

**SEE-ERA.NET Plus Joint Call**

# **SEELEGUMES**

Sustainable preservation of indigenous South East European legumes and their traditional food and feed Products

**Agricultural Institute of Slovenia**

Report, October 2011



**Agricultural Institute of Slovenia**  
Crop and Seed Science Department  
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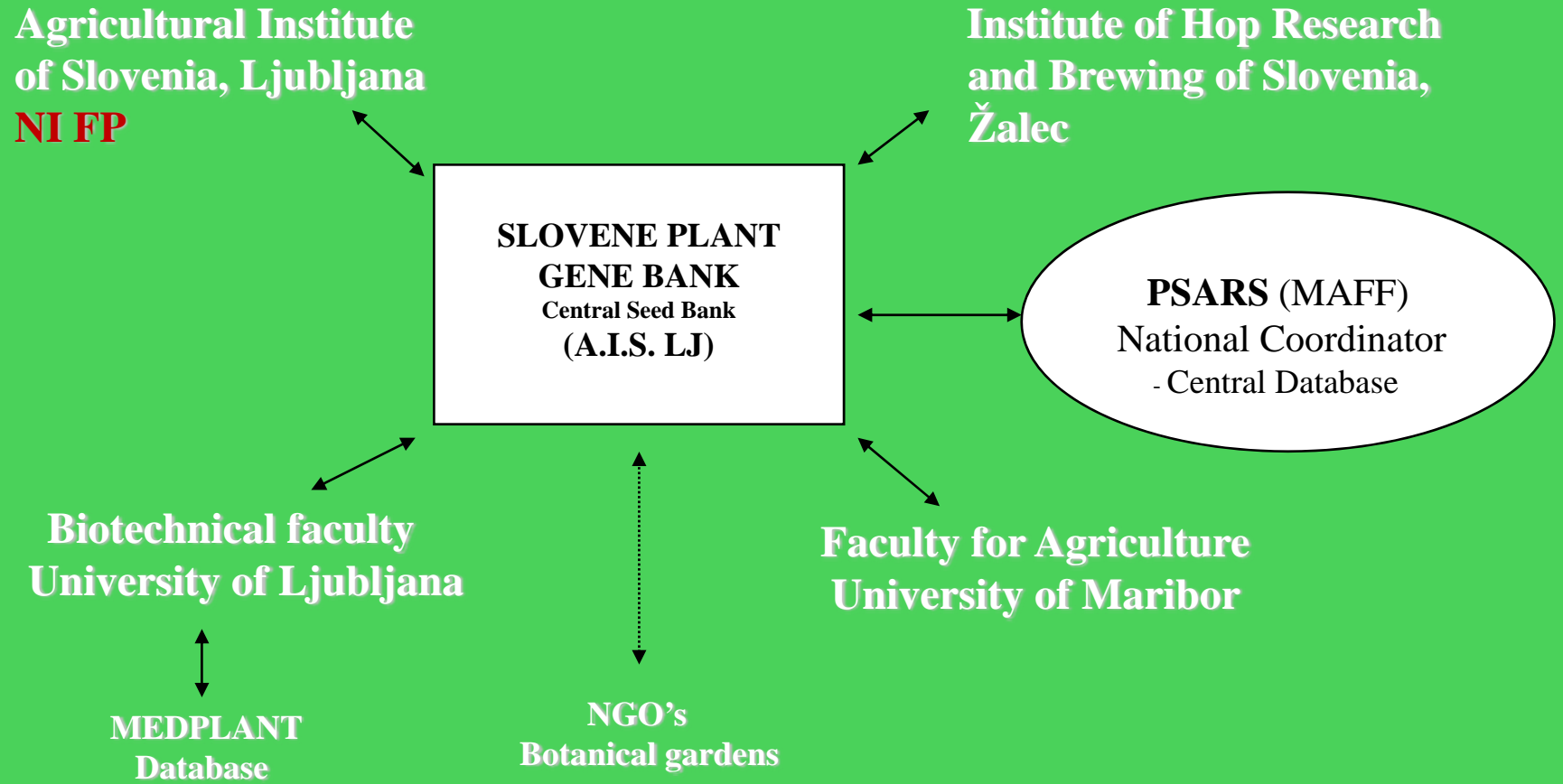
## Role of partner no. 6, KIS

