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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**

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ADDENDUM TO DOCUMENT BMT/14/5

THE USE OF REFERENCE VARIETIES IN VARIETAL DISTINCTNESS: AN APPROACH UNDER  
INVESTIGATION IN THE UNITED STATES OF AMERICA FOR POTENTIAL APPLICATION IN PLANT  
VARIETY PROTECTION

*Document prepared by experts from Monsanto Company*

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The Annex to this document contains a copy of a presentation "The Use of Reference Varieties in Varietal Distinctness: an Approach under Investigation in the United States of America for Potential Application in Plant Variety Protection" made at its fourteenth session of the Working Group on Biochemical and Molecular Techniques and DNS-Profiling in particular (BMT).

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[Annex follows]



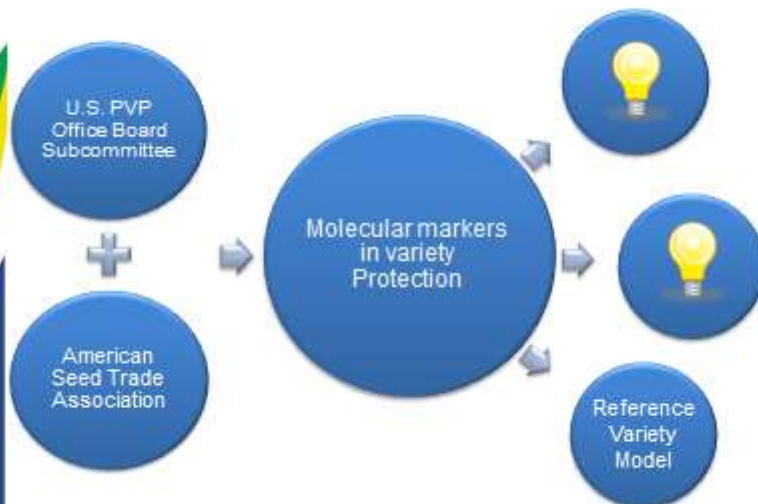
# The use of Reference Varieties in Varietal Distinctness: an Approach under Investigation in the United States of America for Potential Application in Plant Variety Protection

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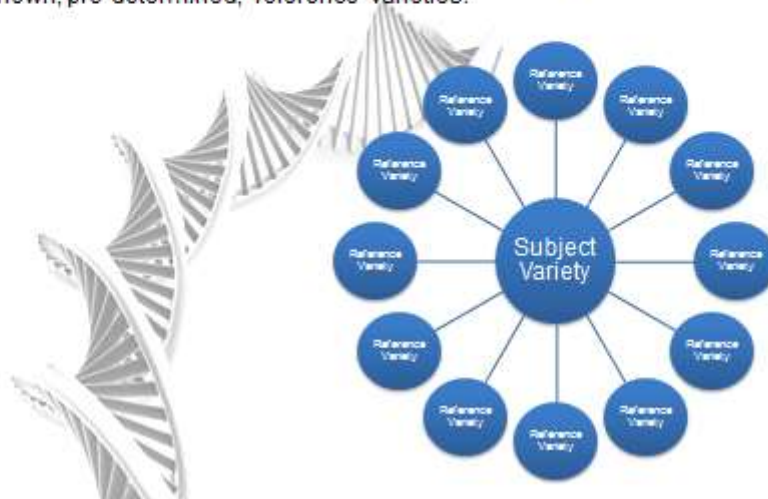
UPOV BMT 14  
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## Coordinated Effort



## Reference Variety Model

Utilize genetic relationship between PVP candidate "subject" varieties and known, pre-determined, "reference" varieties.



## Geographic Analogy



The earth's geographic coordinate system allows for precise placement of every location on earth.

U.S. Cities, Albany, Harrisburg, and Syracuse can be distinguished geographically by their coordinates, latitude and longitude:

<u>City</u>	<u>Latitude</u>	<u>Longitude</u>
Albany	42.6525	-73.757222
Harrisburg	40.28972	-76.875556
Syracuse	43.04694	-75.144444

## Geographic Analogy



Another way to distinguish between, Albany, Harrisburg, and Syracuse is by measuring their distances from well-known 'reference' cities:

Subject City	Boston	New York	Philadelphia	Rochester	Pittsburgh
Albany	223	217	323	319	575
Harrisburg	539	248	151	327	264
Syracuse	423	314	354	120	432

(Kilometers)

Albany and Harrisburg are very similar in their distance from Rochester, but are distinguished from each other by their distances from the other cities.

