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# The Past and Present of Pear Protection Against the Pear Psylla, *Cacopsylla pyri* L.

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### 1. Introduction

The pear psylla, *Cacopsylla pyri* L. (Hemiptera Psyllidae), is known in Europe for its extended infestations which may cause heavy economical losses to most pear growing regions. Pear (*Pyrus sp.* L.) is the second most relevant fruit species of temperate regions: the first one is apple (*Malus domestica* L.) and the third one is peach (*Prunus persica* L.). The top three world regions for pear production are China, Europe and North America. Total European pear production is presently stable around 2.6 million tones, according to 2010 data from the Fruit and Vegetable Service Center (CSO, Ferrara, Italy). In Europe, Italy and Spain are the largest producers, respectively with 35% and 20% of the total production. Pear production in France is decreasing (8%), mainly as a result of fireblight on "Passe Crassane", a highly susceptible variety. Until 2007, in the Netherlands and Belgium there was an increase in production with extensive planting of the "Conference" cultivar, but the first signs of saturation of the "Conference" market started to appear in 2007. The total pear production of the Netherlands and Belgium presently amounts to 9%.

All commercial varieties in Europe belong to the species *Pyrus communis* L. and they are all susceptible to *C. pyri*. The susceptibility increases when orchard techniques are aimed to maximize fruit production, such as the high density of plants per hectare, the large use of fertilizers and the intense irrigation (Fig. 1).

## 2. Damages caused by C. pyri

The damages that *C. pyri* may induce to pear trees are classified in two main types: 1) direct damages, weakening the plant by subtraction of nutrients; when the pest attack is intense, the plant wastes away with reduced production; 2) indirect damages, due to the production of a large amount of honeydew on which sooty molds develop (russetting fruits, Fig. 2), and also to the possible transmission of phytoplasms (Fig. 3).

In the first case the most damaging stages are the nymphs of all instars because of the high amount of honeydew (produced especially in spring and summer) dripping on everything including fruits. Besides lowering the fruit market value, honeydew favours the growth of sooty molds caused by saprophytic fungi, in turn causing indirect injury to the plant. Sooty molds actually induce alterations of photosynthesis, disruption of metabolism, leaf curling and premature loss, together with lower production. Prolonged attacks and intense weakening by *C. pyri* may lead to plant death.



Fig. 1. Pear orchard with high plant density.

Unlike nymphs, adults of C. pyri (similarly to those of the congener species C. pyricola) are responsible for the transmission of the phytoplasma of "pear decline". The phytoplasma is acquired by the psylla upon feeding on an infected plant and transmitted to an healthy one by salivation during feeding on phloem (Carraro et al., 1998). The transmission mechanism is persistent-propagative because the phytoplasm reproduces in the insect body. The acquisition and inoculation of the pathogen requires for the vector insect to feed for 1-2 hours on phloem of infected plants. A latency period in the vector (about 1-2 weeks) follows the acquisition of phytoplasma in which the pathogen circulates and multiplies until it reaches the salivary glands. The first symptom of the disease appears during the summerautumn period, when the leaves exhibit a red-purple hue contrasting with the yellow-green hue of senescent leaves of healthy plants. The leaves also have a stiff lamina with the edges curled upside and the apex folded downwards, and they fall prematurely beginning from the apical ones. In the next spring the infected trees show smaller, light green leaves with upward edges ("transparent canopy"). In some cases a sudden wilting is observed: although still on shoots, leaves become brown and dry. The tree may die within few days or weeks. Some authors (Giunchedi & Refatti, 1997; Davies et al., 1998) showed that some plants undergo recovering during the winter dormancy because of degradation of aboveground phloem elements (Schaper & Seemüller, 1982). Moreover, the quinces Cydonia oblonga Mill used as rootstocks rarely allow the phyoplasma survival between vegetative cycles, unlike P. communis rootstocks which allow phytoplasma reproduction within roots in winter and in the next spring, and the following spreading of the pathogen in aboveground phloem elements. Therefore in pear trees grafted to quince rootstocks the phytoplasma population, in absence of reinfection from overwintering psylla adults, may progressively decrease, disappearing within some years.



Fig. 2. Damages to fruits due to sooty molds.



Fig. 3. Damages caused by phytoplasmas.

## 3. The key to improve C. pyri control is the detailed knowledge of its life cycle

In Europe *C. pyri* shows 4-5 generations per year and overwinters as adult in reproductive diapause. The adult winter forms appear at the beginning of September (Fig. 4) (Civolani & Pasqualini, 2003) and overwinter individually or in small groups sheltered in cracks of the

tree bark, at branch crossing or at the base of buds. As soon as the weather conditions improve, the winter forms leave their shelter and reach the new apical leaves. Here they puncture the buds with their stylets, sucking and excreting fecal droplets which accumulate near the proctiger. In winter the adult male produces active sperms stored in the spermatheca and is therefore ready to mate and inseminate the female. However, the eggs can be fertilized only when oocytes reach maturity (Bonnemaison *et al.*, 1956). In January all females are mature, ready to mate and lay eggs. The main factor affecting the physiology of ovopository apparatus is temperature, which must be higher than 10° C for two consecutive days (thermal quiescence) (Nguyen, 1975).



Fig. 4. C. pyri adult winter form.



Fig. 5. *C. pyri* adult summer form.

From eggs laid in winter months, simultaneously to bud opening, the first instar nymphs emerge, infesting the new vegetation. Later, in April-May, the adult summer forms appear (Fig. 5) whose females lay a large number of eggs (Fig. 6) hatching in the second half of May. The first instar (Fig. 7) and late instar nymphs (Fig. 8) live on the shoots, excreting a large amount of honeydew responsible for heavy damages to the plant. The next generations overlap with all developmental stages until autumn. Aestivation is also observed during the summer months.



