



FIRST MEETING OF THE EUPHRESKO PHYLIB PROJECT TENERIFE, CANARY ISLAND, 20-21TH FEBRUARY 2013

EPIDEMIOLOGY AND DIAGNOSIS OF POTATO PHYTOPLASMAS AND “CANDIDATUS LIBERIBACTER SOLANACEARUM” AND THEIR CONTRIBUTION TO RISK MANAGEMENT IN POTATO AND OTHER CROPS

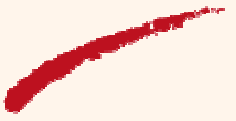


UMT INNOPLANT (FN3PT-INRA)

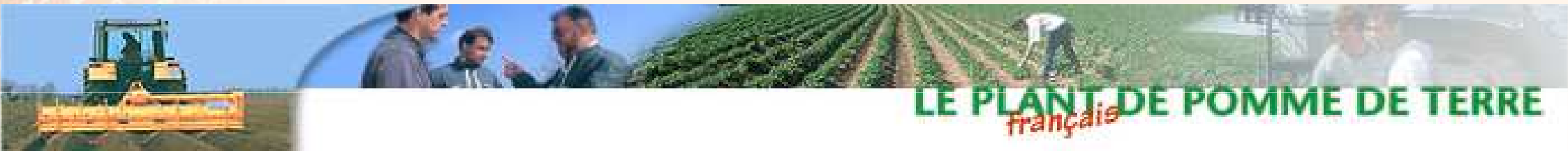
Euphresco PHYLIB meeting FN3PT involvement

Anne-Claire LE ROUX-NIO, FN3PT/INRA **Yves LE HINGRAT, FN3PT/ UMT INNOPLANT, France**
Researcher on potato quarantine diseases National coordinator of the seed potato laboratories
FN3PT leader for the Phylib project Head of R&D and co-leader of the Joint Unit INNOPLANT

Outline of the presentation



UMT INNOPLANT (FN3PT-INRA)



