

# **BREEDING FOR RESISTANCE IN A CHANGING ENVIRONMENT –**

## **DURABLE RESISTANCE: HOPES, PITFALLS, AND MANAGEMENT STRATEGIES**

Moderator: Steven McKeand



# Breeding poplars with durable resistance to *Melampsora larici-populina* leaf rust

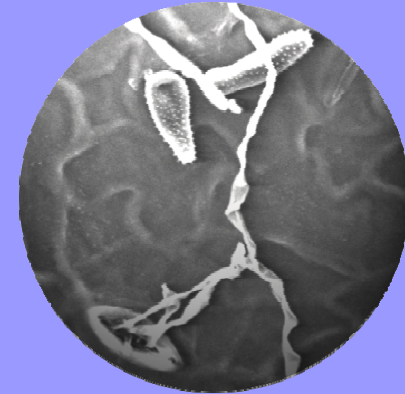
*A multidisciplinary approach to understand and delay pathogen adaptation*

**Arnaud Dowkiw**, Catherine Bastien, Véronique Jorge, Marc Villar, Elodie Voisin, Irinel Mardare, Vanina Guérin INRA, UAGPF, 45166 Olivet Cedex, France; [arnaud.dowkiw@orleans.inra.fr](mailto:arnaud.dowkiw@orleans.inra.fr) ; Patricia Faivre-Rampant, Aloïs Bresson, Frédérique Bitton, INRA-URGV, 91057 Evry Cedex, France; Sébastien Duplessis, Pascal Frey, Annegret Kohler, Cécile Rinaldi, Sandrine Miot, Pierre Gérard, Constance Xhaard, Bénédicte Fabre, Fabien Hacklett, INRA UMR IAM, 54280 Champenoux, France; Christophe Plomion and Céline Lalanne, INRA UMR BIOGECO, 33612 CESTAS Cedex, France.

**Fourth International Workshop on the Genetics of Host-Parasite Interactions in Forestry**

July 31 – August 5, 2011, Eugene, Oregon, USA

# The protagonists



*P. x interamericana*

*P. x euramericana*

*Populus*

*Melampsora larici-populina*

*P. trichocarpa*

X *P. deltoides*

X *P. nigra*



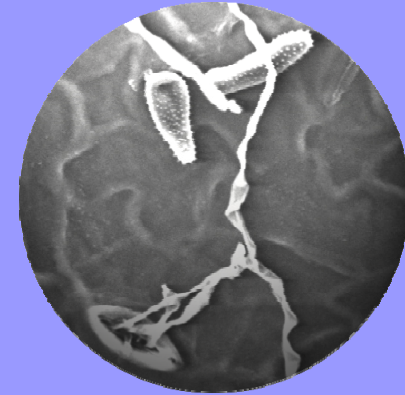
*M. occidentalis*  
*M. x columbiana*  
*M. medusae*

*M. larici-populina*  
*M. allii-populina*

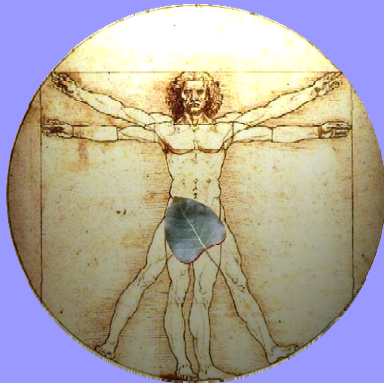
# The protagonists



*Populus*

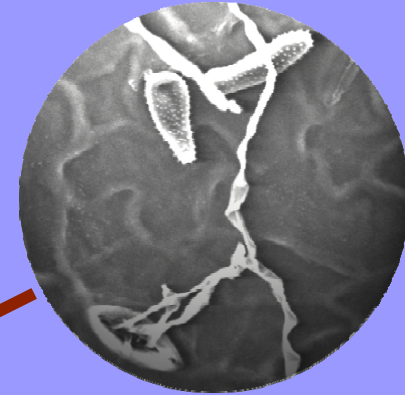


*Melampsora larici-populina*



*Larix*

# The protagonists



*Melampsora larici-populina*

Mixed "epidemic" genetic structure	High (3)	<i>Phytophthora sojae</i>	7	<i>Rhynchosporium secalis</i> <i>Mycosphaerella fijiensis</i> , <i>graminicola</i>	8	<i>Blumeria graminis</i> <i>Bremia lactucae</i> <i>Phytophthora infestans</i> - new populations	9	(3)	Effective population size
			6	<i>Venturia inaequalis</i> <i>Rhizoctonia solani</i> <i>Setosphaeria turcica</i> <i>Phaeosphaeria nodorum</i> <i>Leptosphaeria maculans</i> <i>Pseudocercospora herpotrichoides</i>	7	<i>Puccinia graminis</i> f. sp. <i>tritici</i> – pre 1930's <i>P. coronata</i> f. sp. <i>avenae</i> <i>Sclerospora graminicola</i> <i>Melampsora larici-populina</i> , <i>larici-</i> <i>epitica</i>	7	(2)	
			5		6	7	(1)		
Outcrossing Sexual high genotype diversity ↑ ↓ Inbreeding	Medium (2)	<i>Pratylenchus</i> <i>Heterodera</i>  <i>Armillaria mellea</i>	6		7		8	(3)	Effective population size
			5		6	<i>Ustilago hordei</i> , <i>maydis</i> <i>Tilletia</i>	7	(2)	
4	<i>Sporisorium reilianum</i>	5		6	<i>Sclerotinia sclerotiorum</i>	7	(1)		
Asexual low genotype diversity	Low (1)	<i>Fusarium oxysporum</i> f. sp. <i>melonis</i> , <i>lycopersici</i> , <i>cubense</i> <i>Xanthomonas campestris</i> pv. <i>vesicatoria</i> Soil-borne viruses <i>Meloidogyne incognita</i>	5		6	<i>Melampsora lini</i> <i>Maydisporthe grisea</i> <i>Cladosporium fulvum</i> <i>Puccinia coronata</i> f. sp. <i>avenae</i> – no alternative host <i>P. striiformis</i> <i>P. graminis</i> f. sp. <i>tritici</i> , <i>avenae</i> <i>P. triticea</i> – no alternative host <i>Xanthomonas oryzae</i> pv. <i>oryzae</i>	7	(3)	Effective population size
			4	<i>Colletotrichum graminicola</i> <i>Colletotrichum lindemuthianum</i> <i>Erwinia amylovora</i> Insect dispersed viruses	5		6	(2)	
			3		4		5	(1)	
Reproduction/ mating system	Gene/genotype flow	Low (1)	Medium (2)	High (3)					
		Propagules soilborne, difficult to disperse  ~ 5 meter total dispersal	Propagules waterborne, moderate dispersal  ~100 m – within field	Propagules airborne, easily dispersed  ~10 – 1000 km	Man-aided dispersal may modify risk				

Annu. Rev. Phytopathol. 2002. 40:349-79  
doi: 10.1146/annurev.phyto.40.120501.101443  
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## PATHOGEN POPULATION GENETICS, EVOLUTIONARY POTENTIAL, AND DURABLE RESISTANCE

Bruce A. McDonald and Celeste Linde

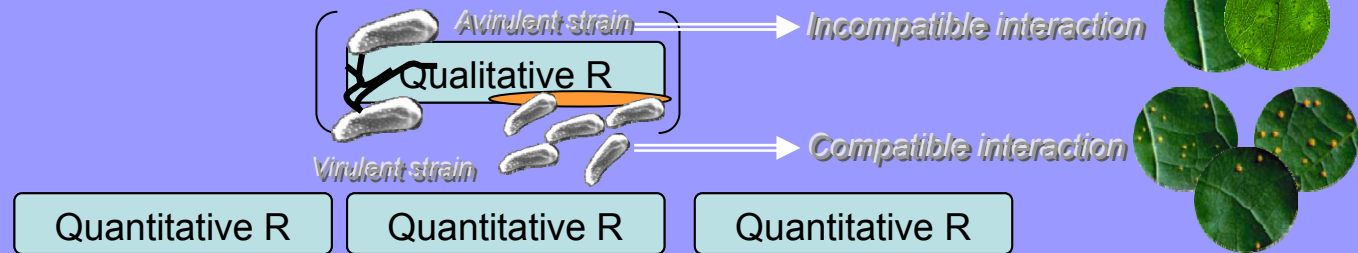
**Figure 4** (See figure on previous page) Scale of evolutionary risk organized according to reproduction/mating system, gene/genotype flow and effective population size. The organization of this diagram is modified from Figure 2 of Brent & Holloman (8). Effective population size ( $N_e$ ) is on a 1-3 scale where 1 is small  $N_e$ , 2 is average  $N_e$ , and 3 is large  $N_e$ . Assignment of total risk value assumes that all effects are additive. This risk model assumes that mutation rates are constant and that selection is efficient for all pathogens. Placement of example pathogens is according to principles explained in the text or from Table 2.

