



BMT/15/22 Rev.
ORIGINAL: English
DATE: June 27, 2016

INTERNATIONAL UNION FOR THE PROTECTION OF NEW VARIETIES OF PLANTS
Geneva

**WORKING GROUP ON BIOCHEMICAL AND MOLECULAR
TECHNIQUES AND DNA PROFILING IN PARTICULAR**

Fifteenth Session

Moscow, Russian Federation, May 24 to 27, 2016

CAN MOLECULAR DISTANCE BE USED AS A CHARACTERISTIC?


Document prepared by an expert from the Netherlands

Disclaimer: this document does not represent UPOV policies or guidance

The Annex to this document contains a copy of a presentation “Can molecular distance be used as a characteristic?” made at its fifteenth session of the Working Group on Biochemical and Molecular Techniques and DNA-Profiling in particular (BMT).


Kees van Ettehoven, Head Variety testing Department, Naktuinbouw

[Annex follows]



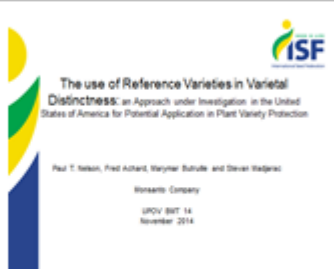
Can molecular distance be used as a characteristic?

June 2016
Kees van Ettehoven
Head Variety testing Department
Naktuinbouw



History

In the BMT meeting of 2014, USA colleagues gave a presentation outlining the possibility to use Reference varieties in varietal distinctness.



History

- Using a geographic analogy, the genetic distance of varieties to a set of standards was used to find a way to communicate on genetic information.
- Further work ongoing in USA



Attempt to transform this approach

- Can we use the USA example in a form that answers to the usual UPOV approach using characteristics and states of expressions to establish distinctness and identify and varieties.

Orchids as model crop

- NL biggest market worldwide for flowers and ornamentals
- Important trade-hub
- Total turn over 2014€ 4,5 billion
 - Cut flowers: € 2,8 billion
 - Plants: € 1,6 billion
 - Garden plants: € 0,3 billion



Ministry of Economic Affairs

Orchids as pot plants

Top trade pot plants in NL:

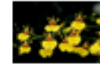
- 1) **Phalaenopsis** € 500 million
- 2) Kalanchoe € 60 million
- 3) Rose € 56 million
- 4) Anthurium € 51
- 5) Chrysanthemum € 36 million
-
- 13) **different orchids** € 22 million



Ministry of Economic Affairs

Orchids as pot plants

- *Phalaenopsis* # one since 2001
- Yearly increase 25-30%, in 2010 10%, 400% in 6 years
- Production still increasing, but is slowing down
- From exclusive and old-fashioned to trendy and modern
- Success factors: keeping quality, flower quality & quantity, easy handling
- Product innovation still going on (*Dendrobium*, *Cymbidium*, *Miltonia*, *Oncidium*, *Paphiopedilum*, *Burrageara*, *Cattleya*, *Vuylistekeria*)



EU-TW PVP Symposium

Ministry of Economic Affairs

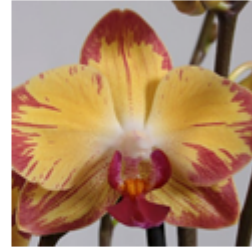
Specifics of orchid testing

- Economically important crop.
- Vegetative crop, multiplication by tissue culture.
- Breeding (crossing) is very easy.
- Breeding centers in Asia but also in USA and the Netherlands.
- Complicated flower with many flower characteristics.



Specifics of orchid testing

- Photographs alone are not sufficient.
- Living reference collection needed.
- Comparison between old references and new material difficult.
- Panels of experts needed.
- Risk to grant right on varieties that are already common knowledge in other parts of the world.



Molecular techniques and orchids

To overcome a number of difficulties, a database with DNA information was created.

Main aim of this database:

- to minimize the risk to grant PBR on existing varieties.
 - to help in the management of the reference collection.
 - to check stability when renewing reference material.
 - to avoid sending plant material over the world.
- Plus fast reference for suspected infringement.