Fig. 6. C. pyri eggs.



Fig. 7. Newly hatched *C. pyri* nymphs



Fig. 8. C. pyri late instar nymphs.

## 4. C. pyri Integrated pest management

In Europe this pest was considered of secondary importance until 1960, but its population rapidly increased simultaneously to heavy use of pesticides in pear orchards and intensive agriculture. The need to control pear infestations required the repeated use of different pesticides and several control strategies, but in most cases the efficiency of control progressively decreased, first because of the great adaptability and survival of the insect to the active ingredients, and second because of the negative effects on beneficial insects caused by excessive use of non-selective toxic ingredients. In this way the psylla population freely increased, leading to conditions in which the pest was hard or impossible to control. Presently the defence against *C. pyri* is mainly based on integrated pest management (IPM), supported by natural control aimed to equilibrate the complex biological relationships of the field community. First of all, *C. pyri* control must follow IPM guidelines to optimize the activity of natural control agents and to reduce the chances of developing pesticide resistance by insect populations.

Among the basic strategies there are the "good agricultural practice" (GAP) techniques, such as reducing excessive plant growth and avoiding overstimulation by fertilizers or incorrect pruning. Also the general strategies to control other pest species may play a relevant role in psylla development and population increase. For example, the technique of mating disruption and the use of granulosis virus (CpGV) employed to control *Cydia pomonella* L. (Lepidoptera Tortricidae), favours the activity of entomophagous fauna by reducing the impact of synthetic products on the orchard, and may also help to reduce the treatments against *C. pyri*. However, in the last decade the chemical control has been the key defence strategy against the pear psylla in intensive pear orchards.

The main strategies of chemical control against *C. pyri* performed in the last 20 years in integrated and conventional farms are listed below. Each strategy has advantages and disadvantages, therefore its efficiency depends both on the active ingredients employed and the weather conditions at the time of treatment.

#### 4.1 Autumn treatment

This treatment, performed at leaf fall, requires pesticides active against adult winter forms such as those belonging to the pyrethroid family (the same principles are also used in late winter against the same generation). These pesticides are completely non-selective, therefore dangerous for the beneficial insects: for this reason the treatment must be performed not too early (in October) when most individuals of *Anthocoris nemoralis* F. (Hemiptera Anthocoridae) are still active on the pear trees, but only at complete leaf fall (late November or early December), when *A. nemoralis* populations have already found shelter in bark crevices while *C. pyri* adult winter forms are still active on plants (Fig. 4) (Civolani, 2000; Civolani & Pasqualini, 2003).

Synthetic pyrethroids exhibit a very high abatement-contacticide action on psylla wintering adults. Table 1 shows the results of tests with the active ingredient ciflutrin: the abatement activity on adults is such that no egg deposition occurs in the next spring. Notwithstanding the good results that may be obtained by pyrethroid treatment in autumn (and also in late winter, as shown below) these strategies, common in France on *C. pyri* and in North America on *C. pyricola*, in the Italian pear growing regions are discouraged: here the pest population, after an initial sharp decline, soon recovers and increases again in May, reaching the economic threshold for spring-summer treatments. This event could be explained by the faster recovery of the pest in spring because the natural control by its predator *A. nemoralis* is limited. Moreover, considering only the pesticide efficiency, the results are strictly dependent on seasonal conditions. Indeed, if an early frost occurs at the beginning of autumn, most psylla adults take shelter early and survive to the late autumn treatments. Given the need to preserve auxiliary insect populations, an untimely autumn treatment with broad spectrum pyrethroids could be very dangerous for them and therefore is strongly discouraged in relevant pear growing areas such as Emilia-Romagna (Northern Italy).

Experimental tests have been performed to evaluate other active principles such as mineral oils and neonicotinoids. According to tests performed in 2001-2002 in the province of Ferrara (Emilia-Romagna), good results were obtained with mineral oil alone or associated to imidacloprid (Table 1) (Civolani, 2000), although for auxiliary insect preservation the use of neonicotinoids is not suggested against *C. pyri* in European pear growing areas, unlike North America where is found in field control guidelines against *C. pyricola*.

	Treatment date	Eggs per 100 bubs	
untreated control		170	
mineral oil	29 November 1999	30	
mineral oil + imidacloprid	29 November 1999	12	
ciflutrin	29 November 1999	0	
mineral oil	3 March 2000	53	
mineral oil + imidacloprid	3 March 2000	65	
ciflutrin	3 March 2000	69	

Table 1. Results of a field test with a pyrethroid and mineral oil with or without a neonicotinoid (Civolani, 2000).

#### 4.2 Late winter treatment

This treatment is presently based on broad spectrum pyrethroids whose purpose is to break down the population of females emerging from winter shelters and about to lay eggs.

Formerly the highly toxic 4,6-dinitro-o-cresol (DNOC) was used, until finally banned in 1999. With a biological activity extended up to 15 days, DNOC was characterized by a strong contact activity especially towards eggs newly laid or about to be laid (Barbieri et al., 1986; Pollini et al., 1992). On pear psylla, the pyrethroids are exclusively active on adults through contact associated to an anti-feeding effect. The pyrethroid treatment is generally performed at pear bud swelling stage or at the latest when they break. The eggs are laid by *C. pyri* at the end of the thermal quiescent period, corresponding to a maximum temperature above 10° C for two consecutive days. The late winter strategy considerably lowers the amount of initial psylla population on pear trees, therefore the first generation is extremely reduced.

Concerning the pesticide activity, the best results of this strategy are obtained after a mild winter, because in these conditions almost all adults leave their shelters at the time of treatment. On the contrary, frost waves at the end of winter interrupt and delay the emergence of adults, reducing the efficiency of the treatment.

