Affairs (DEFRA). Herbs and herb extracts have been implicated as pesticidal and antimicrobial replacements for synthetic materials. The scientific literature is overflowing with references to the pesticidal properties of plant-derived chemicals but the variety of ways in which this data has been generated makes it difficult to assess relative potencies and therefore the most promising candidate materials. In addition to their activity as insecticides, many herb oils have been implicated as potential antiobitics/preservatives, although few reports of controlled quantitative work can be found in the literature. Before any of the legislative and political issues surrounding these issues can be addressed, a good scientific basis for these claims needs to be established. This project is investigating the role of the major and minor components of essential oils of four UK-grown herbs to establish the proof of efficacy over a range of organisms, to establish the active component(s) and to allow prediction of the most promising species and chemotypes for further exploitation.

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# *Genetic resources of medicinal and aromatic plants of Yugoslavia – current situation and further prospects*

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## Introduction

During the last decades the demand for medicinal plant raw material has rapidly increased, especially in developed western countries. This is due to the recent tendency of pharmaceutical companies to process the natural, often cheaper, raw material, but also to a "modern human conscience" for development of alternative healing methods, such as aromatherapy and homeopathy. The increased use of medicinal and aromatic plants (MAPs) in the last decades of the 20th century was motivated by both emotional and rational factors (Lewington 1990; Lange 1996, 1998). (The natural products of MAPs are believed to have no adverse effects, be less toxic and more appropriate for healing). According to recent data of Traffic International (Lange 1998) the import of MAPs into European countries increased by 21% in the period 1992-1996.

Significant and usually secure income generated by the sale of MAPs results in a strong decline in the natural medicinal flora caused by enormous and/or unprofessional exploitation. In ecologically suitable environments of southeastern and eastern European countries, the main exporters of cheap and good quality plant raw material, there is a trend towards the endangering or complete elimination of many important medicinal species. The great floristic richness and mapping of certain medicinal plant habitats in Serbia (Kišgeci *et al.* 1997; Dajić and Kojić 1997, 1998, 1999; Dajić *et al.* 2000; Kišgeci and Sekulović 2000; Dajić and Fabri 2001) enable their exploitation, but simultaneously draw attention to the need for protection of medicinal plant resources in the wild. Therefore this situation requires changes in the legislation of MAP protection and an increase in their production.

## Biogeographic situation of Yugoslavia and elements of biodiversity

Yugoslavia is characterized by a huge geographic and biological diversity reflected in the richness of its indigenous flora which exists as various vegetation formations of terrestrial and aquatic ecosystems.

Yugoslavia is part of the Balkan Peninsula. Its whole territory covers 102 173 km$^2$, with five main biogeographic regions: (i) Mediterranean, (ii) central European, (iii) pontic, (iv) boreal and (v) central-south European mountainous (Meusel *et al.* 1965; Walter 1983; Stevanović 1994).

Dominant ecosystems (biomes) in Yugoslavia include deciduous forests, steppes, coniferous forests, meadows and alpine "tundra", as well as the Mediterranean and sub-Mediterranean vegetation distributed in the coastal region of Montenegro.

Yugoslavia is considered one of the 158 world biodiversity centres, based upon the total number of plant species (including mosses) (more than 4700). The main factors of such floral diversity are: historical background, geographical position, climate, relief (with three main areas: the Panonnian plain, hilly-mountainous region and coastal region of Montenegro), presence of streams, etc.

According to the most recent estimates (Stevanović *et al.* 1995) the flora of Serbia contains 3662 taxa (and an estimated additional 120 taxa), i.e. 3272 species and 390 subspecies, which

[19] Country name changed to Serbia and Montenegro in February 2003. The denomination Yugoslavia (or F.R.Y.) is kept in the present text as it was at time of meeting and preparation of this paper.

makes Serbia a country with very high floristic diversity and density per unit area compared to other European countries. All plant species are included in 141 families and 766 genera.

In addition to its position in the Balkans, Serbia also occupies the southeastern part of the Pannonian plain in the Vojvodina region. It is thus divided into two distinct geographical and orographic entities which are, though separated by the rivers Sava and Danube, interconnected by lowland hilly peripannonian Serbia. These basic geographical and landscape entities have determined the characteristics of the Serbian flora and vegetation, as well as the differences between the plants distributed in these two micro-regions of Serbia.

Montenegro covers a territory of 13 812 km$^2$. A total of 3136 plant taxa (2920 species with 216 subspecies) have been recorded (Stevanović *et al.* 1995) belonging to 804 genera and 151 families. The floristic richness of Montenegro can be explained by various causes, including geography (presence of coasts and mountains), historical and other factors.

According to the total plant species number per territory size (LogS/LogA), Yugoslavia ranks fifth among European countries, Serbia eighth and Montenegro first.

Considering the vegetation diversity of Yugoslavia, over 600 individual plant communities have been identified, including forests (deciduous and coniferous), steppes, Mediterranean vegetation, meadows, pastures, vegetation of sand dunes and saline soils, swamps, alpine tundra, etc.

MAP diversity is also quite high. More than 700 species are considered as medicinal (Sarić 1989), which accounts for 17.29% of the total flora. Among those, 420 plant species are officially registered (10.75% of the total flora), while 279 are to be found in Serbian markets. Medicinal plants of Yugoslavia encompass 89 families, of which the following should be quoted: Lamiaceae (41 species), Asteraceae (40), Apiaceae (20), Ranunculaceae (19), Scrophulariaceae (17), Malvaceae (15), Rosaceae (15), Brassicaceae (10), Polygonaceae (10), etc.

### Significance of natural resources

Meadows and forests are the main resources of indigenous medicinal and aromatic plants.

A recent analysis of MAP distribution in Serbian forests (Obratov *et al.* 2002) showed that in oak communities (ass. *Quercetum frainetto-cerris,* the most common forest community of Serbia), a total of 123 plant species have been identified, including 53 medicinal plants. Some of the most important medicinal plants in oak forests are the following: *Achillea millefolium, Acer tataricum, Ajuga reptans, Campanula glomerata, Crataegus monogyna, Cornus sanguinea, Euphorbia cyparissias, Filipendula hexapetalla, Sorbus aucuparia,* etc.

At higher altitudes, above the oak zone, beech forests (ass. *Fagetum montanum*) are well developed. This association is present on two parent rocks: limestone and silicate. Different plant species are found on different types of parent rock. On limestone 45 medicinal plants were identified, of which 4 are recommended for exploitation: *Veronica officinalis, Tamus communis, Clematis vitalba* and *Corylus avellana.* On silicate parent rock 15 different medicinal plants were identified, of which two are of greater significance: *Calamintha officinalis* and *Asarum europaeum*.

The approximate total number of medicinal plant species present in Serbia is 400, with 122 medicinal plants in beech forests (about 30%).

Meadows and pastures of Serbia have not been sufficiently utilized, although they have great potential for cattle breeding. The total area covered by grasslands in Serbia is about 1.65 million ha. They are widespread in very diverse ecological conditions, occurring at different altitudes and on various forms of relief, soil type, climate, etc. Therefore, meadow vegetation comprises many plant communities differing in floristic structure, stability and type of succession. Besides agriculture, they may serve as a main source for medicinal plant exploitation and as a natural genebank of various populations suitable for further cultivation and selection.

The majority of medicinal plants grow spontaneously in meadows, sometimes forming very abundant groups. Intensive utilization of medicinal plants has led to a diminution of many plant populations and even disappearance of some species.

More than 200 MAPs growing in the meadows and pastures of Serbia were classified into 136 genera of 49 families with a predominance of hemicryptophytes (65.91%) and geophytes (10.23%) (Dajić *et al.* 2000).

The participation of MAPs in the entire floristic spectrum of the best developed meadow/pasture communities of hilly-mountainous regions in Serbia ranges from 21.7% to less than 50% (Dajić *et al.* 2000) (Fig. 1).

