



Plant Protection Service
*Ministry of Economic Affairs, Agriculture and
Innovation*

Overview:

Activities of NPPO-NL on CLso* recent years

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* '*Candidatus Liberibacter solanacearum*' (CLso) and Zebra chip (ZC)



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Overview

- Overview on new (2012) organisation NPPO-NL
- Motivation for activities on CLso
- Inquiry for policy on the disease
- Actions initiated
 - National Survey
 - Extension of information (Datasheet, National work group)
 - Participation in EPPO PRA
 - Validation and Implementation of test methods
 - Initiation of CLso collection



Organisation NPPO – NL (Since 2012)

Ministry: Economic affairs; Agriculture desk

2012: Food & Consumer Product Safety Authority

Division: Agriculture and Nature (= NPPO – NL)

Department: National Reference Centre (NRC)

Teams: **Pests**; **Diagnostic methods**; **Diseases**

Area: **Ent/Nem**; **Molbio/Glassh**; **Bac/Myc/Vir**



Initiation of activities NPPO-NL on CLso

2008:

New Zealand:

Publication of CLso in tomato. Later ZC in potato.

NL tomato breeders/ seed producers asked about risks, symptoms, specific identity, and diagnostics.

Also concerns were laid down by potato breeders.

2009:

Addition of CLso to the EPPO Alert list



Inquiry for policy on CLso (I)

Quick scan on CLso performed 2009.

- Potential disease impact: high (USA, NZ).
- Entry: possible but likeliness seems low
 - No known source of disease agent near to NL (2009)
 - Transmission primarily by vector: *Bactericera cockerelli* (absent in EU)
 - Pathway of clonal (Solanaceae) plant material restricted (200/29/EC)
- Establishment (long term); glasshouses and field potato: Possible
 - Presence of the vector would be requirement (in analogy with NZ)
- Risk: low, as long as the insect vector is absent
 - Quick scan on the insect vector (*Bactericera cockerelli*) needed
 - Survey in tomato to asses pest status/ confirm absence of CLso
 - Maybe a PRA needed (to be decided after Quick scan on the vector)



Inquiry for policy on CLso (II)

Follow up actions initiated 2009:

- Survey in tomato glasshouse crops on CLso

Inspection on visual symptoms

Suspect symptomatic plants to be analysed by testing

- Quick scan for *B. cockerelli* initiated



Inquiry for policy on CLso (II)

Quick scan “potato psyllid” *B. cockerelli* 2009/2010.

- Potential impact: Is a vector to CLso, and causes direct damage
- Entry: possible but uncertainty about the likeliness
 - No known source for dissemination close to NL
 - Pathway on solanaceous plants is secured (2000/29/EC)
 - Many pathways possible (e.g. also other hosts than *Solanaceae*)
 - Example of NZ where it was introduced over large distance overseas.
- Establishment: outdoors maybe transient in summer
 - Possibly invasion of glasshouses with solanaceous crops (i.e. CA, NZ)
 - Migratory habit: Overwintering is possible in e.g. Mediterranean areas
 - Wild hosts in EU areas (nightshade, *Lycium*); Limits to migration range?
- Risk: if established and CLso present, then high impact is foreseen
 - Recommendation for PRA



International developments

2010:

Finland:

Publication of CLso in carrot (*Daucus carota*)

Cooperation on diagnosis with USDA-ARS

Association of CLso with the "carrot psyllid"

Trioza apicalis



Actions initiated in NL on CLso 2010/ 2011

Preparation of datasheet on CLso.

Special attention to symptoms and appearance of vectors

Extension of knowledge on CLso in national bacteriology work group.

Members: Universities, Inspection services, seed companies,

Other stakeholders in the crop protection sector

Participation in EPPO Expert Work Group:

Pest Risk Analysis on CLso and its vectors

Muriel Suffert, Fabienne Grousset

Neil Giltrap, Joe Munyaneza, Anne Nisinen, Tarek Soliman,

Leif Sundheim, Emilio Stefani, Leon Tjou-Tam-Sin,

Dirk Jan van der Gaag

Expansion of CLso survey on tomato to CLso carrot + carrot psyllid

2009 tomato surveyed; Carrot production sites included 2010.