The control strategy based on late winter treatments with pyrethroids was recently disputed first because of the toxicity of the active ingredients involved, but mostly because the sharp reduction of the psylla first generation could starve the anthocorids, interfering with their settlement during early plant growth in spring. To avoid the side effects of pyrethroids, in the last ten years alternative solutions to synthetic pesticides have been repeatedly tested against the overwintering generations, and among them kaolin and some oily compounds. Kaolin, a white, non-abrasive, fine-grained allumosilicate mineral that is purified and sized so that it can be easily dispersed in water, creates a mineral barrier on plants that prevents oviposition and insect feeding (Fig. 9 and Fig. 10) (Puterka et al., 2000). The efficiency of kaolin is shown by the results obtained in two experimental trials in 2001 and 2002 (Pasqualini et al., 2003). These authors found that a double treatment with kaolin affected egg laying of overwintering C. pyri by hindering their anchorage on the leaf surface and inhibiting host-plant acceptance (Table 2). It was also found that the body and wings of some adults became soiled, making insects less mobile and preventing them from reaching the laying site (host location) on plants. Indeed, the kaolin-treated plants were almost completely free of nymphs (Table 2). Due to the high mobility of *C. pyri* adults, the effects of kaolin treatment on the summer population were not assessed in this small plot trial. A larger plot trial was therefore performed in 2004 and again kaolin showed a very high control efficiency (Daniel et al., 2006). At the end of June 2004 some C. pyri individuals were observed in the kaolin treated plot, but the population density tended to be lower than that in the plot treated by a standard spirodiclofen strategy. In kaolin treated plants C. pyri was kept under the economic threshold until harvest (Daniel et al., 2006), therefore it could be an

Some oily compounds could also be used in this period to interfere with egg deposition by *C. pyri* adults. It has been known from a long time that mineral oils and oily compounds could have negative effects on psylla egg deposition (Zwick & Westigard, 1978). A Turkish researcher (Erler, 2004) tested four types of oils, namely cotton seed oil, neem oil, fish-liver oil and summer oil, observing a delay in egg laying of about four weeks for fish-liver oil and summer oil, but of only one or two weeks for cotton seed oil and neem oil, probably depending on the stability of the oily material in open field. Other field trials aimed to interfere with egg laying by *C. pyri* adult winter forms were performed in 2001 and 2002 in Italy with pure mineral oil ("dormant oil"), obtaining a good reduction of the number of eggs laid (Pasqualini et al., 2003) (Table 2).

interesting alternative control strategy for this pest in organic and IPM orchards.



Fig. 9. Pear trees treated with kaolin in late winter.



Fig. 10. *C. pyri* adult with body soiled by kaolin.

	Treatment date	Eggs per 100 bubs	Nimphs per 100 flowers
First trial 2001			
Untreated control	-	136.75	6
kaolin	18 February and 10 March	1	0.25
mineral oil (dormant oil)	18 February.	30	2
Second trial 2002			
Untreated control		77.75	7.5
kaolin	11 and 19 February	0	0
mineral oil	11 February	12	3.5

Table 2. Results of two late winter field trials with kaolin and mineral oil (Pasqualini et al., 2003).

## 4.3 Spring-summer treatments

The treatments against summer generations can be performed towards eggs or nymphs. In the first case chitin inhibitors such as hexaflumuron (banned in 2004), triflumuron (banned in 2009), diflubenzuron and teflubenzuron have been employed. These active ingredients are used against second generation eggs, usually laid in the first decade of May. The treatment is usually performed against *C. pomonella* but shows a secondary effect on psylla. The chitin inhibitors provide the best results on *C. pyri* when they are applied on newly laid eggs (white eggs) or on eggs laid in a short time after the treatment. However, no activity is clearly exerted on eggs laid on the new shoots unreached by the treatment. Some authors observed that chitin-inhibitors show control effects similar to those of specific psyllicides described below: this could be explained by the absence of side effects on C. pyri natural predator, A. nemoralis (Souliotis & Moschos, 2008). However, the most relevant treatment employed against C. pyri in past and present times is the one against juvenile stages. This control strategy is base on specific synthetic active ingredients which are often acaricides, such as amitraz (commercially released in 1975 and banned in 2005), abamectin (commercially released in Italy in 1996) and spirodiclofen (commercially released in Italy in 2007), although in the past generic organophosphorates have been used such as monocrotofos and azinphos methyl.

Abamectin is presently the basic pesticide employed against *C. pyri*. Is should be briefly recalled that abamectin belongs to the chemical family of avermectins, compounds produced by the soil bacterium *Streptomyces avermitilis* (Lasota & Dybas, 1991). The activity of abamectin is mainly directed against young nymphs and secondarily against adults. The best results are therefore obtained when yellow eggs are mostly present and when the hatching peak, that could interfere with the pesticide activity because of honeydew, has not yet achieved (Pasqualini & Civolani, 2006). The product is not systemic but translaminary: the addition of mineral oil improves its penetration and after 24 hours no traces of the compound are found on leaf surface. One treatment timely performed against the second generation often represents the final solution, considering the high activity of the principle in comparison to amitraz (Table 3).