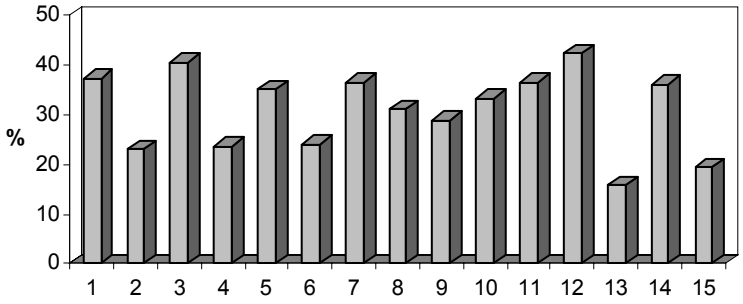


**Fig 1.** Participation of medicinal flora in different hilly meadow communities (%).

1. *Agrostietum vulgaris*
2. *Asperuleto-Agrostietum vulgaris*
3. *Agrostio-Festucetum valesiaceae*
4. *Festucetum valesiaceae*
5. *Rhinantho-Festucetum valesiaceae*
6. *Festuco-Chrysopogonetum grylli*
7. *Thymo-Chrysopogonetum grylli*
8. *Chrysopogonetum grylli*
9. *Chrysopogonetum pannonicum*
10. *Danthonietum calycinae*
11. *Cynosuretum cristati*
12. *Inulo salicinae-Calamagrostietum epigeio*
13. *Artemisio-Salvietum officinalis*
14. *Brometum erecti*
15. *Koelerietum montanae*

Regarding the potential of MAP utilization in Yugoslavia, it should be stressed that except hilly-mountainous region of Yugoslavia, which are rich in various types of forests and meadows, very important MAP resources are found in the northern lowland part of Serbia (Vojvodina) and the coastal region of Montenegro.

The area of Vojvodina is very suitable for MAP production because of its favourable environmental conditions (climate and soil). The commonest cultivated MAPs in this region are: chamomile, dill, mint, sweet marjoram, parsley, tarragon, marshmallow, celery, valerian, lemon balm, poppy, sage, black mustard, coriander, thyme, etc.; a significant number of wild MAPs has been also registered (Kišgeci and Sekulović 2000).

In Montenegro, collecting and processing of MAPs is a long tradition. Among wild MAPs, many of which belong to the Mediterranean and sub-Mediterranean floristic element, the following could be stressed: sage (*Salvia officinalis*), lavender (*Lavandula vera*), thyme (*Thymus* sp.), absinthe (*Artemisia absinthium*), xeranthemum (*Helichrysum arenarium*), oleander (*Nerium oleander*), rosemary (*Rosmarinus officinalis*), olive (*Olea europea*), jasmine (*Jasminum officinalis*), seaside onion (*Scilla maritima*), pyrethrum (*Chrysanthemum cinerariaefolium*) and many others (especially members of the Labiatae).

### *In situ* conservation

Legislation on MAP protection and conservation in F.R.Y. is directly connected with environmental protection in general. In this sense, the work on the ratification and implementation of different IUCN, CITES and UNESCO international conventions (Agenda 21, Rio Declaration, United Nations 1992; European Community Environment Legislation 1992; Bern Convention 1982; Council Regulation No 338/97, 1996; etc.) was of great importance.

Since 1993, legislative means and laws at both federal and republic levels were implemented in the domain of MAP protection and exploitation, such as the *Resolution of policy of environmental protection* (1993), *Resolution of policy of biodiversity maintenance (1994), Law of environment protection* (1993), *Law of protection of organisms rare in the nature* (1993) and *Direction of control, utilization and turnover of wild plant and animal species* (1993, 1996, 1999). All rare species (including MAPs) were categorized into the IUCN groups (EX = extinct, EW = extinct in the wild, CR = critically endangered, VU = vulnerable, etc.).

Some important endangered and vulnerable MAPs of the Yugoslav flora are: *Drosera rotundifolia* L., *Gentiana punctata* L., *Gentiana lutea* L., *Prunus laurocerasus* L., *Acer heldreichii* Boiss., *Pinus mugo* Turr., *Acorus calamus* L., *Adonis vernalis* L., *Helichrysum arenarium* DC., *Lycopodium clavatum* L., *Menyanthes trifoliata* L., *Orchis militaris* L., *Ruta graveolens* L. and *Veratrum album* L.

The collecting of species listed in the Red Book is strictly forbidden (Box 1).

The *Direction of control, utilization and turnover of wild plant and animal species* also lists precisely the quantities of certain species whose collection is allowed (together with proposed collecting methods), as well as the list of species whose collecting is forbidden (not only species from the Red Data Book, but also other endangered and vulnerable MAPs).

After adoption of the *Direction of natural rarities protection*, 17.7% of the species of the total Yugoslavian flora is under state control.

Currently 10 big national parks of Yugoslavia cover the main zones of western Serbia and almost all zones in Montenegro, whereas eastern Serbia and the Mediterranean part of Montenegro are still not sufficiently protected.

Among various activities that have to be undertaken urgently regarding nature protection, MAPs included, the following will be stressed:

- Implementation of the international conventions and partition of responsibility
- Identification of biodiversity components (natural goods of extraordinary significance for F.R.Y.)
- Monitoring
- Information systems
- Sustainable utilization of natural resources
- Conditions of access to MAP genetic resources (export, import, trade)
- Information exchange
- Prevention of massive exploitation through cultivation of endangered MAPs.

**Box 1.** MAPs from the Red Data Book of Serbia

**1. EXTINCT TAXA**

**1.1 Globally extinct taxa**

- *Althaea kragujevacensis* Pančić
- *Althaea vranjensis* Diklić

**1.2 Extinct taxa from Serbia**

- *Aconitum toxicum* Reichenb. subsp. *toxicum*
- *Crocus banaticus* Gaz
- *Iris aphylla* L.
- *Salvia nutans* L.

**2. TAXA THOUGHT TO BE EXTINCT**

- *Achillea ptarmica* L.
- *Aconitum anthora* L.
- *Phlomis pungens* Willd.
- *Stachys serbica* Pančić

**3. CRITICALLY ENDANGERED TAXA**

**3.1. Globally critically endangered taxa**

- *Achillea alexandri-regis* Bornm.& Rudski
- *Artemisia pancicii* (Janka) Ronniger subsp. *pancicii*
- *Crocus rujanensis* Randjelović & D.A.
- *Gentiana pneumonanthe* L. subsp. *nopcsae* T. Wraber
- *Nepeta rtanjensis* Diklić & Milojević

**3.2. Critically endangered taxa**

3.2.1. Taxa with populations containing less than 50 mature individuals

- *Paeonia officinalis* L. subsp. *banatica* Soo
- *Colchicum macedonicum* Košanin
- *Primula auricula L.* subsp. *serratifolia*

3.2.2. Taxa with populations containing between 50 and 250 mature individuals

- *Stachys milanii* Pančić
- *Allium paczoskianum* Tuzson
- *Cypripedium calceolus* L.
- *Scopolia carniolica* Jacq.

3.2.3. Taxa with populations containing over 250 mature individuals

- *Allium cyrilli* Ten.
- *Betula pubescens* Ehrh. subsp. *carpatica* Willd.
- *Glycyrrhiza glandulifera* Waldst. &Kit.
- *Iris humilis* Georgi
- *Sideritis scardica* Griseb.
- *Swertia perennis* L.
- *Achillea ochroleuca* Ehrh.
- *Allium atroviolaceum* Boiss.
- *Artemisia austriaca* Jacq.
- *Helichrysum arenarium* (L) Moench. subsp. *arenarium*
- *Primula halleri* Honckeny
- *Helichrysum plicatum* DC subsp. *plicatum*

### *Ex situ* conservation

It is foreseen that in the near future emphasis will be put on preservation and conservation of various MAP genetic resources. Several state institutions, together with non-governmental organizations and the private sector, have already been included in attempts to maintain MAP diversity, such as the Federal Ministry of Agriculture, Federal Institute for Plant and Animal Genetic Resources which does the collecting, Federal Department for Nature Protection, Institute for Medicinal Plants Research "Josif Pančić" (Belgrade), Institute of Field and Vegetable Crops (Novi Sad) – Department for Medicinal Plants (Bački Petrovac), Faculties of Biology, Faculties of Agriculture (Universities of Belgrade, Novi Sad and Niš), etc.

