Progress report of the AEGIS model crop: *Prunus*July 2008

1. Introduction

Prunus is one of the four model crops chosen for the AEGIS initiative. It was selected on the basis of several criteria set out in the AEGIS Project Document 2005. In particular:

- it includes many crops (such as almond, apricot, cherry, peach and plum) for which <u>Europe</u> can be easily recognized as a <u>relevant region</u> for conservation and use;
- it involves <u>institutions</u> from different sub-regions of Europe;
- the European *Prunus* Database (EPDB) information system is <u>relatively well advanced</u> and allows the possibility of identifying 'most appropriate accessions' (MAAs);
- the ECPGR *Prunus* Working Group has agreed to reach a <u>practical way of sharing</u> responsibilities;
- it is an example of a crop group that is more suited to conservation in a <u>decentralized</u> system, on the basis of existing arrangements and experience;
- it is an example of a crop group that comprises <u>perennial clonal plants</u> that are <u>conserved</u> vegetatively;
- it is an example of a crop group that is not included in Annex I of the International Treaty.

The aim of the AEGIS initiative, so far as *Prunus* is concerned, is to enhance the conservation of *ex situ* accessions, especially those of European origin but also others of importance to European horticulture, breeding programmes or research, via collaborations and agreements that will e.g. minimize barriers to exchange of material, encourage adequate documentation and standards, and reduce duplication. In particular, AEGIS seeks to nominate one primary accession in Europe per appropriate genotype and one reserve, that will be curated to high standards and will be freely available.

The decentralised system of conservation adopted for *Prunus* has the advantages of building on existing collaborations, requiring minimal changes in infrastructure, maintaining genetic resources in the country of origin and maintaining local expertise and collections, features which would not be achieved by a centralized system. It is recognised that one disadvantage of the decentralised system is the more complex management that is needed.

The AEGIS *Prunus* Group previously met in Alnarp and in Rome and chose cherry (*Prunus avium*) as a model to investigate the practicalities of choosing AEGIS accessions from the European *Prunus* Database (EPDB).

The report of the AEGIS *Prunus* Group was endorsed in draft by the ECPGR Working Group on *Prunus* at its Seventh Meeting, 1-3 December 2005, Larnaca, Cyprus. This new document updates that report particularly in the light of subsequent experience and discussion at the Radzików meeting.

2. Establishing selection criteria for the identification of the Most Appropriate Accessions (MAAs)

Selection criteria were proposed by the AEGIS *Prunus* group and agreed by the ECPGR *Prunus* Working Group; they were subsequently revised after discussion with *Malus/Pyrus* and *Vitis* chairs, vice-chairs and database managers at NCG Meeting, 29-31 March 2006, Bonn. Recently, AEGIS has circulated a document on "Draft selection criteria for the identification of MAAs to be included in the European Collection" which has been discussed during the Radzików meeting.

In vegetatively propagated crops such as *Prunus* there are, conceptually, two steps to the choice of MAA. Firstly there is the choice of the appropriate genotypes for further consideration. Secondly there is the choice of the MAAs of those genotypes.

a. Recommended selection criteria for choice of cherry genotypes

The genotypes to be considered for AEGIS should be:

- in the public domain (i.e. designated as such to AEGIS by governments or holders and excluding most breeder's selections);
- originating in Europe or introduced to and important to Europe;
- genetically unique on the basis of available data and/or recorded history;
- agronomically/scientifically and/or historically/culturally important or potentially important
- plant genetic resources, including ornamental genotypes and crop wild relatives (including wild *Prunus avium* even though it is a forest species);

These criteria essentially correspond to the AEGIS 'primary selection criteria' which were approved by the ECPGR Steering Committee and are not crop-specific.

b. Comments on draft priority selection criteria for choice of MAAs

The following set of suggested priority selection criteria has recently been circulated by AEGIS to guide the countries and their genebanks in identifying the accessions for a given crop that the country is prepared to nominate to the respective WG for inclusion in the European Collection. These nominated accessions will then be scrutinized by the respective Working Groups with the intention of identifying the MAAs, in particular by using the secondary selection criteria that have been adopted by the Working Group for a given crop or group of crops, before proposing them as European Accessions to the individual countries for their acceptance. The categories of germplasm for conservation as part of the European Collection are, in decreasing order of priority:

- Accessions that have been collected or bred in the country where they are being conserved in one of its genebanks;
- Accessions of the crop genepool in question that are crop wild relatives;
- *Germplasm accessions that are traditional varieties and/or landraces;*
- *Germplasm accessions that represent old and/or obsolete varieties;*
- Modern varieties, bred with conventional methods;
- Accessions that are breeding lines;
- *Genetic stocks of the crop genepool in question;*
- Accessions that consist of research material like mapping populations, mutants, etc. if different from 7 above.

