



Liberibacter on carrot: what do we know so far?

Anne Nissinen

Anne Lemmetty

Lauri Jauhiainen

Juha-Matti Pihlava

Satu Latvala

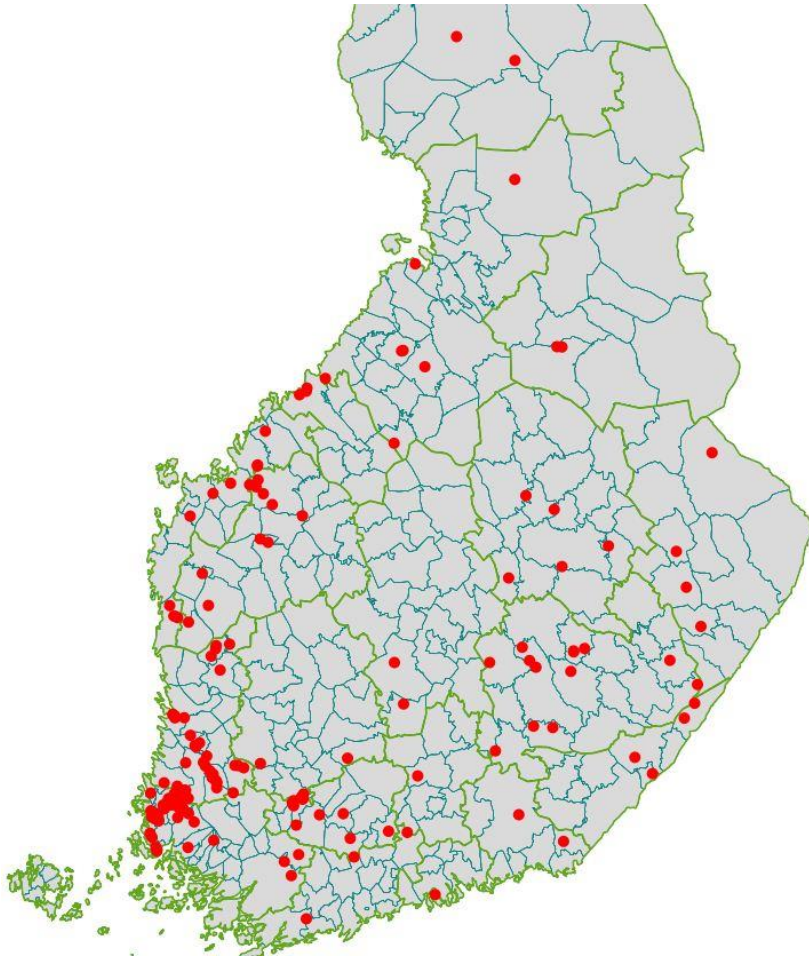
Marika Rastas

Asko Hannukkala

Minna Haapalainen Helsinki University

Minna Pirhonen Helsinki University

Carrot cultivation



Graph: Evira

- 2010: 1606,5 ha, 67257,7 tn
- average yield: 41,8 tn/ha
- organic carrot: 47 ha
- average yield: 39,5 tn/ha
- Carrot psyllid is the major pest
- carrot psyllid occurs south of line Joensuu-Oulu

Carrot psyllid damage



Photo: Anne Nissinen

- Overwintered females cause the most severe damage
- Damage index (0-5): 1h feeding 0,3; 4 h 1,9 and 24 h 2,9 (Markkula et al. 1976)
- Leaf curling occurs in 2 days (1-5 d)
- Carrot psyllid damage is thought to be caused by phytotoxic saliva (Láska 1964) but the toxin is not identified (Markkula & Laurema 1971)