*Ex situ* conservation of MAPs is a very important activity. It includes field collections, herbaria, seeds and *in vitro* cultures in various institutes and faculties and especially in the Bank of Plant Genes of Yugoslavia. So far, 459 different genotypes (cultivars and populations) of 190 MAPs have been collected (Kisgeči and Sekulović 2000).

Besides the MAP field collection already established at the Institute for Medicinal Plants Research in Pančevo, near Belgrade, some attempts are currently being undertaken in order to establish a new collection in the hilly-mountainous region of Serbia where the majority of the indigenous medicinal flora grows.

Additional activities for *ex situ* conservation of MAP genetic resources are conducted by the Biological Institute in Belgrade in terms of *in vitro* propagation of many MAP species such as *Gentiana* sp., *Centaurim umbellatum*, *Iris* sp., *Digitalis* sp., *Blackstonia perfoliata*, *Frangula alnus*, *Satureja* sp. and many others. It is expected that endangered and vulnerable medicinal plants will be more intensively included in programmes of *in vitro* propagation both by this and other scientific institutions.

During the period 2001-2002, the team of scientists at the Institute for Medicinal Plants Research "Dr Josif Pančić" has exploited a number of types of terrain in Serbia and Montenegro in order to fulfil the needs of the Bank of Plant Genes of F.R.Y. and preserve existing plant germplasm. Additional activities were focused on collecting herbarium specimens and plant samples for further reintroduction, chemical screening and creation of pharmacognostic maps.

### Activities required for MAP protection and conservation

Besides the official legislative, multidisciplinary MAP research and the establishment and development of the MAP collection by the Bank of Plant Genes, other activities should be pursued to improve the current situation on MAP protection and conservation, such as:

- Investigations on possibilities for reintroduction and breeding of the most endangered wild medicinal and aromatic plants;
- Reproduction and multiplication of vulnerable and endangered species directly in natural habitats either by sowing or by vegetative propagation;
- Protection of MAP habitats where the most endangered medicinal plants are present;
- Limited and controlled collecting;
- Substitution of MAP collecting with production on plantations and *in vitro* propagation;
- Education programmes for collectors, breeders and other interested parties;
- Supervision and inspection at all levels.

### Production and processing of MAPs in Serbia

The big demand for medicinal and aromatic species keeps increasing as a consequence of irregular and uncontrolled gathering and exploitation. This imposes the need for cultivation, production, breeding and processing of these plants. For example, before the sanctions, Serbia was one of the biggest exporters of medicinal plants (chamomile, St John's worth, gentian, marshmallow, etc.) with a total average amount of about 1000 tonnes of raw material exported per year, and an income of up to 11 million US$ (Dajić 2001). However, this position was lost and other countries from eastern and central Europe (Bulgaria, Hungary, Slovakia, etc.) appeared on the world market. Medicinal plants from Serbia have been exported mainly into European countries (80.26%) and USA and Canada (19.74%) (Sekulović 1999).

Although the collecting of medicinal plants from the wild is the easiest, cheapest and fastest way to obtain desirable amounts of medicinal plant raw material, this approach is not desirable.

There are no accurate data on the quantities of medicinal plants collected annually. Approximate amounts of MAPs gathered from the nature are given in Table 1.

**Table 1.** Wild medicinal and aromatic plants collected in 1999 (source: Stević and Bjelović 2000)

| Species collected | With permission (A) | Without permission (B) | % (B/A) |
|---|---|---|---|
| *Achillea crithmifolia* W.K. | 420 | 38 | 9 |
| *Achillea millefolium* L. | 6538 | 300 | 4.5 |
| *Agrimonia eupatoria* L. | 7713 | 6573 | 84.4 |
| *Althaea officinalis* L. | 10782 | 1802 | 16.7 |
| *Angelica sylvestris* L. | 204 | 204 | 100 |
| *Arctostaphylos uva-ursi* L. | 8942 | 4242 | 47.4 |
| *Arctium lappa* L. | 1350 | 200 | 14.8 |
| *Artemisia absinthium* L. | 6112 | 4812 | 78.7 |
| *Asarum europaeum* L. | 1383 | 33 | 2.3 |
| *Betula pendula* Roth. | 3400 | 450 | 13.2 |
| *Centaurium erythraea* Rafn. | 1538 | 596 | 38.7 |
| *Crataegus oxyacantha* L. | 6600 | 595 | 9 |
| *Equisetum arvense* L. | 4819 | 1019 | 21 |
| *Frangula alnus* Mill. | 3300 | 200 | 6.6 |
| *Hypericum* sp. | 159570 | 112697 | 70.6 |
| *Juniperus communis* L. | 438180 | 70587 | 16.1 |
| *Origanum vulgare* L. | 6103 | 1083 | 17.7 |
| *Plantago lanceolata* L. | 4492 | 1092 | 24.3 |
| *Polygonum aviculare* L. | 941 | 941 | 100 |
| *Primula vulgaris* Huds. | 2374 | 1649 | 69.4 |
| *Pulmonaria officinalis* L. | 2940 | 1663 | 56.5 |
| *Rosa canina* L. | 152926 | 29926 | 19.5 |
| *Sambucus nigra* L. | 27732 | 16587 | 59.8 |
| *Satureja montana* L. | 1325 | 325 | 24.5 |
| *Satureja kitaibelli* Wiercb. | 8945 | 6545 | 73.1 |
| *Taraxacum officinale* Webb. | 4802 | 852 | 17.7 |
| *Teucrium chamaedrys* L. | - | 270 | 100 |
| *Teucrium montanum* L. | 692 | 392 | 56.6 |
| *Thymus* sp. | 13923 | 3403 | 24.4 |
| *Tilia tomentosa* Moench. | 9093 | 1697 | 18.6 |
| *Tussilago farfara* L. | 432 | 432 | 100 |
| *Urtica dioica* L. | 70799 | 51499 | 72.7 |
| *Vaccinium myrtillus* L. | 365736 | 45402 | 12.4 |

Despite the significant natural resources (especially in meadows and pastures), a great number of medicinal plant populations have disappeared, while many plants are endangered or have limited distribution and therefore should be cultivated for the sake of

nature conservation (*Achillea* sp., *Aconitum* sp., *Acorus calamus* L., *Satureja* sp., *Orchis* sp., *Gentiana lutea* L., *Helychrisum arenarium L., Glycyrrhiza glabra* L., *Gypsophila paniculata* L., *Angelica archangelica* L., *Valeriana officinalis* L., *Ruta graveolens* L., *Prunus laurocerasus* L., *Salvia officinalis* L., *Castanea sativa* Mill., *Centaurium erythraea* Rafn., *Colchicum autumnale* L., *Crocus sativus* L., *Daphne alpina* L., D*aphne blagayana* Freyer, *Drosera rotundifolia* L., *Arctostaphylos uva-ursi* Sprengel, *Hyssopus officinalis* L., *Juglans regia* L., *Adonis vernalis* L., *Veratrum album* L., *Menyanthes trifoliata* L., *Pinus mugo* Turr., *Primula* sp., *Vaccinium vitis idaea* L., *Lycopodium clavatum* L., etc.).

Medicinal plant production, especially of species with moderate or low ecological requirements, is totally justifiable in regions where it is not possible or worthwhile to produce the standard crops (rocky, sandy, saline, poor and waterlogged soils, abandoned mountainous pastures, etc.).

