JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY

How Can the Fipronil Insecticide Access Phloem?

ASMAE AAJOUD, MURIEL RAVETON,* DALILA AZROU-ISGHI, MICHEL TISSUT, AND PATRICK RAVANEL

Laboratoire d'Écologie Alpine, Université Joseph Fourier, B.P. 53, F-38041 Grenoble Cedex 09, France

Seeds of sunflower plants coated with the fipronil ^{14}C-insecticide were allowed to grow in the greenhouse. The distribution of the ^{14}C-compounds was studied in each part of the plant after three months. After 83 days of culture small amounts of ^{14}C-compounds were found in the inflorescence (0.6‰ of the seed deposit) which were fipronil itself or its lipophilic or hydrophilic metabolites. The ^{14}C-compounds were found in each part of the inflorescence (bracts, ray and disk florets containing pollen, akenes). The ^{14}C-concentration in the xylem sap evaluated at this stage was much too low to explain the accumulated amount in the inflorescence. Under controlled conditions in a culture chamber, it was then demonstrated that a net phloem transfer of ^{14}C-fipronil occurred from developed leaves to growing organs. This allowed us to suppose that a similar ^{14}C-fipronil phloem transfer could occur toward the inflorescence during its formation. A quantitative evaluation suggests that most of the labeled compounds at this stage were not coming from the leaves but from the roots and stem where storage compounds were hydrolyzed for sustaining inflorescence development.

KEYWORDS: Fipronil; sunflower; inflorescence; phloem

INTRODUCTION

Fipronil [5-amino-3-cyano-1-(2,6-dichloro-4-trifluoromethyl)phenyl-4-(trifluoromethyl sulfinyl) pyrazole] (**Figure 4**) is mainly used in agriculture against soil insects, such as wireworms (*1*). The biochemical target of this insecticide is the γ-amino butyric acid gated chloride channel (*2*), and it has much more potent activity on insects than on mammals (*3*).

Fipronil is used as a seed coating formulation named Regent TS, mostly for maize and sunflower cultures (*4*). Under this formulation, fipronil is applied in solid, crystalline form and is retained in the coat by formulation agents. It is then able to provide an efficient protection of the seed and seedling against soil insect attacks.

At the end of the 20th century the question arose of a possible damage to honey bees, due to insecticides used as seed coating, mainly for sunflower (*5–7*). Such—hypothetical—effect required that the active ingredient (a.i.) be absorbed by the roots of the culture and transported to the inflorescence where it would be found inside pollen grains or nectar, later harvested by honey bees. To produce such an effect, a complex transfer appeared to be required. It has been previously demonstrated that ^{14}C-fipronil was absorbed by sunflower plantlets from an aqueous medium or from soil. Such uptake was carried out by the roots and bound to a xylem transfer, the rate of which was transpiration dependent (*8*). However, such a transfer was unable to solely explain a possible accumulation of a.i. inside the inflorescence. Therefore, the involvement of the phloem pathway seemed necessary.

A large number of herbicides are known for being phloem-transported, such as aminotriazole, glyphosate, phytohormones of the 2,4-D series, or even paraquat (*9–12*). However, the physicochemical properties of these herbicides differ radically from those of the insecticides used as seed coating against soil wireworms (i.e., fipronil, imidacloprid, ...).

A careful examination was therefore required to see whether a phloem transport of such insecticides was possible and whether its result was to accumulate an insecticidal activity inside pollen or nectar in cultivated plants submitted to insect pollination.

The purpose of this report was to study these points in the case of the sunflower culture seed coated with the insecticide fipronil. Two questions were considered: (a) Is it possible under field conditions or in the greenhouse to detect fipronil or its insecticidal derivatives in the inflorescence or pollen of sunflower plants issued from fipronil-coated seeds? (b) Under laboratory conditions, is it possible to demonstrate a phloem-transfer of ^{14}C-fipronil from source to sink?

MATERIALS AND METHODS

Chemicals. Fipronil [5-amino-3-cyano-1-(2,6-dichloro-4-trifluoromethyl)phenyl-4-(trifluoromethylsulfinyl) pyrazole (*13*)] was used (purity: 99.3%, Sigma). The ^{14}C-molecule was uniformly labeled on the aromatic ring with a 741 Bq nmol^{-1} specific activity (Rhône-Poulenc, Ongar, U.K.). Sulfide-fipronil (98.8% purity), amide-fipronil (99.8% purity), sulfone-fipronil (99.7% purity) were synthesized by Rhône-Poulenc, Ongar, U.K. (see the caption of **Figure 4**).