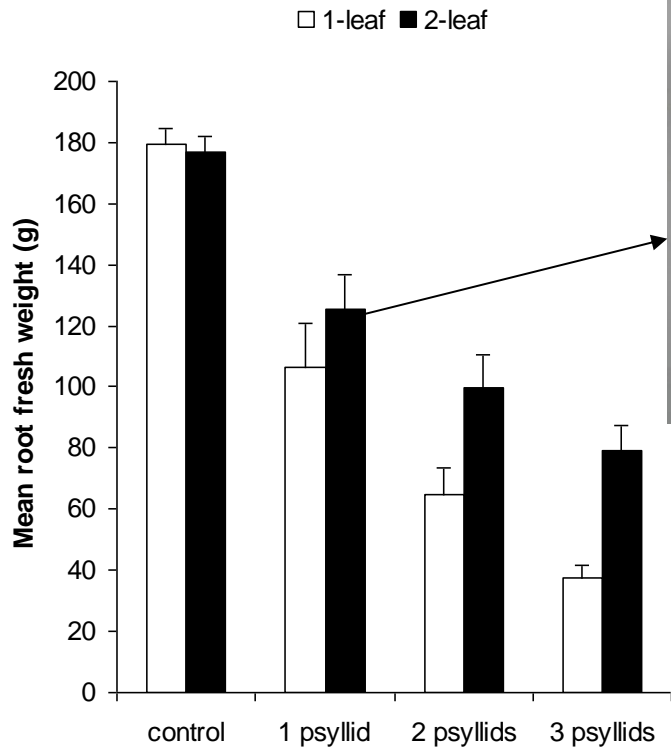
Effect of carrot psyllid feeding intensity on yield 2007



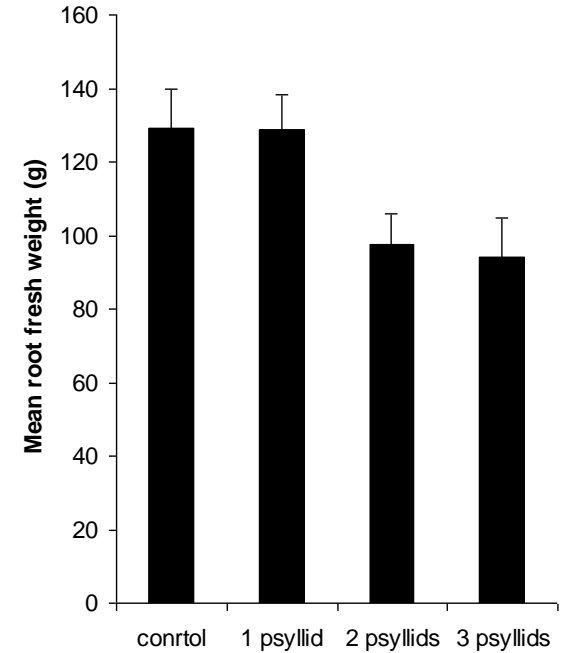
- 1-leaf stage, 2-leaf stage, 4-leaf stage
- 0-3 psyllids
- 4 blocks
- Row-column design (n=20)
- Non-harvested edge rows

Photo: Anne Nissinen

Effect of carrot psyllid feeding intensity on yield 2007



Photos: Anne Nissinen



- Nissinen et al. 2012

Phytoplasma ?

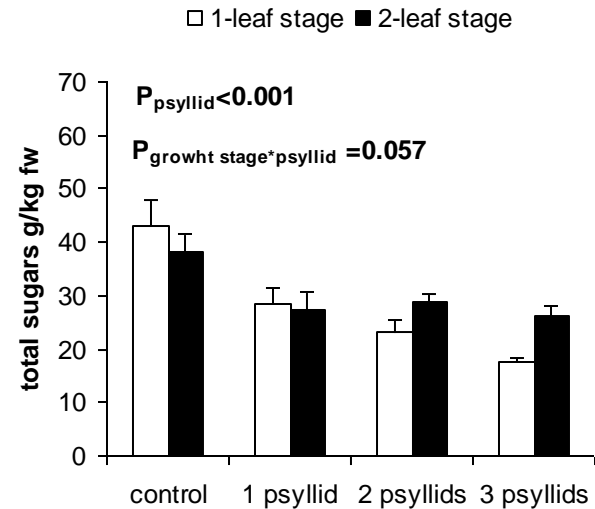
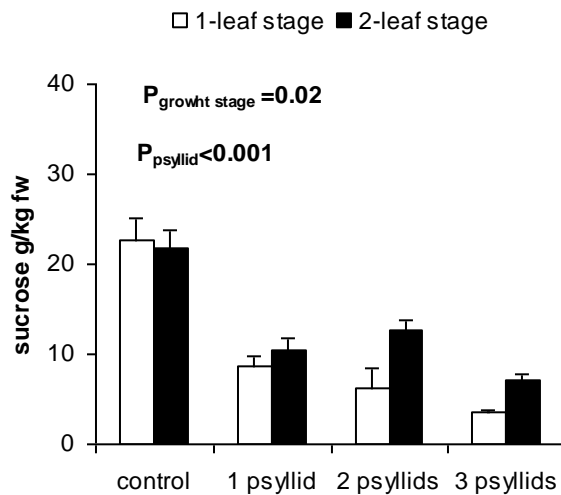