# Breeder's vocabulary and material



**Breeding populations :**

**Resistance phenotypes :**



# Breeder's vocabulary and material






*Populus*



♂

♀

	<i>P. trichocarpa</i>	<i>P. deltoides</i>	<i>P. nigra</i>
	16 F <sub>1</sub> fam. 602 genotypes 	19 F <sub>1</sub> fam. 327 genotypes 	16 F <sub>1</sub> fam. 279 genotypes 
	38 F <sub>1</sub> fam. 2966 genotypes	55 F <sub>1</sub> fam. 2455 genotypes	40 F <sub>1</sub> fam. 1884 genotypes
	6 F <sub>1</sub> fam. 250 genotypes		18 F <sub>1</sub> fam. 936 genotypes

+ some backcrosses and F<sub>2</sub>s

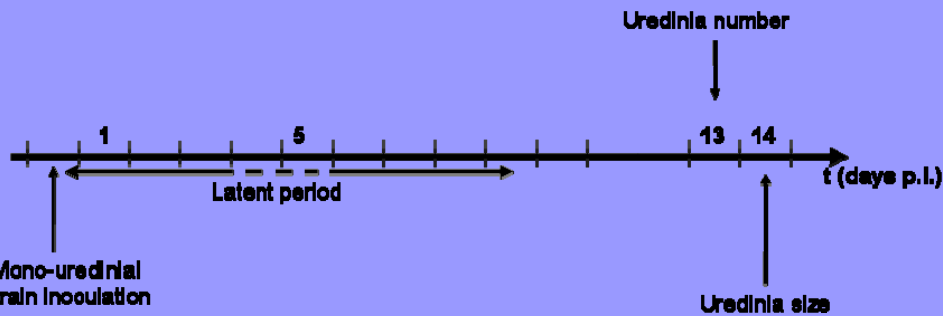
# Breeder's vocabulary and material

## Evaluating resistance

*In the lab*



*In the field*



Rust score on the most infected leaf



# Pathogen adaptation

**A reality**

**Why and how ?**

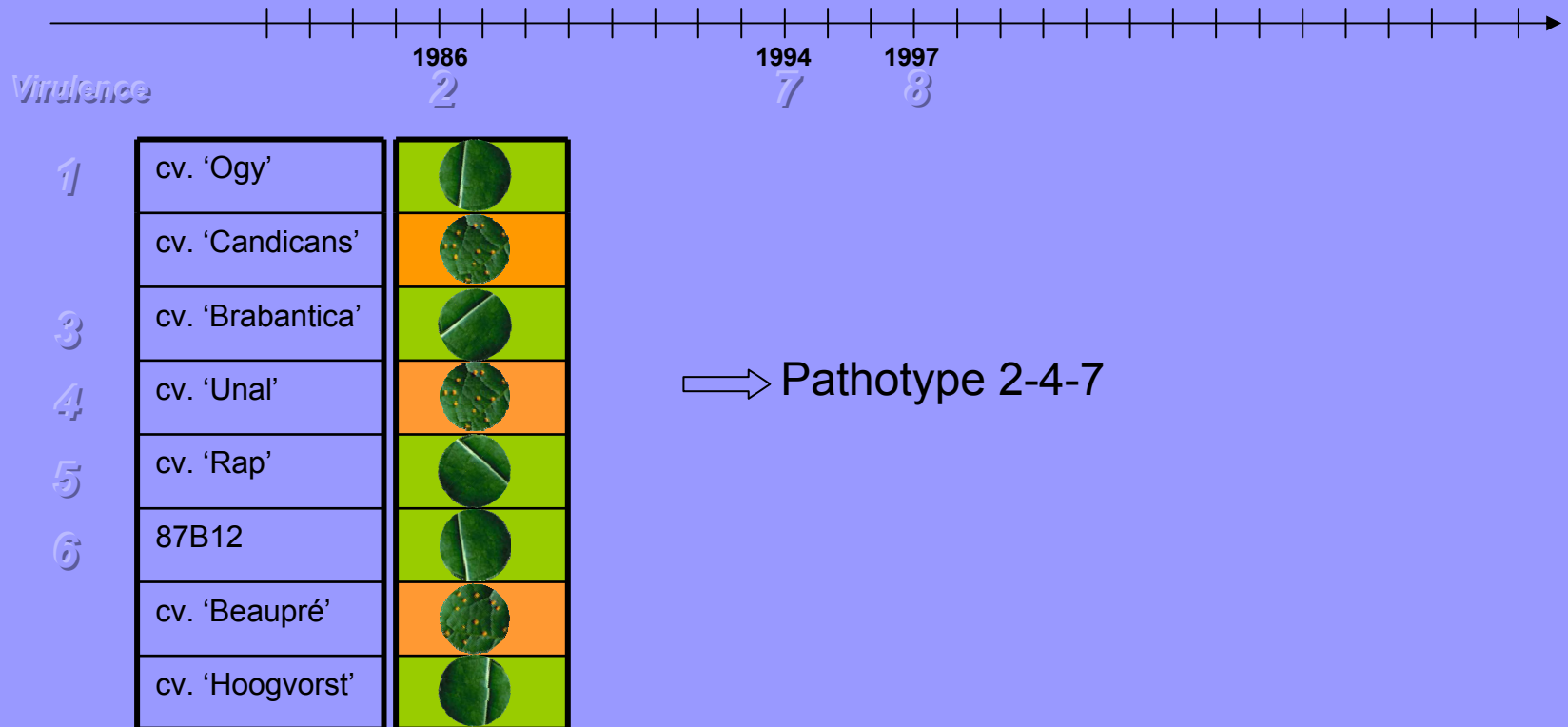
**Breeding strategies**

# Pathogen adaptation A reality

Several qualitative resistances inherited from *P. deltoides* did not prove to be durable



more than a few years after commercial deployment of interspecific hybrids :

