Long-Term Studies of Fungicide Concentrations in Greenhouses. 2. Fungicide Concentrations in Air and on Leaves after Different Exposure Times and under Different Climate Conditions

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The decline of the two fungicides vinclozolin (formulation Ronilan FL) and triadimefon (formulation Bayleton Special) was studied in greenhouses. The decline of vinclozolin was also studied in climate chambers. In greenhouse experiments the fungicides were applied with either a low-volume sprayer or a high-volume sprayer, while in the climate chamber experiments the fungicide was applied with a pipet. Concentrations on leaves 1 day after spraying in the greenhouses with Ronilan FL were 0.87 μg of vinclozolin/cm^2 and after spraying with Bayleton Special close to the detection limit (0.002 μg of triadimefon/cm^2). Concentrations were higher on floors than on leaves. Air concentrations were higher immediately after low-volume spraying than after high-volume spraying but decreased rapidly. Air concentrations of vinclozolin during the first harvest, the third day after application, were below the detection limit (0.4 μg/m^3). Climate chamber experiments showed no significant differences in residue decline rate between different climate conditions, with temperatures in the range of 18−26 °C and a vapor pressure deficit between 0.26 and 0.79 kPa.

Keywords: *Spraying; greenhouse; fungicide; pesticide; residues; decline; vinclozolin*

INTRODUCTION

Chemical control of pests and insects is common in greenhouse cultivations. An estimation of the amount of dislodgeable fungicides on different greenhouse surfaces is interesting from an occupational point of view. Vegetables in greenhouses are normally harvested the third day after fungicide application and then every second day. An earlier entry is sometimes needed for work with the crops. Knowledge of health risks in greenhouses, after respiratory as well as dermal exposure, is limited. In outdoor production of strawberries, Zweig et al. (1985) studied dermal exposure rates and dislodgeable foliar residues after vinclozolin application. Zweig found that the half-life of vinclozolin as dislodgeable foliar residue was 4 days. In greenhouses a study of dislodgeable foliar residues of benomyl was performed by Liesivuori et al. (1988). This study showed that the half-life was 44 h using an initial concentration of 0.95−1.2 μg of benomyl/cm^2. Air concentrations of some phosphorus fungicides in outdoor air have been determined by Garå (1984) using either filter or scrubber techniques. He found that there were no significant differences between the results obtained with the two techniques.

This study was undertaken to increase the knowledge of fungicide exposure and its degradation in greenhouses. Vinclozolin and triadimefon were utilized as model compounds, and complementary investigations of decay rates under well-defined conditions were performed in climate chambers. As far as we know, this is the first investigation of its kind using these two model substances under indoor conditions.

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EXPERIMENTAL PROCEDURES

The rate of decay of fungicides and their concentrations after application were measured in cucumber cultivation in greenhouses under normal growing conditions. The influence of climate factors on the decay rate was studied in climate chambers. Analysis was performed according to the methods previously developed (Papantoni et al., 1995).

Equipment. *Sprayer Equipment.* The sprayers used in the greenhouse experiments were a low-volume sprayer (cold-fogger) and a high-volume, high-pressure sprayer. Both sprayers were manufactured by Wanjet AB, Sweden.

The high-volume sprayer generates droplets through hydraulic pressure with a working pressure ranging from 5 to 15 MPa. During the experiments a pressure of 8 MPa was maintained and the liquid was distributed with four 0.8 mm nozzles placed on a 1 m metal rod connected to a long plastic tube. The pump capacity was 15 L/min, and the sprayer delivered approximately 3500−4000 L of liquid /ha.

The low-volume sprayer is equipped with a fan (370 W) placed behind the nozzle, which is of the air-jet type. Droplets are generated with a twin nozzle; the liquid nozzle is placed inside a turbulent air stream that creates and carries away the droplets while a fan, placed behind the nozzle, distributes the mist in the greenhouse. The liquid is stirred with an air stream to create a uniform concentration.

Climate Measurement Equipment. The temperature was measured with type T thermocouples (Pentronic, Gunnebo, Sweden) (accuracy ±1.5%), the relative humidity with Rotronics YA-100 hygrometers (Rotronic AG, Bassersdorf, Switzerland) (±1% at 25 °C), and the light intensity with pyranometers (Kipp & Zonen, The Netherlands).