The Crop and Seed Science Department covers research in the field of plant breeding and genetics, physiology and technology of field, fodder and vegetable crop production. Emphasis is given to the environment and to humans and animal friendly production, which includes also the Slovene Plant Gene Bank with the mission to collect, evaluate, map, conserve and propagate Slovene autochthonous varieties, old varieties and wild relatives of species important for agriculture.

Botanical and agronomical data on genetic resources from the SRGB are for certain legume species abundant (for instance for Phaseolus beans), on the other hand are fairly scarce for some other species.

The goal of our part of work lies in morphological and genetic analysis of selected in Slovenia underutilized and neglected grain legume species: faba bean (*Vicia faba*), vetches (*Vicia* spp.), vetchlings (*Lathyrus* spp.), lentil (*Lens culinaris*), chickpea (*Cicer arietinum*) and lupins (*Lupinus* spp.).

# Organisational chart



**Agricultural Institute of Slovenia, Ljubljana**

Grain legumes, Allium, Solanum, Triticum, Brassica, Lactuca, forage and fodder crops, Rubus, Vitis, fruit trees

**Biotechnical faculty, Agronomy Dept., University of Ljubljana**

Fagopyrum, Zea mays, fruit trees, forage crops, medicinal and aromatic plants.

**Faculty for Agriculture, University of Maribor**

Prunus, Vitis, Rubus

**Institute for Hop Research and Brewery, Žalec**

Humulus, medicinal and aromatic plants

Institution	Species	Number of accessions
KIS	all	3203
UNI LJ BF	all	1635
UNI MB FKBV	all	346
IHPS	all	143
	<b>SUM</b>	<b>5327</b>

## WP1

Within the WP1 we will organize expeditions to selected regions and check the references in existing national inventory, provide updated information on selected legume species in both wild and agricultural flora. The selected regions of Slovenia and of the territory of SEE may still be rich in plant genetic resources of grain legumes. The effective communications with local farmers requires the representation of plant genetic resources experts. That would help in the more precise documentation of the information on landraces, on their use, and other special characteristics. The joint collecting missions will contribute to the further development of collaboration between the national programmes.

## WP2

Within the WP 2 we will perform morphological and molecular evaluation of the collected species. Field and pot trials will be a basis for morphological characterization and evaluation of genetic resources. As we have already characterized most of the Phaseolus beans accessions we will broaden the evaluation with analysis of protein, mineral and other nutritional compound contents. The selected material of collected grain legume species will be evaluated as well using Microsatellite markers, also called Simple Sequence Repeats (SSR).

All the work described above within WP1 and WP2 will be performed at the Agricultural Institute of Slovenia. The collaboration with other partners is foreseen mainly through the joint collecting missions, characterization and evaluation and data sharing and exchange.

## Results and milestones

Results of the project will consist of:

- Detailed description of all the existing and newly acquired accessions
- Estimation of genetic diversity of grain legume species using morphological, biochemical and molecular markers
- Protocols for appropriate conservation and preservation of grain legume genetic resources
- Preservation and reintroduction of selected accessions.