Photo: Anne Nissinen

- Phytoplasma samples (petioles leaf midribs) were taken at harvest 2007
- Nested PCR P1/16SSr and F2n/R2
- **No phytoplasmas were detected in the greenhouse experiment**
- Field-collected samples 2008: 2/20 were positive for aster yellows sub-group 16Srl-A phytoplasma

Effect of carrot psyllid feeding on sugars

Nissinen et al. 2012



***Candidatus* Liberibacter and ZC on potato**

- Studies conducted in New Zealand, US, and Mexico have associated ZC with a new species of the bacterium ***Candidatus* Liberibacter solanacearum** (or *Ca. Liberibacter psyllauros*)
- This new bacterium is related to, but different from, Liberibacter species that cause **citrus greening disease or Huanglongbing disease** (found in Florida, Brazil, and elsewhere in the world)
- Severely affects other solanaceous crops, including tomato, pepper, eggplant, *Physalis*, and tamarillo

Candidatus Liberibacter on Trioza

Munyaneza et al. 2010

Table 1. PCR testing of field-collected and laboratory-reared carrot psyllids for *Ca. L. solanacearum*. The insects were tested individually or groups of five using primer pairs OA2/OI2c and Lso/OI2c.

Source	Total number of insects	Number of samples	Number of insects per sample	Number of PCR positive samples	
				Primer pair OA2/OI2c	Primer pair LsoF/OI2c
Field-collected psyllids	205	41	5	25	15
Laboratory-reared psyllids	75	15	5	15	15
	30	30	1	21	10

Candidatus Liberibacter on Carrot

Table 2. Carrot plants and roots collected from field, laboratory, and greenhouse were tested for *Ca. L. solanacearum* by PCR, using primer pairs OA2/OI2c and Lso/OI2c.

Source	Plant parts	Symptoms	Number of samples	Number of PCR positive samples	
				Primer pair OA2/OI2c	Primer pair LsoF/OI2c
Field-collected plants	Petioles	Asymptomatic	18	1	1
		Psyllid damage and discoloration	20	16	11
		Psyllid damage only	16	5	0
Laboratory psyllid-exposed plants	Root – Top portion	Secondary roots	10	10	10
	Root – Middle portion	Secondary roots	10	10	10
	Root – Lower portion	Secondary roots	10	10	10
Greenhouse psyllid-free plants	Petioles	Asymptomatic	15	0	0

Munyaneza et al. 2010

Conclusions

- Carrot seems to be very vulnerable to carrot psyllid feeding from cotyledon to 2-leaf stage
- Carrot psyllid feeding lowers the concentration of sugars in the roots > affects the quality
- The symptoms in carrot (i.e. stunted growth, discolouration of leaves) resembled that of zebra chip in potato
- “*Candidatus Liberbacter solanacearum*” was isolated both in carrot psyllids and damaged carrots! (Munyaneza et al. 2010)
- Increased phenolic compound in the tubers of zc potatoes (Navarre et al. 2009) as well as in psyllid-damaged carrot roots (Nissinen et al. 2012)
- Increased sucrose content, but decreased starch concentration in the tubers of ZC potatoes (Gao et al. 2009) whereas decreased sucrose concentration in psyllid-damaged carrot roots (Nissinen et al. 2012)

Field survey 2011



Photo: Anne Nissinen



Photo: Marika Rastas

- 10 commercial carrot fields, 154 samples
- 5 potato fields + volunteer potatoes from carrot fields, 42 samples
- 59 sweep netting samples

