Conservation and Utilization of PGR at ICARDA

Study Case of *Lathyrus* species

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Genetic Resources Section

Bucharest, 10 April, 2019
The Challenges Facing Sustainable Agricultural Development in Dry Areas
Dry Areas: Fragile Dry Eco-Systems

- Physical water scarcity
- Rapid natural resource degradation and desertification
- Groundwater depletion
- Drought
- Salinity
- Climate change
Challenges to Enhancing Food Security in Dry Areas: Biotic Stresses

- Fungi Diseases
- Insect Pests
- Weeds/Parasitic Weeds
Further Challenges in the Drylands: Inadequate Policies and Insufficient Institutions

- Inadequate agricultural policies for sustainable agricultural development
- Insufficient investment in agricultural research and development
Politically volatile region: Importance of economic and political stability

Linkages between the food and financial crisis

Source: Joachim von Braun
ICARDA Vision, Mission and Mandate
**Vision:** Improved livelihoods of the resource-poor in dry areas

**Mission:** To contribute to the improvement of livelihoods of the resource poor in dry areas by enhancing food security and reducing poverty through research and partnerships to achieve sustainable increases in agricultural productivity and income, while ensuring the Efficient and more equitable use and conservation of natural resources.
Research mandate: Cereal improvement

Global Mandate

Barley

Regional Mandate

Bread Wheat

Durum Wheat
Global Mandate

Lentil

Faba bean

Kabuli Chickpea

Grass pea
Research Mandate
Natural resource management in non-tropical dry areas

Water use efficiency

Combating land degradation

Range improvement

Forage Legumes

Small ruminant nutrition
ICARDA’s geographic mandate
Location of Platforms and Thematic Research sites
Middle East and North Africa region encompasses four major centers of diversity and the Mediterranean hot spots of endemic flora
Proposed Model of Plant Genetic Conservation (Maxted et al., 1997a).

Selection of Target Taxa
Project Commission
Ecogeographic Survey / Preliminary survey Mission
Conservation Objectives
Field Exploration
Conservation Strategies

Ex Situ
(Collecting, transfer and storage)

Seed Storage In Vitro Storage Pollen Storage DNA Storge Field Gene bank Botanical Garden

In Situ
(Designation, management and monitoring)

Genetic Reserve On-Farm Home Gardens

Conservation Products
(seed, live & dried plants, in vitro explants, DNA, pollen, data)
Conserved Product Deposition & Dissemination
(gene banks, reserves, botanical gardens, conservation laboratories, on-farm systems)
Characterization / Evaluation
Plant Genetic Resource Utilisation
(breeding / biotechnology)
Utilisation Products
(breeding new varieties and crops, pharmaceuticals, pure and applied research, on-farm diversity, recreation, etc.)
General sequence of operations in the Genebank

Seeds from collecting/acquisition

- Checking database for duplicates
  - NO
  - Check accompanying data
    - YES
    - Seed Health Test under Lab/quarantine areas
      - Accession Registration
        - Quantity of seeds
          - LOW
          - Viability Test
            - LOW
            - Multiplication
          - SUFFICIENT
          - Multiplication
        - SUFFICIENT
      - Request for data (donors, collectors)
        - NO
        - YES
        - Decide what to do with seeds
General sequence of operations in the Genebank (cont’d)

Seed cleaning, drying and viability testing → Packing → STORAGE → Monitoring
(stocks, viability, diversity, etc) → Distribution → Evaluation → Pre-breeding

Characterization

Regeneration

Documentation
Genetic resources: Continuum conservation to utilization

Ex situ Conservation
- Collect
  - Enrich with new accessions
- Conserve
  - Use best practices
- Evaluate
  - Identify valuable traits
- Deploy
  - Use in pre-breeding and breeding

In situ conservation
- Assess
  - Status and Threats
- Designate
  - Selection hot spots
- Manage
  - Management Plan
- Promote
  - ValorizationAwareness
Genetic resources conservation

- Ecogeographic surveys
- Gap analysis
- Collecting missions
- IP issues
- Seed health applications

Germplasm acquisition

Regeneration & characterization

Documentation

Storage

- Database
- Geographic distribution maps
- Catalogues
- Field guides

- Taxon identification
- Multiplication
- Conservation
- Isolation cages for regeneration of out-crossing species