The AEGIS *Prunus* group has problems with this list. All the categories should be included and should be not prioritized (Thus, 'priority selection criteria' is a confusing term and, moreover, could be confused with 'primary selection criteria'). Moreover, in vegetatively propagated crops, most of the categories refer to genotypes and not to accessions.

The AEGIS group recommends the following re-worded categories for inclusion in AEGIS without prioritization:

- Traditional cultivars and/or landraces:
- Old and/or obsolete cultivars:
- Modern cultivars, bred with conventional methods;
- Significant breeding lines;
- Genetic stocks, mutants and seedlings of mapping progenies;
- Crop wild relatives.

c. Recommended secondary selection criteria

The following secondary selection criteria proposed by AEGIS concern the choice of MAA when multiple accessions of acceptable genotypes are available:

- Maintained in "country of origin";
- A known origin (collected and/or bred; pedigree data!?);
- Comprehensiveness of passport information (It is suggested to use the multi-crop passport descriptors);
- Number of regeneration/multiplication cycles (As far as known; otherwise rough estimates would be helpful);
- Health status: virus-free;
- Existence of morphological and/or molecular characterization data;
- Existence of (agronomical) evaluation data;
- Validated accession name (particularly relevant for perennial clonal crops where the same name can be attributed to different accessions; history of individual accessions is important; special attention to be paid to synonyms and homonyms; transparent selection procedure is needed).

The AEGIS *Prunus* group proposes some improvement of wording for the criteria to be considered in the choice of MAAs, as follows:

- Maintained in "country of origin";
- Of known origin, whether collected or bred;
- Accompanied by comprehensive passport information using the EURISCO multi-crop passport descriptors;
- Of high health status, e.g. virus-free;
- Accompanied by morphological and/or molecular characterization data;
- Accompanied by agronomic evaluation data;
- Correctly named (particularly relevant for perennial clonal crops in which synonyms and homonyms are frequent).

Regarding the country of origin criterion, implementation of AEGIS would be hampered by the current lack of participation of many countries in the EPDB. Indeed, if a country chooses not to participate, then accessions from that country cannot be maintained in the country of origin so far as AEGIS is concerned.

Regarding the need for accompanying data, it must be recognised that data sets from many collections are far from complete. The AEGIS group regards the provision of the following EURISCO minimum passport data as obligatory:

- Accession name (ACCENAME)
- Country of origin (ORIGCTY) though the EURISCO definition is not appropriate for vegetatively propagated crops/cultivars
- Institute code (INSTCODE), where the accession is maintained
- Species (SPECIES)

Moreover the AEGIS group recommends that curators complete the following EURISCO passport descriptors and *Prunus* specific descriptors:

- Accession number (ACCENUMB)
- Acquisition date (ACQDATE)
- Donor institute code (DONORCODE) and its decoded name (DONORDESCR)
- Donor accession number (DONORNUMB)
- Other identification (numbers) associated with the accession (OTHERNUMB)
- Breeding institute code (BREDCODE) and its decoded name (BREDDESCR)
- Identification of material (IDENTIF: *Prunus* specific descriptor)

- Virus disease status (VIRUSTATUS: *Prunus* specific descriptor)
- Date of the virus disease status (VIRUSDATE: *Prunus* specific descriptor)

Finally characterization data and photographs should be helpful for selecting the MAAs when several accessions are available.

Regarding the health status criterion, the susceptibility of some *Prunus* crops to virus diseases hinders the selection of MAAs. High health status material free from viruses is desirable, but to create and maintain large numbers of accessions free from viruses would be very expensive. Cryopreservation could be explored as a way of maintaining virus freedom in clean material.

Regarding the validation of the accession name, a specific descriptor called "IDENTIF" is available in the EPDB but the information is seldom provided by curators.

d. General observations and comments on the process of developing the criteria and lessons learnt for other crops

It should be noted that some important *Prunus* collections are not in contact with the EPDB; thus in 2008 there are 2731 cherry accessions in the EPDB and 2434 cherry accessions in EURISCO but only 384 accessions are common to both. If further collections become involved in AEGIS at a later date there may be consequences regarding the re-nomination of AEGIS accessions.

The points made above in relation to *Prunus*, including the caveat about health status, are relevant to other fruit and perennial crops. It should be noted that *Malus* and *Fragaria*, unlike *Prunus*, are Annex 1 crops.