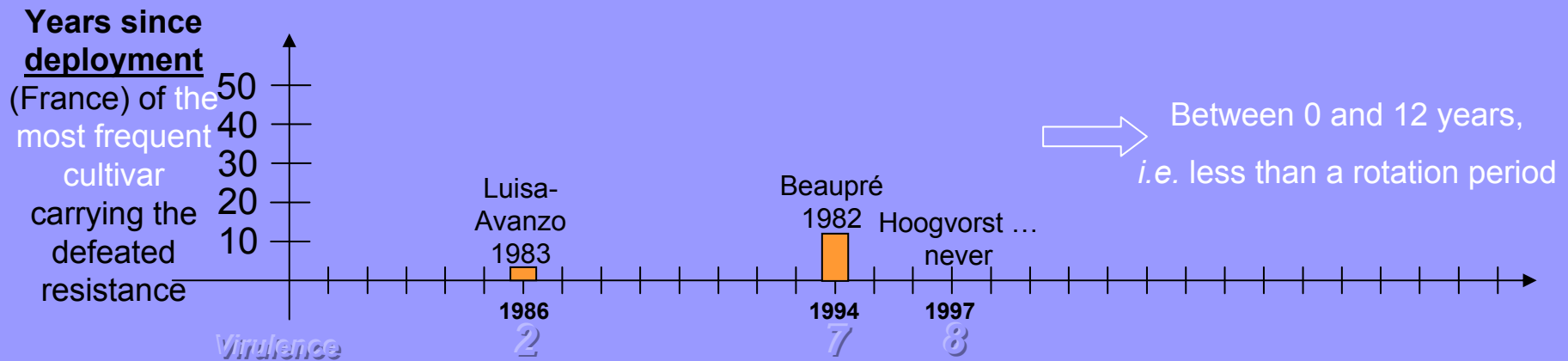
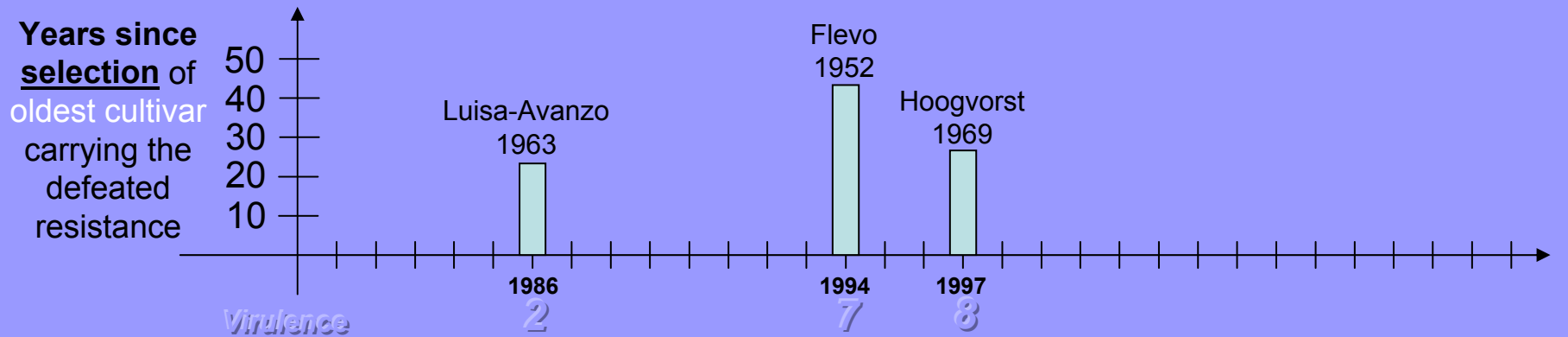


# Pathogen adaptation A reality


Several qualitative resistances inherited from *P. deltoides* did not prove to be durable

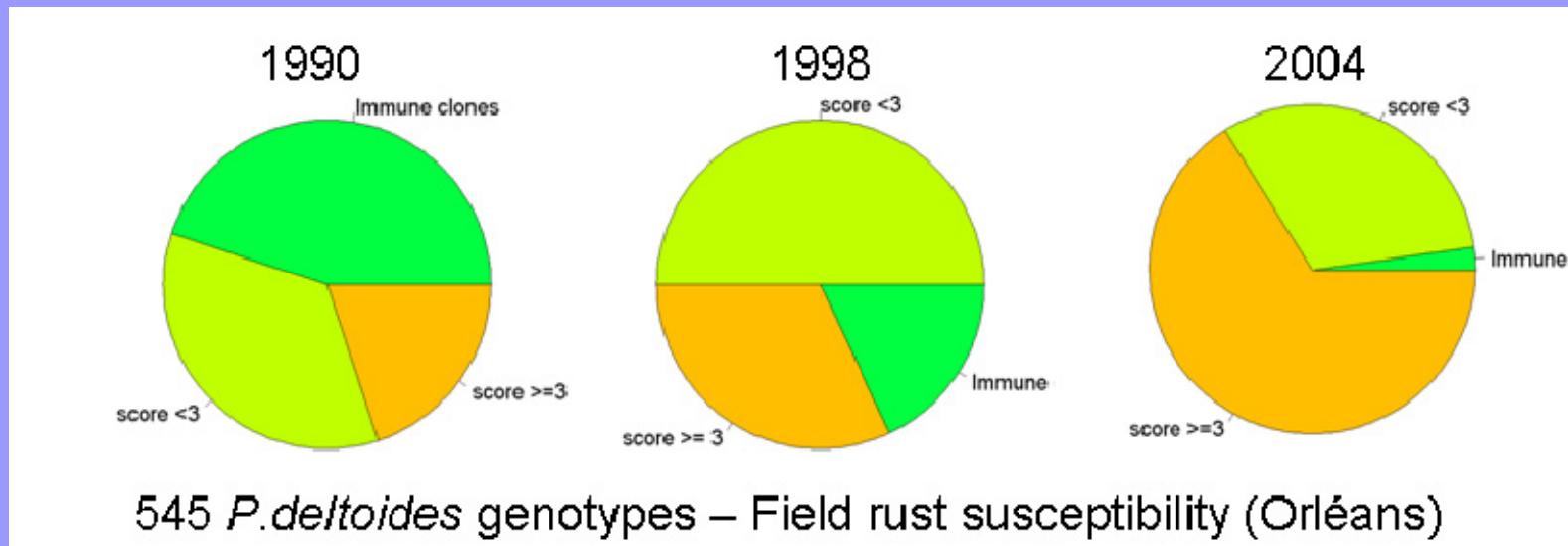


more than a few years after commercial deployment of interspecific hybrids :



# Pathogen adaptation A reality

Most qualitative resistances found in our *P. deltoides* breeding material  
  
are already defeated

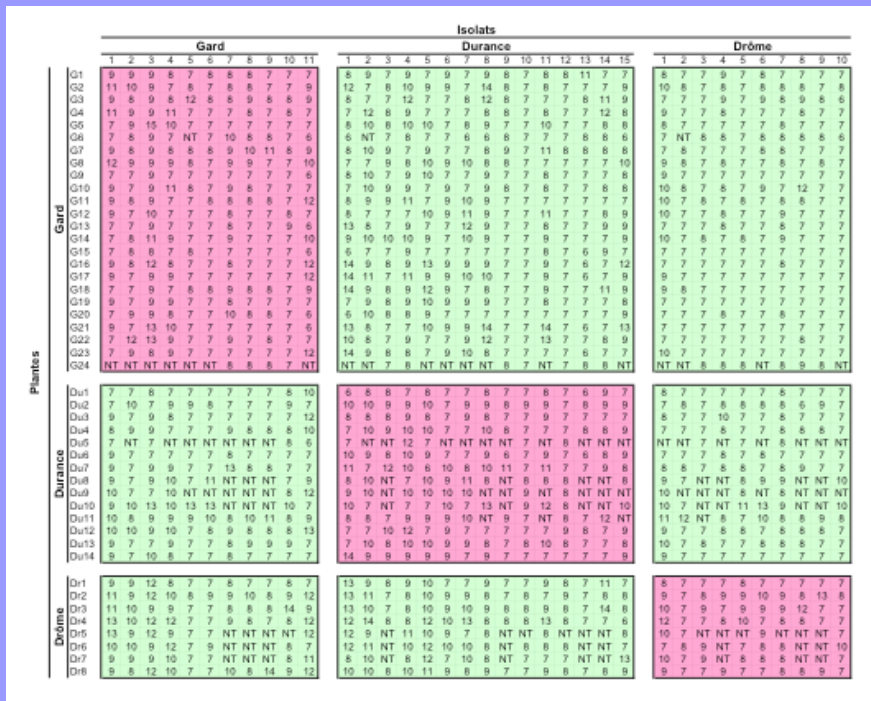


# Pathogen adaptation A reality

Absence of qualitative resistance in the sympatric host *P. nigra*



adaptation as a fatality ?