- Ecogeographic surveys
- Gap analysis
- Collecting missions
- IP issues
- Seed health applications

- Germplasm acquisition
- Regeneration & characterization

- Documentation
- Storage

- Database
- Geographic distribution maps
- Catalogues
- Field guides

- Germplasm acquisition
- Regeneration & characterization

- Documentation
- Storage

- Taxon identification
- Multiplication
- Conservation
- Isolation cages for regeneration of out-crossing species

- Germplasm acquisition
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- Documentation
- Storage

- Database
- Geographic distribution maps
- Catalogues
- Field guides

- Germplasm acquisition
- Regeneration & characterization

- Documentation
- Storage

- Database
- Geographic distribution maps
- Catalogues
- Field guides

- Germplasm acquisition
- Regeneration & characterization

- Documentation
- Storage

- Database
- Geographic distribution maps
- Catalogues
- Field guides
ICARDA Gene Bank Holdings in three different locations

<table>
<thead>
<tr>
<th>Crop/Wild Relatives</th>
<th>No. of Accessions</th>
<th>Crop/Wild Relatives</th>
<th>No. of Accc.</th>
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<tbody>
<tr>
<td>Faba bean BPL</td>
<td>3268</td>
<td>Pisum</td>
<td>6132</td>
</tr>
<tr>
<td>Aegilops</td>
<td>5157</td>
<td>Trifolium</td>
<td>5900</td>
</tr>
<tr>
<td>Barley</td>
<td>30215</td>
<td>Vicia</td>
<td>6561</td>
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<tr>
<td>Bread wheat</td>
<td>15090</td>
<td>Faba bean</td>
<td>6766</td>
</tr>
<tr>
<td>Durum wheat</td>
<td>20516</td>
<td>Chickpea</td>
<td>15195</td>
</tr>
<tr>
<td>Primitive wheat</td>
<td>1276</td>
<td>Lentil</td>
<td>13978</td>
</tr>
<tr>
<td>Wild Hordeum</td>
<td>2575</td>
<td>Wild Cicer</td>
<td>554</td>
</tr>
<tr>
<td>Wild Triticum</td>
<td>1833</td>
<td>Wild Lens</td>
<td>619</td>
</tr>
<tr>
<td>Lathyrus</td>
<td>4451</td>
<td>Range &amp; pasture</td>
<td>7416</td>
</tr>
<tr>
<td>Medicago annual</td>
<td>9133</td>
<td>Others</td>
<td>244</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td></td>
<td></td>
<td><strong>156879</strong></td>
</tr>
</tbody>
</table>

Mostly landraces and unique set of wild relatives
Geo-referencing data for ICARDA’s holdings

<table>
<thead>
<tr>
<th>Germplasm group</th>
<th>No. accessions</th>
<th>Geo-referenced accessions</th>
<th>%</th>
</tr>
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<tbody>
<tr>
<td>Cereals</td>
<td>76906</td>
<td>51075</td>
<td>66.41</td>
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<tr>
<td>Food legumes</td>
<td>40380</td>
<td>15417</td>
<td>38.18</td>
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<tr>
<td>Forage legumes</td>
<td>32177</td>
<td>22713</td>
<td>70.58</td>
</tr>
<tr>
<td>Pastures and range species</td>
<td>7416</td>
<td>6134</td>
<td>82.71</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>156879</strong></td>
<td><strong>95339</strong></td>
<td><strong>60.77</strong></td>
</tr>
</tbody>
</table>
Genetic resources utilization

- In situ conservation
- Ex situ conservation

Ecogeographic surveys & Gap Analysis

Core collection
Reference sets, FIGS sets

Best bet sets

Pre-breeding

Evaluation and gene mining

- Transfer useful genes to elite germplasm
- Evaluation to major biotic and abiotic stresses and good quality attributes

icarda.org
“A limited set of accessions representing, with a minimum of repetitiveness, the genetic diversity of a crop species and its wild relatives”. Frankel, 1984

1. Sampling of a constant number of accessions per region (C strategy),
2. Sampling in proportion to the logarithm of the number of accessions available per region (L strategy),
3. Marker-assisted strategy (M strategy) where the sampling is based on marker allele richness.