1. Presentation of FN3PT
2. Seed potato testing in France
3. Involvement in Phylib project

Organisation and activities of FN3PT

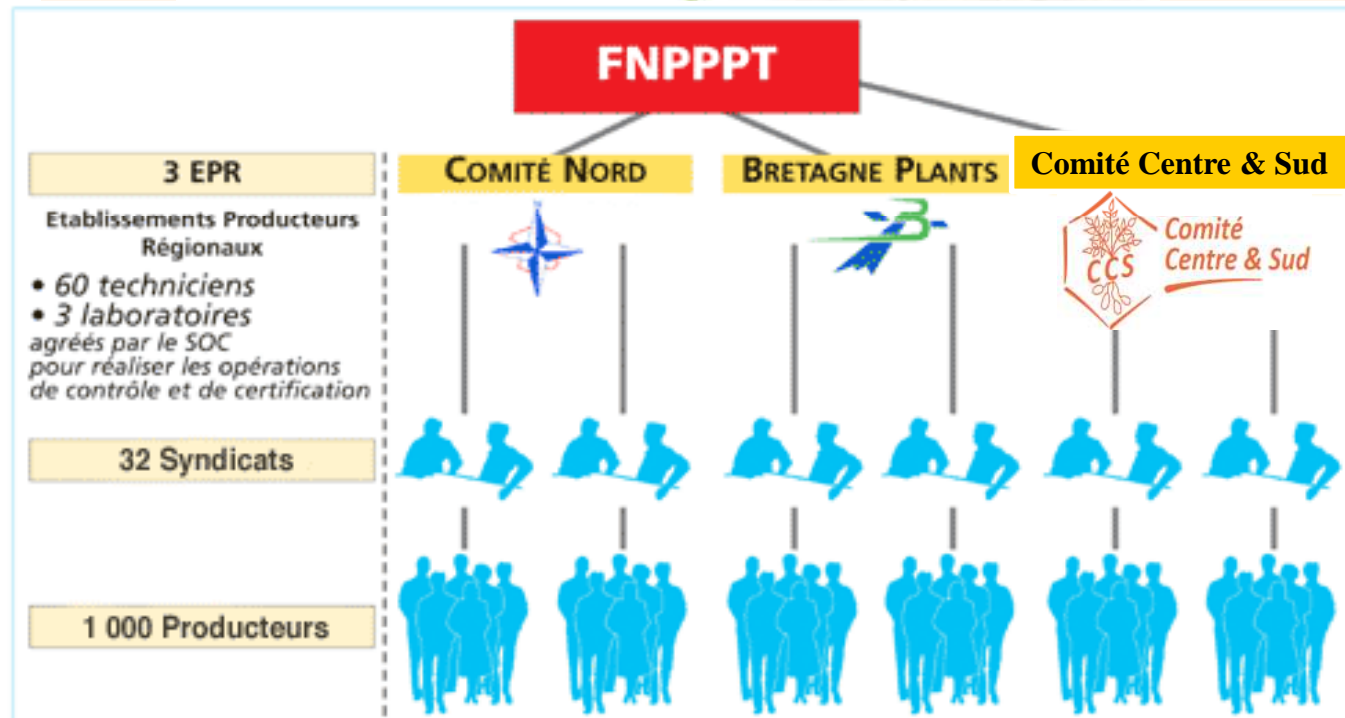


UMT INNOPLANT (FN3PT-INRA)



Main activities of FN3PT :

- Inspection and certification
- Research
- Breeding
- Prebasic production
- Technical support ->growers
- Promotion of the seed sector



3 regional agreed SOC laboratories for seed potato testing in France



UMT

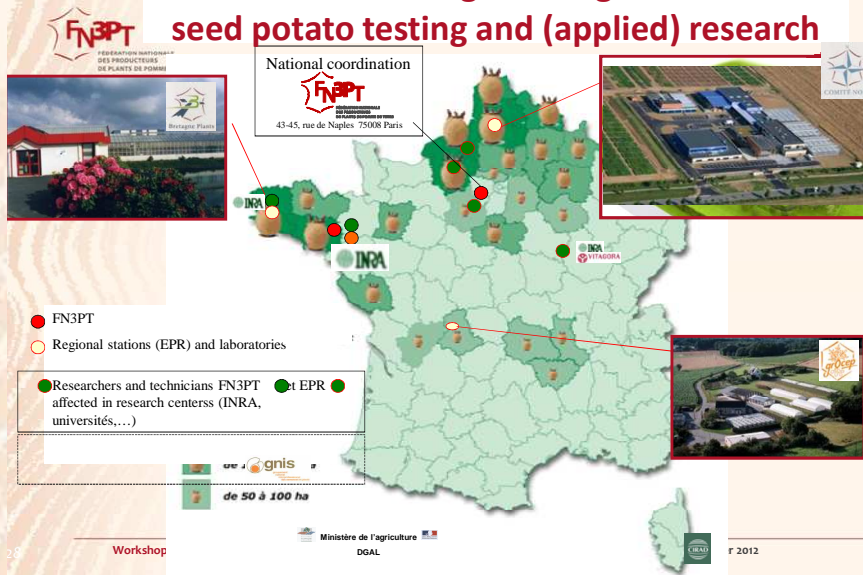


Station Hanvec (BP)



Station Achicourt (CN)

A national and 3 regional organisations for seed potato testing and (applied) research



Phylib meeting, Tenerite, February, 20-21, 2013



Station Lavergne (Grocep)

FN3PT - UMT INNOPLANT REINES



Intensive testing on seed potatoes sur plants

Annual number of analyses on seed potatoes in the FN3PT laboratories (accreditation ISO 17025 and official agreement SOC and DGAL)

Viruses

➤ More than **1,2 million ELISA tests** (1,5 M plants) on postcontrol + voluntary molecular tests

Nematodes

50 000 soil analyses for PCN (> 600 éch. for RKN)

Quarantine bacteria

About **2 x 13 000 analyses, i.e. 2,5 M tubers** tested by IF and/or PCR

Pectinolytic bacteria

> annual survey on blackleg disease (**100 samples/year**) by isolation and CAPS-PCR

Fungi & diagnostic

> **1500 microbiological or molecular tests** for the diagnostic of fungal and other diseases

Genetic fingerprints

> **1 000 samples** to check the identity of potato cultivars by genetic fingerprinting



Seed inspection for Q-diseases

	Field inspection (visual)	Lot inspection (visual)	Sampling for analyses	Routine lab testing
Bacteria				
Brown rot (<i>Ralstonia solanacearum</i>)	Yes	Yes	Yes	Yes
Ring rot (<i>Clavibacter</i>)	Yes	Yes	Yes	Yes
Nematodes				
<i>Globodera pallida</i> and <i>G. rostochiensis</i>	Yes	Yes	Yes	Yes
<i>Meloidogyne chitwoodi</i> and <i>M. fallax</i>	Yes	Yes	Yes	Yes
<i>Ditylenchus destructor</i>	Yes	Yes	If symptoms	On request
Fungi				
<i>Synchytrium endobioticum</i>	Yes	Yes	If symptoms	On request
Viruses				
TSWV	Yes	Yes	If symptoms	On request
Phytoplasmas				
Stolbur	Yes	Yes	If symptoms	On request
Other regulated Q-diseases	Yes	Yes	If symptoms	On request