MAP cultivation is more sustainable than medicinal plant gathering, which is strictly regulated by law. There are several advantages to MAP cultivation, such as the production of raw material of standard quality, use of available machinery, processing facilities and means of production, more rational use of soil resources, yield planning, financial effects, preservation of species whose collection has been prohibited, etc.). In the near future, these plants will therefore be the main enterprise of primary nurseries and small- and medium-size enterprises of the agricultural sector.

Box 2 lists MAPs with known or partially known cultivation technology and Box 3 those of unknown cultivation technology.

**Box 2.** Medicinal and aromatic plants with known or partially known cultivation technology and sold on the Serbian market

| **Species with known cultivation technology** | **Species with partially known cultivation technology** |
|---|---|
| *Althaea officinalis* | *Achillea crithmifolia* |
| *Angelica archangelica* | *Agrimonia eupatoria* |
| *Borago officinalis* | *Cnicus benedictus* |
| *Carum carvi* | *Gypsophila paniculata* |
| *Chamomilla recutita* | *Hypericum barbatum* |
| *Coriandrum sativum* | *Hypericum perforatum* |
| *Cynara scolimus* | *Inula helenium* |
| *Foeniculuma vulgare* | *Iris germanica* |
| *Glycyrrhiza glabra* | *Leonurus cardiaca* |
| *Hyssopus officinalis* | *Malva silvestris* |
| *Juglans regia* | *Marrubium vulgare* |
| *Lavandula* sp. | *Origanum vulgare* |
| *Levisticum officinale* | *Ruta graveolens* |
| *Mentha piperita* | *Saponaria officinalis* |
| *Melissa officinalis* | *Satureja kitaibelli* |
| *Majorana officinalis* | *Satureja montana* |
| *Oenothera biennis* | |
| *Pyrethrum cinerariifolium* | |
| *Rosmarinus officinalis* | |
| *Salvia officinalis* | |
| *Silybum marianum* | |
| *Thymus serpyllum* | |
| *Valeriana officinalis* | |
| etc. | |

**Box 3.** Medicinal plants with unknown cultivation technology sold on the Serbian market.

*Achillea* sp.
*Aconitum* sp.
*Acorus calamus*
*Adonis vernalis*
*Angelica silvestris*
*Allium victorialis*
*Arctostaphylos uva-ursi*
*Artemisia absinthium*
*Aruncus dioicus*
*Atropa belladonna*
*Carlina acaulis*
*Castanea sativa*
*Centaurium erythraea* subsp. *erythraea*
*Colchicum autumnale*
*Convallaria majalis*
*Crocus sativus*
*Daphne alpina*
*Daphne blagayana*
*Drosera rotundifolia*
*Digitalis laevigata*
*Digitalis ferruginea*
*Digitalis grandiflora*
*Digitalis lanata*
*Dryopterix filix-mas*
*Fragaria vesca*
*Gentiana asclepiata*
*Gentiana cruciata*
*Gentiana lutea*
*Gentiana punctata*
*Geum urbanum*
*Helichrysum arenarium*
*Hepatica nobilis*
*Herniaria glabra*
*Herniaria hirsuta*
*Hyosciamus niger*
*Juniperus communis*
*Lycopodium clavatum*
*Menyanthes trifoliata*
*Ononis spinosa*
*Orchis morio*
*Pinus mugo*
*Potentila erecta*
*Primula elatior*
*Primula veris*
*Prunus laurocerasus*
*Pulmonaria officinalis*
*Ruscus aculeatus*
*Sambucus nigra*
*Sanicula europaea*
*Solidago virgaurea*
*Symphytum officinale*
*Teucrium montanum*
*Thymus serpyllum*
*Vaccinium myrtillus*
*Vaccinium vitis-idaea*
*Veratrum nigrum*
*Verbascum densiflorum*

The poor tradition of MAP cultivation in our country – less than 5000 ha under such crops (Table 2) – is not an excuse for low investment and insufficient attention to this agricultural sector. Besides production of medicinal plant remedies, seed production and the sale of transplants can also generate a significant income.

**Table 2.** Estimation of areas cultivated under MAPs in Serbia (source: personal communications from specialists from the Institute for Medicinal Plant Research and private sector)

| Species | Area (ha) | Species | Area (ha) |
|---|---|---|---|
| *Althaea officinalis* L. | 80-100 | *Majorana hortensis* Moench. | 20 |
| *Anethum graveolens* L. | 500 | *Malva silvestris* L. | 10-30 |
| *Angelica archangelica* L. | 10-20 | *Melissa officinalis* L. | 100-300 |
| *Artemisia dracunculus* L. | 20 | *Mentha piperita* L. | 800-1200 |
| *Calendula officinalis* L. | 50-70 | *Ocimum basilicum* L. | 50-70 |
| *Carum carvi* L. | 50-150 | *Oenothera biennis* L. | 20-30 |
| *Chamomilla recutita* (L.) Rausch. | 600-1000 | *Origanum vulgare* L. | 30 |
| *Coriandrum sativum* L. | 50-200 | *Petroselinum sativum* L. | 150-300 |
| *Cynara scolimus* L. | 400-500 | *Pimpinella anisum* L. | 220 |
| *Foeniculum vulgare* Mill. | 200-250 | *Salvia officinalis* L. | 60-100 |
| *Glycyrrhiza glabra* L. | 10-20 | *Sinapis alba* L. | 30-50 |
| *Hypericum perforatum* L. | 15-20 | *Thymus vulgaris* L. | 50-100 |
| *Hyssopus officinalis* L. | 80-100 | *Trigonella foenum-graecum* L. | 30 |
| *Iris germanica* L. | 10-20 | *Urtica dioica* L. | 10-20 |
| *Lavandula angustifolia* Ehrh. | 20 | *Valeriana officinalis* L. | 120-150 |
| *Levisticum officinale* Koch. | 50-60 | | |
| | **Total: 3255-5700** | | |

Other problems with MAP cultivation are: slow acclimatization, heterogeneous seed material, pest/disease-susceptible plant populations and low yields.

Regarding medicinal plant processing, it is known that the world's annual production of essential oils is at least 45 000 tonnes (Verlet 1993). However, Serbian producers do not operate on the world market because of variable yields and quality and lack of technical information to overcome these problems (Menković *et al.* 1997). The largest quantities of essential oils are obtained from species both indigenous and cultivated in Serbia, such as mint, juniper tree, lemon balm, thyme, chamomile, dill, parsley and valerian.

Only recently, mostly in the private sector, more attention has been paid to essential oil distillation from plants such as *Levisticum officinale, Angelica archangelica, Ocimum basilicum* and *Echinacea angustifolia* (Dajić and Stevanović 2001). Unfortunately, it is very difficult to estimate the amount of essential oils produced annually in Serbia, but it is believed to range between a few hundred kg and up to 10 000 kg.

In developed countries, agriculture produces a surplus of traditional crops and this is to be restricted. Council regulation EEC 1765/92 and its updates established a support system for producers of certain arable crops. Although cereal areas should be reduced, it is permitted to use set-aside land for "non-food" crops; medicinal plants and spices belong to this category, which is not restricted, but supported (Németh 2000).

## Current research on MAPs and further prospects and goals

In addition to investigations on MAP biology, diversity and population mapping, significant research is conducted on breeding, selection and introduction of wild MAPs (Institute for Medicinal Plant Research, Belgrade; Department for Medicinal Plants, Bački Petrovac; Faculty of Agriculture, University of Belgrade and Novi Sad).

The main achievements in MAP breeding and selection in F.R.Y. are reflected in the number of domestic cultivars (Table 3).