Spirodiclofen, commercially available since 2007, is the first member of a new chemical family, that of the tetronic acids (Nauen et al., 2000; Nauen, 2005), characterized by a new and original

mechanism of action which interferes with biosynthesis of lipids in the target arthropods. The original mechanism of action of spirodiclofen plays a key practical role by allowing successful rotation strategies with abamectin to limit the risks of occurrence of resistance in *C. pyri* control. The best activity of spirodiclofen occurs when it is targeted on yellow eggs some days before the hatching of first instar nymphs: the active ingredient shows instead a decreasing activity with the advancing of the psylla developmental stages (Table 4). At the stage of yellow eggs the activity of spirodiclofen is improved by addition of mineral oil (Table 4) and the use of spirodiclofen followed by treatment with mineral oil (1000 ml/hl) may represent a good alternative to the treatment with abamectin in presence of high and prolonged infestations. In conditions of low-medium pressure by *C. pyri*, one treatment with spirodiclofen may be enough: the few individuals escaping the treatment may be easily captured by anthocorids, given the good selectivity of the product towards these valuable auxiliary insects (Pasqualini & Civolani, 2007). However, the efficiency of spirodiclofen is often lower than that of abamectin (Table 4) (Pasqualini & Civolani 2007; Boselli & Cristiani 2008; Marčić et al., 2009).

Other active ingredients have been employed on both *C. pyri* in Europe and *C. pyricola* in North America. For example, in *C. pyricola* the differences of abamectin efficiency observed in the field suggested to employ and recommend neonicotinoids in pear IPM programs, among which imidacloprid, introduced in 1995, thiametoxan in 2001, acetamiprid in 2002 and thiacloprid in 2004. Besides *C. pyricola*, the last ingredient is used mainly for codling moth *C. pomonella*.

Presently a new active ingredient, spirotetramat, a lipid biosynthesis inhibitor similar to the tetronic acid derivate spirodiclofen, is under investigation in Europe (Nauen et al., 2008) but already commercially available in North America. Due to its mode of action spirotetramat is especially effective against juvenile stages of sucking pests, psyllid included. In the case of female adults the compound significantly reduces fertility and consequently insect populations. Spirotetramat also exhibits unique translocation properties: after foliar uptake the insecticidal activity is translocated within the entire vascular system. This property allows the protection of new shoots or leaves appearing after foliar application (Nauen et al., 2008): given the high efficiency on *C. pyri* (unpublished data) this active principle could represent a future valuable alternative to abamectin in order to manage the risks of occurrence of resistance in *C. pyri* control.

It is also possible to control nymphs by simply washing the trees with high amounts of water to which insecticidal soaps (fatty acids salts) are added to remove the honeydew (Briolini et al., 1989). Recently some other products have been used, similar to liquid glue and able to trap by a physical mechanism small and scarcely active insects such as almost all juvenile instars of *C. pyri*. These products are synthetic sugar esters (sucrose octanoate) and represent a relatively new class of insecticidal compounds that are produced by the reaction of sugars with fatty acids. (Puterka et al., 2003).

After discussing the active ingredients that could be used against *C. pyri* and the different strategies that could be employed, once again it must be emphasized that an efficient control of *C. pyri* infestations could be obtained by an integrated pest management of the pear orchard which allows a balanced growth of plants and simultaneously favours the growth of populations of natural psylla antagonists.

During the spring growth period a key point is to protect the useful psylla predators, first of all the most important one, *A. nemoralis*. The populations of this anthocorid are low in early spring but increase in the second half of June, insuring the protection of pear trees until harvesting and providing the most relevant contribution against *C. pyri*.

Another aspect that should not be overlooked in *C. pyri* control is the relevant effect of weather conditions on pest populations. A late winter season with mild temperatures favours an early and fast emergence of adults from their winter shelters and a regular egg laying, while the late winter frosts interrupt the adult emergence and egg laying, producing a gradual development of first and second pest generation which interferes with the precise timing of treatments. Cold and rainy periods during blossoming and petal fall interfere with nymph spreading on plants: in this case the nymphs often crowded in the flower calyx, sometimes causing with their feeding activity russet blotches or young fruit drop. On the contrary, high summer temperatures tend to block psylla development because of high egg mortality and slowing of juvenile growth for a long period.

	Treatment date	Nymphs per shoot
First trial (2000, on the variety		
"Conference")		
untreated control	-	16.68
abamectin	6 May	0.15
amitraz	10 May	1.08
Second trial (2000, on the variety "William")		
untreated control	-	46.88
abamectin	6 May	2.23
amitraz	10 May	8.18
Third trial (2004, on the variety "Conference")		
untreated control	-	8.29
abamectin	6 May	0.98
amitraz	10 May	1.39

Table 3. Results of three field trials with abamectin and amitraz on two pear varieties (Pasqualini & Civolani, 2006).

## 5. Evolution of resistance of *C. pyri* to pesticides

As for all phytophagous pests, also for *C. pyri* the repeated use of chemical active ingredients causes the development of resistance. However, in Europe there are less resistance cases documented for *C. pyri* in comparison to those known since 1960 for *C. pyricola* in North America (Harries & Burts, 1965). Among the *C. pyri* resistance events in Europe the best known involve organophosphorates, pyrethroids and chitin inhibitors: in all documented cases a sharp decrease in pesticide activity was observed even after a few years of use. The active ingredient monocrotofos represents the best known case (Berrada *et al.*, 1995). The selection induced by this pesticide around the end of 1980 on some *C. pyri* populations near Toulouse (France) caused an increased resistance up to 140 fold in comparison to the susceptible strain in 30 generations, as shown by laboratory tests. Further tests showed that the mechanisms involved in the onset of resistance to this active