46 *P. nigra* hosts x 36 isolates

3 pops from Southern France

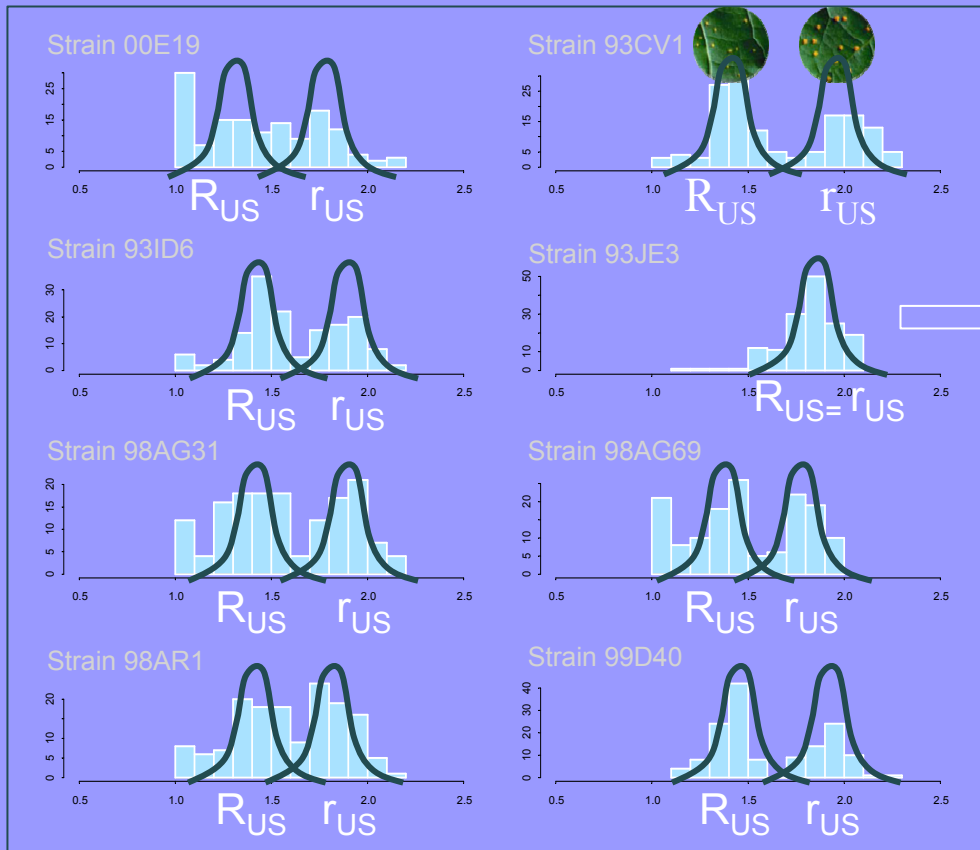


100% of the 1529 tested  
host x isolate interactions  
were compatible

# Pathogen adaptation A reality

Quantitative resistance also can be defeated

Exemple of a major quantitative resistance factor inherited from *P. trichocarpa*



$R_{US}$  defeating strain

Collecting large uredinia on  $R_{US}$  host genotypes in our nursery revealed other  $R_{US}$  defeating strains

- Dowkiw *et al.*  
Phytopathology 2003 Vol. 83 (4)
- Dowkiw and Bastien  
Phytopathology 2004 Vol. 94 (12)
- Dowkiw *et al.*  
Plant Pathology 2010 Vol. 59 (3)

Distributions of clonal means for uredinia size in a *P. X interamericana*  $F_1$  progeny

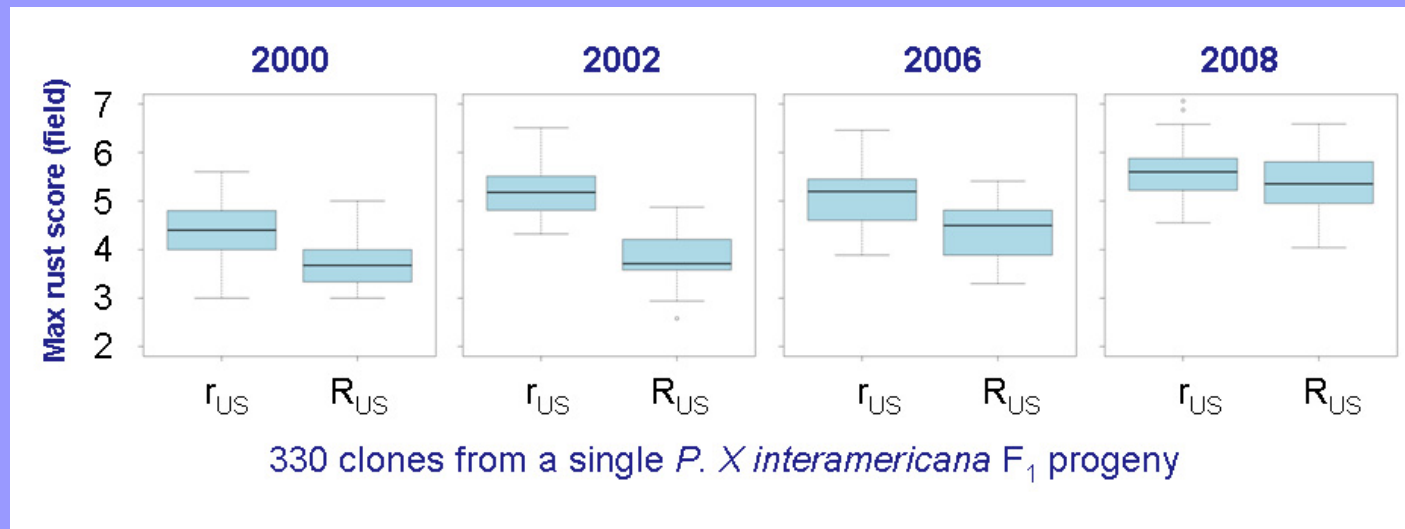
**Fourth International Workshop on the Genetics of Host-Parasite Interactions in Forestry**

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# Pathogen adaptation A reality

Quantitative resistance also can be defeated

Exemple of a major quantitative resistance factor inherited from *P. trichocarpa*



$$\left( \begin{array}{l} r_{US} = r_{US}r_{US} \\ R_{US} = R_{US}r_{US} \end{array} \right)$$

# Pathogen adaptation

**A reality**

**Why and how ?**

**Breeding strategies**



# Pathogen adaptation Why and how ?

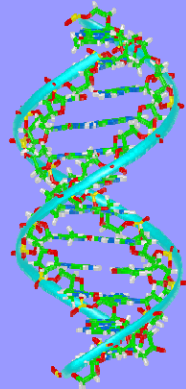
*From genes (host & pathogen) to the environment (incl. management)*



Who are they ?  
How many are they ?  
How do they evolve ?  
...



How does adaptation emerge ?  
How does it maintain and spread ?  
What role do wild/ornamental poplars play ?  
What role does the alternate host play ?  
...

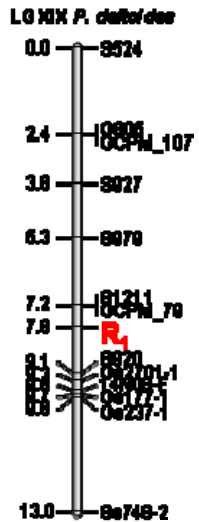


# Pathogen adaptation

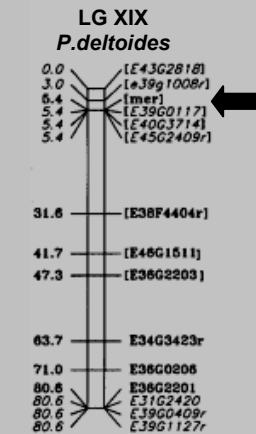
## Why and how ?