**Table 3.** Domestic or improved MAP cultivars in F.R. Yugoslavia (source: Kišgeci *et al.* 1997; Adamović 2000)

| | Species | No. of cultivars | | Species | No. of cultivars |
|---|---|---|---|---|---|
| 1 | *Achillea millefolium* L. | 1 | 26 | *Majorana hortensis* Moench. | 4 |
| 2 | *Acorus calamus* L. | 1 | 27 | *Malva silvestris* L. | 3 |
| 3 | *Agrimonia eupatoria* L. | 1 | 28 | *Melissa officinalis* L. | 3 |
| 4 | *Althaea officinalis* L. | 1 | 29 | *Mentha* x *piperita* L. | 5 |
| 5 | *Anethum graveolens* L. | 2 | 30 | *Mentha spicata* L. | 1 |
| 6 | *Angelica archangelica* L. | 2 | 31 | *Ocimum basilicum* L. | 4 |
| 7 | *Artemisia absinthium* L. | 1 | 32 | *Oenothera biennis* L. | 1 |
| 8 | *Artemisia dracunculus* L. | 2 | 33 | *Origanum vulgare* L. | 1 |
| 9 | *Borago officinalis* L. | 2 | 34 | *Papaver somniferum* L. | 1 |
| 10 | *Calendula officinalis* L. | 1 | 35 | *Pimpinella anisum* L. | 1 |
| 11 | *Carum carvi* L. | 4 | 36 | *Ricinus communis* L. | 1 |
| 12 | *Chamomila recutita* (L.) Rausch. | 3 | 37 | *Ruta graveolens* L. | 2 |
| 13 | *Chrysanthemum cinerariaefolium* L. | 1 | 38 | *Salvia officinalis* L. | 1 |
| 14 | *Cochlearia armoracia* L. | 1 | 39 | *Salvia sclarea* L. | 2 |
| 15 | *Coriandrum sativum* L. | 5 | 40 | *Satureja hortensis* L. | 2 |
| 16 | *Cynara scolimus* L. | 1 | 41 | *Satureja montana* L. | 1 |
| 17 | *Datura innoxia* Mill. | 1 | 42 | *Sideritis scardica* L. | 1 |
| 18 | *Digitalis lanata* Ehrh. | 1 | 43 | *Silybum marianum* (L.) Gaertn. | 1 |
| 19 | *Foeniculum vulgare* Mill. | 5 | 44 | *Sinapis alba* L. | 1 |
| 20 | *Glycyrrhiza glabra* L. | 1 | 45 | *Thymus vulgaris* L. | 4 |
| 21 | *Gypsophila paniculata* L. | 1 | 46 | *Trigonella foenum-graecum* L. | 1 |
| 22 | *Hyssopus officinalis* L. | 2 | 47 | *Urtica dioica* L. | 1 |
| 23 | *Lavandula angustifolia* Mill. | 1 | 48 | *Valeriana officinalis* L. | 3 |
| 24 | *Leonurus cardiaca* L. | 1 | 49 | *Verbascum densiflorum* Bert. | 1 |
| 25 | *Levisticum officinale* Koch. | 1 | | | |
| **Total no. of cultivars = 89** | | | | | |

Since the establishment of the Institute for Medicinal Plant Research "Dr Josif Pančić" (1948), much has been learned about the pharmacological features of MAPs. Other institutions also deal with evaluation of chemical content, active substances and qualitative composition of MAP drugs, as well as their influence on cells of bacteria, fungi and cancer (Faculty of Pharmacy, Faculty of Technology, Faculty of Chemistry, Faculty of Biology, etc.).

The special challenge for the coming decades, in our opinion, is cultivation and/or improvement of new technologies for production of endangered medicinal plants, because many of them are still not cultivated. Among them, the following species could be stressed: St John's wort, eyebright, yarrow, Lady's mantle, gentian, chicory, uva ursi, digitalis, agrimony, borage, centaury and many others. Production technologies have already been developed for some endangered or vulnerable medicinal plants and therefore it is expected that their production will increase in the future (sage, hyssop, valerian, angelica, lemon balm, marshmallow, etc.). It is important to emphasize that many species controlled by the state may be used for cheap and very efficient soil conservation, i.e. for preventing both erosion and leaching of hilly-mountainous soils (e.g. sage, hyssop, lavender, mugwort).

Breeding and further selection of MAPs in Serbia, together with the proper choice of habitat for their production, is of great importance. There are indications that more than half of all cultivated MAP areas are sown with non-registered seeds, resulting in low yields and uncertain drug quality (Dr Slobodan Drazić, personal communication).

Furthermore, the acquisition of both cultivation technologies (where more efforts should be dedicated to weed and pest control) and modern raw processing methods is necessary. It is also important to define the current capacities for MAP production (habitat properties, soil characteristics, climate, irrigation possibilities, availability of machinery and processing facilities), to supply proper seed or seedling material, to cooperate with state and private enterprises and scientific institutions and to analyze the whole financial framework and possible risks.

Application of molecular genetics technologies to MAP studies could provide further possibilities for evaluation of suitable genotypes and quality evaluation of MAP products. No serious research at the molecular level on MAPs has been conducted in F.R.Y. so far. However, the Department of Botany of the Faculty of Agriculture (University of Belgrade) in cooperation with the Royal Botanical Gardens in Kew (United Kingdom) has undertaken a new and promising project considering diversity and evolution of different species of the genus *Thymus*, mostly endemics of southeast Serbia and the Balkan Peninsula. This research will include determination of collected plant material according to cDNA sequencing. It is expected that results will serve for further cultivation and selection of certain thyme species.

At the beginning of 2002 the Ministry of Science and Technology of Serbia approved and started financing several national scientific projects that encompass all aspects of MAPs, both fundamental and applied.

There is indeed hope for future opportunities for more intensive collaboration with neighbouring countries and all other EU countries through common participation in international projects involving multidisciplinary research on MAPs, including the development of a common database.

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# Documentation and databases

## *The Central Database of the Slovene Plant Gene Bank*

***Vladimir Meglič***
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### Introduction

In 1996 the Ministry of Agriculture, Forestry and Food started financing the **Slovene Plant Gene Bank Programme** with the goal to maintain, evaluate, regenerate and preserve Slovenian autochthonous species, ecotypes, populations and landraces of agricultural, medicinal and aromatic plants, forest trees and other woody plants from Slovenian forests. They include Slovenian cultivars, old cultivars, landraces, various populations, clones and lines bred from autochthonous plants and ecotypes from the natural habitat important for food, agriculture and forestry (Černe *et al.* 1998).

In the Slovene Plant Gene Bank Programme working with agricultural, medicinal and aromatic plants (SPGB-AMAP) three institutions are involved:

- the Agronomy Department at the Biotechnical Faculty of the University of Ljubljana is responsible for the *Fagopyrum*, *Zea mays*, fruit trees (*Malus, Pyrus, Juglans, Prunus*), forage crops and medicinal and aromatic plant (MAP) collections;
- the Institute for Hops and Brewery in Žalec is responsible for *Humulus* and medicinal and aromatic plant collections;
- the Agricultural Institute of Slovenia houses collections of grain legumes, *Allium, Solanum tuberosum, Triticum, Brassica, Lactuca* accessions, forage crops, *Rubus* and *Vitis*.

The Programme for conservation of forest genetic resources (Slovenian Forest Gene Bank) is led by the Slovenian Forestry Institute, Ljubljana.

### Multiplication, storage and evaluation

Each year limited numbers of accessions are planted in the field for seed multiplication, characterization and evaluation. During the growth period morphological characteristics and disease susceptibility are recorded (Černe 1999).

When mature, seeds are collected, cleaned and dried. Between 500 and 1000 seeds per accession are stored at +4°C in working collections. In addition samples are prepared for the base collection (long-term storage at –20°C) located at the Agricultural Institute of Slovenia. Genetic resources are also kept in *in vitro* conditions and *in vivo* in permanent plantations for hops, fruit, grapevine and some MAP species.

