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UPOV

INTERNATIONAL UNION FOR THE PROTECTION OF NEW VARIETIES OF PLANTS
GENEVA

WORKING GROUP ON BIOCHEMICAL AND MOLECULAR TECHNIQUES, AND DNA-PROFILING IN PARTICULAR

Thirteenth Session
Brasilia, November 22 to 24, 2011

ADDENDUM

MANAGEMENT OF THE PEACH TREE REFERENCE COLLECTIONS

Document prepared by experts from France, Hungary, Spain and Italy

Management of Peach Tree Reference Collections

CPV.8648 Research & Development Project

Part 1: Bio molecular results

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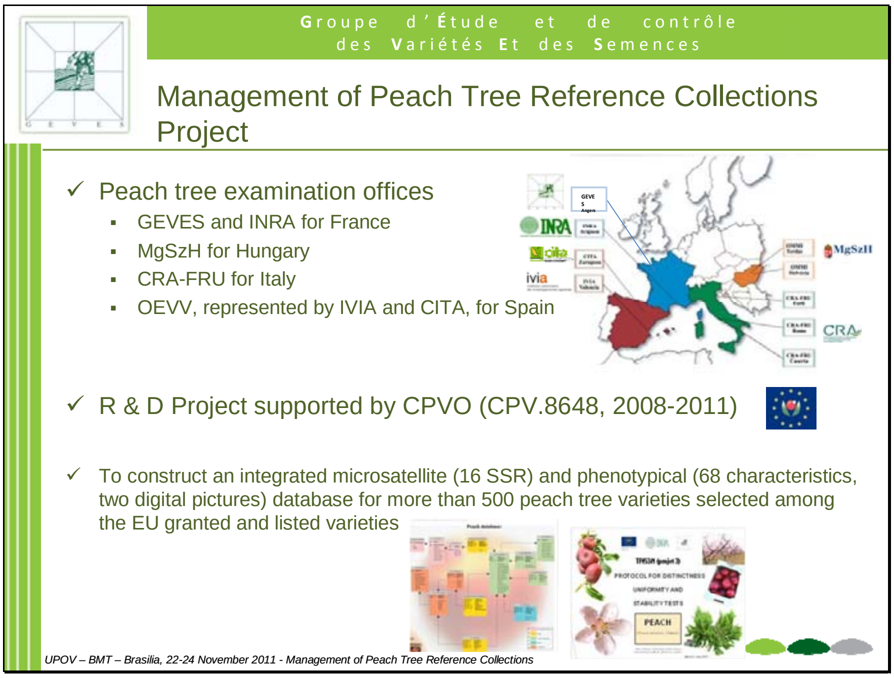


Management of Peach Tree Reference Collections Project

- ✓ Peach tree examination offices
  - GEVES and INRA for France
  - MgSzH for Hungary
  - CRA-FRU for Italy
  - OEVV, represented by IVIA and CITA, for Spain
- ✓ R & D Project supported by CPVO (CPV.8648, 2008-2011)
- ✓ To construct an integrated microsatellite (16 SSR) and phenotypical (68 characteristics, two digital pictures) database for more than 500 peach tree varieties selected among the EU granted and listed varieties

Management of Peach Tree Reference Collections: Biomolecular part

✓ Steps of the project

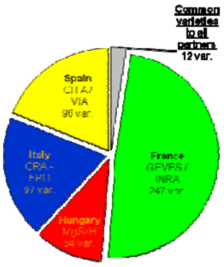
- compilation and production of a representative varieties set of standardized phenotypic data, including morphological descriptions and digital pictures
- selection and testing of a set of SSR markers
- characterisation of the varieties with these SSR markers
- creation and implementation of a database with these data