## Milestone 1: (Month 12)

- Evaluation of existing databases and national inventories
- Preparation for and collecting genetic resources through collecting missions
- Evaluation and recording of material collected
- Preparation of seed material for planting and consecutive trial establishment
- Botanical and morphological characterisation and evaluation
- Genetic analysis of selected accessions using SSRs
- Presentation of results at the meetings and workshops



## Milestone 2: Month 18

- Genetic analysis of selected accessions using SSRs
- Biochemical analysis of selected accessions
- Botanical and morphological characterisation and evaluation
- Estimation of genetic diversity of grain legume species using morphological, biochemical and molecular markers
- Inscription of passport data of newly acquired accessions into existing database

## Milestone 3: Month 24

- Final data evaluation and diversity analysis
- Presentation of results and preparation of the final report
- Presentation of results at the meetings and workshops
- Preparation of publications concerning dissemination of results and various protocols

## Report WP1

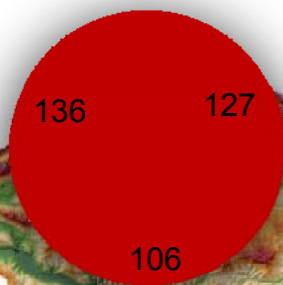
We have made an evaluation of existing databases and national inventories and made wider selection of accessions of different species that are going to be characterised and evaluated during the project.

We have made an inventory of previously visited sites and selected regions for further collecting missions.

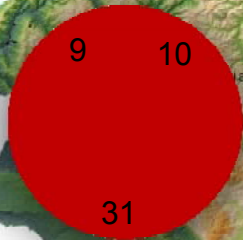
We will organize expeditions to collect genetic legume resources to selected regions in the fall of the 2011. Basic evaluation and recording of material collected will be made after collecting missions are finished.

Along with that we will check the references in existing national inventory and provide updated information on collected legume species.