* To whom correspondence should be addressed. Phone: (+33) 4 76 51 46 80. Fax: (+33) 4 76 51 44 63. E-mail: muriel.raveton@ujf-grenoble.fr.

10.1021/jf800043h CCC: $40.75
Published on Web 04/29/2008

Greenhouse Sunflower Cultivation. Sunflower seeds (98% germination percentage) were coated with unlabeled fipronil (437 μg/seed) and with ^{14}C-fipronil (30 μg/seed; 50000 Bq/seed). Each seed was planted in about 16 kg (total fresh weight (FW)) of compost soil (replicates: 5), and distilled water was added to 35% of the soil weight. During the experiment, water loss by evapo-transpiration was weighted every day and compensated for by spraying distilled water uniformly over the soil surface. The experiment was carried out from April to June.

Phloem Transfer Experiments. Untreated sunflower seedlings were grown in a culture chamber (16 h day, 26 ± 2 °C, 250 μE m^{-2} s^{-1} PAR and 8 h night, 22 ± 2 °C; 78% relative humidity) with 100 g of a humus-clay (1:1 w/w) mixture for one seed. The first leaf was treated with droplets containing ^{14}C-fipronil and 2% DMSO. Then, the treated leaf was maintained in a water vapor saturated atmosphere to avoid droplet dehydration. The experiment was repeated three times. After seven days, the free deposit remaining on the treated leaves was washed with water (+ 2% DMSO).

Plant Extraction Procedures. At the end of the experiment (83 days), plant organs were submitted to three successive extractions with acetone. The residual powders were shown to be deprived of radioactivity.

The acetonic solutions were evaporated to dryness. An aliquot of the dry residue was dissolved with 10 mL of mineralization mixture H_2O_2/perchloric acid/H_2O (1:1:1 v/v/v) in closed polyethylene flasks. After 5−6 days of mineralization at 25 °C, the radioactivity of the mineralization mixture was measured through scintillation counting (1414 Winspectral EG&G Wallac). During the mineralization step, $^{14}CO_2$—possibly released from the flask—was trapped with KOH solution (1 M).

Each measurement was repeated five times. The average value and SE were calculated as shown in all figures.

Transpiration Rate. The weight of each pot containing one plant was measured every day, compared to the weight of a plantless pot; then the loss of water corresponding to plant transpiration was calculated. The transpiration rate was evaluated as water loss per leaf surface unit.

Analyses of the Acetonic Extracts. Fipronil and its metabolites from harvested plant tissue extracts were analyzed using TLC and GC-MS.

TLC was carried out using silica gel plates (G-25 UV_{254}, Macherey-Nagel) with two successive solvents, petroleum ether (boiling point (bp) 40−60 °C)/dichloroethane (1:1 v/v) and petroleum ether/dichloroethane/ethyl acetate (24:24:1 v/v/v) as a second solvent. The quantitative estimation of the different labeled compounds was established using a [^{14}C] thin layer chromatoscanner (Cyclone, Perkin-Elmer). Under these conditions, the measured R_f values were 0.35 for fipronil, 0.80 for sulfide-fipronil, 0.68 for sulfone-fipronil, and 0.064 for amide-fipronil as shown with the use of pure references.

GC-MS measurements were carried out on a HP6840/HP5973 apparatus (Agilent Technologies, Les Ulis, France) equipped with an MDN-12 fused silica capillary column (30 m, 0.25 mm internal diameter, 0.25 μm film; Supelco). The injector was used in the split mode, with a split ratio of 50:1 and an injection volume of 2.5 μL. The oven temperature was kept at 70 °C for 4.5 min, then increased to 240 °C at a rate of 50 °C min^{-1} and kept for a further 20 min. To detect fipronil and its standard metabolites, samples were analyzed in the full scan mode (50−550 mass range). The following retention times were determined for standard chemicals: sulfide-fipronil (11.17 min), fipronil (11.29 min), sulfone-fipronil (13.11 min), and amide-fipronil (16.32 min).