Pest Risk Assessment CLso + vectors 2010 (I)

Estimated probability of entry of CLso:

Very low in absence of a vector

Moderate to low if a suitable *Solanaceae*-vector is already present

Why? EU: regulations (2000/29/EC); Other EPPO area: smaller pathways

Estimated probability of establishment of CLso:

High if *B. cockerelli* is present or introduced simultaneously

Very high if other "EU" psyllids can act as vector for *Solanaceae*

Estimated probability of spread of CLso:

Very low if no vector is present

High if a vector is present or introduced simultaneously

Estimated potential economic effect of CLso:

Massive if *B. cockerelli* is present or introduced simultaneously

Comparable to that in its current area of distribution

Main source of uncertainty:

If a psyllid present in the PRA area could act as *Solanaceae*-vector or as a "bridge" between carrot and *Solanaceae*

In latter case this (2010)-PRA would need to be revised.



Folow up; Pest Risk Analysis CLso and vectors

Conclusions of the EWG were reviewed

EPPO Working Party on Phytosanitary Regulations recommended for regulation

Approval by the Council

Final conclusion:

Inclusion EPPO A1 list (pests absent in EPPO area) of CLso (Solanaceae haplotypes) and its vector *B. cockerelli*



Suspect sample; carrot survey 2011



Typical red color in leaves



Suspect sample; carrot survey 2011



2010-
2012:
no
carrot
psyllid
found



Suspect sample; carrot survey 2011



Notable lateral root growth



Survey on CLso in NL 2009 – 2012 (I)

Crops: Tomato and Carrot

Only suspect fields are sampled for testing

Detection testing by USDA-ARS,
Yakima Agricultural Research Lab – Wapato (USA)



Survey on CLso in NL 2010 – 2012 (I)

Results	# inspected sites		# Samples taken		# Positive tested	
	tomato	Carrot	tomato	Carrot	tomato	Carrot
2009	125	-	0	-	-	-
2010	128	50	0	2	-	0
2011	26	126	0	25	-	0
2012	130	130	0	42	-	-



Survey Stolbur phytoplasma in NL 2011 – 2012

Carrot survey samples CLso 2011, 2012 were also tested on phytoplasma in general, and specific on stolbur phytoplasma. phytoplasma was detected (75%), stolbur phytoplasma not.

ID by sequence analysis (c. 20% of the positive samples)

Target fragments on: the 16S rDNA and the ISR.

Blast results (NCBI):

all highly matching '*Candidatus Phytoplasma asteris*'

For further details phytoplasma: Ko Verhoeven, NPPO-NL



International developments

2011/2012:

Norway, Sweden, Spain, France:

Publication of CLso in carrot (*Daucus carota*).

USDA-ARS assisted the diagnosis in some cases.

Association of CLso with a new psyllid vector
Bactericera trigonica (Spain).



Actions initiated in NL on CLso 2012/ 2013

Participation in EUPHRESKO-IVIA test performance study
CaLsol/100 complete kit of plant print diagnostics S.L.; and
incorporated evaluation of five Real-time PCR platforms

Implementation/validation of methods for testing (planned)
Detection and identification tests; comparison and selection.

Screening of ongoing national CLso survey testing (planned)
Plant sap from survey 2012 frozen (-80°C).

Initiation of CLso collection: Two accession from USDA-ARS



Collection item CLso; graft inoculated plant



tomato

39 days,
21/18°C

Left:
negative
control



Collection item CLso; graft inoculated plant



tomato

39 days,
21/18°C

Left:
negative
control



Collection item CLso; graft inoculated plant



tomato

39 days,
21/18°C

Left:
negative
control



Collection item CLso; graft inoculated plant



tomato

39 days,
25/22°C

Left:
negative
control



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