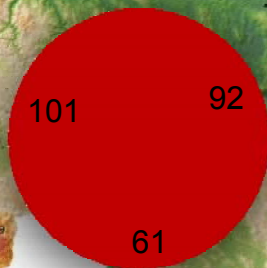
# SVNJULALP 1999



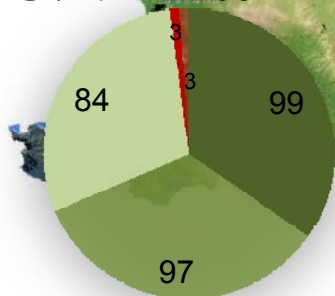
# SVNTG 2008



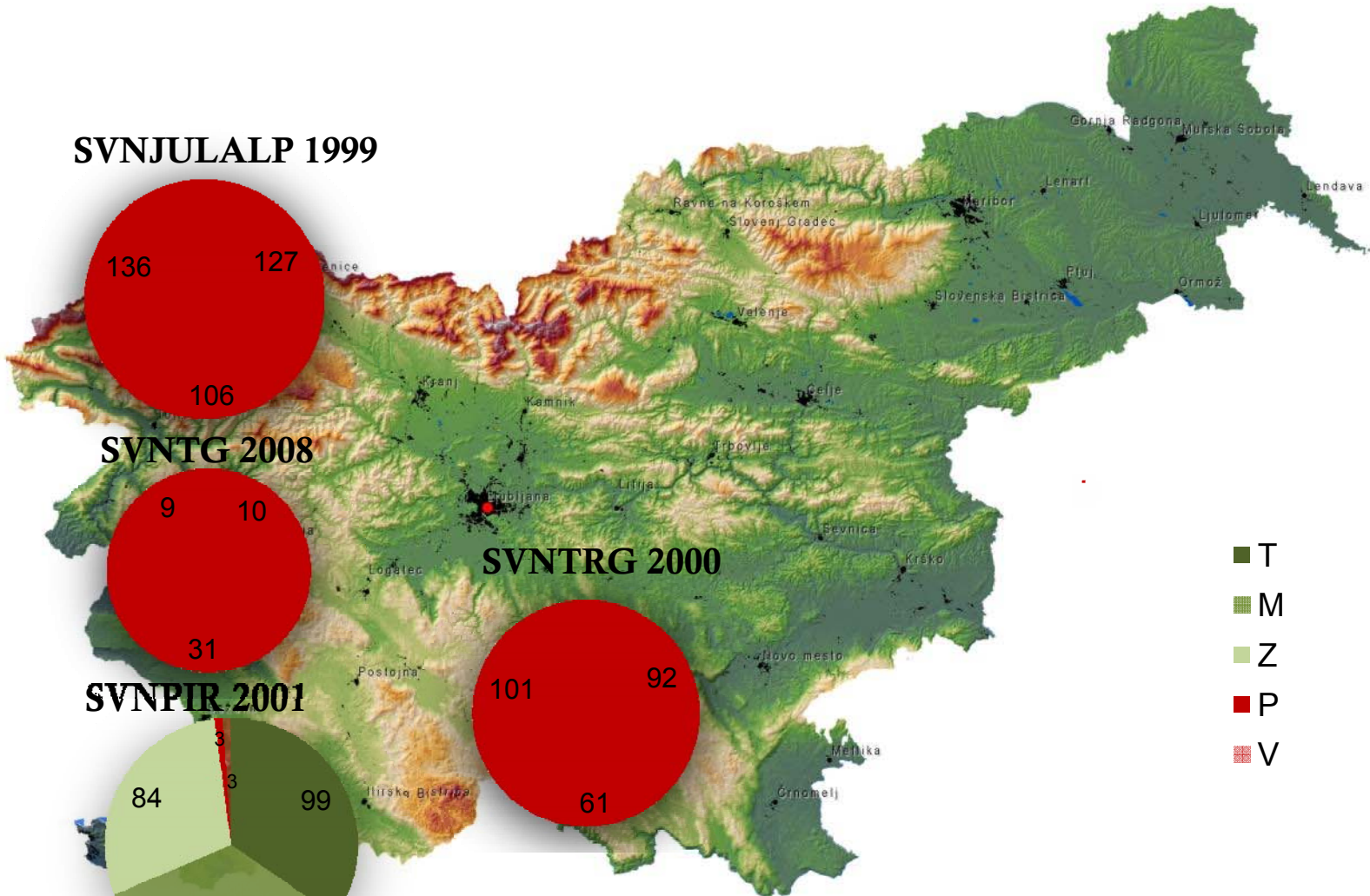
# SVNTRG 2000

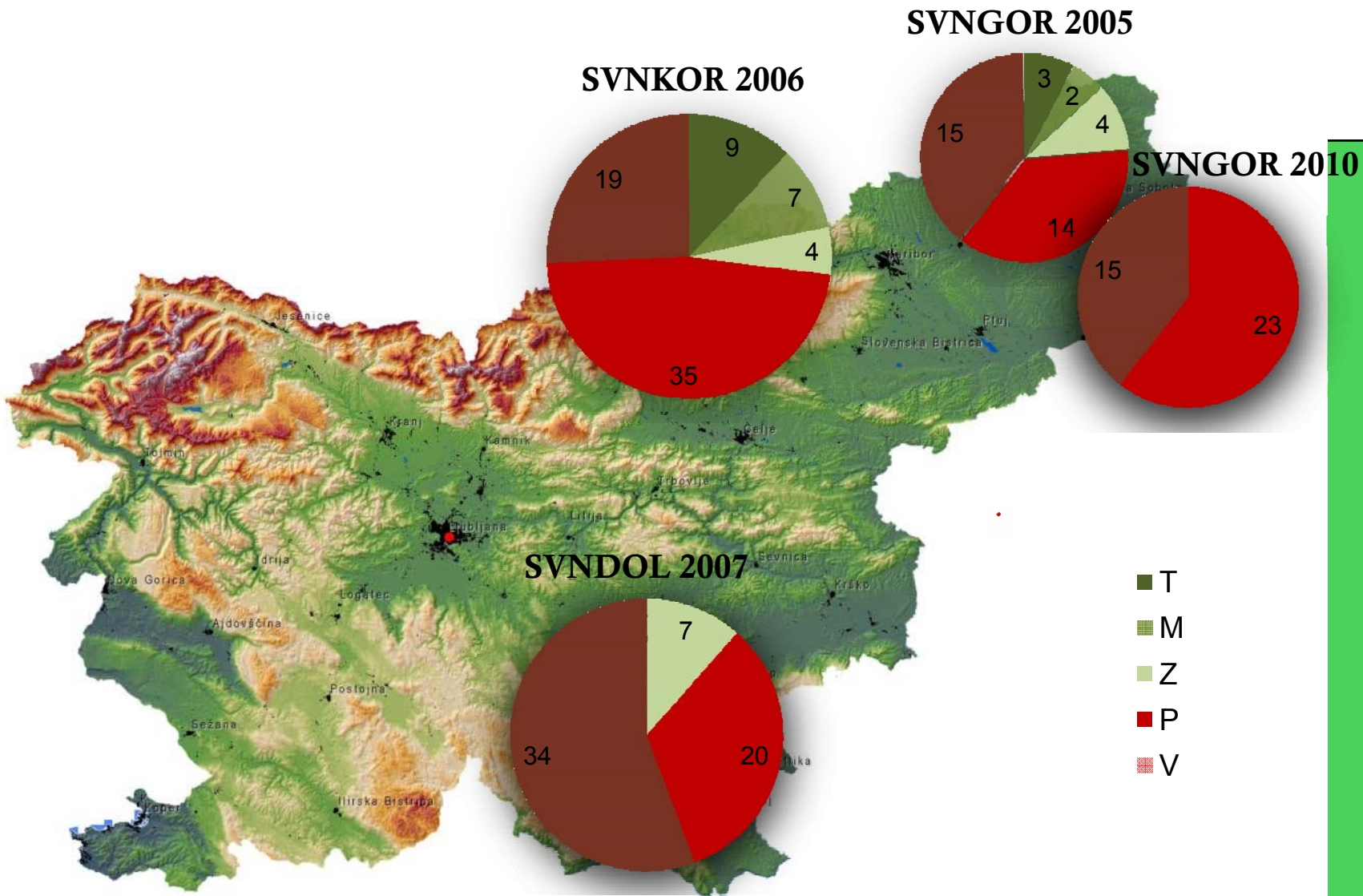


# SVNPIR 2001

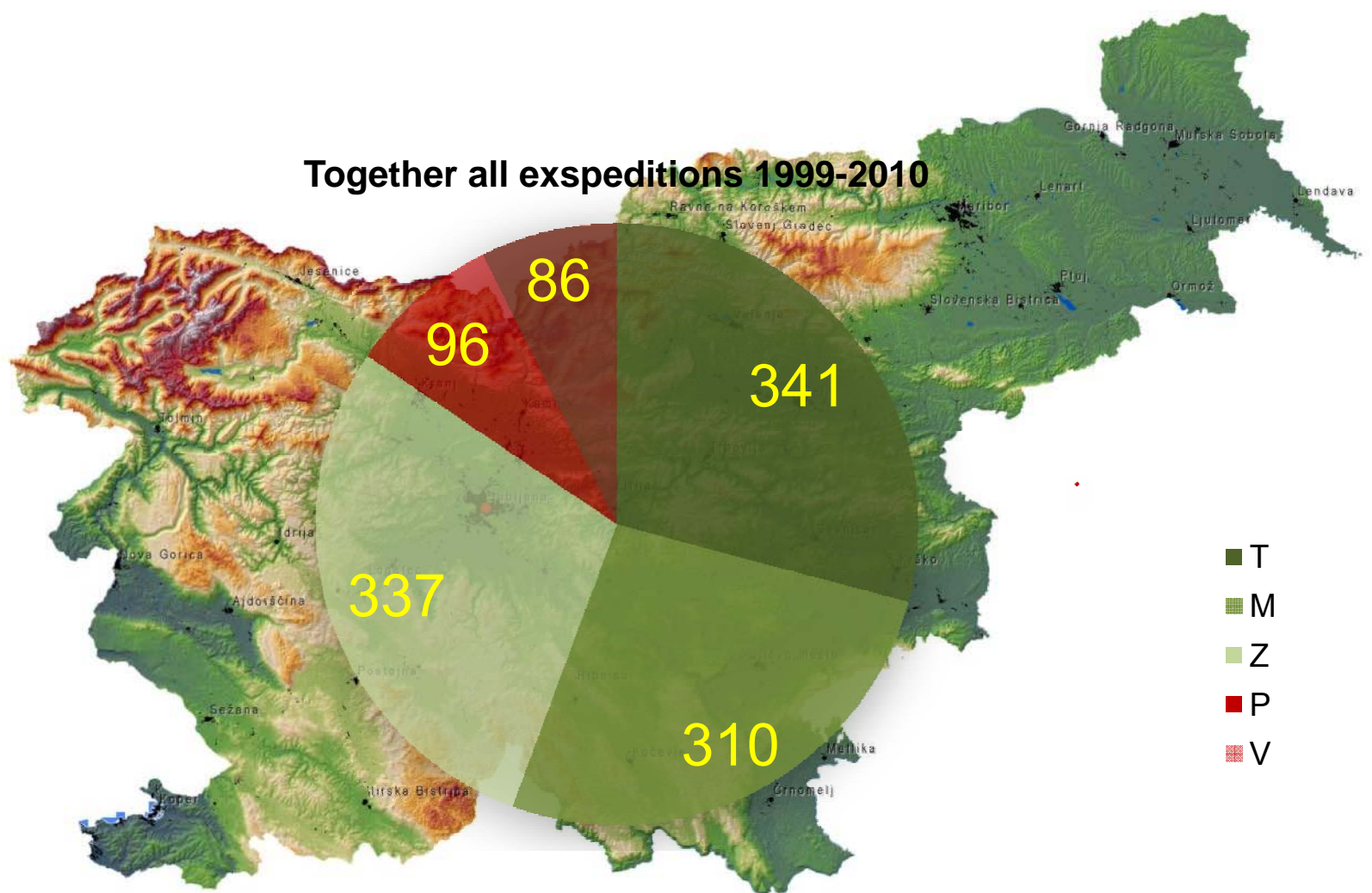


- T
- M
- Z
- P
- V





# Together all expseditions 1999-2010



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Agricultural crop	Species	Number of accessions
Zrnate stročnice	Phaseolus spp.	1116
	Vicia faba	41
	ostale	7
	Skupaj	<b>1164</b>
Krmne rastline	Trifolium sp.	216
	Medicago sp.	43