Training (inspectors, growers) and communication about emergent diseases to prevent their introduction and development



Identification key and data sheets on potato diseases

3 - Coloration violacée

■ Coloration violacée **du bord des feuilles**, liée à la formation de pigments anthocyaniques en réaction à un stress ou à une attaque parasitaire, notamment :

▶ **stress lié au froid** Voir page 155

▶ **stress lié à une infection par un virus** comme le virus de l'enroulement (PLRV), Voir page 50

ou à une infection par un phytoplasme :

▶ **stolbur** Voir page 74

associée à un enroulement des jeunes feuilles, un port dressé des feuilles et la possibilité d'apparition de tubercules et stolons aériens



Phytoplasmas

PHYTOPLASMES

STOLBUR et autres PHYTOPLASMES

(PHYTOPLASME : STOLBUR, AZAR YELLOW, PURPLE TOP...)

1. Feuilles enroulées et violettes, avec tubercules aériens, coupés par le stolbur.
2. Germes filaires sur le tubercule de gauche, signes de phytoplasme de stolbur.

Les phytoplasmes, comme le stolbur, sont des organismes de type procaryotes (sans ADN) - des organismes vivants sans membrane nucléaire et sans cellule eucaryote en dehors d'un organisme hôte.

Agents responsables et transmission

La diversité des phytoplasmes et de leur gamme d'hôtes rend leur classification difficile. Maintenant, on les regroupe selon leur proximité génétique plutôt qu'en fonction de leurs symptômes.

On distingue habituellement 3 groupes principaux de phytoplasmes sur la pomme de terre :

- phytoplasmes provoquant des maladies de type **jaunisse** (stolbur, azar yellow, purple top) que l'on distingue selon les symptômes, la gamme d'hôtes ou le polymorphisme de l'ADN ;
- phytoplasmes provoquant des maladies de type **batai de sorcière** (Witches' broom).

Les maladies à phytoplasmes sont observées en Europe centrale, mais aussi en Europe du Sud, au Moyen-Orient, aux États-Unis, en Australie, en Asie, etc.

Les vecteurs principaux sont des insectes de la famille des cicadelles (*Moszkowski sp.*, *M. pascu* sp., *Rosselia sp.*), mais la transmission est possible aussi par une plante parasite, la oustille.

Le stolbur a besoin de plantes réservoirs (pomme, framboise, aubergine, aubergine, bettade, datura, liban des champs, etc.) pour servir d'hôte car la transmission par la semence semble inexistant. À la différence du batai de sorcière.

FICHES DESCRIPTIVES DES MALADIES, AVISAGERS ET DÉSORIRES SUR POMME DE TERRE

PHYTOPLASMES

Description des symptômes

Les deux groupes de phytoplasmes provoquant sur la pomme de terre les symptômes suivants :

- **Groupe jaunisse (stolbur et azar yellow) :**

Sur feuillage :

- **pigmentation violacée** des extrémités (photo 1) ;
- **de coloration jaune avec enroulement** des jeunes feuilles, port des feuilles dressé.

Par temps sec, les plantes flétrissent et mourant. Le développement de la maladie est lent en cas de temps frais et humides.

Sur les parties souterraines :

- possible **apparition de tubercules et stolons aériens** ;
- **pourriture sèche** à l'extrémité des racines ;
- **les tubercules** perdent du poids et deviennent **mous**, pourrissant ensuite en conservation. Les tubercules sous de plantes malades donnent des **germes filaires** et des pousses faibles (photo 2).

- **Groupe batai de sorcière :**
- **prolifération de nombreuses tiges grêles et érigées.**

Détection et lutte

La détection des phytoplasmes peut être réalisée par différentes méthodes : sondage biologique (greffage sur plantes indicatrices), sérologie pour certaines souches, tests moléculaires de type PCR (amplification universelles phytoplasmes ou spécifiques du stolbur) et microscopie électronique.

Le stolbur fait partie des parasites de quarantaine de l'Union Européenne, ce il fait l'objet de mesures de lutte obligatoire.