### Collecting missions

Seed samples and passport data were obtained with the help of local elementary and agricultural schools, the Agricultural Advisory Service, newspaper ads and farmers. Most of the people who sent us samples filled out a questionnaire which provided necessary data and some additional information on local names and growing practices. Collecting efforts are being continued through collecting missions throughout Slovenia. An important part of the collecting missions are joint missions with the Czech Republic and Croatia.

**Documentation**

The Agronomy Department of the Biotechnical Faculty, University of Ljubljana, the Agricultural Institute of Slovenia, the Institute of Hop Research and Brewing and the Slovenian Forestry Institute form the Slovenian Plant Gene Bank (SPGB) and work with species used in agriculture, forestry and for food. These institutions are responsible for *ex situ* germplasm collections stored in the form of seeds, *in vitro* and *in vivo* collections. Each institution holds a database for its working collection. With a need for a uniform and centralized documentation and information system, a computer program was used (Žitnik *et al.* 2000) to unite the four separate databases. This will enable easier and faster access to the complete information for all users, better management of germplasm resources in the Central Plant Gene Bank and exchange of information with other ECP/GR and EUFORGEN genebank databases.

Each local database of the Slovenian Plant Gene Bank holds only data of the particular institution. These local databases are incorporated into the central database.

The contents of the Central Plant Gene Bank (CPGB) are divided into 5 categories:

- multicrop passport descriptors,
- additional passport descriptors,
- characterization data,
- evaluation data, and
- central seed bank holdings.

The multicrop passport descriptors are the same for all plant species. The other four categories are specific for one species or group of species. Therefore, we have one database for the multicrop passport descriptors and several databases for the four other categories.

The central database is constructed using the computer program MS Access 2000. It consists of one Access database containing five separate databases corresponding to the five categories above. The central database has been built using three different ways of entering data:

- data existing in other computer formats (Dbase, Excel, etc.) are transformed directly into the central working database by the database manager;
- new data are entered into the local Access databases at each institute and a copy of the local database is sent by email to the database manager;
- at present the preferred way of entering data is via a computer program written specifically for this purpose, which enables members of the Slovenian Plant Gene Bank to enter data automatically into the central database via the Internet.

The protocol for data entry has been written. Data are entered using a password assigned to each institution involved and a unique SRGB[20] accession number is assigned to the data set automatically.

Other users will be able to browse through the read-only, publicly available, version of the central database (http://www.kis.si/srgb/) (Žitnik *et al.* 2000).

At present multicrop passport descriptor information is available for 3099 accessions in the Central Plant Gene Bank Database (agricultural and forest plants combined). We are planning to add to the CPGB database a specialized database on medicinal and aromatic plants. This database, called MEDPLANT, is being developed at the Biotechnical Faculty at the University of Ljubljana, but unfortunately it is not a public database and its use will have to remain restricted.

[20] SRGB = Slovenska Rastlinska Genska Banka (Slovenian Plant Gene Bank)

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# Research

## *Molecular tools for determining genetic variability* [21]

***Branka Javornik***
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This presentation deals with modern approaches in determining genetic variability, in which three categories of genetic markers are applied – morphological, biochemical and molecular. Morphological markers (such as plant height, leaf shape, colour, etc.) are among the oldest markers used in the evaluation of genetic variability. However, they are not sufficiently specific and informative because different gene expression in different environments causes wide variability of phenotypic characters in individuals. Similarly, biochemical markers (such as albumin content, isoenzymes or essential oil contents in an individual organism) can be considered to be non-specific due to the wide variability of biochemical characters, which are strongly influenced by an individual's environment.

The development of recombinant DNA in the 1980s enabled the development and use of molecular markers, thus providing a modern tool for determining genetic variability.

Molecular markers show variability among individuals on the DNA level, which is not influenced by the environment.

Different genetic markers (e.g. RFLP, AFLP, RAPD, SSR, SCAR) have different properties, (dominant and co-dominant markers, different coverage of the genome) and different advantages and disadvantages (e.g. specificity, cost, ease of analytical interpretation of the resulting data). However, they are highly informative about genetic variability among individuals, populations and cultivars. Their use is universal for all organisms. Molecular markers can be considered to be essential tools in cultivar identification (DNA typing), assessment of genetic variability and relationships, management of genetic resources and biodiversity, studies of phylogenetic relationships and in genome mapping.

The main aim of the management of genetic resources is to ensure that as much as possible of the existing genetic diversity of species is conserved. The effectiveness of this depends to a large extent on the genetic information available on the germplasm under study.

Molecular markers provide genetic information of direct value for *ex situ* and *in situ* conservation. For *ex situ* conservation, the acquisition of data on the diversity of collections is important. Molecular markers can be used (i) to identify valuable genetic variation that is under-represented in a collection sample; (ii) to identify duplicate accessions and to monitor changes in genetic structure as accessions are generated; (iii) to assess the available genetic diversity for each species; (iv) to provide more accurate and detailed information than is available using classical phenotypic data; (v) to identify traits and types quickly.

On a more fundamental level, molecular marker information may lead to the further identification of useful genes contained in a collection. Molecular data on diversity may provide essential information to develop core collections that accurately represent the entire collection. For *ex situ* conservation, molecular markers may be used in: the determination of identity and similarity of accessions or individuals; the measurement of the structure of diversity among individuals, accessions, populations and species; and the detection of particular allele or nucleotide sequences in a taxon, genebank accession or *in situ* population.

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[21] Summary of the presentation made at the meeting.

# *Genetic variability of native populations of oregano in Slovenia*

***Jelka Šuštar-Vozlič***
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## Introduction

Many species belonging to the *Origanum* genus, rich in essential oils, phenolic compounds and flavonoids, have been used for thousands of years as spices and in traditional medicine. Nowadays, a number of other properties are reported as well (e.g. antifungal, antibacterial, antioxidant, antiviral, nematicidal). The taxonomy of the genus *Origanum* is rather complicated and is still a current issue of debate. The genus is characterized by large differences in morphological and chemical properties resulting in the distinction of 49 taxa and 43 species as well as 18 hybrids (Kintzios 2002). The most comprehensive revision of the genus was done by Ietswaart (1980), who divided the taxa in ten sections, the monospecific section *Origanum* being the most widely distributed.

In Slovenia, *O. vulgare* subsp. *vulgare* occurs wild in nature and autochthonous populations differ largely in their morphological as well as biochemical characteristics (Baričevič 1997). They are characterized by a low content of essential oils (up to 0.5%); nevertheless they show a considerable antioxidant activity. Large variability in the morphological as well as in the biochemical properties could be a reflection of either environmental differences, a product of microclimatic variations found in Slovenia, or of genetic differences between the populations and clones, or a combination of both factors.

Classical methods of estimating genetic diversity among groups of plants have relied upon morphological or chemical characters, but these characters can be influenced by environmental factors. By looking directly at the genetic material itself, molecular markers represent a powerful and potentially rapid method for the characterization of diversity *per se* within the *in situ* and *ex situ* conservation (Ford-Lloyd 2001). However, molecular studies dealing with medicinal and aromatic plants (MAPs) are rare in comparison with other cultivated plants. This is probably due to the presence of large amounts of secondary metabolites and essential oils in MAP tissues, which inhibit DNA amplification in PCR reaction (Khanuja *et al.* 1999; Mizukami and Okabe 1999). There have been only two molecular studies on the genus *Origanum* reported until now. A molecular systematic study of the family Lamiaceae using *rbcL* gene sequences was made by Kaufamnn and Wink (1994) with two *Origanum* species (*O. vulgare* and *O. laevigatum*) and *Majorana hortensis,* among others. Some authors have treated *M. hortensis* as a member of the *Origanum* genus, but the results of this study show that it differs significantly from the two *Origanum* species and was therefore suggested to be a separate species. A study using RAPD markers for the comparison of essential oils and genetic relationship of *Origanum* x *intercedens* with its parental taxa in the island of Crete was performed (Gounaris *et al.* 2002).