RESULTS

Fipronil Distribution in Sunflower Plants. An 83-day-long culture of sunflower plants was carried out on soil from fipronil coated seeds. At the end of this culture, 40 leaves and one inflorescence were formed, giving 678 ± 84 g FW of total aerial parts. The weight of the inflorescence was 163 ± 5 g. During the last 15 days of culture, the plant transpiratory rate was estimated at 20 ± 4 mL day^{-1} dm^{-2} leaf area. Slight variations of this transpiratory flux occurred as a function of the plant

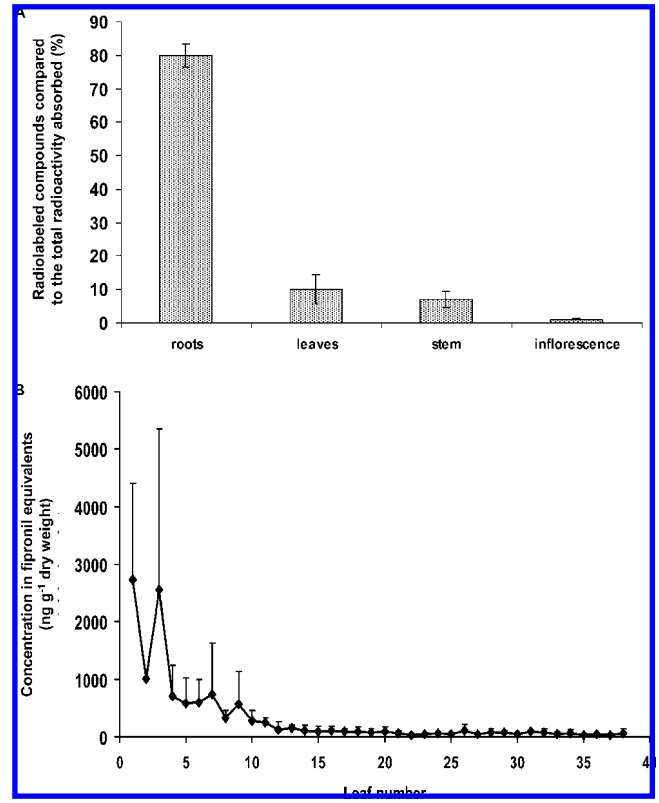


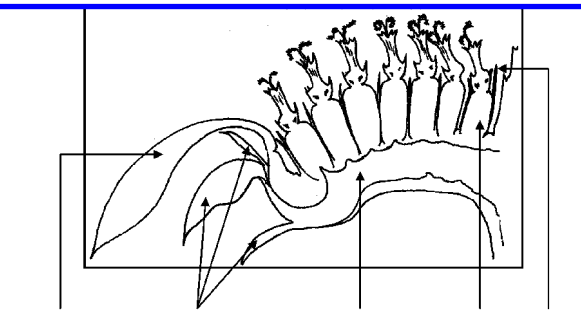
Figure 1. Equivalent-fipronil distribution in sunflower plants at day 83 of cultivation. (**A**)Distribution of radiolabeled compounds in plant organs (five replicates ± SE). (**B**)Concentration of radiolabeled compounds in leaves from the oldest one (no. 1) to the youngest one (no. 38) (five replicates ± SE).

age. The average value of the total amount of water lost per plant through transpiration during the 83-day experiment was 56 ± 4 L.

The ^{14}C-compounds distribution after the 83-day culture was as follows: 55 ± 18% were inside the soil, 36 ± 20% remained in the seed coat, and 9 ± 2% were inside the plant.

Figure 1A shows the distribution of ^{14}C-compounds between the different parts of the sunflower plant. The organs which concentrated an important quantity of radiolabeled compounds were roots (80 ± 3% of the inner content) while aerial parts accumulated only 18.5 ± 3.1%. Leaves contained quite a high amount of ^{14}C-compounds (10 ± 4%), but the distribution was not homogeneous depending on leaf age (**Figure 1**B). The first leaves (nos. 1−4) showed a high ^{14}C-compound concentration, reaching 5 nmol g^{-1} dry weight. This concentration decreased to 1.3 nmol g^{-1} in the leaves nos. 5−9 and was only 0.13 nmol g^{-1} in the leaves nos. 23−38. Therefore, the concentration inside the first leaves was approximately 38 times higher than in the last ones.