Passport data of accessions to be used to develop groups based on the geographical origin of the accessions and the sampling is to carry out randomly within these groups. The clustering methods include molecular and morphological to guide the sampling (Schoen and Brown, 1993).
A rational approach to exploiting large genetic resource collections

Efficient and effective methods to mine genebanks for useful traits to breeders

Deliver priority trait best bet subsets to requestors
Geographical distribution of the two sets (wet set, blue circle and dry set, green triangle) of Faba bean selected using FIGS.
Characterization/Evaluation

Characterization/preliminary evaluation using international set of descriptors
- In the field
- In the plastic house
- Isolation cages for cross-pollinating species

In-depth evaluation in collaboration with Germplasm Program

Molecular characterization
Crop Genebank Knowledge Base

Forage legume genetic resources

Contact person for Forage legumes: Jean Hanson, ILRI, Ethiopia

Contributors to the section on best practices for management of forage legume genetic resources: ILRI, Ethiopia; Jean Hanson; Joe B. Carruth;建昂贵; ICARDA; Syrma Ahmed Amm; Kenneth Street; Ali Shehadeh; Nafisa Rashid; F. Ahmad; 86CFLP, Australia; Richard Smaith; BIOLR, International; Alidou Abadje; Ethiopia (Alexandra Henge); CIAT, Cali, Colombia; Daniel B€epoeb, Marthe Caste;

Compilation of best practices

Information on genebank management of forage legumes was gathered from current genebank practices, accumulated experience, literature and the websites of major genebanks (ICARDA, ILRI, CIDA, CIAT, CAB, INRA, USDA, U.S. Department of Agriculture, USDA-VSS and BGRU- Munich) and compiled into tables of procedures. This provided the basic information for selection, justification and recommendations that were then compiled, distinguishing information for seed banks and field banks. This information was finally uploaded into the website, complemented with relevant photos and revised and validated by the expert group.

Importance and origin

Over 1600 species of legumes (from about a total of 17,000 legume species worldwide) can be used as feed for livestock, although only about 60 species have been developed and widely used as cultivated forage. The use of forage legumes is estimated to be as old as 11,000 years, with some species first being used as grain for human consumption and more recently only used for fodder or pasture, or vice versa (Miller, 1983). Although tropical forage legumes have been grazed in natural habitats for many years, they have only been under cultivation for as little as 50 years, while some like Centrosema pubescens, Lalage purpurea and Paranica javanica were used as cover crops and green manure at least a century earlier. Tropical legumes may have originated from tropical forests and natural grasslands and were later adapted to a variety of environments. Currently genebanks can be found from the tropics to the Arctic regions, from high mountain regions to deserts and from rocky slopes and deserts to swamps (Williams, 1983).
Conservation field guide to Grasspeas
**L. sativus L.**


**Description** - Annual, slender, ascending plant. Vegetative parts glabrous when green. Plants 10–70(--100) cm. Stems winged. Leaflets present, 1 per leaf, pinnate. Leaf rachis not laminate, tendrilous. Leaflets linear, or lanceolate, apex mucronate, 20–100 mm long, 1.5–11 mm wide, venation parallel. Stipules lanceolate, or lanceolate-acuminate, base semi-sagittate, margin entire, glabrous, 1–1.5 times as broad as stem. Stipules shorter than petiole. Peduncles 1–40(–45) mm long, longer than leaf. Pedicel 5–8 mm long. Flowers 1 per inflorescence, concolorous. Corolla white, or violet, or blue. Flower 14–20 mm long. Standard with more than 5 conspicuous veins, apex strongly emarginate. Wings white, or blue, or violet. Calyx glabrous, 7–10 mm long, tube not gibbous, teeth unequal, straight, longer than tube. Calyx lower teeth longer than tube. Style 5–6 mm, twisted, linear. Ovary oblong. Legume beaked, broadly-oblong. Legume 9–12 mm long, 6–8 mm wide, glabrous. Lower suture not ciliate. Mature legume indelicient. Amygdaloid pods not present. Legume valves gibbous, eglanular. Upper suture broadly winged. Seed surface smooth. Seeds per pod (2–)3–4(–5). Hilum 1.5 mm long.