Dans les régions où les phytoplasmes sont présents, la lutte préventive consiste à détruire les cicadelles vectrices (insecticides) et les adventices, à éviter le voisinage de plantes hôtes et à utiliser des plants certifiés.

FICHES DESCRIPTIVES DES MALADIES, AVISAGERS ET DÉSORIRES SUR POMME DE TERRE

1 - Rust-coloured or brown spots

■ **Rust spots**, irregularly shaped and of variable size, usually scattered in the centre of the tuber:

▶ **internal brown spot (physiological rust)** See page 174



■ **Spraing with diffuse internal rust spots** occasionally associated with necrotic arcs or lines:

▶ **tobacco rattle virus (TRV)** See page 62

▶ or **potato mop-top virus (PMTV)** See page 48



■ **Network of rust punctuations around the vascular ring, net necrosis**, (quite a rare event), restricted to some cultivars, due to:

▶ **potato leaf roll virus (PLRV)** See page 46



■ **Brown stripes or streaks** inside the flesh of the tuber, often coinciding with the medullary rays of the tuber. Upon slicing and frying to produce potato chips, tubers exhibit very dark blotches, stripes, or streaks.

Zebra chip

"Zebra Chip" disease

This disease has been documented in several potato-growing areas, such as Central and North America (in 8 States), and more recently in New Zealand. So far, it has not been reported to be present in Europe. Recent research strongly suggests the disease to be caused by a bacterium, *Candidatus Liberibacter solanacearum*, and vectored by the potato (or tomato) psyllid, *Bactericera cockerelli*.



IDENTIFICATION GUIDE TO THE ORIGIN OF SYMPTOMS

➤ tests on doubtful samples by specialized labs

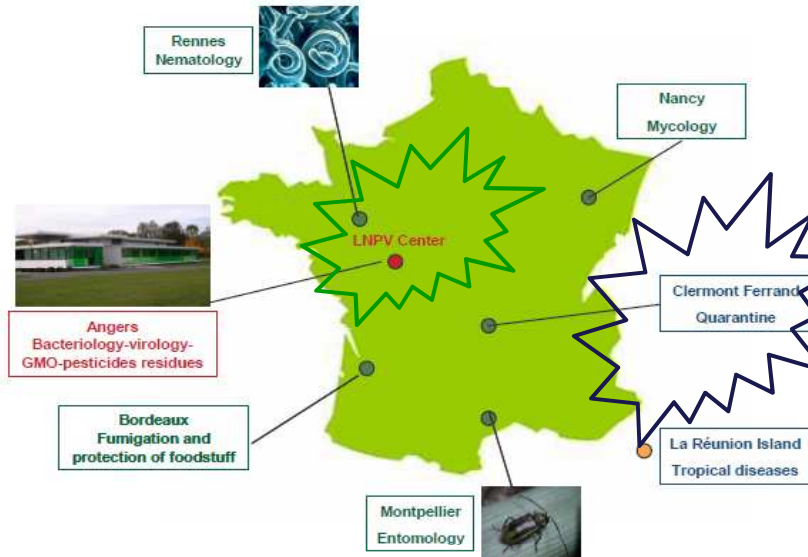
Testing for Phytoplasmas and other rare Q diseases



*Compulsory or voluntary tests
on request by specialized labs*

UMT INNOPLANT (FN3PT-INRA)

National Plant Health Laboratories (ANSES-LSV)



- **Introduction outside EU** : systematical full Q-testing at LSV potato Quarantine unit

Analyses on doubtful symptoms :
Reference LSV lab (or agreed lab)

Voluntary surveys on genebanks,
genealogical scheme.. : agreed or
approved lab

The new LNPV organisation lead to a new quality management approach to facilitate and improve:

Remarques :