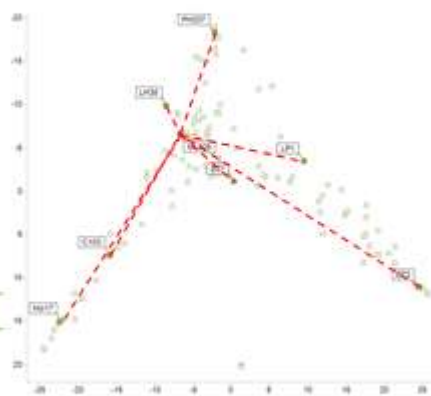
## Distance Application to Genotypes

Varieties do not have fixed "coordinates".

However, genetic distances can be easily computed.

Genetic similarities coefficients, with a known set of 'reference' varieties, can be used to distinguish between genotypes.

Subject Variety	B37	B73	C103	LH33	LP1NRHt	Mo17	PH207
NK764	0.474	0.704	0.395	0.397	0.594	0.368	0.486
DK4676A	0.469	0.459	0.372	0.405	0.497	0.376	0.413
ML606	0.484	0.426	0.438	0.732	0.428	0.483	0.484
NK740	0.441	0.341	0.655	0.485	0.400	0.890	0.433
LH132	0.542	0.838	0.371	0.418	0.504	0.355	0.485
NQ208	0.549	0.444	0.428	0.462	0.479	0.445	0.843
PH777	0.425	0.409	0.437	0.464	0.412	0.445	0.401



## Distance Application to Genotypes

Genetic similarity coefficients fit nicely into the existing PVP morphological framework.

Each reference variety is treated as a new 'trait'.

PVA	Variety Name	Kernel Type	Days to 50% silk	Heat to 50% silk	Plant Height	Ear Height	Inferred cob length	No. of Tillers	No. of Ears	557 Sim	575 Sim	C105 Sim	LP1 Sim	Mo17 Sim	PH207 Sim
8400057	PH071	DNT	066	151.0	166	064	006	0	1.25	0.485	0.487	0.406	0.595	0.597	0.657
8000011	PH19	DNT	066	141.1	175	061	012	0	1.25	0.450	0.442	0.451	0.594	0.425	0.445
8000067	LH59	DNT	065	136.0	203.5	76.4	01.0	0	2.54	0.487	0.401	0.455	0.455	0.435	0.469
7600047	LH1	DNT	069	162.5	176	05.6	01.1	0	1	0.752	0.467	0.592	0.469	0.422	0.545
8000066	LH56	DNT	062	155.6	160.2	49.2	01.5	0	1.75	0.467	0.559	0.455	0.405	0.441	0.455
7600019	LP1	DNT	069	152.0	215	10.6	01.2	1.5	1.25	0.445	0.515	0.424	1.000	0.595	0.456
7900077	Seagull-17	DNT	064	165.0	17.0	07.0	01.2	0	1	0.444	0.551	0.630	0.406	0.965	0.455
8200062	LH51	DNT	067	146.5	207	06.4	01.5	0	1	0.462	0.551	0.445	0.597	0.912	0.447
8200065	LH74	DNT	061	154.4	169	07.2	01.0	1.5	1.75	0.470	0.724	0.575	0.666	0.565	0.446
8200064	LH19	DNT	062	154.4	22.0	06.7	01.2	0	1.25	0.556	0.665	0.566	0.495	0.554	0.474
8200002	LH145	DNT	060	129.4	207	06.2	01.5	0	1.25	0.449	0.479	0.449	0.705	0.409	0.449

## Identification of Reference Varieties

Reference varieties must be relevant to the pool of subject varieties under consideration.

Subject City	Boston	New York	Philadelphia	Rochester	Pittsburgh	Sacramento	Fresno
Albany	225	217	525	319	575	4005	3946
Harrisburg	559	345	151	327	264	5799	5719
Syracuse	425	514	554	120	452	3806	3747
Los Angeles	4174	3940	3647	3635	3455	552	550
San Francisco	4335	4135	4055	3795	3641	120	261





## Model Limitations


- Data reduction = loss of information.
- The reference variety model will sometimes fail to distinguish between genetically distinct varieties.
  1. Fingerprints can be used to make direct variety comparisons.
  2. Morphology remains the primary means of determining distinctness by the PVPO.



## Model Features

1. Simplicity in theory and computation.
2. Breeders maintain control of fingerprints.
3. The U.S. PVPO does not need to store, maintain, or safeguard fingerprints.
4. The model fits nicely into the existing PVP framework.
5. Genetic similarity coefficients do not replace existing morphological descriptors.

## Ongoing Efforts

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1. Reference variety identification.
  2. Definition of thresholds for distinctness.
  3. Application across crops.

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