The inflorescence accumulated a low amount of ^{14}C-compounds reaching 0.66 ± 0.1% of the radioactivity absorbed by the whole plant (this amount corresponding to 0.6‰ of the seed deposit; **Figure 1**A). The distribution of radioactivity within the sunflower inflorescence organs (ray-florets, disk-florets, pappus of hairs, capitulum bracts, and flat receptacle, **Figure 2**) was worth measuring in order to evaluate the potential of pollen contamination. Therefore, after a dissection of all these organs, radioactivity was measured as shown in **Table 1**. Inside the inflorescence, radioactivity reached 3.68 ± 2.58 pmol g^{-1} FW, with around 70% of the radioactivity accumulated in sterile organs such as capitulum bracts and receptacle. The ray- and



	Number	Fresh weight (g)	Exchange area with the atmosphere (cm^2)
Capitulum bracts	49	9	165
Ray florets	60	13	324
Disk florets	4160	54	1387
Pappus of hairs	4160	35	300
Receptacle		52	80
Total		**163**	**2256**

Figure 2. Anatomic structure of the sunflower inflorescence.

Table 1. ^{14}C-Equivalent-Fipronil Distribution inside Inflorescence Organs at Day 83 of Cultivation[a]

	radioactivity (%)	concentration (pmol g^{-1} fw)
capitulum bracts	39 ± 18	10.92 ± 7.73
ray florets	12 ± 4	7.98 ± 5.40
disk florets	2 ± 2	3.80 ± 1.23
akene in formation	18 ± 9	1.84 ± 1.23
receptacle	29 ± 13	2.45 ± 2.58
global inflorescence	100	3.68 ± 2.58

[a] Five replicates ± SE.

disk-florets which contained the stamen with pollen did not show a specifically high ^{14}C-compounds concentration (ranging between 4 and 8 pmol g^{-1} FW).

Fipronil Transfer to Sunflower Inflorescence. In a first approach it was supposed that inflorescence contamination could occur via a xylem transfer of fipronil from roots to inflorescence. In order to obtain confirmation, the inflorescence transpiratory fluxes were measured (**Table 2**). The evaporation of water occurred either via stomata located on receptacle and capitulum bracts or through the thin cuticle of aerial tissues (ray florets, pappus of hairs, ...). **Table 2** shows the amount of labeled compounds (Qt + QT) brought by the xylem sap into the inflorescence with the water used for cell constitution (QT) and for transpiration (Qt). This amount only represented less than 4% of the total radioactivity present in this organ. Therefore, inflorescence contamination by a phloem way was to be considered.

The experimental device used here was designed to show whether a phloem transfer of ^{14}C-fipronil could occur, from the first photosynthetically active leaves to the sinks (young-developing leaves and roots).

For studying phloem migration of an a.i. from the source leaves, it was necessary to obtain an intense and long-term penetration of the a.i. into the source leaves, where a superficial deposit had been done (*14*). For this purpose, an absolute requirement was to avoid fipronil crystallization in the droplets

Table 2. Transpiration Rates of the Sunflower Inflorescence[a]

Inflorescence		
fresh weight (g)	water content W_c (mL)	total water transpirated per inflorescence V (mL)
163 ± 5	130.4 ± 3.3	19.2 ± 7.4

Estimated Contamination of the Inflorescence				
Cs[b] (pmol mL^{-1})	Qt[c] (pmol)	QT[d] (pmol)	C_c[e] (pmol g^{-1} fw)	C_c/C_m[f]
0.15 ± 0.04	2.88	19.5	0.14	0.04

[a] Five replicates ± SE. [b] Cs: concentration of ^{14}C-compounds in the xylem sap (*13*). [c] Qt: quantity of ^{14}C-compounds corresponding to the transpiration flux [Qt = Cs × V]. [d] QT: Quantity of ^{14}C-compounds transferred to the inflorescence with the constitutive organ water [QT = Cs × W_c]. [e] C_c: calculated concentration of ^{14}C-compounds transferred into the inflorescence by the xylem sap flux [C_c = (Qt + QT)/fresh weight]. [f] C_m: measured total concentration of ^{14}C-compounds (cited in **Table 1**).

deposited on the leaf cuticle (11.5 nmol fipronil per leaf). This was obtained through the addition of 2% DMSO in the aqueous solution, DMSO being a good solvent for fipronil with a high bp, avoiding its volatilization. Furthermore, the treated leaves were enclosed in shut Petri dishes on whose side a hole was made through which the leaf petiolae could be let out (**Figure 3**). Under these conditions, a high relative humidity was obtained in the atmosphere surrounding the treated leaves, and the deposited droplets containing ^{14}C-fipronil never dried.