	Vicia sp.	42
	Lotus sp.	35
	Lolium sp.	31
	Festuca sp.	102
	Dactylis	116
	Phleum	45
	Travniške zeli	112
	ostale	242
	Skupaj	<b>984</b>
Poljščine	Zea mays	13
	Triticum	6
	Secale	12
	Papaver	26
	Panicum	15
	ostale	63
	Skupaj	<b>135</b>
Zelenjadnice	Lactuca sp.	227
	Allium cepa	31
	Brassica oleracea	11
	Diplotaxis spp.	18
	ostale (Cichorium, Valerianella, Solanum Lyc.)	16
	Skupaj	<b>303</b>
Krompir	<i>Solanum tuberosum</i> L.	34
Druge vrste krompirja	<i>Solanum</i> sp.	18
	<i>Solanum</i> sp. diploidne populacije	14
	Skupaj	<b>66</b>

Agricultural crop	Species	Number of accessions
Vinska trta (kloni lastnih selekcij)	Vitis	39 (Ivanjokovci, Vrhpolje)
Vinska trta (stare sorte)	Vitis	50 (Amp. vrt BF - Kromberk pri N.G.)
	Skupaj	<b>89</b>
Jablana	Malus sp.	245 (na Brdu)
Jagoda	Fragaria sp.	56 (na Brdu)
	Skupaj	<b>301</b>
Malina	Rubus sp.	29 (na Brdu)
Robida	Rubus sp.	7 (na Brdu)
Črni ribez	Ribes sp.	53 (na Brdu)
Rdeči ribez	Ribes sp.	13 (na Brdu)
Beli ribez	Ribes sp.	6 (na Brdu)
Kosmulja	Ribes sp.	12 (na Brdu)
Ameriška borovnica	Vaccinium corymbosum	50 (na Brdu)
Brusnica	Vaccinium macrocarpon	2 (na Brdu)
	Skupaj	<b>161</b>
	<b>SKUPAJ</b>	<b>3203</b>