	Treatment date	Nymphs per shoot
First trial (2005, on the variety "Conference")		
untreated control	-	83.25
spirodiclofen	19 May (yellow eggs)	36.25
spirodiclofen + mineral oil	19 May (yellow eggs)	15.50
abamectin + mineral oil	19 May (yellow eggs)	0.75
Second trial (2002, on the variety "Abbé Fétel")		
untreated control	-	25
spirodiclofen	30 April (white eggs)	6
spirodiclofen	14 May (yellow eggs)	11
spirodiclofen	17 May (first hatching)	19
spirodiclofen	22 May (20-30 % hatching)	
amitraz	14 May (yellow eggs)	2
Third trial (2007, on the variety "Beurré Bosc")		
untreated control	-	66.5
spirodiclofen	27 April	1.5
spirodiclofen and abamectin	27 April and 9 May	0
spirodiclofen and mineral oil	cal oil 27 April and 9 May 0.	
abamectin	27 April	0.5
Abamectin and spirodiclofen	27 April and 9 May	0

Table 4. Results of three field trials with spirodiclofen on three pear tree varieties (Pasqualini & Civolani, 2007; Boselli & Cristiani, 2008).

ingredient were the enhanced activity of cytochrome P450 monooxygenase (MFO) and also the changes in acetyl cholinesterase, since the susceptibility to the pesticide could not be fully recovered by pretreating *in vivo* the adults with piperonylbutoxide (PBO), a specific inhibitor of MFO (Berrada *et al.*, 1994).

In 1994 in the Avignon region (France) the survey for *C. pyri* resistance was extended to 16 active ingredients belonging to five pesticide families, by topical laboratory tests on adults (Fig. 11). The tests showed that the resistance rates (RR) were extremely low for the family of carbamates (one- to 2.4-fold), relatively low for the family of pyrethroids (4.7- to 6.2-fold). For the family of organophosphorates insecticides, the resistance rates among the active ingredients were very different: lower than one (0.2-fold) for parathion-methyl, low for mevinphos and malathion (3.5- and 2.5-fold, respectively), and higher for chlorpyriphosethyl (10.2-fold), monocrotophos (26-fold), azinphos-methyl (62.2-fold) and phosmet (179.7-fold). In the same Avignon area, tests on resistance selection were performed in laboratory with the organophosphorate azinphos-methyl, which was frequently used in high amounts against *C. pomonella* and could indirectly cause selection also in *C. pyri*. The RR observed ranged from 10 to 40-fold in comparison to wild populations: these values were

considerably lower than those reached by monocrotofos (Bues *et al.*, 2000). Further tests showed that the selection by azinphos-methyl produced a strong cross resistance with phosmet and monocrotofos (155 fold). By the same tests no cross resistance was shown for azinphos-methyl with amitraz, pyrethroids, carbamates and other organophosphorate pesticides such as chlorpyriphos and mevinphos (Bues *et al.*, 2000). The same authors started genetic studies by crossing the resistant and susceptible strains and showed that the resistance was autosomically inherited and semi-dominant in expression (Bues *et al.*, 2000). They also advanced by backcross the hypothesis that the resistance factor was monogenic. However, the result that the resistance to azinphos-methyl is semi-dominant is different from what previously obtained in Oregon by Van de Baan (1988) on pyrethroids: this author crossed two populations of *C. pyricola*, one susceptible and the other 240-fold resistant to the active ingredient fenvalerate (a pyrethroid), showing that the resistance was in this case semi-recessive (Van de Baan, 1988). Probably there are different mechanisms involved in the resistance to different pesticide families.

As mentioned before, the pyrethroids are another pesticide family largely employed in France against *C. pyri* and in North America against *C. pyricola* during leaf fall and late winter, before egg laying by overwintering adults. In laboratory topical tests on overwintering adults, some of these active ingredients showed very variable RR: for example, in tests performed in Avignon in 1994 the observed RR were 4 or 6-fold higher in comparison to the susceptible laboratory population (Buès *et al.*, 1999), but further studies in 1996 in the same southern Rhone valley, on a field population collected in Pont Saint Esprit, showed that the RR was 42.9-fold higher (Buès *et al.*, 1999). Later, the same authors confirmed similarly high resistance values to the active ingredient deltametrin in some Southern France populations in which the RR was 30-fold, always with the adult winter forms less susceptible in comparison to the same stage of the adult summer forms (Buès et al., 2003).

Concerning the mechanism of action producing resistance to pyrethroids, laboratory tests showed that the addition of PBO caused an recovery of pyrethroid efficiency almost complete: this shows that other mechanisms of induction of resistance could be secondarily involved.

Therefore the detoxifying enzyme MFO is involved in the mechanism of action of both organophosphorate and pyrethroid active ingredients.

Around 1995, in a region of West Switzerland a lower susceptibility was observed to teflubenzuron, active ingredient belonging to the family of chitin inhibitor (Schaub *et al.*, 1996). More recently this resistance to teflubenzuron was also observed in the Czech Republic, according to tests performed in 2004 and 2005 (Kocourek & Stará, 2006). The mechanisms of resistance to teflubenzuron have not yet been completely investigated.

As previously mentioned, abamectin is the most efficient and most used active ingredient against *C. pyri*: it was used against this pest for the first time in 1996, with only one spring treatment and only in some orchards, also because amitraz was an alternative until 2005, the year in which the ingredient was banned. Probably after the ban on this active ingredient the number of treatments per orchard and the area of employment were increased: this trend was also observed in the fruit growing area of Lleida, Girona and Huesca in Spain (Miarnau et al., 2010), and Ferrara and Modena in Italy. The current high selection pressure with this active ingredient, repeatedly applied over both geographical areas could induce selection for resistance.

Following the sudden outbreak of *C. pyri* populations in some fruit growing area in Emilia-Romagna in 2005, abamectin tests for resistance were performed for the first time only on overwintering adults (Fig. 11), but no resistance was detected, although LC<sub>50</sub> and LC<sub>90</sub> values appeared related to the time of adult field collection (Civolani et al., 2007).