From genes (host & pathogen) to the environment (incl. management)

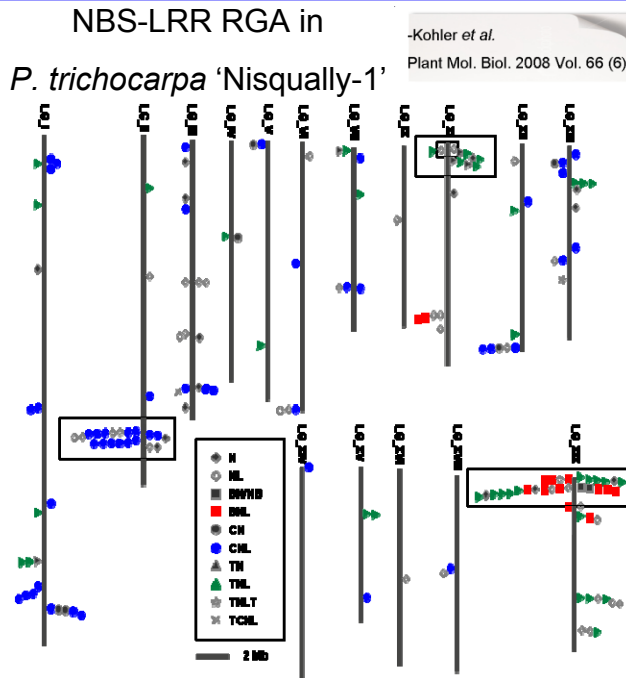
Qualitative resistance inherited from *P. deltoides*



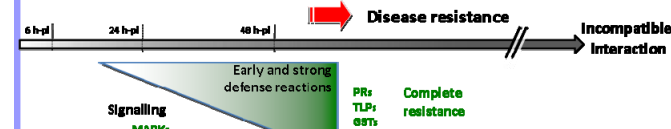
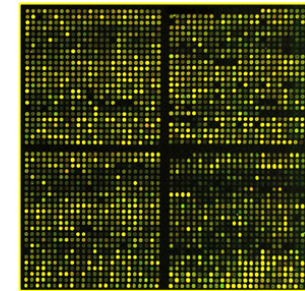
- Jorge *et al.*  
New Phytol. Vol. 167 (1)



Cervera *et al.*, Genetics, 2001



## Transcriptomics



- Rinaldi *et al.*  
Plant Physiol. 2007 Vol. 144

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# Pathogen adaptation

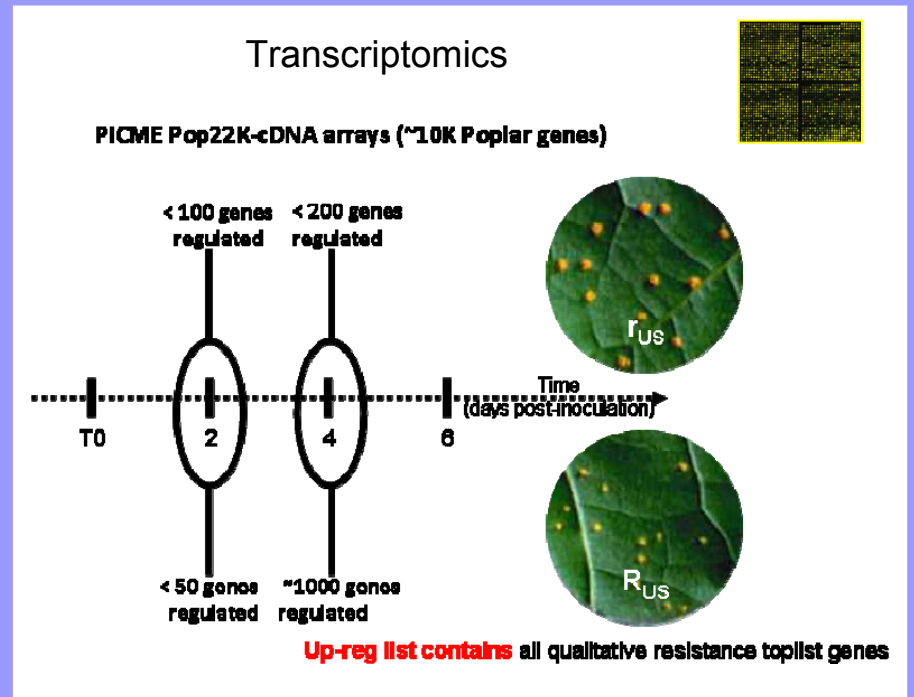
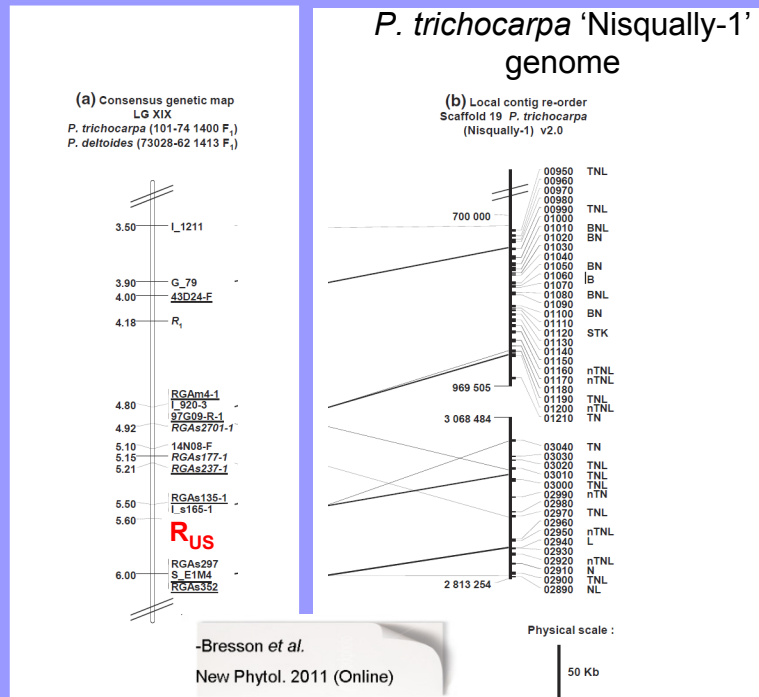
## Why and how ?

From genes (host & pathogen) to the environment (incl. management)

Quantitative resistance inherited from *P. trichocarpa*



host



# Pathogen adaptation Why and how?

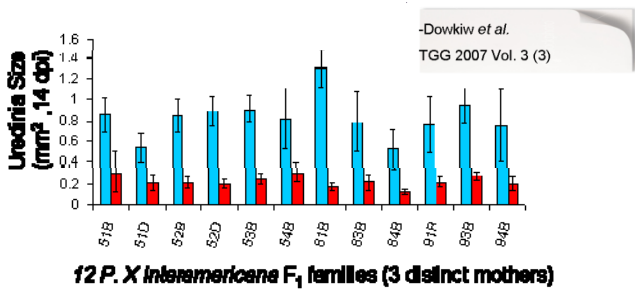
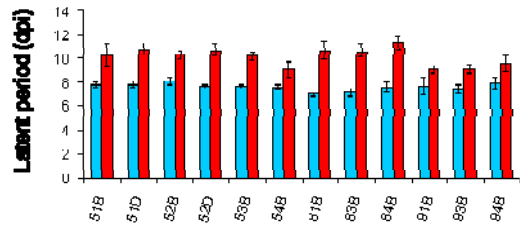
From genes (host & pathogen) to the environment (incl. management)



host

Other quantitative resistance factors

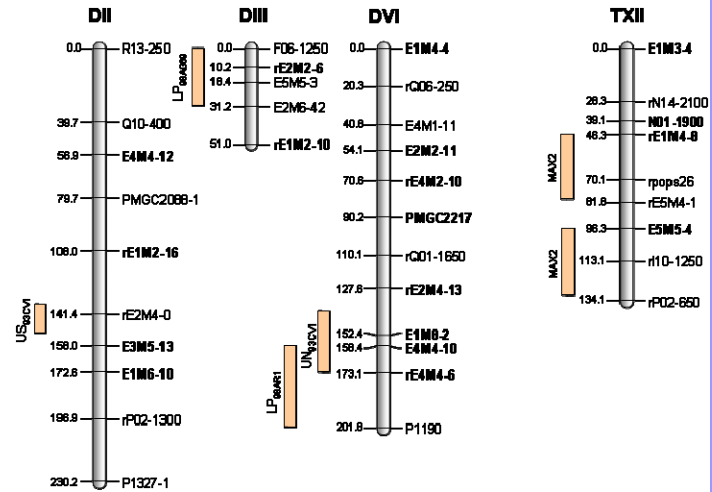
## R-gene associated QR factors



12 *P. X interamericana* F<sub>1</sub> families (3 distinct mothers)