Varieties included in the project

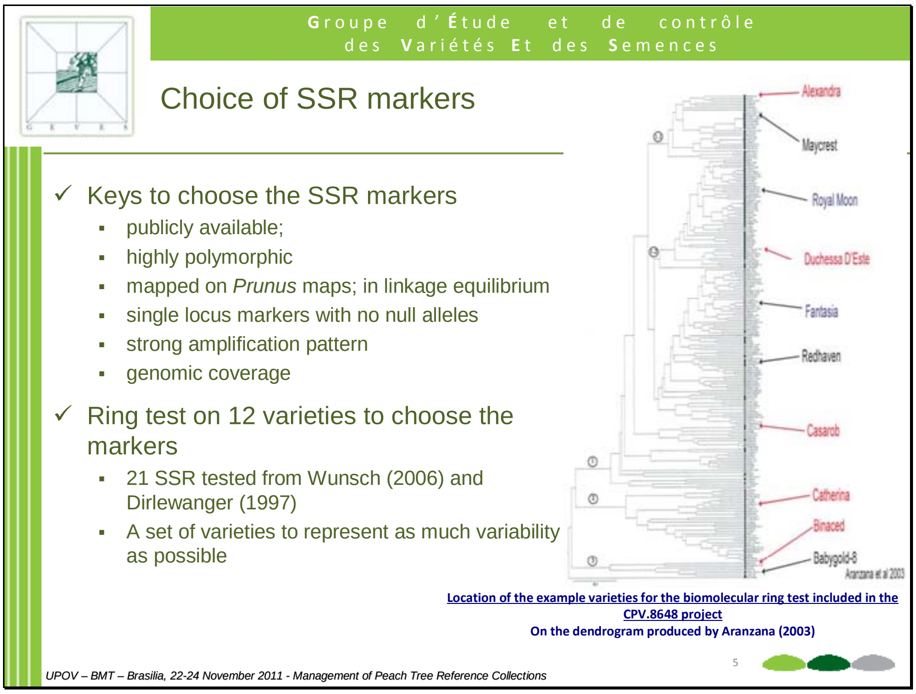
✓ Keys to choose the varieties

- Registered in a National List (trading authorization), and / or protected at a national or European level (Plant Breeder Rights)
- Physically available in at least one of the partners' orchards
- Priority to the latest material

	France		Hungary	Italy	Spain	
	GEVES	INRA	MgSzH	CRA-FRU	IVIA	CITA
Common varieties (phenotypic harmonization purposes)	11		12	12	12	10
Specific varieties studied by office (without common varieties)	247		54	97	21	75
TOTAL	**258**		**66**	**109**	**33**	**85**



Repartition between partners of the number of varieties included in the CPV.8648 project



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Choice of SSR markers

- ✓ Keys to choose the SSR markers
  - publicly available;
  - highly polymorphic
  - mapped on *Prunus* maps; in linkage equilibrium
  - single locus markers with no null alleles
  - strong amplification pattern
  - genomic coverage
- ✓ Ring test on 12 varieties to choose the markers
  - 21 SSR tested from Wunsch (2006) and Dirlewanger (1997)
  - A set of varieties to represent as much variability as possible

Location of the example varieties for the biomolecular ring test included in the CPV.8648 project

On the dendrogram produced by Aranzana (2003)

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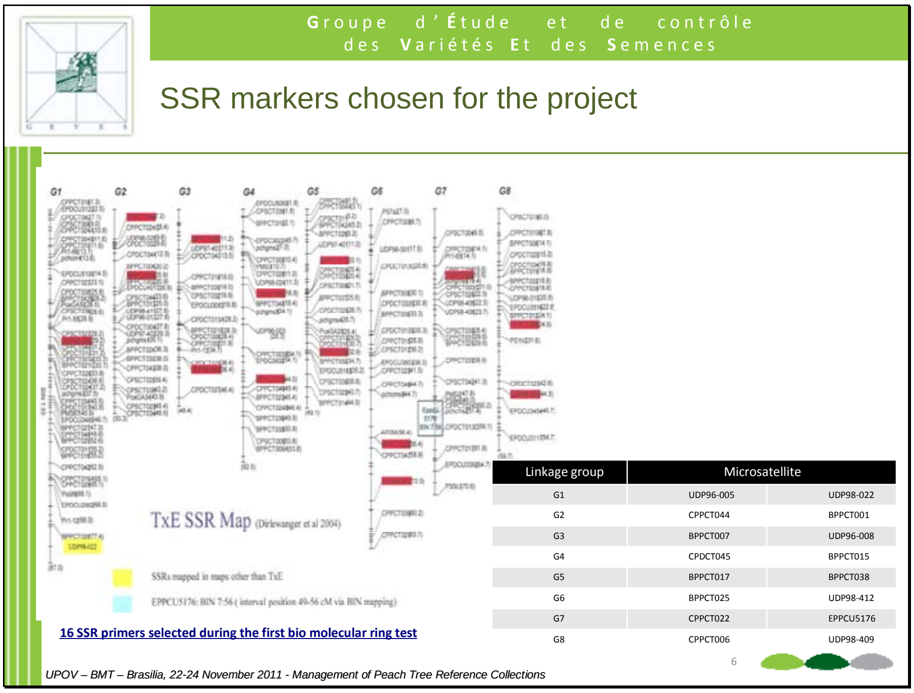