## Report WP2 a

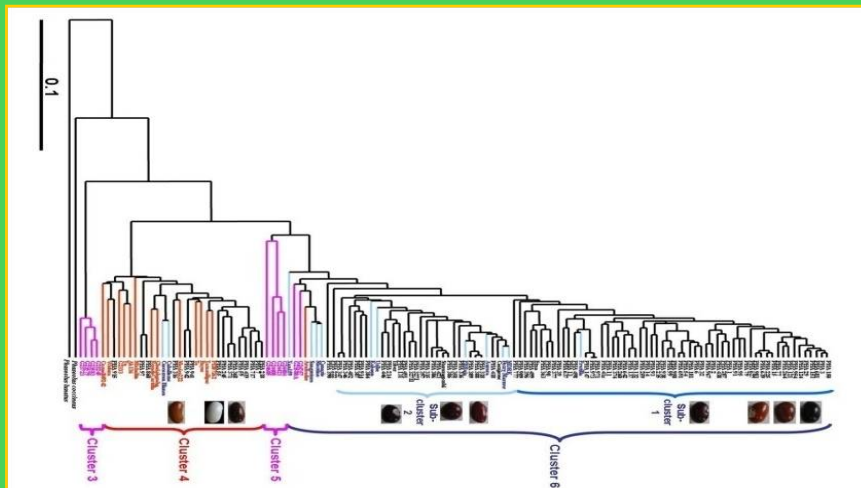
We started to perform morphological and molecular evaluation of the collected species and selected accessions. Field and pot trials will form a basis for morphological characterization and evaluation of genetic resources. Seed material was prepared for planting and consecutive trial establishment. Twenty plants per accession were prepared in styrofoam plates for consecutive planting in pots and in the field (Picture 1). Botanical and morphological characterisation and evaluation for the first year was conducted according to IPGRI(Bioversity International)/FAO descriptors in the greenhouse and in the field (Picture 2 and 3). Herbarium sheets of leaves were prepared for selected species (Picture 4) enabling us to perform morphological measurements (e.g. width and length) later in the season.





## Report WP2 b

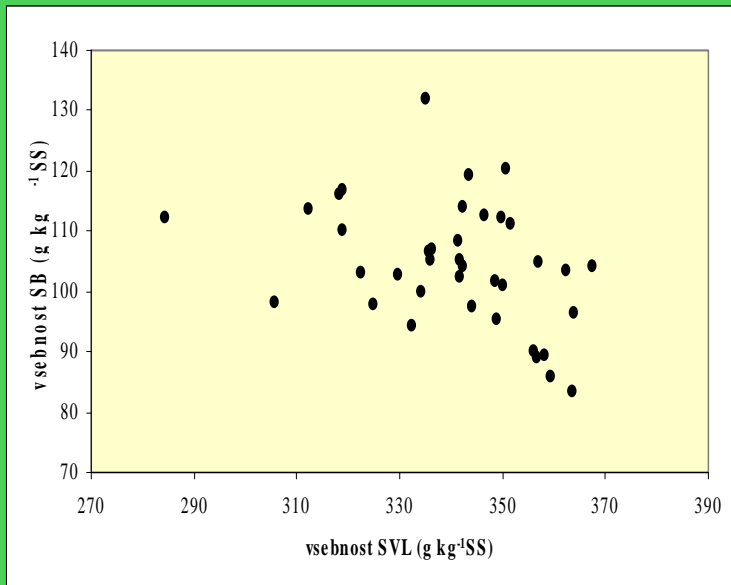
Genetic analysis of selected accessions using Microsatellite markers has begun on selected accessions of Phaseolus. Methodology was prepared and shared with other collaborating organisations (Serbia) performing similar evaluations, in order to unify methods. As well seeds of selected accessions from collaborating institutions (Macedonia, Republika Srpska), that are not going to perform this analysis, were acquired.