Genotypes with/without a defective qualitative resistance inherited from *P. deltoidea*

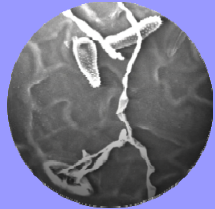
## Small QTLs



- Jorge et al. New Phytol. Vol. 167 (1)

# Pathogen adaptation Why and how ?

*From genes (host & pathogen) to the environment (incl. management)*



**PNAS**

**Obligate biotrophy features unraveled by the genomic analysis of rust fungi** 2011 Vol. 108 (22)

Sébastien Duplessis<sup>h,1,2</sup>, Christina A. Cuomo<sup>h,1,2</sup>, Yao-Cheng Lin<sup>5</sup>, Andrea Aerts<sup>4</sup>, Emilie Tisserant<sup>6</sup>, Claire Veneault-Fourrey<sup>6</sup>, David L. Joly<sup>6,7</sup>, Stéphane Hacquard<sup>h,4</sup>, Joëlle Amselem<sup>1</sup>, Bradi L. Cantarel<sup>9</sup>, Readman Chiu<sup>h</sup>, Pedro M. Coutinho<sup>8</sup>, Nicolas Feu<sup>h,5</sup>, Matthew Fiedl<sup>h</sup>, Pascal Frey<sup>6</sup>, Eric Gelhaye<sup>6</sup>, Jonathan Goldberg<sup>h</sup>, Manfred G. Giabherr<sup>8</sup>, Chinnappa C. Kodira<sup>h,8</sup>, Arnegret Kohler<sup>6</sup>, Ursula Kües<sup>1</sup>, Erika A. Lindquist<sup>8</sup>, Susan M. Lucas<sup>8</sup>, Rohit Mago<sup>8</sup>, Evan Maucel<sup>8</sup>, Emmanuelle Morin<sup>6</sup>, Claude Murat<sup>6</sup>, Jasmyn L. Pangillan<sup>8</sup>, Robert Park<sup>8</sup>, Matthew Pearson<sup>8</sup>, Hadi Quecaveville<sup>1</sup>, Nicolas Rouhier<sup>6</sup>, Sharadha Sakthikumar<sup>h</sup>, Asaf A. Salamov<sup>8</sup>, Jeremy Schmutz<sup>8</sup>, Benjamin Selles<sup>6</sup>, Harris Shapiro<sup>8</sup>, Philippe Tanguay<sup>6</sup>, Gerald A. Tuskan<sup>4,1</sup>, Bernard Henrissat<sup>6</sup>, Yves Van de Peer<sup>8</sup>, Pierre Touzé<sup>6</sup>, Jeffrey G. Ellis<sup>8</sup>, Peter N. Dodds<sup>1</sup>, Jacqueline E. Schein<sup>h</sup>, Shaobin Zhong<sup>h,7</sup>, Richard C. Hamelin<sup>8</sup>, Igor V. Ananko<sup>8</sup>, Leif S. Mortensen<sup>8</sup>, and Francis Martin<sup>h,1</sup>

MPMI Vol. 24, No. 7, 2011, pp. 808–818. doi:10.1094/MPMI-01-11-0006. © 2011 The Authors

***Melampsora larici-populina* Transcript Profiling During Germination and Timecourse Infection of Poplar Leaves Reveals Dynamic Expression Patterns Associated with Virulence and Biotrophy**

Sébastien Duplessis, Stéphane Hacquard, Christine Delaruelle, Emilie Tisserant, Pascal Frey, Francis Martin, and Arnegret Kohler

INRA, Institut national de la Recherche Agronomique, UMR 1136 INRA/Université Nancy, Interactions Arbres/Micro-organismes, Centre INRA de Nancy, 54290 Champenoux, France

Only candidate effectors for the moment : >1000 small secreted proteins, 74% are species-specific

Several homologs of genes encoding haustorially expressed secreted proteins and avirulence factors identified in *M.lini*

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# Pathogen adaptation Why and how ?

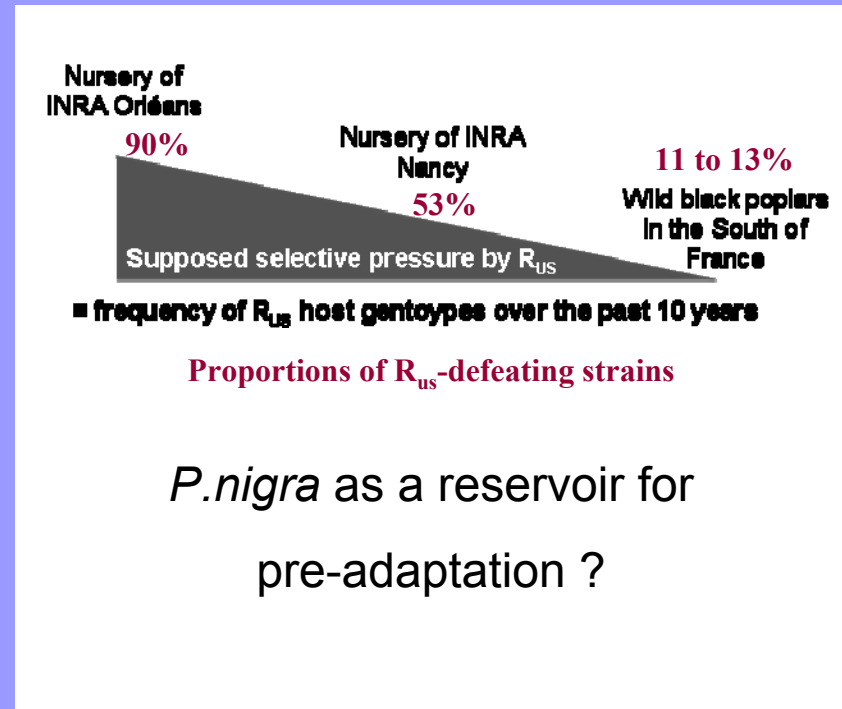
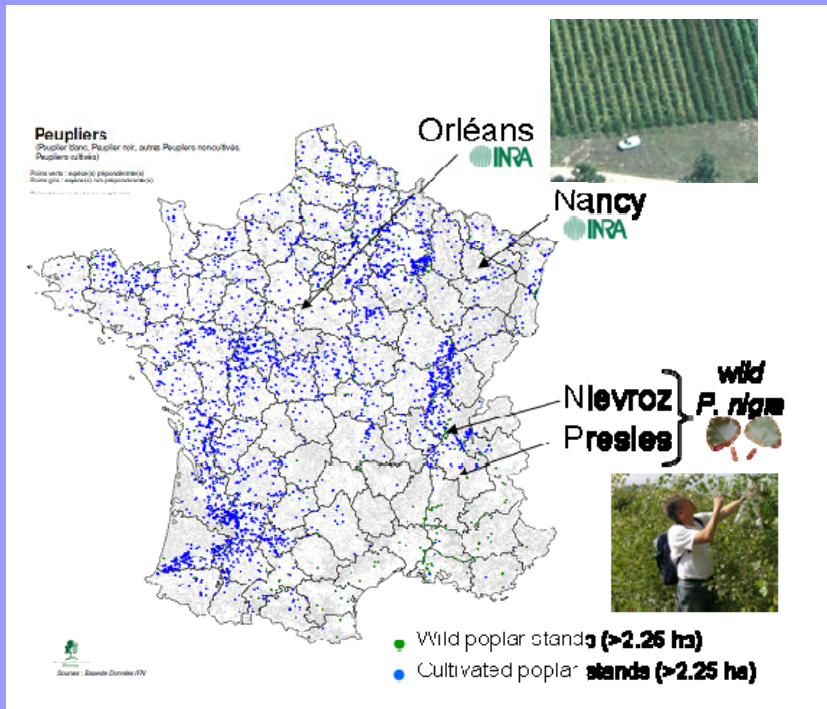
*From genes (host & pathogen) to the environment (incl. management)*

How does adaptation emerge ?

How does it maintain and spread ?

What role do wild/ornamental poplars play ?