Transmission trial in greenhouse 2011



Photos: Anne Nissinen



- 10 potatoes were exposed to feeding of 20 carrot psyllids for 7 days
- At the same time 10 carrots were exposed to feeding of 5 carrot psyllids for 7 days
- 5 unexposed controls in both
- Plants were tested for liberibacter 1,5 months after exposure with primer pair OA2/O12c
- All the psyllid samples were positive
- 5 of the exposed carrots were positive for liberibacter
- **None of the potatoes were positive (leaf and tuber samples)**

Transmission trial in greenhouse

- 12 insect cages, 3 control
- Psyllids from 3 different populations
- Exposure to 30 carrot psyllids for 11 days
- in each cage one potato seedling and two carrot seedlings
- Psyllids were still observed on potato



Photo: Anne Nissinen

Transmission trial in greenhouse

- No particular symptoms on potato
- Plants and psyllids tested with primer pair OA2/O12c
- **No CLs positive potatoes**
- 5 carrot samples were positive
- Of the three different psyllids populations used 80%, 44% and 29% individuals were tested positive for liberibacter



Photos: Anne Nissinen



Field survey 2012



Photo: Marika Rastas

- 6 commercial potato fields,
- 9 carrot fields of which 5 with volunteer potatoes
- 100 plant samples
- 75 sweep netting samples
- Under analyses

Damage intensity



Photos: Anne Nissinen

- Does the amount of bacterial titer in the psyllid affect the damage intensity?
- 3-l pots, drip irrigation
- Exposure for 3 days
- Field-collected population 2012
- Mostly females (31)
- Only 15 males
- Number of leaves counted and height measured weekly
- At harvest root fresh and dry weight, leaf weight, root diameter and length
- qPCR from both psyllids and plants



