

The importance of psyllids (Hemiptera Psyllidae) as vectors of phytoplasmas in pome and stone fruit trees in Austria

Christa LETHMAYER, Hermann HAUSDORF, Betty SUAREZ-MAHECHA, Helga REISENZEIN

Austrian Agency for Health and Food Safety, Institute for Plant Health, Spargelfeldstraße 191, 1220 Vienna, Austria

Abstract

To study the occurrence and distribution of psyllids as potential vectors of European stone fruit yellows (ESFY), pear decline (PD) and apple proliferation (AP) a survey was conducted in Austrian orchards during the last years. Specimens were collected by using the beating tray method in apricot, pear and apple tree orchards. The obtained psyllids were analysed by PCR and RFLP assays for the presence of phytoplasmas. Molecular analyses showed few infections of *Cacopsylla pruni* with 'Candidatus Phytoplasma prunorum' and few individuals of *Cacopsylla pyricola*, *C. pyri* and *C. pyrisuga* were carrier of 'Candidatus Phytoplasma pyri'. The data presented in this study are a preliminary report because analyses of apple psyllids are still in progress.

Key words: *Cacopsylla* spp., European stone fruit yellows, pear decline, apple proliferation.

Introduction

European stone fruit yellows (ESFY), pear decline (PD) and apple proliferation (AP) are quarantine diseases associated with phytoplasmas ('Candidatus Phytoplasma prunorum', 'Ca. P. pyri' and 'Ca. P. mali'), which are responsible for great economic losses in fruit production (Seemüller and Schneider, 2004). The spread of these phytoplasmas is due to infected planting material or insect vectors, especially psyllids (Hemiptera Psyllidae). The occurrence of these diseases has been described in Austria by Richter (1999), Spornberger *et al.* (2006), Steffek and Altenburger (2008). A literature review revealed a lack of data on potential vectors in Austria. The aim of this study was to gain more information on the occurrence of these potential vectors and the phytoplasma infection status of psyllids from pome and stone fruit trees in Austria.

Materials and methods

Investigation sites were located in the Eastern part of Austria - 10 apricot orchards in Lower Austria, Burgenland and Vienna, 5 pear orchards and 3 apple orchards in Lower Austria. Samples were collected by using the beating tray method with 100 beats respectively 100 branches (trees) per sampling date and orchard. Psyllid captures were done in the period from March to July, on apricot trees in 2005 and 2006, on pear and apple trees in 2009 and 2010. Collected psyllids were identified according to Ossiannilsson (1992) and Burckhardt and Jarausch (2007).

Molecular analyses for phytoplasma infection of psyllids were carried out with 1 to 8 individuals per sample taken for testing: CTAB-method for the DNA-extraction of psyllids (Maixner *et al.*, 1995), qualitative PCR for the detection of phytoplasmas in the psyllid samples using universal primers fU5/rU3 (Lorenz *et al.*, 1995) and then for nested PCR using phytoplasma specific primer pairs P1/P7 primer (Deng and Hiruki, 1991; Schneider

et al., 1995) and f01/r01 primer (Lorenz *et al.*, 1995) respectively. RFLP assays using restriction enzymes *SspI* and *RsaI* were applied to discriminate among the three fruit tree phytoplasmas AP, PD and ESFY (Tedeschi *et al.*, 2009).

Results

The plum psyllid *Cacopsylla pruni* was found in all investigated apricot orchards. The first report of *C. pruni* on apricot trees in Austria was mentioned in Lethmayer and Hausdorf (2005). Interesting was the high number of *Cacopsylla melanoneura* on apricot trees at some investigation sites which was due to hawthorn hedges near the apricot orchards. The three pear psyllid species *Cacopsylla pyricola*, *Cacopsylla pyri*, *Cacopsylla pyrisuga* and the hawthorn psyllid *C. melanoneura* were the main species on pear trees. *C. pyricola* was the most abundant species. On apple trees the psyllids *C. melanoneura* and *Cacopsylla picta* were mainly captured.

Molecular analyses of *C. pruni* showed five positive samples (with 30 individuals in total) out of 37 tested samples (with 142 individuals in total) with 'Ca. P. prunorum'. These samples originated from four sites in Lower Austria. All individual of *C. melanoneura* caught on the apricot and pear trees tested negative for phytoplasmas. All three pear psyllid species, *C. pyricola*, *C. pyri* and *C. pyrisuga*, were found infected with 'Ca. P. pyri' comprising 16 positive samples (with 48 individuals in total) out of 33 tested samples (with 118 individuals in total). Positive samples were obtained at three pear sites in Lower Austria. First analyses showed that the all psyllids collected on apple were infected with 'Ca. P. mali'. An overview of the molecular analyses is given in table 1.

Other studies have already confirmed that the psyllid species which were tested positive in our study are vectors of the respective phytoplasmas (reviewed by Jarausch and Jarausch, 2010).