Besides adults (Table 5) (Fig. 11), in 2007 and 2008 the abamectin tests in Emilia-Romagna were extended also to eggs and nymphs (Fig. 12 and Fig. 13), the stage target in *C. pyri* field treatments (Civolani et al., 2010). On adults, the abamectin topical tests performed in autumn 2007 and 2008 did not show significant differences among all populations tested, but the LC<sub>50</sub> values were apparently related to the adult collection dates, as previously reported (Civolani et al., 2010).

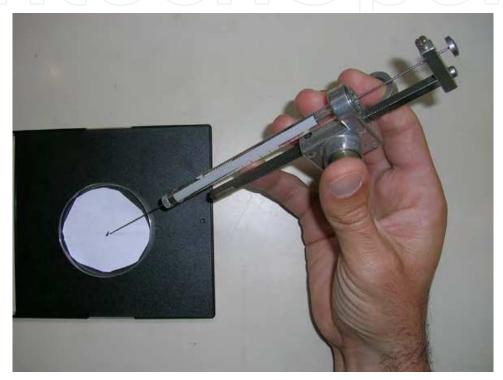


Fig. 11. Topical application of the insecticide solution on *C. pyri* adult winter form with hand-held manual micro-applicator.

The egg spray test ( $LC_{50}$  and  $LC_{90}$ ) did not show relevant differences, although  $LC_{50}$  and  $LC_{90}$  values were always lower in the organic farm than in all others (Table 6). The results of leaf dip tests performed on young and old larvae were generally similar to the egg spray ones (Civolani et al., 2010).

Overall, the tests data indicate that no apparent resistance to abamectin has been developed up to now in *C. pyri* populations of Emilia-Romagna.

In 2009 and 2010 new cases of loss of efficiency of abamectin reappeared in some orchards in the province of Modena (Emilia-Romagna). In 2010 other tests were again performed on a C. pyri population which underwent 7 abamectin field treatment in 2009. However, in this case also the results of  $LC_{50}$  and  $LC_{90}$  did not show significant differences and the RR was just above 2 (unpublished data).

From 2004 to 2006 tests were carried out also in Spain to monitor the evolution of susceptibility to abamectin in orchards where the number of treatments changed from less than two until 2005 to an average of three after the ban on amitraz, with some orchards

undergoing 6-7 treatments. The tests were performed both on adults and nymphs and no evidence of a high RR was found (Miarnau et al., 2010). However, there are some Spanish populations of *C. pyri* that show low susceptibility in adults as well as in nymphs: these populations come from the fields with the highest number of abamectin treatments per year. As in Emilia-Romagna, these cases also indicate a high risk of selection for resistance to abamectin, especially if the number of treatments per year is high and there are no alternative products to use in a resistance management program.



Fig. 12. Cages for potted pear plants to keep adults of different population separated.



Fig. 13. Potted young pear plants on which psylla eggs were laid and then treated with abamectin.

Population (locality)	Field atmategy	2007		2008	
Population (locality)	Field strategy	$LC_{50}$	RR	$LC_{50}$	RR
Buondì-Vezzani (Diamantina)	organic	3.13		5.27	
Minotti (S. Martino)	integrated	4.98	1.59	3.24	0.61
Scagnolaro (Francolino)	integrated	2.80	0.89	4.16	0.78
Bonora (Boara)	integrated	/-/12		5.95	1.12
Celati (Francolino)	integrated	-	7 Y	4.23	0.80
Marchetti (Diamantina)	conventional	<i></i>	$\bigcirc$ /	5.31	1

Table 5. Response of *C. pyri* overwintering adult populations to topical applications of abamectin (2007 and 2008) in the province of Ferrara (Civolani et al., 2010). The values of  $LC_{50}$  are expressed in mg  $l^{-1}$  of abamectin.

Population (locality)	Eigld atmatager	2007		2008	
1 opulation (locality)	Field strategy	$LC_{50}$	RR	$LC_{50}$	RR
Buondì-Vezzani (Diamantina)	organic	0.20		0.15	
Minotti (S. Martino)	integrated	0.43	2.15	0.29	1.93
Scagnolaro (Francolino)	integrated	-	-	0.15	1
Bonora (Boara)	integrated	-	-	0.27	1.80
Celati (Francolino)	integrated	-	-	0.34	2.26
Marchetti (Diamantina)	conventional	-	-	0.44	2.93

Table 6. Response of *C. pyri* to egg spray applications (2007 and 2008) in the province of Ferrara (Civolani et al., 2010). The values of LC<sub>50</sub> are expressed in mg l-1 of abamectin.

Spirodiclofen is a recently introduced active ingredient in *C. pyri* control. Different LC<sub>50</sub> values were observed for spirodiclofen in tests again performed in Emilia-Romagna, regardless of its limited use as an alternative to abamectin. This active ingredient was tested on three populations of the province of Ferrara in 2006 and 2007 by laboratory assays on adults and eggs.

The topical tests on overwintering adults showed high susceptibility differences for this active ingredient between the adult population collected in the organic farm and those collected in the integrated (Minotti) and conventional (Marchetti) farms (Table 7).

Donulation (locality)	Eigld atmatages	2006		2007	
Population (locality)	Field strategy	$LC_{50}$	RR	$LC_{50}$	RR
Buondì-Vezzani (Diamantina)	organic	19.09	1	89.01	-
Minotti (S. Martino)	integrated	2582.60	135.28	1931.70	21.,69
Marchetti (Diamantina)	conventional	25.93	1.358	943.72	10.60

Table 7. Results obtained in the topical test with spirodiclofen on C. pyri overwintering adults (2006 and 2007). The values of LC<sub>50</sub> are expressed in mg  $l^{-1}$  of spirodiclofen.

