



**BMT/9/12 Add.**

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**INTERNATIONAL UNION FOR THE PROTECTION OF NEW VARIETIES OF PLANTS**  
GENEVA

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

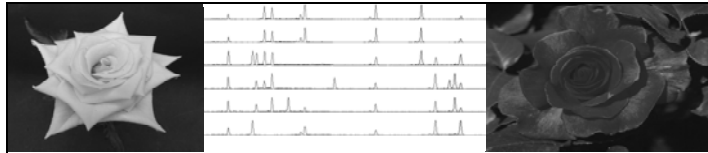
**Ninth Session**

**Washington, D.C., June 21 to 23, 2005**

**ANALYSIS OF A DATABASE OF DNA PROFILES OF  
734 HYBRID TEA ROSE (*ROSA HYBRIDA*) VARIETIES**

*Document prepared by experts from the Netherlands*

1. The BMT agreed that, where agreed by the relevant experts, the presentations made at the meeting should be made available in the BMT document section of the UPOV website, as addenda to the relevant documents. This document contains a copy of the presentation given by Mrs. Lysbeth Hof (Netherlands), based on document BMT/9/12, and prepared by Mr. Ben Vosman (Netherlands).



## Analysis of a database of DNA profiles of 734 hybrid tea rose (*Rosa hybrida*) varieties

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& Ben Vosman



### Rose

- Most important ornamental crop
- More than 25,000 varieties of modern rose (Cairns, 2000)
- More than 10,000 hybrid tea varieties
- Rose list 2002: 13,000 varieties in commercial trade
- Large collection of roses in “common knowledge”

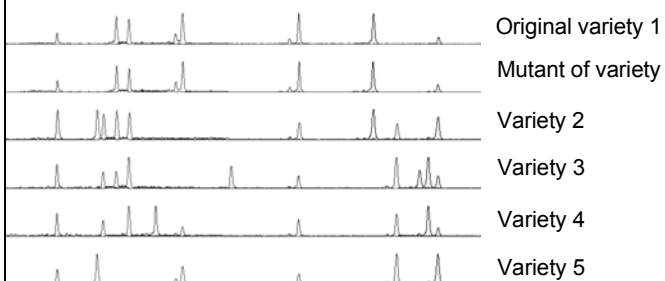


### The markers:

- Large set of microsatellite markers developed
- 24 markers described (TAG (2003) 106: 277-286)
- For this study 11 markers used (in 4 assays)
- Data on 734 varieties, including duplicates



### Microsatellite analysis of rose (3 loci)



### Topics:

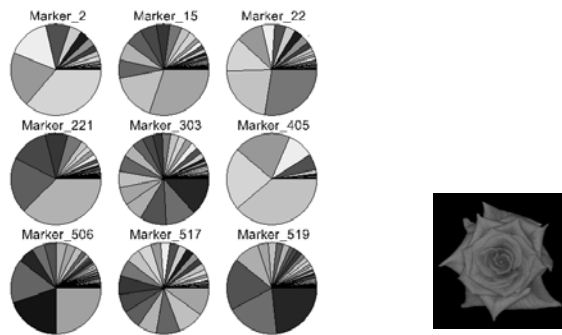
- Discriminative power of the markers
- Reproducibility of the results
- Correlation between molecular and DUS characteristics (option 2 approach).

### Marker characteristics:

Locus	Number of alleles	Number of allelic phenotypes	PIC value based on allelic phenotypes	Frequency of most common allelic phenotype	Number of different alleles in allelic phenotype with highest frequency
RhAB15	6	28	0.72	0.29	2
RhAB201	4	15	0.67	0.23	2
RhAB22	7	23	0.52	0.31	2
RhAB40	9	79	0.76	0.19	2
RhB303	6	37	0.76	0.12	3
RhD221	6	32	0.67	0.31	2
RhE2b	7	32	0.54	0.37	1
RhEO506	6	34	0.72	0.20	2
RhM405	4	9	0.73	0.4	4
RhO517	5	27	0.77	0.12	3
RhP519	6	32	0.71	0.22	3

Based on 407 different varieties

### Occurrence of allelic phenotypes:



### Discriminative power:

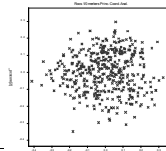
- All seedling varieties had a unique DNA profile
- Pairwise genetic similarities (Jaccard) of seedling varieties was < 0.9
- Mutants had a genetic similarity of 1 with original variety

### Reliability of the database:

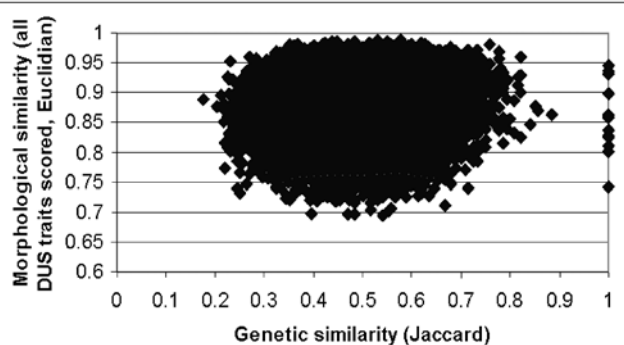
- Number of assays that need to be repeated
  - Without multiplexing in PCR 15 - 18%
  - Multiplexed PCR 3 - 4%
- Error rate 1/1000 alleles  
(based on duplicated samples and mutants)

### Genetic structure within the set of varieties

- Analysed using Fst (population differentiation)
  - No structure when varieties submitted within one year are considered a population ( $F_{st}=0.0007 \pm 0.0005$ )
  - No structure when varieties submitted by a breeder are considered a population ( $F_{st}= 0.0056 \pm 0.0011$ )
  - PCA also shows no structure
- Conclusion: All breeders basically use the same gene pool
- No substructure



### Genetic versus overall morphological



### Correlation with flower color

- Most important distinguishing trait
- UPOV color grouping (classes 1-19, 34, 40, 46-47, 50)
- Question we considered:
  - Does a higher genetic similarity between two varieties increase the probability that these varieties are in the same color group (have the same color)?

### Correlation genetic similarity and flower color class

Bold = significantly higher  
than the background level  
(Mantel test,  $p < 0.001$ ).

### Conclusions:

- Markers show a high discriminative power
  - All seedling varieties can be uniquely identified
  - Mutants are identical to original variety
- Reliability of the data stored in the database is high
  - Error rate 1/1000 alleles
  - Multiplexing reduces error
- No correlation between genetic similarities based on morphological and molecular characters
  - an option 2 approach is not realistic for rose

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