What role does the alternate host play ?



# Pathogen adaptation Why and how ?

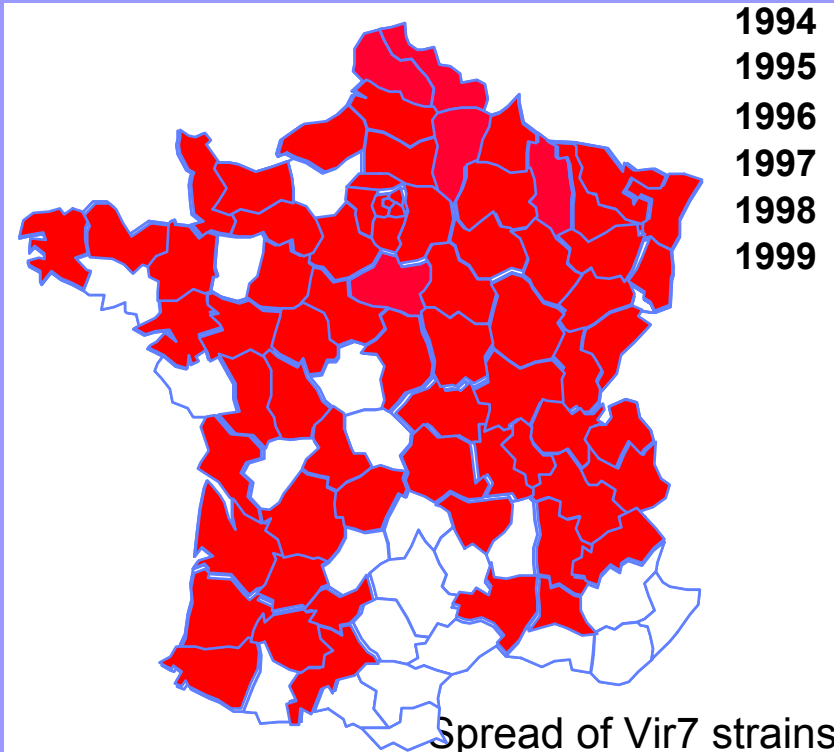
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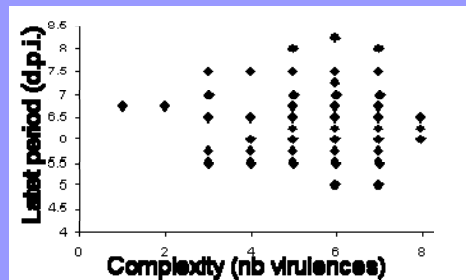
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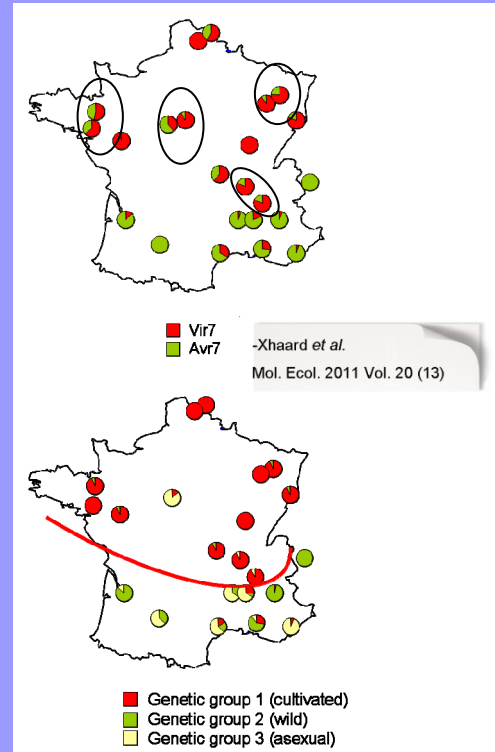


First pathotypes identified containing Vir7

1996	1999
3-4-7	4-7
3-4-5-7	1-2-3-4-5-7
1-3-4-5-7 (98%)	
1-3-4-5-6-7	



200 isolates isolated from larch



# Pathogen adaptation

**A reality**

**Why and how ?**

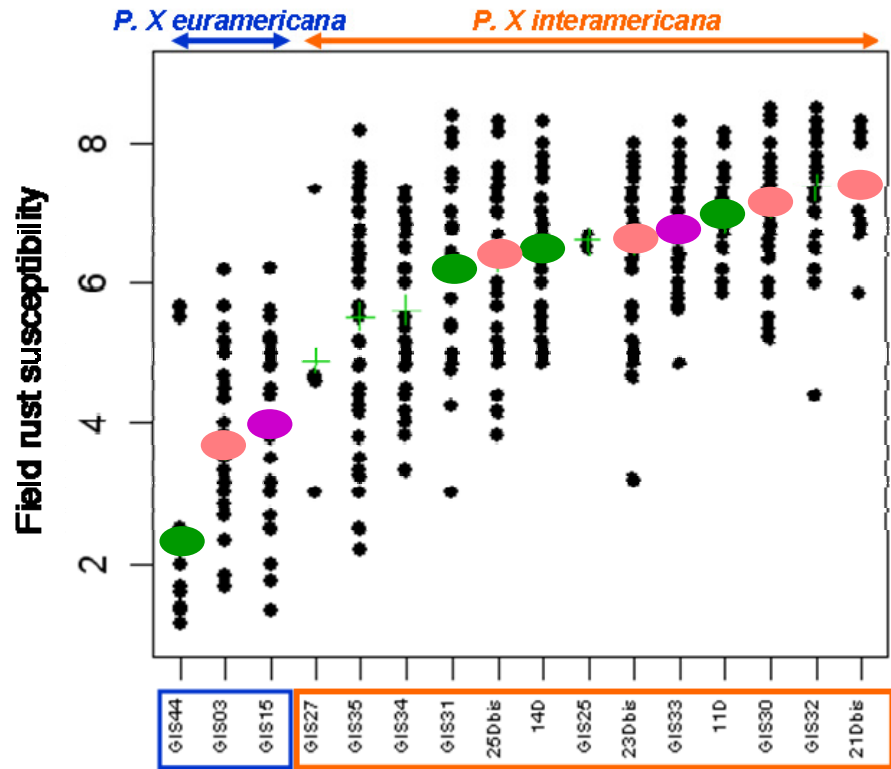
**Breeding strategies**



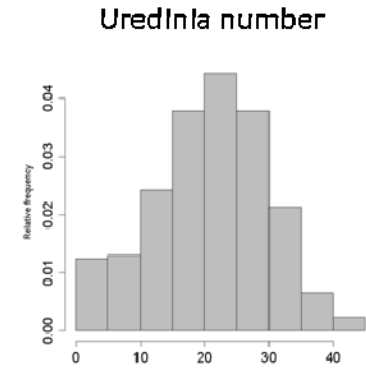
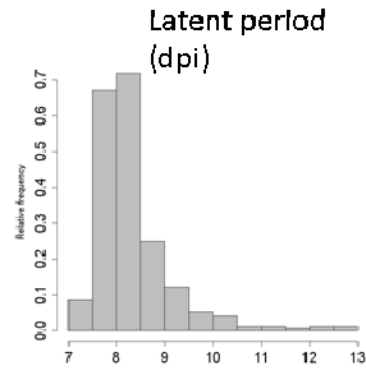
# Pathogen adaptation

## Breeding strategies

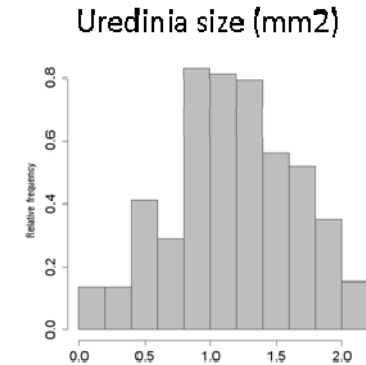
Using co-evolved quantitative resistance from



16 F<sub>1</sub> families involving common *P. deltoides* female parents



348 *P. nigra* genotypes  
Lab test (1 strain)