3. Establishing the list of MAAs

a. The procedure followed, including the respective roles of associated institutions, the countries (i.e. National Coordinators), the Central Crop Database manager and the Working Group

The following procedure had been agreed by the AEGIS *Prunus* group at the meeting in Rome in June 2005 and subsequently approved by the ECPGR *Prunus* Working Group.

In nominating the initial AEGIS accessions, the <u>first step</u> is for the EPDB Manager to update the current accession database, contacting current participants for revisions and seeking new contacts. The <u>second step</u> is for the Database manager to collate the data relating to the accessions offered so that, for the different identities, it can be seen at which sites they are held and what data are available. The <u>third step</u> is for the Database manager and the ECPGR *Prunus* Working Group (or crop-specific sub-committees) to review the data with a view to nominating the primary and reserve AEGIS accessions and for the Database manager to register the AEGIS status of the chosen accessions and the reasons for choosing them. The <u>fourth step</u> would be for the Database manager to notify the collection holders and AEGIS Secretariat of the decisions.

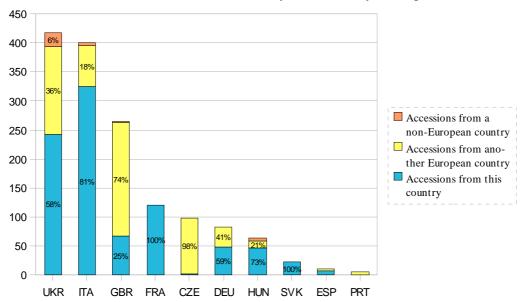
However, as no countries have yet signed to AEGIS, this procedure could not be implemented. Moreover problems of synonymy and trueness-to-name were apparent. As a consequence, a subgroup of cherry experts was formed in order to collaborate with the Database manager for the identification in the EPDB of likely synonyms among the cherry accessions. The subgroup met at Gembloux in June 2008 and addressed the synonymy problems of 636 preselected accessions, attributing them to 178 duplicate groups each representing a single genotype referred to by an agreed euonym. Then the EPDB manager was able to select a list of genotypes, using the primary

criteria, and subsequently to prepare a list of MAAs of those genotypes by applying some of the secondary criteria.

b. List of MAAs generated for Prunus based on the EPDB

The generation of a preliminary list of MAAs has been put into practice in advance of the implementation of AEGIS, using the EPDB as a tool. This list will be revised after the update of the data sets and probably after the signature of the countries involved.

Among the 2731 sweet cherry accessions that had been already registered in EPDB, 668 accessions were excluded when preparing the preliminary list (267 unnamed accessions, 70 protected cultivars, 259 UPOV reference cultivars from non-European countries, 5 mislabelled accessions, 67 others). Among the 2063 accessions remaining, 1116 were considered as unique accessions and the other 947 accessions were attributed to 367 duplicate groups (including the 178 duplicate groups identified by the subgroup of cherry experts). As a result, the preliminary list contained 1483 primary MAAs (1116+367) and 367 reserve accessions or safety duplicates. The following diagram shows that the primary MAAs were maintained by ten countries of the twelve involved in the EPDB. Moreover, 60% of MAAs were maintained by their country of origin.



Preliminary MAAs for *Prunus avium* – host countries and countries of origin

When the MAAs have been finalized, the EPDB manager will complete the following AEGIS descriptors that were presented to the Larnaca meeting and that will need to be harmonized with the ECPGR Doc & Information Network:

- Offered to AEGIS System (Yes/No);
- Year of inclusion in AEGIS System;
- Year of exclusion from AEGIS System;
- AEGIS status: 1 primary; 2 reserve; or 3 undetermined (or AEGISSTAT field suggested by the ECPGR Documentation & Information Network);
- AEGIS reasons for choosing the accession (and/or for excluding?).

c. Experiences with the use of the selection criteria while establishing the list

The EPDB was easy to use for the application of selection criteria. Among the passport descriptors already in the EPDB, six were identified as relevant for the designation of AEGIS accessions on the criteria previously mentioned: Accession name; Synonyms; Country of origin; Curator institute; Donor institute: Protection status.

However, several problems are apparent. First no countries have yet offered accessions to the EPDB system. Furthermore many partners could not or did not send updated data and the Database Manager cannot reasonably ask for updates every year. In addition there are many cultivar synonyms and some homonyms. There are also some examples of unlikely data sets and dubious identities. And MAAs could not easily be selected if candidate accessions had non-comparable data sets.