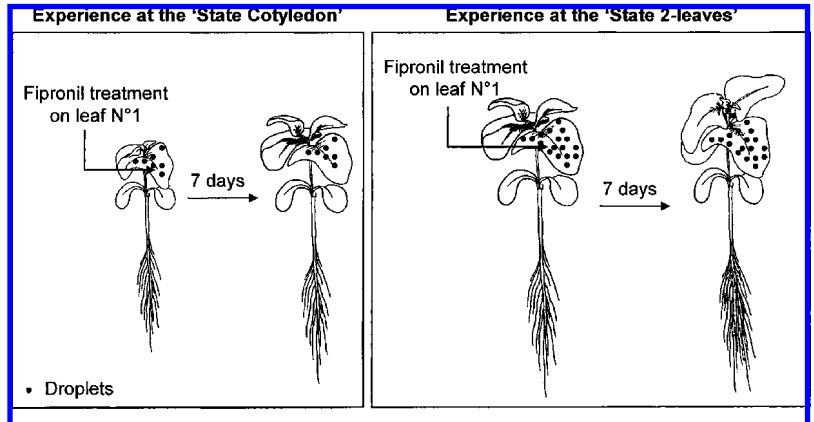
The radioactivity percentage that penetrated into the plant was 79.1 ± 19.9% of the deposit, corresponding to 9 ± 2.3 nmol per treated leaf. The average uptake flux was 1.3 nmol $leaf^{-1}$ day^{-1}.

The experiments were repeated at two development stages of the sunflower seedling. (a) The first chosen stage was named 'State cotyledon': leaf no. 1 was growing, and leaf no. 2 had a very small size when leaf no. 1 was treated (9 days of culture). b) The second stage was named 'State 2-leaves': leaf no. 1 reached its normal size, and leaf no. 2 was developing when the treatment of leaf no. 1 took place (12 days of culture). **Figure 3** shows the results of these experiments 7 days after treatment.

The results show (1) that a phloem transfer of fipronil was possible, from source to sink, inside the seedling, and (2) that the rate of this transfer varied a lot with the physiological state of the plants. A high rate was obtained toward the stem and roots, from the expanding leaf no. 1 ('State cotyledon'). This leaf afterward rapidly lost its exporting activity.

Origin of Fipronil Phloem Transfer. In the flowering sunflower plants developing in the greenhouse, the radioactivity concentration of the inflorescence was 3.68 ± 2.58 pmol g^{-1} FW and 7.08 ± 2.31 pmol g^{-1} FW in the physiologically active leaves (nos. 23−38). The concentration ratio (7.08/3.68 = 1.9) demonstrated clearly that the relative high concentration reached in the inflorescence could not be explained by a phloem transfer from these leaves.

It was therefore supposed that another tissue could be responsible for a phloem mobilization of fipronil. The first tissue which may act in this way was the pith of the stem. The measurement of radiolabeled compounds in the stem tissues showed a low but non-negligible concentration in the pith zone (2.33 pmol g^{-1} FW; **Table 3**). The global quantity of ^{14}C-compounds measured in pith after 83 days of cultivation was about 290 pmol. Therefore, the role of the stem pith in a potential phloem fipronil transfer toward inflorescences seemed low.



	Distribution of radiolabeled compounds (% of the ^{14}C-compounds penetrated)	
	Plantlets 'State cotyledon'	Plantlets 'State 2-leaves'
New leaves	3 ± 0.5	0.85 ± 0.115
Stems	10 ± 0.5	0.1 ± 0.002
Roots	16 ± 0.5	0.15 ± 0.05
Total	29	1

Figure 3. Distribution of equivalent-fipronil via a phloem transfer from leaves to sink tissues.