No reference protocol -Universal primers and specific stolbur primers

No positive results on potato

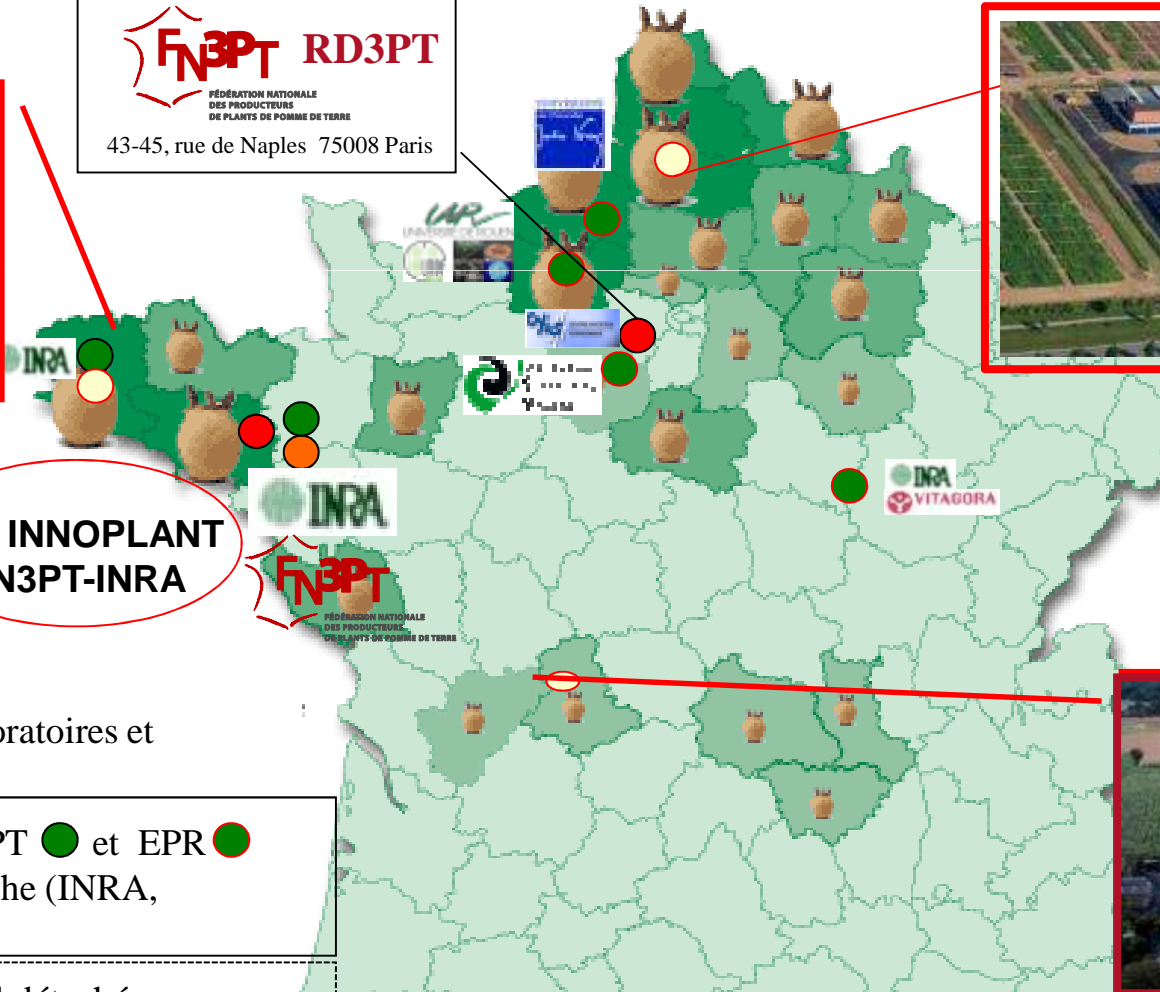


FN3PT research on seed potato



Coordination nationale
FN3PT RD3PT
 FÉDÉRATION NATIONALE
 DES PRODUCTEURS
 DE PLANTS DE POMME DE TERRE
 43-45, rue de Naples 75008 Paris

UMT INNOPLANT (FN3PT)



UMT INNOPLANT
 FN3PT-INRA

- FN3PT
- Stations régionales (OP) : laboratoires et stations expérimentales
- Chercheurs et personnel FN3PT ● et EPR ● détachés en centres de recherche (INRA, CNRS, universités,...)
- Chercheurs et personnel GNIS détachés en centres de recherche (INRA)

Ainsi que du personnel spécialisé sur le terrain pour l'appui technique

Et en partenariat avec



56 people – budget 4,5 M€

Joint Research Unit with INRA : INNOPLANT
 Qualified as the French technical institute for seed potatoes



National FN3PT research team/programmes

Markers-assisted Selection,
genetic resistance to diseases
variétale, empreinte



Ing. Yves LE HINGRAT

Researchers implanted at



UMT INNOPLANT (FN3PT)

Tec. Amandine Méar

Ing. Sylvie MARHADOUR

**Coordination
R & D**

Ing. Laurent GLAIS

Détection, characterization
and epidemiology of
**potato viruses, with focus
on PVY**



Tec. Angélique
Laurent

**Ing. Valérie
HELIAS***

Detection, characterisation
and epidemiology

**Pectobacterium
(Erwinia) and
Dickeya spp.**

13 personnes

Virology

**Production of
antibodies ELISA**

Tec. Maryse GUILLET

Thèse CNRS-FN3PT
Yannick Raoul des
Essarts



Frédéric BOULARD



**Ing. Anne-Claire
LE ROUX-NIO**



**Quarantine
diseases**

**Tuber
blemishes**



Post doc Florence
FAUREZ

Potato quarantine diseases
(*Ralstonia solanacearum* et
Clavibacter mich. sepedonicus,
Meloidogyne, ZC ...)