**Habitat** - Field crop and weed, s1–1520 m.

**Geographical distribution** - Native:
- **AFRICA**
  - *East Tropical Africa*: Tanzania
- **Eastern Asia**: Indonesia

**Uncertain Origin:**
- **ASIA-TEMPERATE**
  - *Western Asia*: Afghanistan, Iran; Iraq; Palestine; Jordan, Lebanon, Syria
Ecogeographic Survey and Gap Analysis of *Lathyrus* species
• National, regional and international efforts are ongoing for collecting and conserving *ex situ* the genetic resources, new approaches are needed to fill the gaps in the existing collections (Amri *et al.*, 2008, unpublished report). There has been relatively little effort in conserving *in situ/on-farm* the landraces and wild relatives of major crops as these were not targeted with most of the existing genetic reserve (Guarino *et al.*, 1995; Hawkes *et al.*, 2000, Amri *et al.*, 2008 unpublished report).

• One such novel approach to help prioritize conservation action is genetic gap analysis.

• Burley (1988) proposed four steps to identify the gaps in conservation efforts:
  • identifying and classifying biodiversity;
  • locating areas managed primarily for biodiversity;
  • identifying biodiversity that is under-represented in the managed areas; and
  • setting priorities for conservation action (Jennings, 2000).
The approach of conservation gap analysis proposed by Maxted et al. (2008) is based on comparing natural diversity with current conservation actions to identify the gaps to revise the conservation strategy. He recommended four steps for gap analysis:

- identification of priority taxa,
- identification of ecological breadth and complementary hotspots using distributional data,
- matching the identified ecogeographic breadth and complementary hotspots with the existing conservation actions, and
- ending with the formulation of a revised in situ and ex situ conservation strategy.
• Gap analysis can also be applied to taxonomic and genetic diversity and its distribution in existing wild populations, as illustrated in the “Ecogeographic Survey and Gap Analysis for Different Sections of Lathyrus L. Shehadeh et al., 2013.

• The Lathyrus gene pool is an ideal candidate for this application of a gap analysis due to its adaptation to harsh environments and the agricultural importance of some species, such as grass pea as food and feed for poor people. A review of ex situ conservation efforts of Lathyrus was done through the Lathyrus conservation strategy undertaken in 2007 by the Global Crop Diversity Trust in collaboration with ICARDA (GCDT, 2007),
### Importance of *Lathyrus* genus

- Useful crops and ornamental plants.
- About 160 species belonging 13 sections;
- Four main cultivated species: *L. sativus*, *L. cicera*, *L. odoratus* and *L. ochrus* included in sections Lathyrus and Clymenum.
- *Lathyrus* species, may play a key role in adapting to climate change;
- Improvement in yields and development of low or free ODAP varieties.

<table>
<thead>
<tr>
<th>Species</th>
<th>Use</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>L. annuus</em></td>
<td>Pulse, Fodder</td>
</tr>
<tr>
<td><em>L. aphaca</em></td>
<td>Fodder</td>
</tr>
<tr>
<td><em>L. blepharicarpus</em></td>
<td>Pulse</td>
</tr>
<tr>
<td><em>L. cicera</em></td>
<td>Pulse, Fodder</td>
</tr>
<tr>
<td><em>L. clymenum</em></td>
<td>Fodder</td>
</tr>
<tr>
<td><em>L. gorgoni</em></td>
<td>Forage</td>
</tr>
<tr>
<td><em>L. hirsutus</em></td>
<td>Horticulture</td>
</tr>
<tr>
<td><em>L. latifolius</em></td>
<td>Pulse, Fodder</td>
</tr>
<tr>
<td><em>L. ochrus</em></td>
<td>Horticulture</td>
</tr>
<tr>
<td><em>L. odoratus</em></td>
<td>Forage</td>
</tr>
<tr>
<td><em>L. pratensis</em></td>
<td>Horticulture</td>
</tr>
<tr>
<td><em>L. rotundifolius</em></td>
<td>Forage</td>
</tr>
<tr>
<td><em>L. sativus</em></td>
<td>Pulse, Forage</td>
</tr>
<tr>
<td><em>L. sylvestris</em></td>
<td>Forage</td>
</tr>
<tr>
<td><em>L. tingitanus</em></td>
<td>Fodder</td>
</tr>
<tr>
<td><em>L. tuberosus</em></td>
<td>Tubers</td>
</tr>
</tbody>
</table>

Ali Shehadeh, 22 November 2017
Methodology

- Taxonomic, ecological, geographic and conservation information for *Lathyrus* were collated;
  - Passport data associated with herbarium specimens,
  - Germplasm accessions data;
  - Secondary data from media sources.