Lokus	Mikrosatelitska ponovitev	Nukleotidno zaporedje parov oligonukleotidov v smeri –	začetnih Ta (°C)	Fluoresc entna oznaka	D (bp)
ATA2	(TAA) <sub>20</sub> (GAA) <sub>10</sub>	1: CGTCTGTGTTTGCTAGTA 2: GAGAGAGGTGAAATTGAAAG	47	FAM	129
ATA3	(TAA) <sub>9</sub>	1: CCGAAGACATAACTGATGAT 2: TGAGTACGTGCTGGAACCTT	57	JOE	133
ATA4	(TTA) <sub>8</sub>	1: CAGATTAGTCTTTTGTTCAT 2: GCGTCATGAAACAAGAA	52	JOE	154
ATA5	(TTN) <sub>42</sub>	1: TCCGTAATTGGAACGGAACA 2: CCGATTTTCTTTTGGTCGCT	62	FAM	191
ATA6	N=A, G, T (TAT) <sub>19</sub>	1: TGATTTGTCTAACACTTCAC 2: GGAGATGATTGCATGTAG	57	TAMRA	143
ATA7	(ATA) <sub>11</sub>	1: ATAAATCTATTGAGTCTAG 2: AACAAAGTCAATCTAAAG	49	FAM	136
ATA9	(AAT) <sub>12</sub>	1: AAGCCACTGTAGCTGGAAGC 2: GCCCTTCTACTACCACTCTA	60	JOE	191
ATA10	(ATT) <sub>11</sub>	1: TTGTATCCAAAGATTATTA 2: GACAATAATAAAAAATGGTTT	48	TAMRA	106
ATA16	(TAA) <sub>14</sub>	1: CAAAATGGAAGAAAATGTC 2: TGTTGTAATCAAATCTTG	47	FAM	155
GATS91	(GA) <sub>17</sub>	1: GAGTGCGGAAGCGAGTAGAG 2: TCCGTGTTCTCTGTCTGTG	60	FAM	229
BM170	(CT) <sub>5</sub> CCTT(CT) <sub>12</sub>	1: AGCCAGGTGCAAGACCTTAG 2: AGATAGGGAGCTGGTGGTAGC	57	JOE	179
BM183	(TC) <sub>14</sub>	1: CTCAAATCTTACTGGTGACG 2: TCTTACAGCCTGCAGACATC	57	FAM	149
BM210	(CT) <sub>15</sub>	1: ACCACTGCAATCCTCATCTTTG 2: CCCTCATCCTCCATTCTATCG	60	TAMRA	166

## Report WP2 c

Already characterized Phaseolus accessions will be evaluated for protein, mineral and other nutritional compound contents. In order to do that, we have performed initial analysis of two accessions, to validate analytical methods. Characterisation of red clover accessions (morpf., quality, NIR).



Analytical number		2028/2011		2029/2011	
Accession		ČEŠNJEVE C		GB 740	
Parameter		In sample	In DM	In sample	In DM
DM	g/kg	925,2	1000	904,3	1000
Moisture	g/kg	74,8	0	95,7	0
Raw ash	g/kg	39	42	40	45
Kalcij - Ca	g/kg	1,08	1,17	0,91	1,01
Magnezij - Mg	g/kg	1,41	1,53	1,37	1,51
Kalij - K	g/kg	15,1	16,3	16,7	18,5
Fosfor - P	g/kg	5,25	5,67	4,14	4,57
Baker - Cu	mg/kg	8	9	6	7
Železo - Fe	mg/kg	47	51	63	70
Mangan - Mn	mg/kg	10	10	10	12
Cink - Zn	mg/kg	22	24	28	31