Tec. Emilie HUCHET 21, 2



Ing. Karima BOUCHEK*
**Tuber blemishes (*Streptomyces*,
Rhizoctonia...) and crop management**

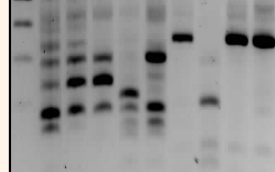
* En cofinancement : GNIS-Arvalis



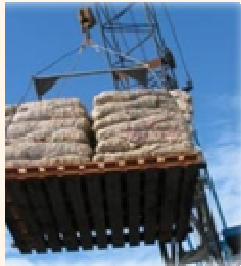
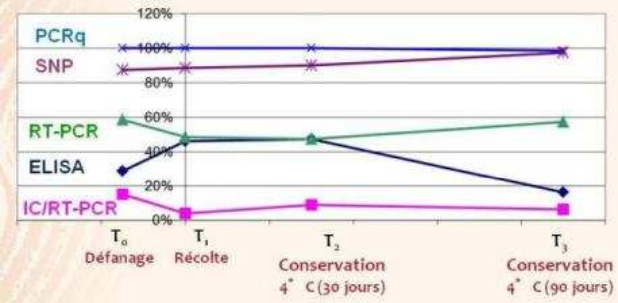
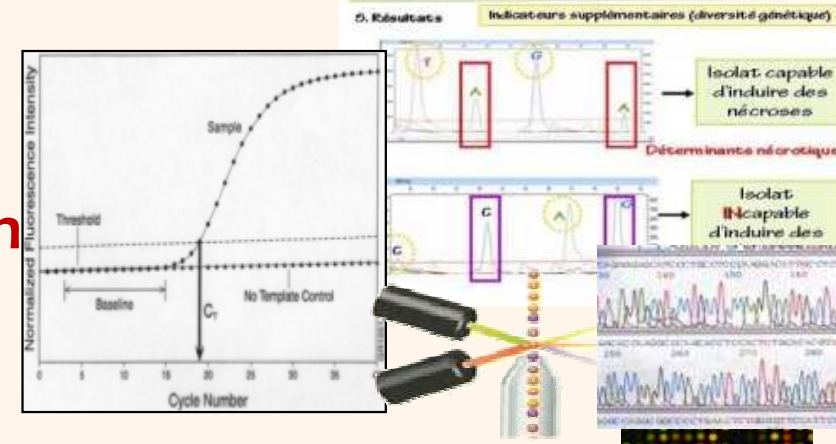
Development and validation of techniques for the detection, characterisation or quantification of potato diseases

UMT INNOPLANT (FN3PT-INRA)

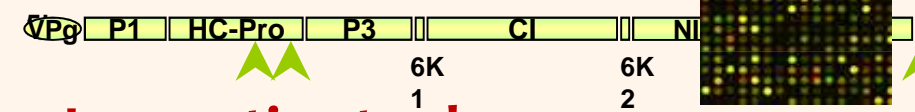
Objective : Developing new tools for the certification of seed potatoes, adapted to the evolution of techniques and of pathogens



Tools for the diagnostic, detection, characterization and quantification in plants and complex matrix

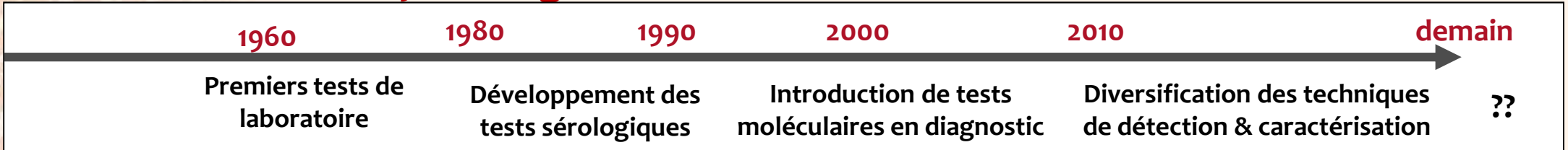


Transfert



Innovative tools, multiplexing

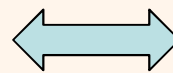
Early testing



French network of laboratories (seed potatoes)