The aim of the present study was to develop a protocol for DNA isolation and to optimize the PCR reactions for further evaluation of genetic diversity of wild oregano in Slovenia using RAPD markers.

## Materials and methods

Nine populations of *O. vulgare* subsp. *vulgare* from Slovenia conserved in the Slovene national genebank for medicinal and aromatic plants were included in the study. One population of *O. vulgare* subsp. *hirtum* and one population of *O. vulgare* subsp. *vulgare* obtained from the genebank in Georgia were selected as outgroups. The number of plants tested per population varied from two to five. The list of accessions included in the study is presented in Table 1.

**Table 1.** Accessions of *O. vulgare* L. included in the study

| Population | No. of plants | Subtaxa | Origin |
|---|---|---|---|
| 9 | 2 | *O. vulgare* L. subsp. *vulgare* | ORI 15/91, Georgia |
| 9/1 | 2 | *O. vulgare* L. subsp. *hirtum* (Link) Ietswaart | Greece |
| 9/3 | 4 | *O. vulgare* L. subsp. *vulgare* | Jesenice |
| 9/4 | 5 | *O. vulgare* L. subsp. *vulgare* | Žirovniška planina |
| 9/6 | 4 | *O. vulgare* L. subsp. *vulgare* | Stari vrh nad Škofjo Loko |
| 9/7 | 4 | *O. vulgare* L. subsp. *vulgare* | Završnica |
| 9/8 | 5 | *O. vulgare* L. subsp. *vulgare* | Mežakla |
| 9/9 | 5 | *O. vulgare* L. subsp. *vulgare* | Nanos, Lijak |
| 9/11 | 3 | *O. vulgare* L. subsp. *vulgare* | Slovenske Konjice |
| 9/12 | 5 | *O. vulgare* L. subsp. *vulgare* | Blato |
| 9/14 | 5 | *O. vulgare* L. subsp. *vulgare* | Štampetov most, Vrhnika |

For the isolation of DNA, different types of plant material (fresh, frozen, young, old, lyophilized leaves, leaves dried at 40°C, plants grown *in vitro*) were taken and different protocols were tested: two modified CTAB protocols (Šuštar-Vozlič and Javornik 1999; Z. Liber, personal communication 2000), SDS protocol (Beye and Raeder 1993) and a commercial kit (Dneasy Plant Mini Kit, Qiagen).

PCR amplifications were carried out in 25 µl mixture with DNA content and $Mg^{2+}$ concentrations being optimized. Different types of *Taq* DNA polymerases were also tested. Twenty-five arbitrary 10-mer primers (Operon Technologies) were applied.

A preliminary study of genetic variability using RAPD molecular markers was made. For data analysis and the construction of a dendrogram, NTSYS-pc software was used (Rohlf 1993). UPGMA cluster was performed using the RAPD data.

## Results and discussion

Various types of plant material and a number of different protocols for the isolation of DNA were tested in order to obtain good quality DNA for PCR reactions. When fresh or frozen leaves of *O. vulgare* subsp. *vulgare* collected in autumn were used for the isolation of DNA, no positive result in PCR reaction was obtained regardless of the isolation protocol being used. This was probably due to the accumulation of large amounts of secondary metabolites in old plant material, as previously reported (Khanuja *et al.* 1999). DNA purified with DNeasy Plant Mini Kit (Qiagen) from plants grown *in vitro* gave the best results in PCR reactions. A positive PCR reaction was also obtained when DNA was isolated from leaves taken from the greenhouse or dried at 40°C. It was found out that the age and quality of plant material were of major importance for a successful PCR amplification. The results of PCR reactions with different types of plant material used and different isolation protocols applied are presented in Table 2.

Because of a limited number of samples from individual populations and a small number of polymorphic RAPD profiles generated after electrophoresis of PCR products, the results of statistical analysis showed that samples belonging to the same populations, as well as outgroups on the dendrogram, did not usually cluster together.

In the present study a protocol for DNA isolation was developed and PCR reactions were optimized. In further investigations, genetic analysis will be performed on a larger number of samples from each population of *O. vulgare* subsp. *vulgare*, and possible correlations between genetic variability, morphological and biochemical properties will be assessed.

**Table 2.** Plant material, isolation protocols and resulting PCR reaction: (+) = successful amplification, (o) = partly successful amplification, (-) = unsuccessful amplification

| Plant material | | Isolation protocol | PCR reaction |
|---|---|---|---|
| Fresh (frozen) leaves, collected in autumn | | Protocol Liber with modifications, CTAB method | (-) |
| Fresh leaves, collected in May | | Dneasy Plant Mini Kit (Qiagen) | (-) |
| Leaves from greenhouse | | Protocol Liber | (-) |
| | | SDS buffer | (-) |
| | | Qiagen Kit | (+) |
| *In vitro* grown plants | - fresh | Qiagen Kit, SDS buffer | (+) |
| | - frozen | Qiagen Kit | (-) |
| | - lyophilized | | (o) |
| | - dried at 40°C | | (+) |
| | - treated with hexan and methanol | | (o) |

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# APPENDICES

## Appendix I. List of priority species/genera

The Vice-Chair compiled the proposals from the Working Group members and made a priority list of 10 model species/genera, taking into consideration the following criteria:

- The species must be important from the point of view of both biological and chemical diversity;
- The species must be important from the point of view of medicinal plant production (either from cultivation and/or collection);
- There must be some common interest in the genebank conservation of the chosen species for different European regions (at least 1/3 of countries for each);
- The conservation and characterization/evaluation results achieved could be used as a model for the conservation of other MAP species.

### Final 10 species/genera proposed for modelling

| Species/Genus | Number of proposals | Countries interested in the study |
|---|---|---|
| *Achillea* spp. | 10 | Bulgaria, Estonia, Finland, Hungary, Israel, Italy, Lithuania, Romania, Serbia and Montenegro, Slovenia |
| *Artemisia* spp. | 8 | Bulgaria, Estonia, Hungary, Israel, Italy, Nordic Countries, Romania, Serbia and Montenegro |
| *Carum carvi* | 8 | Estonia, Finland, Hungary, Lithuania, Nordic Countries, Romania, Serbia and Montenegro, Slovenia |
| *Gentiana* spp. | 6 | Italy, Lithuania, Macedonia, Poland, Serbia and Montenegro, Slovenia |
| *Hypericum* spp. | 9 | Bulgaria, Estonia, Hungary, Italy, Lithuania, Macedonia, Nordic Countries, Romania, Serbia and Montenegro |
| *Melissa officinalis* | 8 | Bulgaria, Cyprus, Hungary, Israel, Nordic Countries, Romania, Serbia and Montenegro, Turkey |
| *Mentha* spp. | 9 | Bulgaria, Cyprus, Finland, Hungary, Israel, Nordic Countries, Romania, Serbia and Montenegro, Turkey |
| *Origanum* spp. | 14 | Bulgaria, Cyprus, Estonia, Finland, Hungary, Israel, Italy, Lithuania, Macedonia, Nordic Countries, Romania, Serbia and Montenegro, Slovenia, Turkey |
| *Salvia* spp. | 11 | Bulgaria, Cyprus, Hungary, Israel, Italy, Lithuania, Macedonia, Romania, Serbia and Montenegro, Slovenia, Turkey |
| *Thymus* spp. | 12 | Bulgaria, Cyprus, Estonia, Hungary, Israel, Italy, Lithuania, Macedonia, Nordic Countries, Romania, Serbia and Montenegro, Slovenia |