The results obtained by the spray test on yellow eggs before hatching, using the field doses of spirodiclofen, show low activity on the population collected in the Minotti integrated farm (both on first and second psylla generation). The activity levels in the Minotti farm, expressed as percentage of nymph mortality, are very different from those detected in the other two farms. However, the values detected in the conventional Marchetti farm are unexpectedly similar to those obtained in the organic farm (Table 8): these results were surprising because spirodiclofen was never employed before in none of these farms (unpublished data).

Population (locality)	Field strategy	Nymph mortality % on eggs laid by overwintering adults	Nymph mortality % on eggs laid by adult summer forms
Buondì-Vezzani (Diamantina)	organic	100	94,91
Minotti (S. Martino)	integrated	30.62	77.95
Marchetti (Diamantina)	conventional	-	94.81

Table 8. Results obtained in the spray test with spirodiclofen on *C. pyri* eggs laid by overwintering and summer adults in 2007. The results are expressed as percentage of nymph mortality.

## 6. Natural and biological control of C. pyri

In open field and especially in the pear orchard the techniques of biological control are not common because the fruit growers have always aimed to favour and exploit the development of the wild auxiliary insects, thus performing strategies of natural control. Antochoris nemoralis, common in all Europe, is known as the main predatory species of pear psylla. This species overwinters as an adult (Fig. 14) and starts to lay eggs in spring, inserting them under the leaf epidermis. The development of the juvenile forms occurs in 5 stages. A. nemoralis preys on both eggs and nymphs of psylla and in Emilia-Romagna typically shows three generations. Although generally preferring psylla, this anthocorid may feed on other insects and on the pear trees its activity against aphids and the pear sawfly Hoplocampa brevis Klug (Hymenoptera Tenthredinae) is very interesting. Laboratory tests showed an average predation of about 300 psylla nymphs during the entire life of an adult, which lasts about 60 days. The presence of A. nemoralis in pear orchards mainly depends on the type of control strategy applied in the farm. As previously mentioned, by limiting the use of pesticides to the minimum required and preferring the selective ones (see next chapter), it is favoured the development of the wild A. nemoralis populations which become a relevant factor to control the pest. Indeed, a dynamic equilibrium develops between the predator and C. pyri populations, often leading to the solution of the problem without the need for specific chemical treatments, or with treatments only reduced to tree washing. The main problem of this strategy is nevertheless the slow initial development of the predator population, which must have the prey available (in this case C. pyri) to rapidly increase in number within the pear orchard. Therefore, in the initial part of the season (around May-June) it is necessary to tolerate some amounts of the pest in the orchard to obtain later a good number of predator anthocorids on the trees. In other words, this means

that when the presence of *C. pyri* rapidly increases in the third decade of May, usually we can expect a rapid increase of the predator population about two-three weeks later. This requires for the fruit grower to tolerate a relatively high presence of the pest and "resist" to the temptation to perform specific treatments. Since 1990, other field conditions added to these problems of the natural control, such as the increased chemical treatments against the codling moth, *C. pomonella*. These treatments caused a general weakening of the wild *A. nemoralis* populations, disrupting a very fluctuating natural equilibrium. Another problem recently emerged is that even in equilibrated pear orchards, not undergoing any heavy chemical treatment, for some reason the presence of predators remains low or undergoes high fluctuations over the years or even according to the seasons.

The previously mentioned limits of natural control led to artificially introduce anthocorids in the pear orchard, buying them from biofactories and thus performing a true biological control technique. The aim is to obtain a more numerous presence of the predator in the critical periods, anticipating the reproduction mechanism of the population which could occur naturally but with some delay. The introduction of predators is performed at the end of winter, between the end of March and the beginning of April. About 1000 adult individuals of *A. nemoralis* are placed per hectare of the pear orchard, in three consecutive weekly introductions. This biological control technique was common around 2000 in integrated pear orchards where the active ingredient amitraz was employed for the spring-summer control of *C. pyri*, then it was slowly neglected because of the limited results and the relatively high costs.



Fig. 14. Adult of *A. nemoralis*.

## 7. Selectivity of pesticides against A. nemoralis

In the last 20 years new families of pesticides have been developed with generally lower toxicity towards beneficial species in comparison to the previous ones. These pesticides are more suitable for the new techniques of integrated pest control (IPM) which preserve the contribution of beneficial insects to the natural control against pests, especially against *C. pyri*. Therefore it was necessary to perform tests on the new active ingredients, to verify their different toxicity degree and obtain the data required to improve their use.

Several studies have been recently performed to evaluate the selectivity of the new pesticides towards auxiliaries found in the pear orchards, first of all *A. nemoralis* (Fig. 14), the most relevant in the natural control against *C. pyri*. The active ingredients recently investigated include those directly employed against *C. pyri* (psyllicides) and also those largely employed on other key pests (non-psyllicides).

Concerning the psyllicides, in different toxicity tests performed in Emilia-Romagna since 1997 abamectin showed a medium degree of toxicity against the *A. nemoralis* population, mostly against first and second instar nymphs in comparison to adults. In some cases the mortality of nymphs reached 50% and, according to field data, the total population appears limited in comparison to untreated controls for about two weeks after the treatment. This result must nevertheless take into account the lower presence of prey as food for *A. nemoralis* (Pasqualini & Civolani 2007). Other products specific for *C. pyri*, such as amitraz (now banned), mineral oil, or insecticide soaps did not show relevant effects on the predator population (Civolani & Pasqualini, 1999; Pasqualini et al., 1999). Concerning spirodiclofen, the tests on toxicity towards *A. nemoralis* were performed again in Emilia-Romagna in years 2004-2006. The results show that the anthocorid populations undergoing treatment with spirodiclofen have similar development to the untreated ones, unlike abamectin for which the population first sharply decreases, then recovering according to the amount of prey available (Pasqualini & Civolani, 2007).

