Persistence of Pirimicarb in Peaches and Nectarines

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The fate of pirimicarb residue on field and greenhouse peaches and nectarines was studied. Pirimicarb showed similar behaviors in both fruits. The decrease in residue during the course of the experiment was mainly due to the dilution effect caused by fruit growth. In the greenhouse the active ingredient (ai) was stable, while in the field it was degraded moderately. Pirimicarb metabolites, which are usually formed from ai degradation in plants, were never found during the experiment. The data from a laboratory test to evaluate the influence of volatilization and photodegradation on pirimicarb would indicate that pirimicarb loss cannot be ascribed to volatilization.

Keywords: *Pirimicarb; residues; peaches; nectarines*

The green aphid (*Myzus persicae* Sulz.) is the most common pest of peach trees, its choice plant. This insect can cause considerable damage to growing plants unless proper treatment is carried out at the right time. Among the most effective active ingredients (ai) against aphids, pirimicarb, 2-(dimethylamino)-5,6-dimethylpyrimidin-4-yl dimethylcarbamate (I) (Figure 1), is especially suitable when the treatment is to be carried out at high ambient temperature. Furthermore, pirimicarb has a selective action against aphids and is widely employed in integrated control programs. It has contact and respiratory actions, and thanks to its translaminar action, it can reach the aphids even on the undersides of leaves (Tomlin, 1994). According to the literature (FAO/WHO, 1977) pirimicarb is taken up by the roots and translocated through the xylem and does not penetrate into the fruits. After spraying, it rapidly dissipates in the plants by volatilization and photochemical and metabolic degradation. The major degradative products are the carbamate-containing metabolites **II** and **III**, while the carbamate-containing metabolite **IV** and the hydroxypyrimidines **V**–**VII** are the minor products (Figure 1). In the FAO/WHO monograph on pirimicarb, the above metabolic pathway is reported for this ai in peaches, but no information is given on the metabolites that are formed or on their quantity.

This work was aimed at contributing to the knowledge of the metabolic pathway of pirimicarb in peaches and nectarines in different cultivation conditions, considering that in maximum residue limit (MRL) evaluation the sum of the active ingredient and its metabolites **II** and **III** is taken into account in some countries (e.g. Spain), as indicated by FAO/WHO. Furthermore, a laboratory test was carried out to evaluate the influence of volatilization and photodegradation on pirimicarb degradation after exposure to direct sunlight.

EXPERIMENTAL PROCEDURES

Materials and Methods. The trial was carried out at an experimental farm of the Centro Regionale Agrario Sperimentale located near Uta (Cagliari, Italy), in a 1000 m^2 greenhouse and a contiguous field. The orchard was planted in 1986 with 6666 trees/ha and a planting space of 1.5 × 1.0 m. Irrigation was by a drop system. The varieties were Maraviglia peaches and Armking nectarines. A random block scheme was used, with four replications, and each block contained 20 trees.

The environmental conditions were continuously recorded with SM 3800 automatic weather stations (SIAP, Bologna, Italy) in the greenhouse and field.

Treatments were made with 10 hL/ha of a liquid formulation of Pirimor, containing 17.5% ai, applied with an F 320 portable motorized sprayer (Fox Motori, Bologna, Italy) in the dose recommended by the manufacturer (150 g/hL). Treatments were carried out on April 9 and 22 and May 26, 1993, in the greenhouse and field, respectively.

Random 20-fruit samples were collected from each block. Samplings started about 2 h after the last treatment (on the dry plants) and were repeated after 1, 3–4, 7–8, and 14 days. The last sampling was carried out at commercial ripening of fruits.

The stones were removed, and the pulp was chopped and homogenized with a food cutter.

Extraction Procedure. A 20-g portion of the homogenized sample was weighed in a 100-mL screw-capped test tube; 8 g of sodium chloride, 40 mL of methylene chloride, and 40 mL of acetone (for **I**–**IV**) or 1-butanol (for **V**–**VII**) were added, and the tube was agitated in a rotating stirrer for 15 min. The phases were allowed to separate, the organic layer was isolated, and a 10-mL aliquot was dehydrated with anhydrous sodium sulfate (2 g for **I**–**IV** or 5 g for **V**–**VII**). A 4-mL aliquot of the organic layer was transferred into a 10-mL beaker and evaporated to dryness under a nitrogen stream. The residue was recovered with 1 mL of acetonitrile and allowed to percolate through a previously activated (2 × 3 mL of acetonitrile) NH_2 cartridge. The cartridge was further eluted with 2 mL of acetonitrile. For the determination of pirimicarb and metabolites **II**–**IV**, the acetonitrile eluates were collected, evaporated to dryness under a nitrogen stream, recovered with 1 mL of the mobile phase, and injected for HPLC analysis. For the determination of **V**–**VII**, the acetonitrile eluates were discarded and the cartridge was eluted with 2 mL of methanol. The methanolic eluate was collected, evaporated to dryness under a nitrogen stream, taken up with 1 mL of the mobile phase, and injected for HPLC analysis.

