

Proposal for the identification of 500 old potato clones having unreliable variety name by means of fingerprinting using 12 microsatellite (SSR) markers to assist in setting up the AEGIS collection for potato cultivars.

### **1. Problem statement**

The true identity (cultivar name) of potato clones in different collections is not always clear or correct. This is hampering the selection of the Most Appropriate Accessions (MAA's) for the AEGIS collection.

- In particular for old potato cultivars the clone can be mislabelled, as reported by H. Campbell from SASA (Frese & Hoekstra, 2009).
- Some variety names have been used more than once (e.g. Gloria 1921, 1937, 1972) and it is not always known to the curator what the true identity of the clone in his collection is.
- Based on SSR data, K. Dehmer (IPK, Germany) found for old blue/purple fleshed potato varieties that different names may be synonyms for the same clone.

*Summary provided by K. Dehmer: a set of 15 SSR markers was applied onto 26 blue fleshed accessions of the IPK Genebank. Only seven different SSR patterns/genotypes were identified. Four unique genotypes were represented by one GLKS accession each, while the other three genotypes were attributed to three duplication groups consisting of thirteen, five and four GLKS accessions, respectively.*

### **2. Justification and rationale**

The selection of the Most Appropriate Accessions (MAA's) by the ECPGR Potato Working Group for the AEGIS collection will be based on the passport data provided by the collection holders. Correct data on the identity of the individual clones and knowledge about synonymy are crucial for this process. This project will provide accurate identifications for clones having questionable name labels (of potato germplasm selected from several European collections).

### **3. Background**

In particular clones of presumably old potato cultivars can be mislabelled. This may be caused by incorrect information from the germplasm donor, or errors/interchanges made in following maintenance years. The classical differentiation of cultivars based on morphological characteristics is a highly skilled and time-consuming task.

To assist in granting Plant Breeders' Rights for new potato varieties, a standard fingerprinting method has been developed (Reid & Kerr, 2007; Reid et al. 2009). It is a rapid and robust method for variety differentiation using nine microsatellite (SSR) markers. Over 1,000 cultivars have successfully been differentiated so far. Obviously, somaclonal variants and mutants cannot be separated from the original cultivar. The set of markers was expanded to twelve to give an added level of discrimination. All potato varieties maintained by SASA have been fingerprinted. SASA's potato SSR profile database is currently not public. This well established method will be applied for this AEGIS project.

The ECPGR European Potato Cultivar Database [EPCD] ([www.europotato.org](http://www.europotato.org)) currently lists information on 5,264 (presumable) different clones (incl. 4,000 cultivars as well as 159,000 observations) provided by 51 contributors. Those variety names that were used more than once for different genotypes (e.g. Gloria), are listed with year of release. When the identity is unclear then the abbreviation of the data donor is included in the name label.

*The Multi-Crop Passport Descriptor list (MCPD) as well as EURISCO do not include a field for year of first release of varieties. In 2010 GRIN adapted the format of the downloadable passport data to provide this information. Obviously it is included in the EPCD.*

#### **4. Main objective and specific objectives**

Establishment of the AEGIS collection for a part of the potato cultivars by means of fingerprinting old potato clones with questionable identity, to confirm or correct cultivar names.

#### **5. Materials and methods**

Material: 500 clones from different European collections ( $\neq$  SASA). Background knowledge: the SASA potato SSR database containing profiles of >1000 cultivars. Method: fingerprinting by means of 12 microsatellite (SSR) markers used in four multiplex reactions. Twelve markers (for 500 clones) will be used, giving the best discrimination. The use of the bare minimum of 9 markers (for 600 clones) will not be considered now additional funding for this fingerprinting work has become available from ECPGR, after a reallocation of funds from the Potato WG. This allows to increase the number of fingerprinted clones from 200 to 500.

*Commercial labs offered to test a higher number of samples, but then a harmonization exercise would have been necessary, to allow the results to be compatible with those in the SASA database of potato SSR profiles, which can be a fairly drawn out and expensive process.*

#### **6. Expected outputs**

The initial product of this project will be fingerprints of old potato clones from different European potato collections. Comparison with the SASA's extensive database on potato cultivar SSR profiles will identify mislabelling or confirm the genetic uniqueness of the clone, when no match with the database was found.

#### **7. Benefits and impact**

These results will significantly benefit the selection of potato MAA's for AEGIS. Curators will get essential information about the identity (and uniqueness) of the investigated germplasm. For the fingerprinted varieties a European Potato Collection will be proposed for AEGIS, indicating which accessions would be the Most Appropriate for inclusion.

## 8. Innovation

New are the fingerprints of previously not investigated genotypes (old varieties), which will be added to the database. Presumable parentage and offspring can be checked, when available in the database. The resulting SSR profiles from this project will be made public.

## 9. Application of results

Using the fingerprints, identities of old cultivars will be checked. The current identity will be re-identified, confirmed or recognized as a unique genotype when no match was found. Furthermore, new synonyms (or mutants) may be discovered, genetic distances can be calculated and presumable parentage and offspring may be checked, when available in the database. Last but not least: the selection of MAA's for AEGIS will be supported significantly and for the fingerprinted varieties a European Potato Collection will be proposed, indicating which accessions would be the Most Appropriate for inclusion.

## 10. Workplan

	Month 1-2	Month 3-6	Month 7-8	Month 9-10	Month 11-12
CGN (RH) and SASA (HC) selecting clones based on the ECPGR potato database and indications from curators	X				
Several curators (depending on the selection made in month 1-2) picking and drying leafs and sending samples to SASA		X			
SASA fingerprinting 500 clones			X		
SASA (AR) analysis and communication of results				X	
CGN (RH) establishment of Eur. Potato Collection for AEGIS for the fingerprinted varieties					X

## 11. Budget

The requested budget will primarily be used for DNA extraction and the fingerprinting work.

	project	in kind	Total
CGN staff time	1,000	1,000	2,000
potato curators staff time		1,000	1,000
curators - supplies	800		800
SASA staff time	16,000	1,000	17,000
“ lab supplies	9,000		9,000
<b>Total</b>	<b>26,800</b>	<b>3,000</b>	<b>29,800</b>

## 12. Contributions offered by applicant

- CGN (R. Hoekstra) and SASA (H. Campbell) will check the ECPGR potato cultivar database ([www.europotato.org](http://www.europotato.org)), request lists of questionable cultivar identities from curators and select 500 accessions (clones) from different European potato collection holders [non from SASA, because its collection is already fully fingerprinted, nor CGN (maintains no cultivars)].
- Curators from different European potato collections (e.g. IPK, INRA and others) will communicate the questionable cultivar identities within their collection, collect leaves from the ultimately selected clones, dry them on silica gel and send the dried leaf samples to SASA.
- Beside DNA extraction from the leaf samples and performing the routine fingerprinting using 12 SSR markers (100% project funded), A. Reid (SASA) will compare the fingerprinting results with the extensive SSR profile database and draw conclusions on the identity of the germplasm. SASA & CGN will inform the donors of the samples as well as the ECPGR potato cultivar database manager and make the resulting SSR profiles public.
- CGN (R. Hoekstra) will identify the proposed European Potato Collection for AEGIS concerning the fingerprinted varieties.

## 13. Bibliography

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