Concerning the non-psyllicides, those belonging to the chitin inhibitors, usually characterized by a long period of action, generally show a low toxicity on *A. nemoralis*, even if some of them, such as flufenoxuron, induce a heavy reduction of the anthocorid populations (Girolami et al., 2001, Pasqualini & Civolani 2002).

Among other non-psyllicides employed in pear orchards against the most relevant pests, there is spinosad, a pesticide of natural origin, whose activity derives from a toxin produced by *Saccharopolyspora spinosa* (Bacteria Actinomycetales), the indoxacarb belonging to the family of the oxadiazines, then methoxyfenozide and tebufenozide, synthetic molecules belonging to the family of moulting accelerator compounds (MAC), and the organophosphorates azinphos-methyl (banned in 2007), chlorpyrifos, chlorpyrifos-methyl and phosmet, all employed against relevant lepidopteran pests such as *C. pomonella, Cydia molesta* Busck and *Pandemis cerasana* Hübner. All these active ingredients have been more or less tested for toxicity against *A. nemoralis* and the results did not show relevant toxic effects on this predator (Civolani & Pasqualini, 1999).

As previously mentioned, the neonicotinoids are not employed in Italy and Europe as specific psyllicides, on the contrary of what happens in United States where they are employed as an alternative to abamectin against *C. pyricola*. However, a large amount of the above active ingredients are used against other pests, such as aphids and the pear sawfly *H. brevis*: the most relevant are imidacloprid, acetamiprid and tiametoxan, while against the codling moth *C. pomonella* the most frequently used is thiacloprid. All those active

ingredients have significant toxic effects on *A. nemoralis*, thus they must be employed only for very few treatments along the year.

### 8. Conclusions

Concerning the strategies and methods to control *Cacopsylla pyri* (Hemiptera Psyllidae), and also their effects on the beneficial insects and the development of resistance, during the past decade Italian and European populations of *C. pyri*, in significant decline, have been apparently less capable to induce damage: this could be due to the success of defence programs based on integrated pest management (IPM). For pear, the IPM involves pest population control and auxiliary insect protection, associated to availability of new chemical or microbiological agents specifically targeted to pear key pests (mainly *Cydia pomonella*).

As far as known about *C. pyri* infestation, the biological control alone is not successful in preventing damage, especially when caused by second generation nymphs that feed on shoots and leaves in late spring and summer; thus chemical pest control strategies are also employed. For example, in Northern Italy the most common defense strategy against *C. pyri* in pear orchards involves chemical treatments on second-generation eggs or nymphs. Traditional treatments with chitin inhibitors, mainly aimed against *C. pomonella*, have also some secondary effects on *C. pyri*.

Specific treatments against *C. pyri* nymphs involve amitraz (now banned), abamectin and spirodiclofen. During spring and summer the treatment with the last two active ingredients, in addition to non-chemical treatments such as tree washing, is usually successful in limiting the honeydew damages caused by *C. pyri*.

During autumn and winter, the *C. pyri* management strategies involve treatments with synthetic pyretroids after leaf fall, to limit adult overwintering population, or at the end of winter, when females are ready to lay eggs. However, these treatments during autumn and winter, common in France and North America, are rarely employed in Italy because of their high toxicity against populations of auxiliary insects (such as Anthocoridae) which could still be present in the pear orchards. In late winter it is also possible to perform a non-chemical treatment against *C. pyri* by distribution of an aqueous suspension of kaolin on trees, in order to obtain a physical barrier to egg laying.

Each one of the above strategies shows favorable and unfavorable aspects in terms of efficacy, side effects on beneficial insects, timing of application and environmental conditions.

As for all phytophagous pests, also for *C. pyri* the repeated use of chemical active ingredients causes the development of resistance. Indeed, several insecticides employed in the past to control the pear psylla showed a sharp decline in activity because of resistance development. After the sudden outbreak of *C. pyri* populations in some fruit growing area of Ferrara and Modena (Italy) and Lleida, Girona and Huesca (Spain), abamectin tests were performed on winter form adults and nymphs. The results did not show relevant resistance effects, although LC<sub>50</sub> and LC<sub>90</sub> values were always higher in populations where abamectin treatment was repeated several times in the year. Overall, the results indicate that no apparent resistance to abamectin has been yet developed in *C. pyri* populations of the most important European areas of pear growth: nevertheless, the pear orchards in which *C. pyri* outbreaks recently occurred are presently under close investigation and careful survey.

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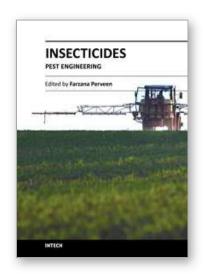
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#### **Insecticides - Pest Engineering**

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This book is compiled of 24 Chapters divided into 4 Sections. Section A focuses on toxicity of organic and inorganic insecticides, organophosphorus insecticides, toxicity of fenitrothion and permethrin, and dichlorodiphenyltrichloroethane (DDT). Section B is dedicated to vector control using insecticides, biological control of mosquito larvae by Bacillus thuringiensis, metabolism of pyrethroids by mosquito cytochrome P40 susceptibility status of Aedes aegypti, etc. Section C describes bioactive natural products from sapindacea, management of potato pests, flower thrips, mango mealy bug, pear psylla, grapes pests, small fruit production, boll weevil and tsetse fly using insecticides. Section D provides information on insecticide resistance in natural population of malaria vector, role of Anopheles gambiae P450 cytochrome, genetic toxicological profile of carbofuran and pirimicarp carbamic insecticides, etc. The subject matter in this book should attract the reader's concern to support rational decisions regarding the use of pesticides.

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