- The *ex situ* conservation status of the genus was assessed to provide a priority ranking for future collection missions;

- Patterns of species richness were analyzed and *in situ* hotspots identified;

- Target IUCN-recognized protected areas are identified as potential sites to establish genetic reserves.
Ecogeographic survey

Taxonomic, ecological, geographic and conservation information for *Lathyrus* were collated from ICARDA, Global Biodiversity Information Facilities (GBIF) and herbarium specimens survey made during the project.
Ecogeographic survey data

- Ecogeographic survey data
- Ecological data
- Geographic data
- Curatorial data
- Taxonomic data
- Descriptive data
<table>
<thead>
<tr>
<th>Step</th>
<th>Details</th>
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</thead>
<tbody>
<tr>
<td>Standardization to a single format</td>
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<tr>
<td>Duplicate observations identified and removed</td>
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</tr>
<tr>
<td>Occurrences identified outside of the natural range of the species were not considered in the final analysis</td>
<td></td>
</tr>
<tr>
<td>Unknown locations removed</td>
<td></td>
</tr>
<tr>
<td>Basic statistics describing the taxonomic, geographic, curatorial and ecological data were derived</td>
<td></td>
</tr>
<tr>
<td>Geo-reference data was checked</td>
<td></td>
</tr>
<tr>
<td>Each data field was indexed, and errors and invalid entries were manually corrected</td>
<td></td>
</tr>
</tbody>
</table>
A total of 157,959 records were gathered

61,081 unique herbarium and germplasm of 97 *Lathyrus* species were derived

18,147 unique herbarium and germplasm accessions of 37 priority species were used in this study
Gene pool concept of *Lathyrus*

**Primary gene pool**
- GP1A (Cultivated forms of *L. sativus*)
- GP1B (Wild forms of *L. sativus*)

**Secondary gene pool**
- L. chrysanthus
- L. gorgoni
- L. marmoratus
- L. pseudocicera
- L. amphicarpos
- L. blepharicarpus
- L. chloranthus
- L. cicera
- L. hierosolymitanus
- L. hirsutus

**Tertiary gene pool**
- Other *Lathyrus* species
Gap analysis

Conservation gap analysis is based on comparing natural diversity with current conservation actions to identify the gaps to revise the conservation and utilization strategies.

- Collecting to fill the gaps in the existing *ex situ* collections;
- New collecting missions (Targeted collections);
- *In situ* Conservation of crop wild relatives;
- On-farm conservation of the landraces of major crops.
Steps of gap analysis

1. Identification of priority taxa
2. Identification of ecological layer and complementary hotspots
3. Matching the identified complementary hotspots with the existing conservation actions, and
4. Formulation of a revised *in situ* and *ex situ* conservation strategy
<table>
<thead>
<tr>
<th>Country</th>
<th>Total species</th>
<th>Priority species</th>
<th>% of unique collections</th>
<th>Total species / country</th>
<th>Priority species / country</th>
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</thead>
<tbody>
<tr>
<td>Turkey</td>
<td>1662</td>
<td>652</td>
<td>2.72</td>
<td>57</td>
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<td>Syria</td>
<td>1784</td>
<td>1105</td>
<td>2.92</td>
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<td>Greece</td>
<td>1194</td>
<td>617</td>
<td>1.95</td>
<td>30</td>
<td>15</td>
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<tr>
<td>Spain</td>
<td>3263</td>
<td>1690</td>
<td>5.34</td>
<td>43</td>
<td>14</td>
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<tr>
<td>France</td>
<td>15,820</td>
<td>5314</td>
<td>25.9</td>
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<tr>
<td>Lebanon</td>
<td>174</td>
<td>109</td>
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<td>Italy</td>
<td>399</td>
<td>209</td>
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<td>Palestine</td>
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<td>11</td>
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<td>10</td>
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<td>Finland</td>
<td>509</td>
<td>51</td>
<td>0.83</td>
<td>9</td>
<td>2</td>
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<tr>
<td>Others</td>
<td>716</td>
<td>528</td>
<td>1.17</td>
<td>15</td>
<td>9</td>
</tr>
<tr>
<td>Total</td>
<td>61081</td>
<td>18147</td>
<td></td>
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</tr>
</tbody>
</table>
Regression of *Lathyrus* priority species against the number of records for each country.