The AEGIS group foresees problems when a second round of nomination of MAAs is undertaken, using additional and enhanced data sets.

d. Lessons learnt for other crops

The general problems experienced with *Prunus* are likely to be encountered with the other ECPGR fruit crops *Malus*, *Pyrus* and *Vitis*. The previous report of the AEGIS *Prunus* group was presented to the chairs, vice-chairs and database managers of the *Malus/Pyrus* and *Vitis* Working Groups and a brief update was provided to these other parties at the Fruit Synonyms meeting. So these groups are aware of the direction in which the *Prunus* group is moving. The AEGIS group would draw attention to three specific points.

Clarification of the existing synonyms is very important to help identify the MAAs to be included in the AEGIS European Collection.

Clarification of the EURISCO descriptors 'Country of origin' (for cultivars) and 'Accession name' (for wild accessions) is essential.

The flow of information between curators, ECCDB, EURISCO and National Focal Points (NFPs) needs to be clarified. The EPDB manager will contact the Doc & Information Network.

4. Establishing the quality management system (AEGIS quality system - AQUAS)

a. General observations on establishing a AQUAS for model crop

Establishing a QMS for the AEGIS European *Prunus* Collection is desirable. However, it is important to be pragmatic rather than doctrinaire and to recognise that the different participating collections can achieve satisfactory standards in different ways. The general principles are more important than over-prescriptive protocols.

b. Comments on the proposed principles and elements of the AQUAS (see discussion paper)

The AEGIS group considered the design of the AQUAS (ie monitoring, reporting, roles...), following the 'PDCA' approach and proposed the following process for discussion:

PLAN

- Definition of minimum technical standards by the AEGIS *Prunus* group
- Agreement of standards by the Prunus WG

DO

- Acceptance of standards by National Coordinators and participating curators by signing the Memorandum of Understanding
- Implementation of standards by participating curators

CHECK

• <u>Self-checking</u>: curators to maintain 'light' evidence of compliance with AEGIS minimum standards of conservation and distribution

and/or

• External checking by WG or National Coordinators (to be decided): curators to confirm, when requested by the 'checker', compliance with AEGIS minimum standards of conservation and distribution – perhaps by completing a simple 'tick box' questionnaire

ACT

- National Coordinators to arrange capacity building where appropriate
- Curators to respond to helpful suggestions to improve operations
- Curators to make comments to improve the system

c. Recommendations on 'generic' technical (genebank management) standards, ie for the Fruit Network

The process described above and the standards presented below should with minor modifications be appropriate for *Malus/Pyrus* and *Vitis* as well as other *Prunus* species.

d. Recommendations on crop-specific technical standards

The AEGIS *Prunus* group considered the "Information required for Field Genebanks" document that had been circulated by the AEGIS secretariat. For various reasons this model did not appear to be suitable. The group developed the following standards from the guidelines that were approved by the *Prunus* WG in Larnaca in December 2005.

Prunus (cherry) technical standards for AEGIS

Minimum standards proposed by AEGIS Prunus group for endorsement by the Prunus WG

Receipt and propagation of new material.

- New accessions should be received with minimum passport data (agreed by *Prunus* WG) and with phytosanitary documents and Material Transfer Agreements where appropriate.
- New accessions should be added to genebank register and given an accession number.
- During propagation care should be taken to ensure correct labelling and to use virus-tested compatible rootstocks.
- Accession passport data should be provided to EPDB manager when trees are planted in the field collection or greenhouse.

Maintenance.

- Four trees of each accession should be maintained, two at the primary genebank and two at the reserve genebank.
- Isolation of AEGIS accessions is not obligatory.
- The planting site should be suitable with respect to soil and exposure and security.
- The trees should be adequately labelled and/or a plot plan maintained.
- The trees should be managed well enough to provide characterization data and scion wood for distribution and/or re-propagation.
- The trees should be inspected visually and/or tested immunologically/molecularly for freedom from quarantine pests and diseases.
- The trees should be protected against quarantine pests and diseases.

Re-propagation.

- When trees are re-propagated, care should be taken to ensure correct labelling.
- For tree propagation, virus-tested compatible rootstocks should be used.

• New plants should be authenticated by traceability procedures or by morphological inspection or by molecular fingerprinting.

Despatch and disposal.

- Two sticks per accession should be provided in response to reasonable requests.
- Propagating material should be despatched with labels and minimum passport data (agreed by *Prunus* WG), together with appropriate phytosanitary paperwork and, if necessary, a Material Transfer Agreement
- Before any AEGIS accessions are grubbed, two years' notice should be given to the European *Prunus* Database manager or the National Coordinator or the AEGIS Secretariat (to be decided).