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SSR markers chosen for the project

Linkage group	Microsatellite	
G1	UDP96-005	UDP98-022
G2	CPPCT044	BPPCT001
G3	BPPCT007	UDP96-008
G4	CPDCT045	BPPCT015
G5	BPPCT017	BPPCT038
G6	BPPCT025	UDP98-412
G7	CPPCT022	EPPCU5176
G8	CPPCT006	UDP98-409

16 SSR primers selected during the first bio molecular ring test

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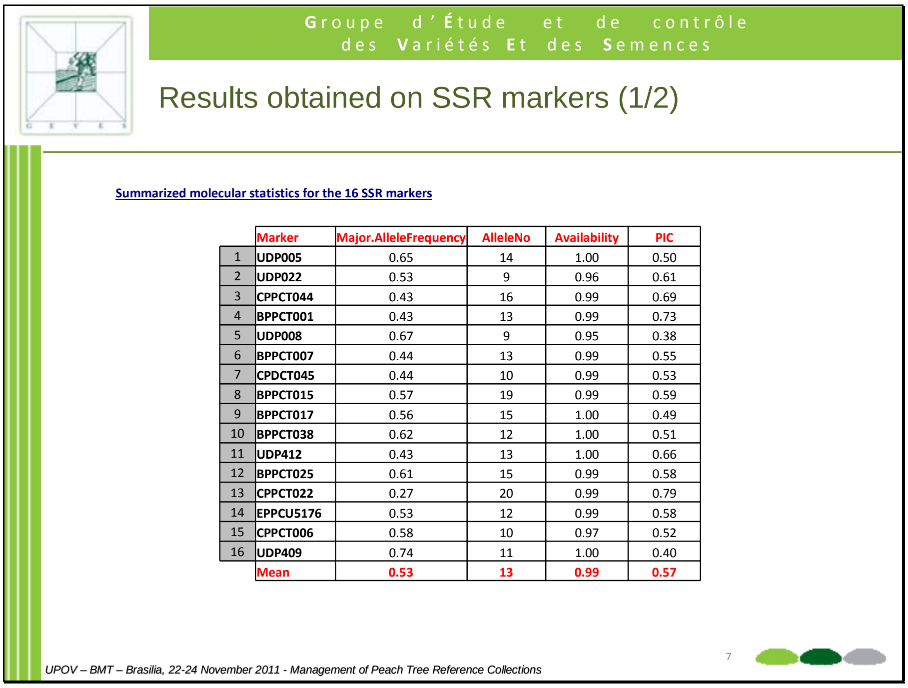

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Results obtained on SSR markers (1/2)

Summarized molecular statistics for the 16 SSR markers

	Marker	Major.AlleleFrequency	AlleleNo	Availability	PIC
1	**UDP005**	0.65	14	1.00	0.50
2	**UDP022**	0.53	9	0.96	0.61
3	**CPPCT044**	0.43	16	0.99	0.69
4	**BPPCT001**	0.43	13	0.99	0.73
5	**UDP008**	0.67	9	0.95	0.38
6	**BPPCT007**	0.44	13	0.99	0.55
7	**CPDCT045**	0.44	10	0.99	0.53
8	**BPPCT015**	0.57	19	0.99	0.59
9	**BPPCT017**	0.56	15	1.00	0.49
10	**BPPCT038**	0.62	12	1.00	0.51
11	**UDP412**	0.43	13	1.00	0.66
12	**BPPCT025**	0.61	15	0.99	0.58
13	**CPPCT022**	0.27	20	0.99	0.79
14	**EPPCU5176**	0.53	12	0.99	0.58
15	**CPPCT006**	0.58	10	0.97	0.52
16	**UDP409**	0.74	11	1.00	0.40
	Mean	**0.53**	**13**	**0.99**	**0.57**