\[
y = 0.0012x + 8.0027
\]

\[R^2 = 0.0991\]

Ex situ conservation priorities

Geo-referenced passport data associated for *Lathyrus* species used to identify gaps in current *ex situ* conservation

- **High priority:** <100 accession *ex situ* conserved
- **Medium priority:** >100<500 accessions
- **Low priority:** > 500 accessions
### Ecogeographic data set of the priority species included in the analysis

<table>
<thead>
<tr>
<th>Species</th>
<th>Accessions in Global <em>Lathyrus</em> Database</th>
<th>Accessions in EURISCO</th>
<th>Accessions in USDA</th>
<th>Total germplasm accessions</th>
<th>Georeferenced accession &amp; herbaria samples</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>cicera</em></td>
<td>214</td>
<td>558</td>
<td>42</td>
<td>814</td>
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In situ species richness and complementarity analysis

Patterns of specific richness, based on the germplasm accession and herbarium specimen data, were analyzed

• Hotspots of species diversity identifies

Complementarity analysis is carried out to identifying the minimum number of 100 x 100 km² grid cells that will capture the maximum number of species.

• Putative reserves were selected

Species richness and complementarity were analysed for all *Lathyrus* sections

Only the sections Lathyrus and Clymenum are presented here
Species richness for all unique herbarium specimens and germplasm accessions of *Lathyrus* species
Species richness for unique herbarium and germplasm accessions of 37 priority *Lathyrus* species
Location of complementarity analysis for priority *Lathyrus* species diversity hotspots
Species richness for priority species of section Lathyrus
Location of complementarity analysis for priority section Lathyrus species diversity hotspots
Species richness for priority species of section Clymenum
Location of complementarity analysis for priority section Clymenum species diversity hotspots
Location of priority species complementary hotspots with associated IUCN recognised protected areas
A Case Study for Grass peas

Grass pea (Lathyrus sativus L.), known as Chickling pea or Indian pea, annual legume crop of economic and ecological significance in South Asia and Sub-Saharan Africa, and to a limited extent in some countries of CWANA.

L. sativus is widely cultivated for human consumption, fodder and green manure. Considered by farmers as an important low risk aversion crop.

It is nutritionally equivalent with other grain legume species; But, may contain 0.1-2.5% ODAP which have been found to be neurotoxins, the causative agent of crippling, irreversible neurological disorder, Lathyrism.
Genetic diversity of Lathyrus sativus using Shannen index
Genetic diversity of *Lathyrus sativus* landraces using Shannon index.
Genetic diversity of wild *Lathyrus sativus* using Shannon index
Conclusions

The research illustrates how the existing geo-referenced passport data associated for *Lathyrus* species can be used for a comprehensive gap analysis;

Gap analysis is an effective tool to fully address the need for a more comprehensive and complementary conservation strategy that encompasses both *in situ* and *ex situ* applications;

The study of herbarium and germplasm accessions’ passport and collection data coupled with ecogeographic analyses can quantify the completeness of current *in situ* and *ex situ* conservation actions and identify gaps in conservation diversity at both the taxon and geographic level that in turn helps in the prioritisation of future conservation actions;

This basic methodology could be successfully applied to assist the development of national and regional conservation strategies for crop wild relative diversity;

West Asia, mainly Syria and Turkey are confirmed as the centres of diversity for *Lathyrus* priority species;

*Lathyrus* diversity and its threats should continuously be assessed and monitored.
Thank you