From data to characteristics

- Molecular data from the existing database were used to design a model using the genetic distance between applications and a set of standard varieties

Similarity to states of expression

From similarity (Jaccard) distance to state of expression

	Ref number	Breeder's ref	Type	OPS A		OPS B		OPS C	
				Similarity	score	Similarity	score	Similarity	score
	OP00000		166124 White medium	50.00	5.26.29	644.82	5		
	STANDARD		145124 White medium	22.52	2.2.27	922.52	7		
	OP00000		166424 White large	21.25	7.24.48	640.00	6		
	OP571		120224 White large	21.25	7.25.61	716.07	6		
	OP581		120644 White large	21.62	6.27.27	715.00	7		
	STANDARD	Phalinea	White large	24.27	6.29.02	724.27	6		
	STANDARD	PHALFLACAN	White spotted	15.79	6.12.89	612.82	8		
	OP542		115021 White spotted	12.51	6.5.41	67.69	9		
	STANDARD	PHALDOTWEL	Yellow	16.92	6.7.89	622.22	8		
	OP00092	phaloudac	Yellow	6.22	6.11.50	614.71	8		
	STANDARD	PHALCATEM	Yellow	6.57	6.12.90	611.77	8		
	OP00094	phalorafom	Yellow	17.14	6.16.75	612.89	8		
	STANDARD	phalobando	white violet	27.27	7.14.71	622.52	7		
	OP5272	phalbasfos	white violet	19.45	6.14.29	612.16	8		
	STANDARD	PHALIFSIG	Hankeljn	17.95	6.26.47	724.22	7		
	OP00077	Ganlin Pearl	Hankeljn	17.65	6.6.82	614.29	8		
	OP51110		277521 Hankeljn	10.91	6.6.82	615.26	9		
	OP00087	Pikacchi	Purple red	15.79	6.12.89	615.79	8		
	STANDARD	Phalobas	Purple red	26.18	4.20.00	621.87	8		

Characteristics in UPOV format

3 applications

OPS A			OPS B			OPS C		
Char 75.1	Genetic distance to 14134	7 long	Char 75.1	Genetic distance to 14134	9	Char 75.1	Genetic distance to 14134	7
Char 75.2	Genetic distance to Phalcinco	6 medium to long	Char 75.2	Genetic distance to Phalcinco	7	Char 75.2	Genetic distance to Phalcinco	6
Char 75.3	Genetic distance to Phallican	6 long to very long	Char 75.3	Genetic distance to Phallican	6	Char 75.3	Genetic distance to Phallican	6
Char 75.4	Genetic distance to Phalidoowl	6 long to very long	Char 75.4	Genetic distance to Phalidoowl	9	Char 75.4	Genetic distance to Phalidoowl	6
Char 75.5	Genetic distance to Phalcatom	9 very long	Char 75.5	Genetic distance to Phalcatom	6	Char 75.5	Genetic distance to Phalcatom	6
Char 75.6	Genetic distance to Phalobando	7 long	Char 75.6	Genetic distance to Phalobando	6	Char 75.6	Genetic distance to Phalobando	7
Char 75.7	Genetic distance to Phalifib	6 long to very long	Char 75.7	Genetic distance to Phalifib	7	Char 75.7	Genetic distance to Phalifib	7
Char 75.8	Genetic distance to Phalcosap	4 short to medium	Char 75.8	Genetic distance to Phalcosap	6	Char 75.8	Genetic distance to Phalcosap	9

Comparison between the 3 applications

- Application of the normal UPOV rules possible?

		OPS A	OPS B	OPS C
Char 75.1	Genetic distance to 14134	7	9	7
Char 75.2	Genetic distance to Phalcinco	6	7	6
Char 75.3	Genetic distance to Phallican	6	6	6
Char 75.4	Genetic distance to Phalidoowl	6	9	6
Char 75.5	Genetic distance to Phalcatom	9	6	6
Char 75.6	Genetic distance to Phalobando	7	6	7
Char 75.7	Genetic distance to Phalifib	6	7	7
Char 75.8	Genetic distance to Phalcosap	4	6	9



Suitable as characteristic?

- UPOV TG/1/3
- 2.4.2 The 1991 Act of the UPOV Convention makes this clear by stating in Article 1(vi) that a variety is a plant grouping that can be “defined by the expression of the characteristics resulting from a given genotype or combination of genotypes” and can be “distinguished from any other plant grouping by the expression of at least one of the said characteristics.”



Suitable as characteristic?

- Can the genetic distance in itself be considered as the expression of a given genotype or more appropriate a combination of a number of genotypes?



Type of characteristic?

- Genetic distance to a certain standard with the states 1 absent to very short 3 short, 5 medium, 7 long, 9 very long can be considered as QN, MG and should have a (+) with a clear explanation of the method in chapter 8.



Usefulness of such characteristics

- Use of this approach offers the opportunity to exchange DNA information between examination offices through the variety description in a meaningful way, without exchanging the actual DNA information.
- DNA characteristics are more independent from environment, observers etc. than morphological characteristics
- No need to develop databases for this approach.
- Cost effective



Usefulness cont.

- This approach makes the examination offices less dependant on the (few) examination offices that have access to software needed to run similarity tests.
- Possible downsides
Standards may lead to already suspected groups (varieties of same breeder)
Harmonisation of method possible?



Matters to settle

- Discussion on the principles
- Choice of standard varieties per species
- Further tests to check in other species