## Appendix II. Abbreviations and acronyms

| | |
|---|---|
| AARI | Aegean Agricultural Research Institute, Menemen, İzmir, Turkey |
| AFLP | amplified fragment length polymorphism |
| ARI | Agricultural Research Institute, Nicosia, Cyprus |
| ARO | Agricultural Research Organization, Bet-Dagan, Israel |
| BPGV | Banco Português de Germoplasma Vegetal, Portugal |
| BSBCP | Bulgarian/Swiss Biodiversity Conservation Programme |
| CBD | Convention on Biological Diversity |
| CCEWNH | Convention on the Conservation of European Wildlife and Natural Habitats (Bern Convention) |
| CGIAR | Consultative Group on International Agricultural Research |
| CHM | Clearing House Mechanism |
| CITES | Convention on International Trade in Endangered Species of Wild Fauna and Flora (Washington Convention) |
| CMS | Convention on the Conservation of Migratory Species of Wild Animals (Bonn Convention) |
| CPGB | Central Plant Gene Bank of Slovenia |
| CWANA | Central and West and North Africa region |
| DEFRA | Department of Food and Rural Affairs, United Kingdom |
| DRAAL | Direcção Regional de Agricultura do Algarve, Portugal |
| DRAEDM | Direcção Regional de Agricultura de Entre Douro e Minho, Portugal |
| ECCDB | European Central Crop Database |
| ECP/GR | European Cooperative Programme for Crop Genetic Resources Networks |
| EPGRIS | Establishment of a Plant Genetic Resources Infra-Structure |
| ESAB | Escola Superior Agrária de Bragança, Portugal |
| ESAC | Escola Superior Agrária de Coimbra, Portugal |
| ESAPL | Escola Superior Agrária de Ponte de Lima, Portugal |
| EU | European Union |
| EUFORGEN | European Forest Genetic Resources Network |
| EURISCO | European Internet Search Catalogue |
| FAO | Food and Agriculture Organization of the United Nations |
| GC | gas chromatography |
| GC-MS | gas chromatography-mass spectroscopy |
| GPA | Global Plan of Action for the Conservation and Sustainable Utilization of Plant Genetic Resources for Food and Agriculture |
| HPLC | High Performance Liquid Chromatography |
| ICMAP | International Council of Medicinal and Aromatic Plants |
| IFAD | International Fund for Agricultural Development, Rome, Italy |
| IGB | Israeli Gene Bank, Bet-Dagan, Israel |
| INIA | Instituto Nacional de Investigação Agrária, Portugal |
| INIA | Instituto Nacional de Investigaciones Agrarias, Spain |
| IPGR | Institute of Plant Genetic Resources, Sadovo, Bulgaria |
| ISAFA | Istituto Sperimentale per l'Assestamento Forestale e per l'Alpicoltura (Forest and Range Management Research Institute), Villazzano, Italy |
| ISF | International Seed Federation, Nyon, Switzerland |
| IUCN | International Union for the Conservation of Nature |
| MAFF | Ministry of Agriculture, Forestry and Food of Slovenia |
| MAO | Ministry of Agriculture of Israel |

| | |
|---|---|
| MAPs | medicinal and aromatic plants |
| MOS | Ministry of Science of Israel |
| NGB | Nordic Gene Bank, Alnarp, Sweden |
| NGO | non-governmental organization |
| NHC | National Herb Centre, Banbury, United Kingdom |
| PCA | principal component analysis |
| PCR | polymerase chain reaction |
| RAPD | random Amplified Polymorphic DNA |
| RFLP | restriction fragment length polymorphism |
| RICP | Research Institute of Crop Production, Prague–Ruzyne, Czech Republic |
| SCAR | sequence characterized amplified region |
| SGGW | Warsaw Agricultural University, Poland |
| SPGB | Slovenian Plant Gene Bank |
| SSR | simple sequence repeats |
| UC | Universidade de Coimbra, Portugal |
| UNCED | United Nations Conference on Environment and Development |
| UNEP | United Nations Environment Programme |
| UNESCO | United Nations Educational, Scientific and Cultural Organization, Paris, France |
| WHO | World Health Organization, Geneva, Switzerland |
| WRI | World Resources Institute, Washington DC, USA |
| WWF | World Wide Fund for Nature |

# Appendix III. Agenda

***First meeting of the ECP/GR Working Group on Medicinal and Aromatic Plants***
***12-14 September 2002, Gozd Martuljek, Slovenia***

**Wednesday 11 September**
Arrival of participants
Welcome reception

**Thursday 12 September**

**Conservation of medicinal and aromatic plants (MAPs) – needs and strategy**

| | |
|---|---|
| 09:00 – 09:15 | Introduction |
| | **Opening of the meeting / Welcome addresses** *(M. Černe, National Coordinator for ECP/GR, D. Baričevič, University of Ljubljana, and J. Glavač, Ministry for Food, Agriculture and Forestry of Slovenia)* |
| 09.30 – 09:50 | **The role of ECP/GR and IPGRI in MAP conservation** *(E. Lipman, IPGRI)* |
| 09.50 – 10.10 | **MAP conservation strategies** (*P. Skoberne, Ministry of the Environment of Slovenia*) |
| 10:10 – 10:30 | **Sustainable use of MAPs and the European Plant Conservation Strategy** (*S. Honnef and S. Schmitt, WWF/TRAFFIC*) |
| *10:30 – 11.00* | *Coffee Break* |
| 11.00 – 11.30 | **Origin of medicinal plants in Central Europe –an ecological approach** (*E. Schneider, PhytoConsulting)* |
| 11.30 –12.30 | **Country reports / short presentations** *(Reports from Austria, Bulgaria, Croatia, Cyprus, Estonia, Germany, Hungary, Israel, Italy, Lithuania, Macedonia F.Y.R., Nordic countries, Poland, Romania, Slovenia, Turkey, United Kingdom, F.R. Yugoslavia)* |
| *12:30 – 14:00* | *Lunch* |
| 14.00 – 15.30 | **Country reports –** continued |
| *15:30 – 16:00* | *Coffee break* |
| 16.00 – 18.00 | **Country reports –** continued |

**Friday 13 September**

**MAP conservation and evaluation databases**

| | |
|---|---|
| 09:00 – 09:30 | **The European Internet Search Catalogue, EURISCO and the EPGRIS project** *(E. Lipman, IPGRI)* |
| 09:30 – 09:45 | **The Slovenian Gene Bank database and the National Inventory** *(V. Meglič, Agricultural Institute of Slovenia)* |
| 09:45 – 10:15 | **Presentation of the MEDPLANT database** *(D. Baričevič, Slovenia)* |

| | |
|---|---|
| *10:15 – 10:45* | *Coffee break* |
| 10:45 – 12:30 | ***Discussion and workplan*** |
| *12:30 – 14:00* | *Lunch* |

**Methodological approaches in MAP conservation and evaluation**

| | |
|---|---|
| 14:00 – 14:30 | **Molecular tools for determining genetic variability of natural populations – the example of *Origanum***<br>*(B. Javornik, Biotechnical Faculty of University of Ljubljana, Slovenia)* |
| 14:30 – 14:50 | **Genetic variability of native populations of oregano in Slovenia**<br>*(J. Šuštar-Vozlič, Agricultural Institute of Slovenia)* |
| 14:50 – 15:30 | ***Discussion*** |
| *15:30 – 16:00* | *Coffee break* |
| 16:30 – 18:00 | ***Discussion and workplan*** |

**Saturday 14 September**

| | |
|---|---|
| 8:30 – 15:00 | **Excursion** (visit of the Central Slovene Genebank for Plant Genetic Resources, Agricultural Institute, Ljubljana) |
| 9:00 – 15:00 | ***Drafting of the report*** *(only delegates involved in the drafting)* |
| *15:00 – 16:00* | *Lunch* |
| 16:00 – 18:00 | **Presentation of the draft report and adoption of recommendations** |
| | **Election of the Chair and Vice-Chair** |
| | **Closing remarks** |
| *20:00* | *Social dinner* |

**Sunday 15 September**

Departure of participants

# Appendix IV. List of participants

## *First meeting of the ECP/GR Working Group on Medicinal and Aromatic Plants 12-14 September 2002, Gozd Martuljek, Slovenia*

### Chairperson

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