Characterization (if funded).

- Data sets should be completed using characterization descriptors of the *Prunus* WG.
- Molecular fingerprints should be obtained using molecular markers approved by the SSR workshop held at East Malling in December 2006.
- For each accession, a photograph of the fruits should be obtained, using the protocol recently approved by the Synonymy workshop held in Gembloux in June 2008).
- Characterization data should be supplied to EPDB manager for inclusion in EPDB.

e. General comments and observations

It is highly desirable that all participating genebanks in AEGIS should follow these standards, but minor deviations could be approved in consultation with the WG.

5. Observations on the framework and tool for the assessment of operational costs for collection maintenance

The AEGIS group discussed the practicalities of cost-benefit analysis of field genebank collections. Whereas dedicated genebanks may be able to provide annual cost per accession, working genebanks may find it difficult to provide such figures because of split projects and NGO collections may be impossible to cost because of their inputs of uncosted voluntary work.

In addition, the group highlighted the additional economic considerations important for implementing AEGIS as shown below.

Activity	By whom?
1. Complete minimum passport data in EPDB	Curators and DB manager
2. Choose genotypes for AEGIS	DB manager?
3. Choose MAAs (primary + reserve if available)	Cherry committee
4. Propagate safety duplicates where necessary and plant them (where?)	Curators, DBM
5. Check the identity of primary and reserve accessions: ie photo, morphological, molecular	Relevant curators
6. Check health status of chosen accessions and notify DBM	Relevant curators, DBM
7. Update AEGIS MAAs list and acquire new candidates	DBM, relevant curators
8. Implement realistic AQUAS in relevant genebanks	Relevant curators
9. Distribute material in response to requests	Relevant curators

Additional activities that will incur costs include meetings (inauguration, 1 year review, 3 year review), publications, newsletter, promotion and seeking funding.

The group also discussed the cost savings and additional costs that would result from the implementation of AEGIS. The cost savings included reduced duplication, eventually. Additional costs included implementation costs, mentioned above, and maintenance of the AEGIS system. Improved quality would represent better value for money.

6. Proposal on the involvement of all the relevant stakeholders of the European Region in establishing and operating the European Collection for *Prunus* (including services to be provided, rationalization aspects and coordination)

The AEGIS group suggested that the National Coordinators should be responsible for coordinating participation of the genebanks (public and NGOs) in their country and encouraging further involvements. It is also important to promote AEGIS to potential users by appropriate publicity.

7. Proposed "general workplan", whenever possible costed, for the model crop *Prunus* Working Group activities

The following workplan is envisaged for the implementation of AEGIS and could be included in the workplan of the *Prunus* WG for Phase VIII.

Action	Carried out by	Estimated cost
Meetings		
Inauguration meeting	Participating curators and AEGIS group	25,000€
1-year review meeting	Participating curators and AEGIS group	25,000€
3-year review meeting	Participating curators and AEGIS group	25,000€
Data enhancement		
Passport, morphological data and	Participating curators	10€ per accession + 1,500€
photos for about 1850 MAAs		for other needs (20,000€)
Fingerprinting of about 1850	Several laboratories	25€per accession (46,250€)
MAAs		
Health status monitoring	Several laboratories	25€per accession (46,250€)
Safety duplication of 1200 accessions		
Propagation of safety duplicates	Curators or nurseries	7€per accession (8,400€)
Establishing orchard (2x1200	Curators	30 to 50€ per accession
trees) probably at a single site		(48,000€approx.)
Publicity		
Publications and newsletter		5,000€
Promotion (12 countries)		6,000€

Estimated total amount: 254,900€

Janos Apostol (Hungary), Emilie Balsemin (EPDB manager, France), Daniela Giovannini (Italy), Kenneth Tobutt (UK) July 2008

References

Responses of the ECP/GR Working Group on *Prunus* regarding AEGIS Activities 4.1-4.5 and 4.7 and 4.9 Maggioni L., Lipman E., 2005. Report of a Working Group on *Prunus*. Seventh Meeting, Larnaca, Cyprus, 1-3 December 2005, 137 p.

Maggioni L., Lipman E., 2006. Report of the ECP/GR Network Coordinating Groups, Bonn, Germany, 29-31 March 2006, 84 p.

Sharing of long-term conservation responsibilities as a possible model for "A European Genebank Integration System" (AEGIS)

Draft selection criteria for the identification of Most Appropriate Accessions to be included in the European Collection (AEGIS).