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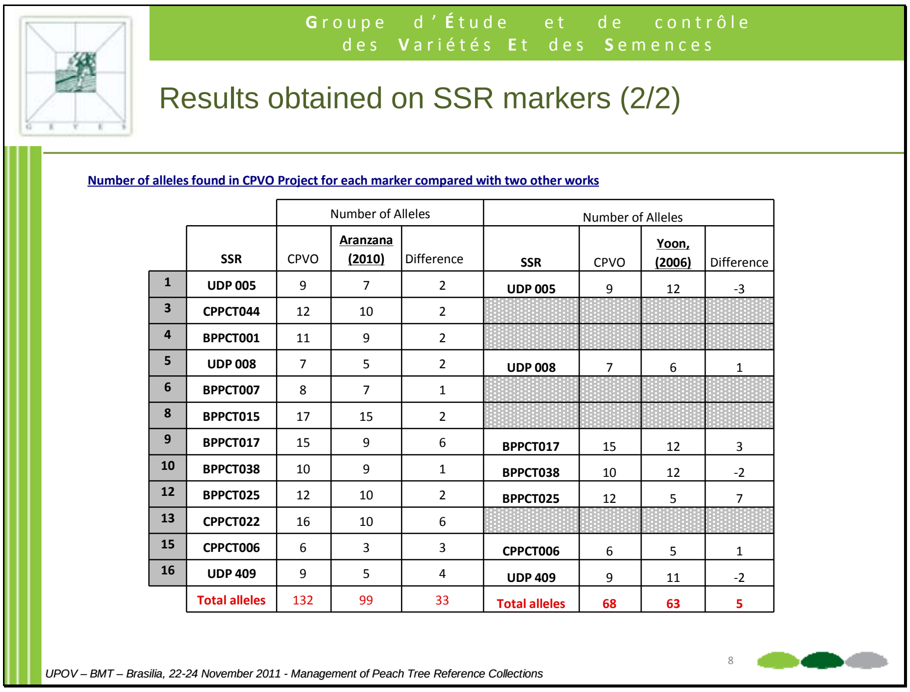

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Results obtained on SSR markers (2/2)

Number of alleles found in CPVO Project for each marker compared with two other works

		Number of Alleles				Number of Alleles		
	SSR	CPVO	Aranzana (2010)	Difference	SSR	CPVO	Yoon, (2006)	Difference
1	**UDP 005**	9	7	2	**UDP 005**	9	12	-3
3	**CPPCT044**	12	10	2	▨	▨	▨	▨
4	**BPPCT001**	11	9	2	▨	▨	▨	▨
5	**UDP 008**	7	5	2	**UDP 008**	7	6	1
6	**BPPCT007**	8	7	1	▨	▨	▨	▨
8	**BPPCT015**	17	15	2	▨	▨	▨	▨
9	**BPPCT017**	15	9	6	**BPPCT017**	15	12	3
10	**BPPCT038**	10	9	1	**BPPCT038**	10	12	-2
12	**BPPCT025**	12	10	2	**BPPCT025**	12	5	7
13	**CPPCT022**	16	10	6	▨	▨	▨	▨
15	**CPPCT006**	6	3	3	**CPPCT006**	6	5	1
16	**UDP 409**	9	5	4	**UDP 409**	9	11	-2
	Total alleles	132	99	33	**Total alleles**	**68**	**63**	**5**