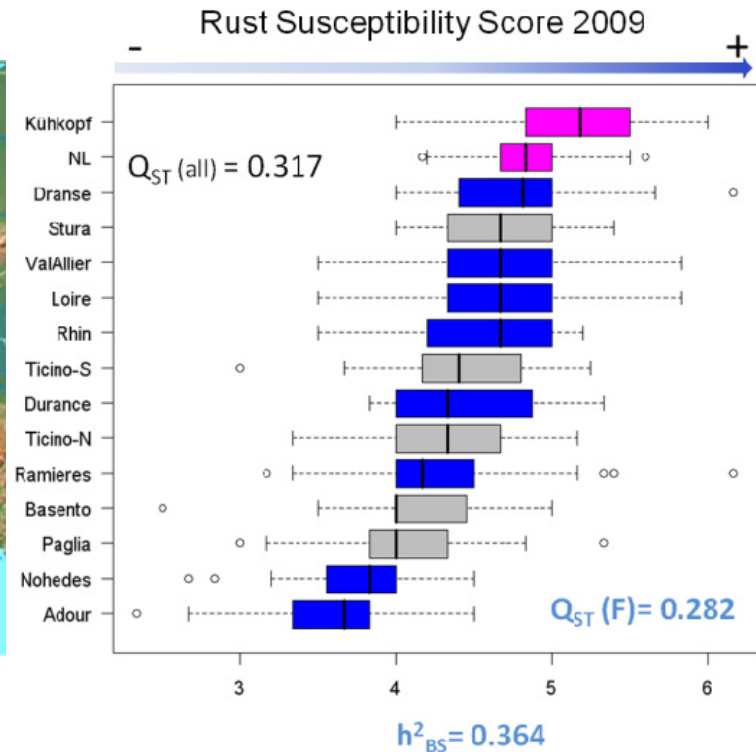
# Pathogen adaptation

## Breeding strategies

Using co-evolved quantitative resistance from *P. nigra*



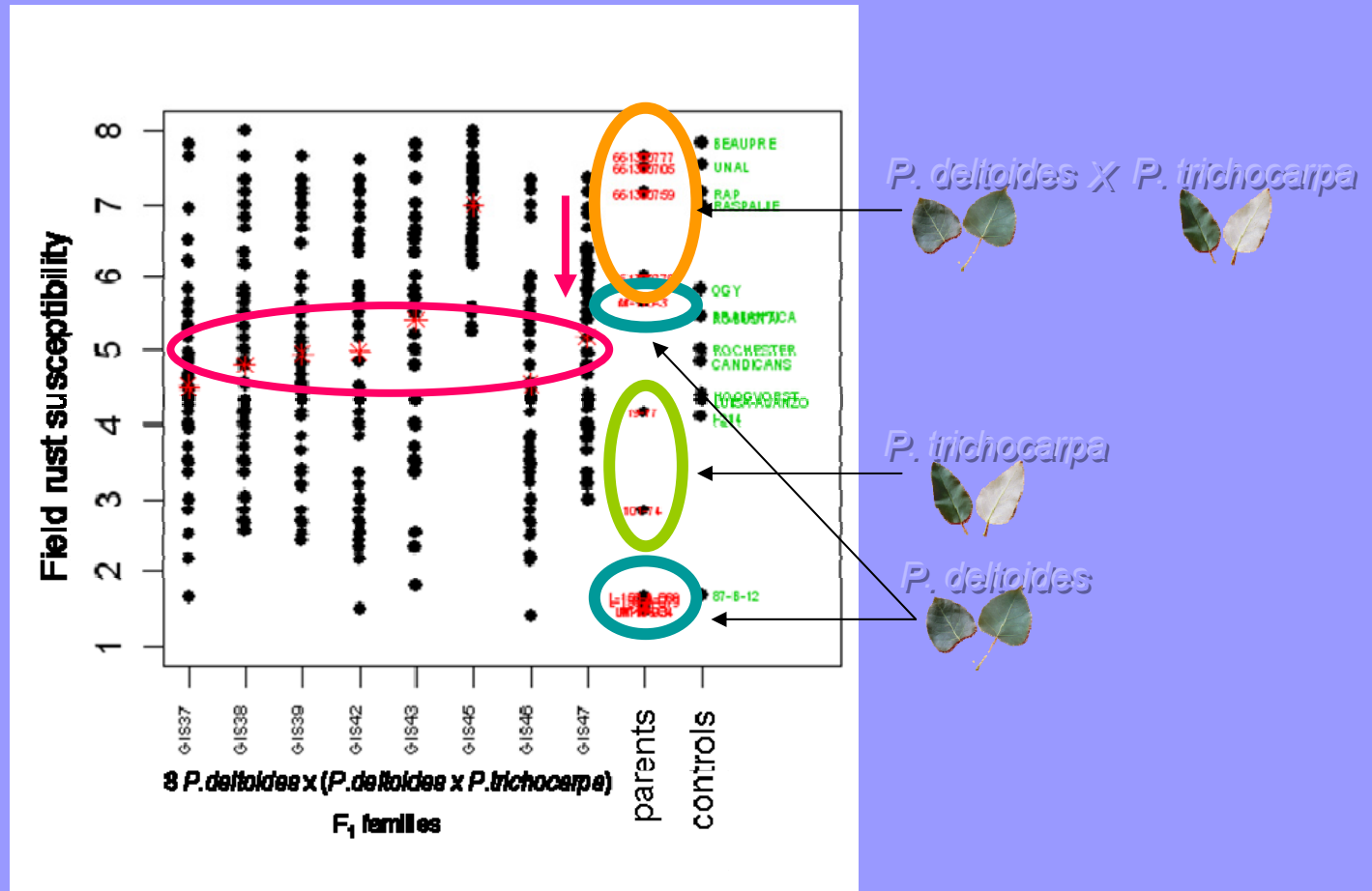
Evoltree *P. nigra* Collection (1100)



# Pathogen adaptation

## Breeding strategies

Backcrossing to a parental species

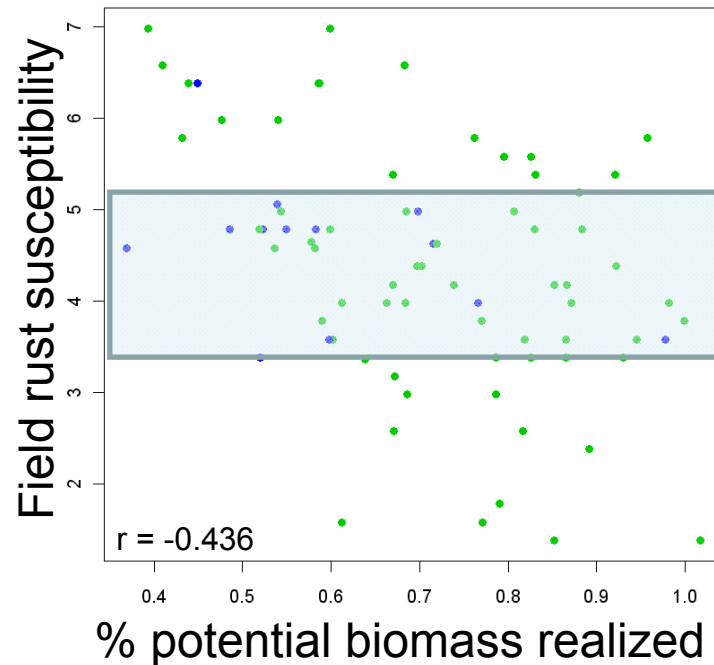


# Pathogen adaptation

## Breeding strategies

Living with the pathogen ...

Using  
'fungicide treated vs.  
non-treated'  
field experiments  
(cloned material)





Resistance ?  
Escape ?  
Tolerance ?

# Pathogen adaptation

## Thank you for your attention



The research leading to these results received funding from (e.g.)

- the European Community's Seventh Framework Program (FP7/ 2007-2013) under the grant agreement n° 211868 (Noveltree Project ) 
- the French « Bureau des Ressources Génétiques » and the French Ministry for Agriculture and Fisheries (Programme 142, sous-action 27, CTPS) 

- INRA

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