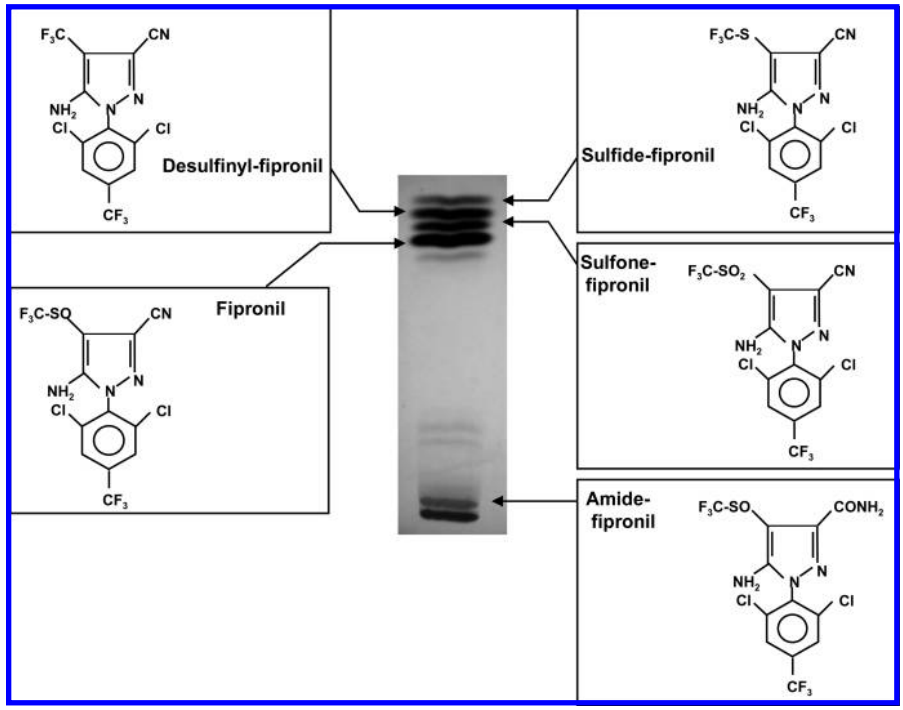


Figure 4. ^{14}C-fipronil and its metabolites present in sunflower inflorescences after 83 days of cultivation. Three successive solvents were used for this silice TLC: solvents no. 1 and no. 2, then the following mixture (no. 3), petroleum ether (bp 40−60 °C)/dichloroethane/ethyl acetate (24:24:5 v/v/v). Sulfide-fipronil: 5-amino-3-cyano-1-(2,6-dichloro-4-trifluoromethyl)phenyl-4-(trifluoromethylsulfide)pyrazole. Sulfone-fipronil: 5-amino-3-cyano-1-(2,6-dichloro-4-trifluoromethyl)phenyl-4-(trifluoromethylsulfone)pyrazole; Amide-fipronil: 5-amino-3-amide-1-(2,6-dichloro-4-trifluoromethyl)phenyl-4-(trifluoromethylsulfinyl)pyrazole.

The second organ 'source' could be the roots through a remobilization of the fipronil root stock. Indeed at day 83 of cultivation, roots were able to concentrate approximately 80% of the total fipronil absorbed by a sunflower plant. This quantity found in the roots represented 77.5 ± 35.4 nmol (**Figure 1**A).

Nature of the ^{14}C-Labeled Compounds Accumulated Either in the Young Leaves or in the Inflorescence. During the 83-day-long experiment, with a low rate transfer from the coating to soil and to plant organs, it was shown that fipronil was partly submitted to structural changes, mainly yielding the sulfone-, the sulfide-, the desulfinyl-, and the amide-fipronil

Table 3. ^{14}C-Compound Repartition in Stem Tissues at Day 83 of Cultivation[a]

	radiolabeled compounds (pmol g^{-1} fw)
suber	33.01 ± 2.12
xylem zone	9.69 ± 1.96
pith	2.33 ± 1.23

[a] Five replicates ± SE.

derivatives (**Figure 4**). The amide derivative has a lower K_{ow} than the parent compound and has only a limited insecticidal activity (*15*). It was therefore necessary to know what ^{14}C-labeled structures were present inside the inflorescence and also inside the youngest leaves.

Figure 4 shows an autoradiogram of an inflorescence extract separated on TLC. All the known fipronil metabolites were present, and a considerable proportion of fipronil itself could be seen.