Damage symptoms

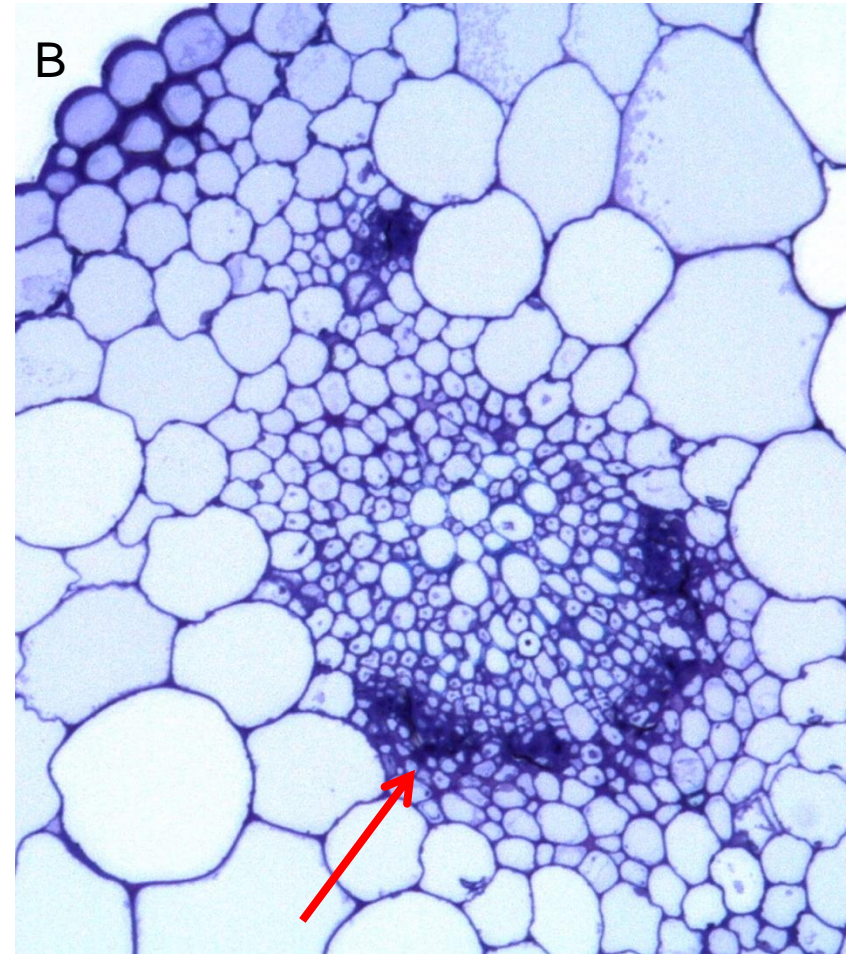
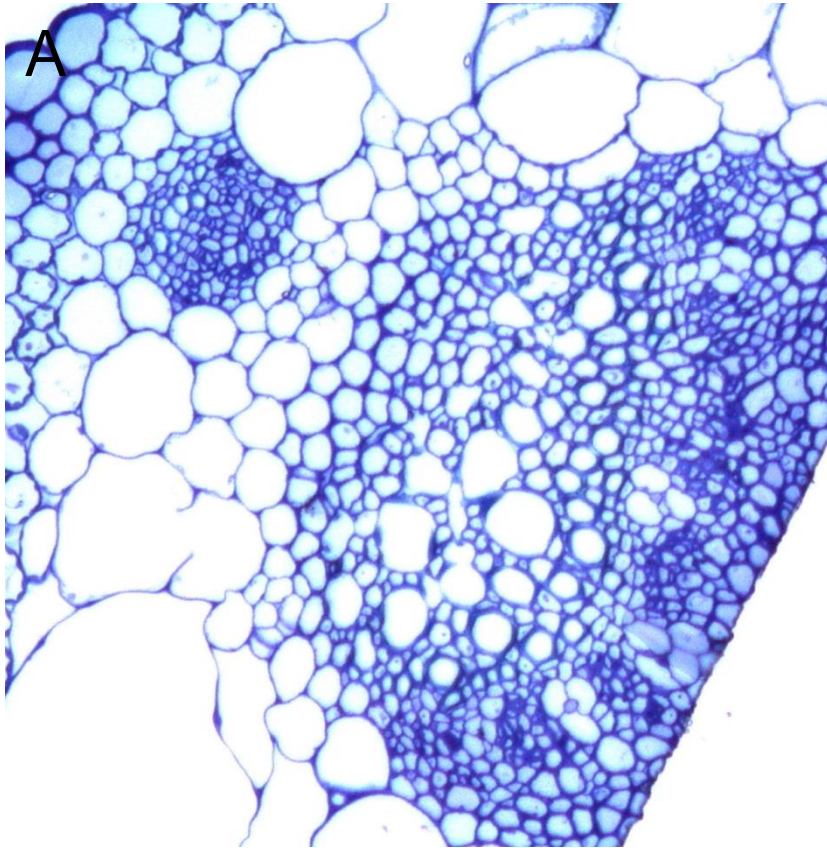
Uninfected female



Infected female



Photos: Annen Nissinen



Photos : Minna Haapalainen

Histological staining of the main vascular bundle of carrot petiole:
A) healthy control, B) symptomatic carrot. Note the dark
colouring of the phloem tissue in B.

Conclusions

- In greenhouse no transmission of CLs to potatoes by carrot psyllids was shown
- Carrot psyllid feeding and the bacterial titer both affect carrot root weight
- Bacterial titer has no effect on leaf weight
- Carrot psyllid feeding and number of nymphs increased the number of damaged leaves, but the bacterial titer had no effect on it, which suggest that the leaf curling is caused by insect feeding (discussed in Nissinen et al. 2012) and the leaf discoloration by the bacteria.
- When the number of discolored leaves increases, it probably decreases the photosynthesis (see Gao et al. 2009) which affects the root weight
- Blockage of phloem tubes by the bacteria affects the translocation of sugars from source leaves which also may reduce root weight

Future prospects 2013

- Transmission experiment with dodder
- Transmission experiment with carrot psyllids
- Field survey on potatoes continues
- Pesticide testing?

Acknowledgements



Photo: Anne Nissinen

- Senja Räsänen
- Senja Tuominen
- Outi Järvinen
- Jaana Grahn
- Aila Siren
- Panpan Jiang
- Timo Väätäinen
- Timo Jaska
- Niko Jalava

Acknowledgements



JOSEPH E. MUNYANEZA, TONJA W.
FISHER, VENKATESAN G. SENGODA,
STEPHEN F. GARCZYNSKI

USDA-ARS Yakima Agricultural
Research Lab