Apparatus and Chromatography. A Waters Sep-Pak vacuum manifold (Millipore, Milford, MA) with NH_2 cartridges (500 mg, 3 mL) (Isolute, IST, Mid Glamorgan, U.K.) was used for sample purification.

An HP Model 1050 liquid chromatograph (Hewlett-Packard, Avondale, PA), consisting of a pump, an autosampler with a 100-μL loop, and a variable wavelength UV–vis detector was

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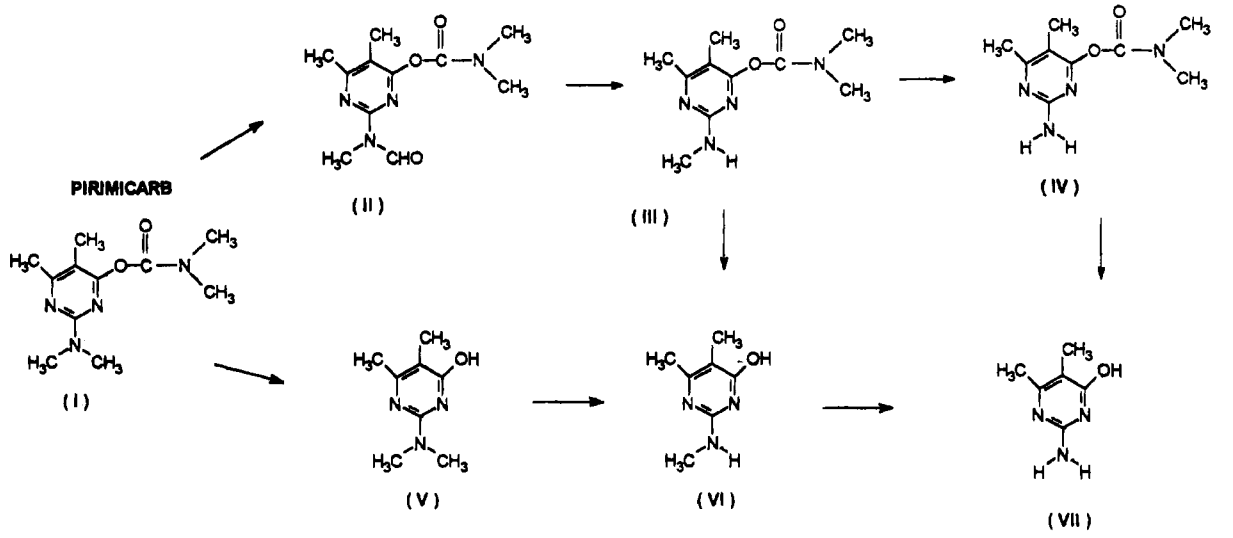

Figure 1. Pirimicarb (**I**) and six of its metabolites (**II**–**VII**).

employed. The chromatograph was connected to an HP 3394 reporting integrator (Hewlett-Packard).

Spherisorb S_5-C8 and S_5-NH_2 columns (250 × 4.6 mm i.d., 5 μm) (Phase Separations, Waddinxveen, Netherlands) were employed, respectively, for the determination of **I**–**IV** with a 25:75 mixture of acetonitrile and buffer solution (10^{-2} M monobasic potassium phosphate with 5 mL/L of glacial acetic acid) and for the determination of **V**–**VII** with a 94:6 mixture of acetonitrile and water. In both cases the flow rate of the mobile phase was 1 mL/min. The detector wavelength was 236 nm for pirimicarb, 245 nm for **II**, 230 nm for **III** and **IV**, and 295 nm for **V**–**VII**. Standard curves for pirimicarb and its metabolites were constructed by plotting peak heights *vs* concentrations; good linearities were achieved, respectively, in the range 0.03–1.50 ppm for **I**–**IV** with correlation coefficients between 0.9999 and 1.0000 and in the range 0.05–1.50 ppm for **V**–**VII** with correlation coefficients between 0.9997 and 0.9999.

Chemicals. Acetone, acetonitrile, 1-butanol, methanol, and methylene chloride were HPLC grade solvents (Carlo Erba, Milan, Italy); glacial acetic acid, monobasic potassium phosphate, sodium chloride, and anhydrous sodium sulfate were of analytical grade (Carlo Erba). Double-distilled water was filtered through a Milli Q apparatus (Millipore, Molsheim, France) before use. Pirimicarb and its metabolites were analytical standards supplied by ICI Solplant (Milan, Italy). Stock standard solutions were prepared at the concentration of 200 mg/L in acetonitrile, except for metabolite **VII**, which was dissolved in methanol; work standard solutions were obtained by dilution with the mobile phase.