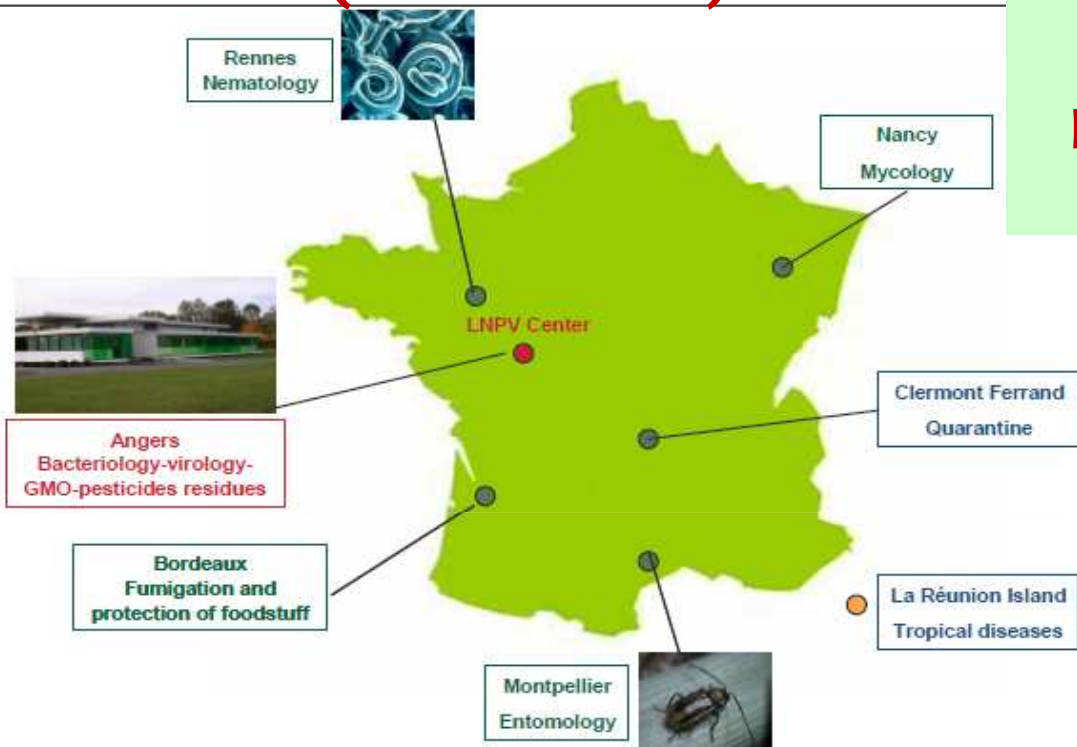
UMT INNOPLANT (FN3PT-INRA)