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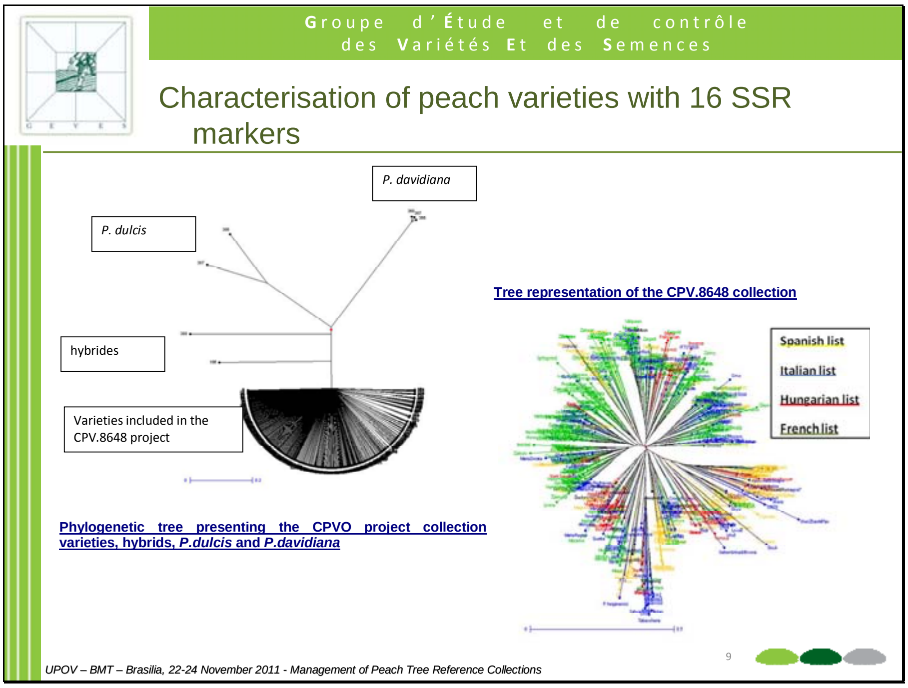

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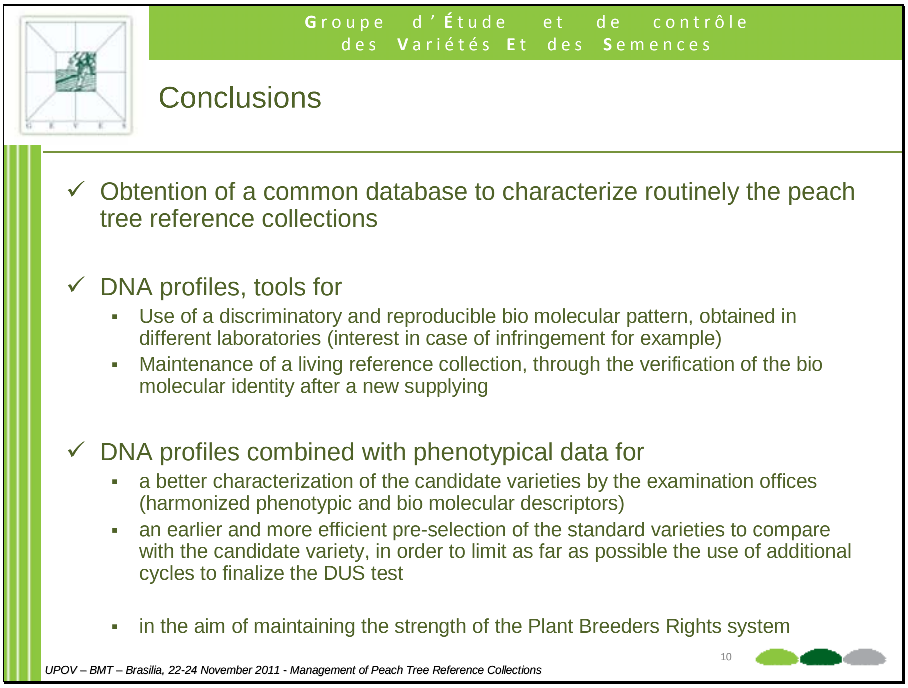
Characterisation of peach varieties with 16 SSR markers

Tree representation of the CPV.8648 collection

Phylogenetic tree presenting the CPVO project collection varieties, hybrids, *P.dulcis* and *P.davidiana*



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Conclusions

- ✓ Obtention of a common database to characterize routinely the peach tree reference collections
- ✓ DNA profiles, tools for
  - Use of a discriminatory and reproducible bio molecular pattern, obtained in different laboratories (interest in case of infringement for example)
  - Maintenance of a living reference collection, through the verification of the bio molecular identity after a new supplying
- ✓ DNA profiles combined with phenotypical data for
  - a better characterization of the candidate varieties by the examination offices (harmonized phenotypic and bio molecular descriptors)
  - an earlier and more efficient pre-selection of the standard varieties to compare with the candidate variety, in order to limit as far as possible the use of additional cycles to finalize the DUS test
  - in the aim of maintaining the strength of the Plant Breeders Rights system

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Thank you for your attention !

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