Recovery Assays. Untreated fruit samples (20 g) were fortified with appropriate volumes of standard solutions to reach concentrations of 1.5, 1.0, 0.5, 0.1, and 0.03 (0.05 for **V**–**VII**) ppm for pirimicarb and its metabolites. Average recovery from four replicates showed values ranging from 86% to 107% for carbamates **I**–**IV** and from 75% to 85% for hydroxypyrimidines **V**–**VII**, with a maximum CV of 7.

Photodegradation and Volatilization Laboratory Test. One-milliliter portions of 1.7 ppm pirimicarb solutions in acetone were poured into Petri dishes (o.d. 4 cm) and vials (o.d. 1 cm) and evaporated under a nitrogen stream at ambient temperature. The uncovered dishes and screw-capped vials were then exposed to direct sunlight. Every 30 min (for a 4-h period) one dish and one vial were removed from the sunlight. The residue contained in the dish was dissolved with 1 mL of the mobile phase and injected for HPLC analysis. The vial was stored at −25 °C for 1 h to allow condensation of the vapors eventually present, and the residue was taken up with 1 mL of the mobile phase and injected for HPLC analysis. The test was carried out in duplicate; the ambient temperature ranged between 22 and 35 °C during the experiment. Control samples were stored in the dark and analyzed at the beginning and at the end of the test. Chromatographic analyses of control samples showed no degradation of pirimicarb.

Table 1. Pirimicarb Residues (Parts per Million ± SD, n = 4) on Peaches and Nectarines in the Greenhouse and Field

days after treatment	peaches		nectarines	
	fruit wt, g	residues	fruit wt, g	residues
		Greenhouse		
−0	24 ± 1	0.36 ± 0.17	21 ± 1	0.27 ± 0.06
0		1.31 ± 0.32		1.52 ± 0.26
1	25 ± 1	1.35 ± 0.14	22 ± 3	1.32 ± 0.23
4	29 ± 4	1.11 ± 0.25	22 ± 2	0.83 ± 0.17
8	31 ± 1	0.86 ± 0.13	29 ± 2	0.83 ± 0.14
14	52 ± 5	0.59 ± 0.13	54 ± 6	0.45 ± 0.14
26			95 ± 6	0.24 ± 0.08
		Field		
0	51 ● 6	0.62 ± 0.12	54 ± 9	0.51 ± 0.21
1	59 ± 8	0.51 ± 0.21	55 ± 7	0.51 ± 0.04
3	66 ± 2	0.47 ± 0.17	67 ± 8	0.27 ± 0.08
7	83 ± 6	0.40 ± 0.10	82 ± 10	0.22 ± 0.03
14	107 ± 18	0.17 ± 0.05	94 ± 4	0.17 ± 0.05
21	119 ± 17	0.10 ± 0.03	115 ± 20	0.09 ± 0.03

RESULTS AND DISCUSSION

The analytical method employed allows the determination of pirimicarb and six of its metabolites (**II**–**VII**), though only the ai was detected in all of the fruit samples analyzed. In the fruits from the greenhouse, where two treatments had been carried out with a 13-day interval, pirimicarb residue before the last treatment was *ca.* 0.3 ppm, both in peaches and in nectarines (Table 1). After treatment, the residue increased by *ca.* 1 ppm in both cases and then progressively decreased in a similar trend in peaches and nectarines. Nevertheless, considering the fruit growth and its dilution effect, the ai residue was practically unchanged during the experiment. Pirimicarb residue in fruits from the field was almost half that in the greenhouse, probably because the average weight of the fruits in the field was almost twice the weight of the fruits from the greenhouse. In the field the decrease in residue was also similar in peaches and nectarines (Table 1). The dilution effect caused by the fruit growth alone could not account for the decrease of pirimicarb residue from its initial levels to 0.1 ppm at harvest. Other factors such as volatilization or photodegradation may have contributed significantly to residue dissipation. The

Table 2. Microclimatic Conditions[a] in the Greenhouse (G) and Field (F) during the Period of Residue Evaluation

weeks after treatment	rel humidity, %		temp, °C		rainfall, mm
	G	F	G	F	F
1	79.6	69.4	18.2	19.6	
2	73.2	62.4	20.3	23.3	
3	73.1	65.3	20.1	21.7	5.2
4	73.0	60.7	22.0	19.4	
5	65.4	69.4	22.9	23.2	3.6

[a] Weekly averages.