DISCUSSION

The 83-day experiment in the greenhouse demonstrated that 0.6‰ of the ^{14}C initially labeling the benzenic ring of fipronil in the coating of sunflower seeds was transferred into the inflorescence. A main proportion of this label was present in this organ as inside the leaves, under the form of the a.i. or of its insecticidal metabolites.

The inflorescence is a massive organ, with a weight (163 g FW) higher than the fresh weight of the leaves (106 ± 17 g) and with a complex anatomical structure. The labeled mixture distributed without important changes among the different parts of this inflorescence.

Two main ways were suspected to be involved in the transfer of fipronil or its metabolites from the roots to this organ: the xylem and the phloem pathways. The first way has been previously shown to act for the transfer from the roots to the leaves and to depend upon the rate of leaf transpiration and upon the concentration of the labeled products in the xylem sap (*8*).

The inflorescence formation occurs after two months of culture. At this stage, the estimated fipronil concentration in the xylem sap was very low (0.15 ± 0.04 pmol mL^{-1}), giving therefore a low ^{14}C-compounds concentration in the new leaves. Furthermore, the transpiration rate of the inflorescence at different development stages was measured and was very low, in marked contrast with the leaves. As a consequence, the xylemian water movement toward the inflorescence could only import into it a small percentage (4% of the radioactivity measured in the inflorescence) of the ^{14}C-amount which was effectively measured inside it.

The second way, that of the phloem sieve tubes, seemed therefore to be involved. Such an hypothesis required to demonstrate clearly that phloem transport of fipronil was theoretically possible. This was achieved using sunflower plantlets and ^{14}C-fipronil leaf uptake. As a matter of fact, a pure phloem transfer of the a.i. was shown, from the leaf source to sinks, mainly represented by the nonphotosynthetic, growing organs. The best stage, for this demonstration, was when the seed storage compounds were exhausted and when leaf no. 1 began to export organic compounds.

Thus the ^{14}C-content of the inflorescence was suggested to be mainly the result of a phloem transfer. Furthermore, it was shown that it did not only concern hydrophilic, inactive metabolites.

The source involved in such a transfer might be the physiologically active leaves (leaves nos. 23−38). However, their ^{14}C-content seemed too low to agree with the rate of ^{14}C-accumulation inside the inflorescence. It is therefore suggested that a new phloem source opened at the stage of inflorescence formation. This main source appeared to be the first root having a high ^{14}C-content. The stem pith emits large amounts of hydrophilic carbohydrates, able to be loaded into the sieve tubes. However, it had a relatively low radiolabeled content. This suggests that a complex exchange of radiolabeled compounds might occur between the root and the pith at the stage of inflorescence development.

As a whole, when using sensitive methods, one can see that small amounts of relatively lipophilic compounds can be phloem transported. This transport is based upon passive, physicochemical mechanisms, which involves K_{ow}, partition equilibrium between lipophilic and hydrophilic compartments, diffusion inside water, and export in a water stream which is physiologically regulated in the case of phloem.

Such a mechanism evidently occurs in the case of lipophilic herbicides, as was demonstrated years ago for dinoterb (*16*). However, as only low amounts of a.i. were transported by this process, they were physiologically meaningless. In contrast, some herbicides are specifically transported into the phloem through the action of membrane transporters, namely for glyphosate (*17*), or acidic trapping (*18*) for ionizable compounds with pK_a values between 4 and 6 and intermediate K_{ow}. The specific herbicide activity of such compounds depends upon their phloem loading which has been thoroughly studied previously (*19*).

The complex and low rate transfer from seed coating to inflorescence in sunflower was demonstrated here in the case of the fipronil insecticide. Its consequences on honey bees' physiology require specific studies.

ACKNOWLEDGMENT

We are grateful to Anne-Laure Tissut for review of the English text.

LITERATURE CITED

(1) Colliot, F.; Kukorowski, K. A.; Hawkins, D. W.; Roberts, D. A. Fipronil: a new soil and foliar broad spectrum insecticide. *Brighton Crop Prot. Conf. Pests Dis.* **1992**, *1*, 29–34.

(2) Cole, L. M.; Nicholson, R. A.; Casida, J. E. Action of phenylpyrazole insecticides at the GABA-gated chloride channel. *Pestic. Biochem. Phys.* **1993**, *46*, 47–54.

(3) Bloomquist, J. R. Chloride channels as tools for developing selective insecticides. *Arch. Insect Biochem.* **2003**, *54* (4), 145–156.