Table 1. Results of the RFLP analyses of the *Cacopsylla* samples taken in fruit tree orchards in Austria.

date	orchard/ fruit tree	psyllid species	positive (+)/negative (-) tested for		
			ESFY	PD	AP
2005	apricot	<i>C. pruni</i>	+	-	-
2006	apricot	<i>C. pruni</i>	-	-	-
2006	apricot	<i>C. melanoneura</i>	-	-	-
2009	pear	<i>C. pyricola</i>	-	+	-
2009	pear	<i>C. pyri</i>	-	+	-
2009	pear	<i>C. pyrisuga</i>	-	+	-
2009	pear	<i>C. melanoneura</i>	-	-	-
2009	apple	<i>C. melanoneura</i>	-	-	-
2010	apple	<i>C. melanoneura</i>	-	-	-
2010	apple	<i>C. picta</i>	-	-	-

C. pyrisuga has been found infected with 'Ca. P. pyri' (Kucerova *et al.*, 2007) but its ability of transmission is still not verified (Jarausch and Jarausch, 2010). Due to the geographical position of Austria it is interesting which of the two psyllid species found on apple can be identified as main vector for transmission of AP in Austria. First investigations did not indicate a particular vector capacity for 'Ca. P. mali' by *C. melanoneura* or *C. picta*. However, due to the low number of investigated apple psyllids further analyses are necessary to clarify the vector role and therefore this issue is still in progress.

Discussion

One of the main phytosanitary measures for preventing phytoplasma diseases spread is the control of their vectors. The use of insecticides in sustainable production methods is restricted. Therefore, knowledge on the vectors (mainly psyllids), their distribution and biology is strongly needed for control strategies, especially for new approaches on integrated control strategies.

Acknowledgements

We are very grateful to all growers who made their orchards available to us for carrying out the investigations and to Christoph J. Mayer for advice in psyllids identification. The present work has been carried out in the frame of COST action FA0807 "Integrated Management of Phytoplasma Epidemics in Different Crop Systems".

References

- BURCKHARDT D., JARAUSCH W., 2007.- Bestimmungsschlüssel für Psylliden auf Rosaceen in Mitteleuropa.- [online] URL: <http://www.psyllidkey.info> [accessed 20 March 2011].
- DENG S., HIRUKI C., 1991.- Genetic relatedness between two non-culturable micoplasmalike organisms revealed by nucleic acid hybridization and polymerase chain reaction.- *Phytopathology*, 81: 1475-1479.
- JARAUSCH B., JARAUSCH W., 2010.- Psyllid vectors and their control, pp. 250-270. In: *Phytoplasmas: Genomes, Plant Hosts and Vectors* (WEINTRAUB P. G., JONES P., Eds).- CAB International, Wallingford, UK.

- KUCEROVÁ J., TALÁČKO L., LAUTERER P., NAVRÁTIL M., FIALOVÁ R., 2007.- Molecular tests to determine 'Candidatus Phytoplasma pyri' presence in psyllid vectors from a pear tree orchard in the Czech Republic - a preliminary report.- *Bulletin of Insectology*, 60(2): 191-192.
- LETHMAYER C., HAUSDORF H., 2005.- Der Pflaumenblattsauger: Überträger der Steinobstvergilbungskrankheit.- *Besseres Obst*, 7: 8-9.
- LORENZ K.H., SCHNEIDER B., AHRENS U., SEEMÜLLER E., 1995.- Detection of the apple proliferation and pear decline phytoplasmas by PCR amplification of ribosomal and non ribosomal DNA.- *Phytopathology*, 85: 771-776.
- MAIXNER M., AHRENS U., SEEMÜLLER E., 1995.- Detection of the German grapevine yellows (Vergilbungskrankheit) MLO in grapevine, alternative hosts and a vector by a specific PCR procedure.- *European Journal of Plant Pathology*, 101: 241-250.
- OSSIANNILSSON F., 1992.- *The Psylloidea (Homoptera) of Fennoscandia and Denmark*.- Brill Verlag, Leiden, 347 pp.
- RICHTER S., 1999.- Apricot chlorotic leaf roll - first report in Austria, diagnosis and epidemiology of a quarantine pest.- *Mitteilungen Klosterneuburg, Rebe und Wein, Obstbau und Früchteverwertung*, 49(6): 245-249.
- SCHNEIDER B., SEEMÜLLER E., SMART C. D., KIRKPATRICK B. C., 1995.- Phylogenetic classification of plant pathogenic mycoplasma-like organisms or phytoplasmas, pp. 369-380. In: *Molecular and Diagnostic Procedures in Mycoplasmaology, Vol. I* (RAZIN S., TULLY, J. G., Eds).- Academic Press, San Diego, CA, USA.
- SEEMÜLLER E., SCHNEIDER B., 2004.- 'Candidatus Phytoplasma mali', 'Candidatus Phytoplasma pyri' and 'Candidatus Phytoplasma prunorum', the causal agents of apple proliferation, pear decline and European stone fruit yellows, respectively.- *International Journal of Systematic and Evolutionary Microbiology*, 54: 1217-1226.
- SPORNBERGER A., STEFFEK R., RÖSLER M., 2006.- Neues zu Feuerbrand, Birnenverfall und Apfeltriebsucht – Bericht einer internationalen Streuobsttagung in Dossenheim.- *Besseres Obst*, 2: 12-14.
- STEFFEK R., ALTENBURGER J., 2008.- Eine Quarantänekrankheit erkennen: Dem Birnenverfall auf der Spur.- *Besseres Obst*, 9: 4-5.
- TEDESCHI R., LAUTERER P., BRUSETTI L., TOTA F., ALMA A., 2009.- Composition, abundance and phytoplasma infection in the hawthorn psyllid fauna of northwestern Italy.- *European Journal of Plant Pathology*, 123: 301-310.

Corresponding author: Christa LETHMAYER (e-mail: christa.lethmayer@ages.at), Austrian Agency for Health and Food Safety, Institute for Plant Health, Spargelfeldstraße 191, 1220 Vienna, Austria.