Table 3. Residues (Parts per Million) of Pirimicarb and Its Metabolites II and III after Exposure to Direct Sunlight in Petri Dishes and Scew-Capped Vials

time, h	pirimicarb		metabolite **II**		Metabolite **III**	
	dish	vial	dish	vial	dish	vial
0	1.67	1.62				
0.5	1.04	1.36	0.07	0.08	0.06	0.07
1	0.53	1.05	0.18	0.12	0.14	0.12
1.5	0.17	0.60	0.18	0.22	0.12	0.17
2	0.06	0.35	0.12	0.18	0.03	0.13
2.5	0.05	0.27	0.21	0.14	0.07	0.09
3	0.05	0.26	0.16	0.16	0.06	0.10
3.5	0.03	0.26	0.13	0.21	0.04	0.14
4	nd[a]	0.09	0.18	0.28	0.04	0.10

[a] nd, not detected.

environmental conditions recorded during the experiment were similar in the greenhouse and field (Table 2), except for a higher relative humidity in the greenhouse and a moderate rainfall (*ca.* 9 mm) in the field. Since the below-described laboratory experiment leads to us exclude the ai evaporation and since a washing away effect cannot be attributed to the recorded rainfall, these environmental parameters should not have affected the residue decrease that occurrred in the field.

The results of the laboratory test for the evaluation of pirimicarb photodegradation and volatilization are reported in Table 3. Pirimicarb was initially present at a concentration of 1.67 ppm in samples in Petri dishes. After 2 h, it showed a residue of 0.06 ppm (*ca.* 96% reduction), slowly decreasing and disappearing (<0.01 ppm) after 4 h. In the vials, the ai degradation rate was lower, the residue being 0.35 ppm after 2 h (79% reduction) and 0.09 ppm after 4 h (95% reduction). This different behavior could be attributed to the difference in irradiated surfaces in dishes (o.d. 4 cm) and vials (o.d. 1 cm). Moreover, UV light absorption by glass vials could have affected the degradation rate of pirimicarb. The same could have occurred due to the reduced UV light transmission by greenhouse glass, thus justifying the greater stability of pirimicarb in peaches and nectarines grown in the greenhouse. The rapid degradation of pirimicarb, both in uncovered dishes and in capped vials, should therefore be ascribed to photodegradation rather than volatilization. From the very first sampling, 30 min after the beginning of the laboratory test, metabolites **II** and **III** were detected, but their concentrations, which were of the same order of magnitude in both dishes and vials, were always below 20% of the initial ai concentration during the course of the experiment.

Metabolites **II** and **III** were separately subjected to a photodegradation test carried out in Petri dishes and vials: the initial concentration was 0.8 ppm for both compounds. In a 4-h period, **III** was degraded by *ca.* 80% and **II** by *ca.* 25%. Metabolite **II** did not change into **III**, as reported in the literature for plants (FAO/WHO, 1977). As **II** and **III** progressively degraded, no peaks of the corresponding hydroxypyrimidines were detected during chromatographic analyses.

The data from the photodegradation test showed that pirimicarb rapidly degraded under these laboratory conditions, leading to metabolites **II** and **III** following parallel and not consecutive pathways. Owing to their low concentration and low degradation rate, it can be reasonably assumed that **II** and **III** were not the only products of the transformation of pirimicarb.

CONCLUSIONS

Pirimicarb showed similar behaviors in both peaches and nectarines. It was stable in the greenhouse and underwent a moderate degradation in the field. The presence of metabolites (**II**–**VII**) was never detected, though it was allowed by the analytical method. Therefore, in the case of peaches and nectarines, only pirimicarb will be important in residue evaluation, different from asparagus and lettuce, where pirimicarb metabolites account for more than the ai (Szeto *et al.*, 1984; Szeto *et al.*, 1985; Cabras *et al.*, 1990). In the literature (FAO/WHO, 1977), volatilization is referred to as the primary means of pirimicarb loss, but this was not confirmed by our investigation. Another trial on lettuce (Cabras *et al.*, 1990) showed that the decrease in pirimicarb concentration had to be ascribed to the rapid transformation of the ai into its metabolites **II** and **III** and not to its volatilization. Considering that pirimicarb is rapidly degraded by sunlight, its stability in peaches and nectarines could be ascribed to the diffusion of the ai in the waxy layer of the fruit surface that could absorb or reflect the solar radiation responsible for photolysis, thus not allowing pirimicarb degradation. The rapid degradation of pirimicarb in lettuce depends on its translaminar action, for which the ai enters the leaves and is enzymatically degraded.

In case of repeated pesticide treatments, the remarkable stability of pirimicarb in peaches and nectarines could lead to AI accumulation in the fruits, thus causing high residues at harvest time. Therefore, as shown by data in this investigation or reported in other papers (Flori *et al.*, 1992; Banfi *et al.*, 1992), in countries (*e.g.* Italy) where a rather low maximum residue limit has been established for pirimicarb (0.2 ppm, as opposed to 0.5 ppm, indicated by Codex Alimentarius and FAO/WHO), the likelihood of residues above the legal limit at harvest time is rather high.

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