(4) Turnbald, K. M.; Chen, Y. A.; Summit, N. J. U.S. Patent 5,849,320-A, 1998.

(5) Tingle, C. C. D.; Rother, J. A.; Dewhurst, C. F.; Lauer, S.; King, W. J. Fipronil: Environmental fate, ecotoxicology, and human health concerns. In *Reviews of Environmental Contamination and Toxicology*; Ware, G. W., Ed.; Springer: New York, 2003; Vol. 176, 1−66.

(6) Chauzat, M. P.; Faucon, J. P.; Martel, A. C.; Lachaize, J.; Cougoule, N.; Aubert, M. A survey of pesticides residues in pollen loads collected by honey bees in France. *J. Econ. Entomol.* **2006**, *99* (2), 253–262.

(7) Halm, M. P.; Rortais, A.; Arnold, G.; Taséi, J. N.; Rault, S. New risk assessment approach for systemic insecticides: the case of honey bees and imidacloprid (Gaucho). *Environ. Sci. Technol.* **2006**, *40* (7), 2448–2454.

(8) Aajoud, A.; Raveton, M.; Aouadi, H.; Tissut, M.; Ravanel, P. Uptake and xylem transport of fipronil in sunflower. *J. Agric. Food*

Chem. **2006**, *54*, 5055–5060.
(9) Long, J.; Basler, E. Some factors regulating auxin translocation in intact bean seedlings. *Plant Physiol.* **1973**, *51* (1), 128–135.
(10) Lichtner, F. T. Amitrole absorption by bean (*Phaseolus vulgaris* L.) roots. *Plant Physiol.* **1983**, *71*, 307–312.
(11) DiTomaso, J. M.; Hart, J. J.; Kochian, L. V. Compartmentation analysis of paraquat fluxes in maize roots as a means of estimating the rate of vacuolar accumulation and translocation to shoots. *Plant Physiol.* **1993**, *102*, 467–472.
(12) Delétage-Grandon, C.; Chollet, J. F.; Faucher, M.; Rocher, F.; Komor, A.; Bonnemain, J. L. Carrier-mediated uptake and phloem systemy of a 350-Dalton chlorinated xenobiotic with an alpha-amino acid function. *Plant Physiol.* **2001**, *125* (4), 1620–1632.
(13) Buntain, I. G.; Hatton, L. R.; Hawkins, D. W.; Pearson, C. J.; Roberts, D. A. Eur. Patent 0,295,117-A1, 1988, p 40.
(14) Tissut, M.; Delval, P.; Mamarot, J.; Ravanel, P. In Plants, Herbicides, and Weeding, 2nd ed.; CTIFL: Paris, France, 2006; 199−207.
(15) Raveton, M.; Aajoud, A.; Willison, J. C.; Aouadi, H.; Tissut, M.; Ravanel, P. Photo-transformation of the insecticide fipronil: identification of novel photo-products and evidence for an alternative pathway of photo-degradation. *Environ. Sci. Technol.* **2006**, *40*, 4151–4157.
(16) Belbachir, O.; Matringe, M.; Tissut, M.; Chevallier, D. Physiological actions of dinoterb, a phenol derivative. I. Physiological effects on the whole plant and on tissue fragments of pea. *Pest. Biochem. Physiol.* **1980**, *14*, 303–308.
(17) Arnaud, L.; Sailland, A.; Lebrun, M.; Pallett, K.; Ravanel, P.; Nurit, F.; Tissut, M. Physiological behavior of two tobacco lines expressing EPSP synthase resistant to glyphosate. *Pestic. Biochem. Physiol.* **1998**, *62*, 27–39.
(18) Kleier, D. A. Phloem mobility of xenobiotics: I. Mathematical model unifying the weak acid and intermediate permeability theories. *Plant Pysiol.* **1988**, *86* (3), 803–810.
(19) Grayson, B. T.; Kleier, D. A. Phloem mobility of xenobiotics. IV. Modelling of pesticide movement in plants. *Pestic. Sci.* **1990**, *30*, 67–79.

Received for review January 7, 2008. Revised manuscript received March 14, 2008. Accepted March 17, 2008. We are grateful to the Ministère de l'Écologie et du Développement Durable of France for financial support.

JF800043H