**National Plant Health Laboratories
(ANSES-LSV)**

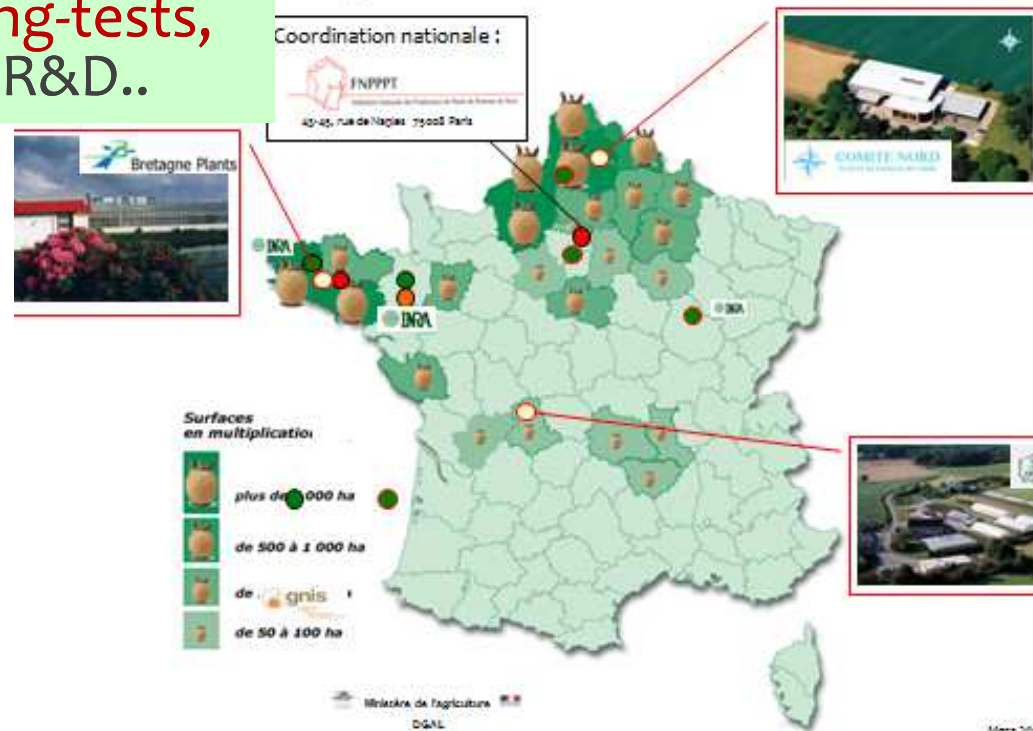


Agreed seed potato labs

Methods,
Training,
Ring-tests,
R&D..



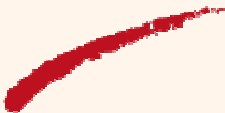
seed potato laboratories in France



The new LNPV organisation lead to a new quality management approach to facilitate and improve:

Euphresco PHyLIB - FN3PT involvement :

1. DETECTION Ls



UMT INNOPLANT (FN3PT-INRA)

	TASK	DESCRIPTION	COMMENTS	LAB(S) INVOLVED	COMMENTS	France FN3PT (INRA)
2 LIBERIBACTER SOLANACEARUM AND VECTORS						
2.2 LIBERIBACTER SOLANACEARUM						
WP 2 Detection and Diagnosis/ Identification	Evaluate current Ls tests for detection in hosts/ vectors	Conventional PCR and real time PCR		UK (SASA), CA(CFIA), NL, FR (FN3PT)	ances have successfully used 1289F & 1364R + 1311T probe for Ls detection	3,4
	Ring test of target in host tissue			CA (CFIA), NL, FR (FN3PT)		3,4

Euphresco PHyLIB FN3PT involvement :

2. VARIETAL SUSCEPTIBILITY (potato)

UMT INNOPLANT (FN3PT-INRA)

TASK	DESCRIPTION	COMMENTS	LAB(S) INVOLVED	COMMENTS	France FN3PT (INRA)	
2 LIBERIBACTER SOLANACEARUM AND VECTORS						
2.2 LIBERIBACTER SOLANACEARUM						
WP 2 Detection and Diagnosis/ Identification	Evaluate current Ls tests for detection in hosts/ vectors	Conventional PCR and real time PCR		UK (SASA), CA(CFIA), NL, FR (FN3PT)	ances have successfully used 1289F & 1364R + 1311T probe for Ls	3,4
	Ring test of target in host tissue			CA (CFIA), NL, FR (FN3PT)		3,4
WP 3 Epidemiology	Assessment of the behavior of different cultivars of potato against Ls ?	Potato cultivars (or other hosts) ?		FR (FN3PT) ?		3,4 ?



3. TRAINING AND SURVEY ?

UMT INNOPLANT (FN3PT-INRA)

TASK	DESCRIPTION	COMMENTS	LAB(S) INVOLVED	COMMENTS	France FN3PT (INRA)
2 LIBERIBACTER SOLANACEARUM AND VECTORS					
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WP 2 Detection and Diagnosis/ Identification	Evaluate current Ls tests for detection in hosts/ vectors	Conventional PCR and real time PCR	UK (SASA), CA(CFIA), NL, FR (FN3PT)	ances have successfully used 1289F & 1364R + 1311T probe for Ls detection	3,4
	Ring test of target in host tissue		CA (CFIA), NL, FR (FN3PT)		3,4
	Assessment of the behavior of different cultivars of potato against Ls ?	Potato cultivars (or other hosts) ?	FR (FN3PT) ?		3,4 ?

- *Training of inspectors*
- *Application of protocols on surveys*

Collaboration



UMT INNOPLANT (FN3PT-INRA)

- with Anses but also foreign partners
- Need discussion and collaboration in
 - ✓ improved detection protocols
 - ✓ ring-testing and positive material
 - ✓ expertise in potato evaluation

THANK YOU

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