UPOV

International Union for the Protection of New Varieties of Plants

E

Working Group on Biochemical and Molecular Techniques and DNA-Profiling in Particular

BMT/17/12

Seventeenth Session
Montevideo, Uruguay, September 10 to 13, 2018

Original: English
Date: August 31, 2018

CONSTRUCTION OF A EUROPEAN POTATO DATABASE WITH VARIETIES OF COMMON KNOWLEDGE AND ITS IMPLEMENTATION IN THE POTATO DUS TESTING SYSTEM PART II: GENERATION OF MOLECULAR DATA*

Document prepared by experts from the United Kingdom and the Netherlands

1. In 2004, the Centre for Genetic Resources/Plant Research International (CGN/PRI) and SASA collaborated to produce, from the public domain, a harmonized set of 9 SSR markers for potato variety differentiation which yield results that are both robust and easy to interpret. This set of 9 markers has been used for the European Potato Database since 2006 (see documents BMT-TWA/POTATO/1/4, BMT/10/5, BMT/11/9 and BMT/11/10 for background). During the initial stages both laboratories carrying out the molecular work used a similar capillary based platform making harmonization 'relatively' easy. The first European potato project ran from 2006 to 2008 and typed around 900 varieties in the EU Common Catalogue. A few years elapsed between the end of this first project and two subsequent projects. During this time, the SSR analysis of potato varieties in the Netherlands moved to Naktuinbouw and was set up using a different gel based platform. Between 2008 and 2011, the United Kingdom and the Netherlands both continued to independently add varieties to their database and in 2012, a follow up project was arranged to update the database with varieties added to the Common Catalogue by all of the original project partners. A second project (an European Potato database as centralized collection of varieties of common knowledge) ran between 2014 to 2015 and included all 9 entrusted Examination Offices for potato in the EU. The third and final project followed on from this (Construction of a European Potato database with varieties of common knowledge and its implementation in the potato DUS testing system). During these subsequent projects, the United Kingdom and the Netherlands laboratories had to undertake additional harmonization due to the differing platforms used in the two labs.

2. The experience of the two labs is that reproducibility is generally high but that discrepancies can occur between the labs and that these generally fall into five categories.

Category 1. There is a new allele (for example, the allele-bin for 2028 F, actually contained 2 separate alleles now called F and K). These differences are normally resolved fairly easily by the labs agreeing on the presence of a new allele. – **3 %**

Category 2. An allele was miscalled by one lab (can either be missed completely or assigned an incorrect letter). These are normally resolved fairly easily by both labs checking their data and agreeing on the correct call. – **44 %**

Category 3. There is a genuine difference in the profiles obtained by the two labs. Fortunately a rare occurrence, usually the result of a sample with poor DNA quality that does not amplify well (2017/PL-006 was such a sample but between the two labs and two tubers it was possible to obtain a complete profile for this variety). Alternatively, this can be caused by a mixture of varieties, this is very easy to spot as the profiles are very different. There were only two instances of this in 753 candidate samples and these data have not been counted for this analysis.

* Part I "Construction, maintenance and use of the common database", prepared by an expert from Germany, is presented in document BMT/17/11

Category 4. An allele is called as questionable by one lab (the lab cannot decide whether to call an allele or not) and IS called by the other lab. – **17 %**

Category 5. An allele is called as questionable by one lab (the lab cannot decide whether to call an allele or not) and is NOT called by the other lab. – **36 %**

3. A considerable number of discrepancies were observed as shown in the following table. Better harmonization was reached since 2013 but there are still 20-25% of samples where clarification between the labs is necessary, at least for one maker.

Year	# samples	# discrepancies	discrepancy %
2013	121	74	61.2
2014	208	82	39.4
2015	156	32	20.5
2016	147	36	24.5
2017	121	27	22.3
2018	116	16	13.8
Total	869	267	30.7

Discrepancies were not linked to specific markers or to specific alleles. Discrepancies of category 1, 2 and 3 are critical and have an effect on the calculation of the similarity values. Errors of category 4 and 5 are less critical. Both labs consider a category 4 discrepancy as not being a problem as the lab calling the allele as questionable thinks that there might be an allele present but it falls slightly below a predetermined threshold in the analysis software and the other lab definitely calls the allele as present. However, category 5 is more worrying as one lab thinks there might be an allele present but the other lab does not.

Both labs have developed a set of decision rules. These are used as guidance in the interpretation and scoring of the alleles. Labs are aware from experience which alleles are more reliable on the ABI capillary system that is used by SASA and which of the alleles are always more clear to score on the LICOR gel-based system used by Naktuinbouw. There is not a better or more preferred machine/system. Both systems contribute equally to the accuracy of the DNA profiles.

[End of document]