Analytical Equipment. The air-sampling equipment used in the greenhouse experiments was a combination of a Teflon filter (0.8 μm, Millipore, Bedford, MA) and an impinger, connected in series to a Gilair S-C pump (Bicapa AB, Kungälv, Sweden).

Equipment for surface sampling in both greenhouses and climate chambers has previously been described by Papantoni

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Table 1. Experiments A−D[a] Performed in 1991−1994

expt	spraying technique	dosage per greenhouse ground area: amt of vinclozolin in Ronilan FL (mg/m^2)	dosage per greenhouse ground area: amt of triadimefon in Bayleton Special (mg/m^2)	date for application
A:1	low volume	100	2.38	Sept 13, 1991
A:2	high volume	100	1.64	Sept 20, 1991
B:1	low volume	177		Aug 20, 1992
B:2	high volume	208		Aug 14, 1992
C:1	low volume	182		May 13, 1993
C:2	high volume	208		May 21, 1993
D	low volume	143		April 7, 1994

[a] Experiment A was a pilot study performed in 1991. Spraying was performed using high- and low-volume sprayers with Ronilan FL (500 g of vinclozolin/L) and Bayleton Special (50 g/kg triadimefon) in tomato and cucumber cultivations, respectively. Sampling was performed during the first week after application. The total volume of Ronilan sprayed was 106 mL and the total volume of triadimefon was 25 and 17.3 g, respectively, in experiments A:1 and A:2. Experiments B were made in 1992. Spraying was performed with high- and low-volume sprayers with Ronilan FL (500 g of vinclozolin/L) in cucumbers. Sampling was made during the first 4 weeks after application. Experiments C were made in 1993. Spraying was performed using high- and low-volume sprayers with Ronilan FL (500 g of vinclozolin/L) in cucumber cultivation. Sampling was made during the first 4 weeks after application. Experiment D was made in 1994. Spraying was performed using a low-volume sprayer with Ronilan FL (500 g of vinclozolin/L) in cucumber cultivation. Sampling was made the first day after application on the upper- and underside of the leaves.

et al. (1995). The final analytical equipment was an HPLC system consisting of an LKB 2150 pump, an LKB 2151 variable-wavelength detector (both from LKB, Bromma, Sweden), and an HP3388 integrator (Hewlett-Packard, Avondale, PA).

Chemicals. The fungicide formulations used were Ronilan FL, containing the active ingredient vinclozolin at a concentration of 500 g/L (BASF, Limburgerhof, Germany), and Bayleton Special, containing the active ingredient triadimefon at a concentration of 50 g/kg (Bayer, Monheim, Germany).

Experimental Methods. *Spraying Procedure in Greenhouses.* When using the low-volume fogging equipment, the fan was started 1−2 h before application and the liquid was mixed and put into the tank immediately prior to spraying. Spraying was performed automatically without human presence and stopped by a timer. The spray delivery rate was 6 L/h, and the volume was 0.005−0.011 L/m^2 of greenhouse ground area. The concentrations in the formulations using the low-volume sprayer were 8.5−16 g of vinclozolin/L and 9.9 g of triadimefon/L. The spraying volume varied between 2.5 and 6 L in the experiments.

When the high-volume sprayer was used, the operator placed the machine in the main aisle and walked down the side aisles toward the main aisle. The metal tube with the nozzles was directed toward the plants and moved up and down as the operator walked backward through the aisle avoiding the spray cloud. In high-volume spraying the concentration of fungicide was 32−77 times lower than in low-volume spraying, yielding a volume of 100−209 mg of vinclozolin/m^2 of greenhouse ground area. Dosages, calculated from the quantities of fungicide placed in the sprayer tank and on greenhouse surfaces, are given in Table 1.

Application Procedure in Climate Chambers. Application of the vinclozolin emulsion was carried out on cucumber leaves marked in advance with a circle. Fifty microliters of vinclozolin emulsion was applied with a pipet on an area equal in size to the sampling area. In experiments P1−P3 the vinclozolin solution was placed on three different sites on each leaf. In experiments P4 and P5 four sampling areas were used on each leaf. Sampling was made on randomly selected leaves.

Analysis Procedure. A filter and an impinger were connected in series to a pump (see Figure 1).

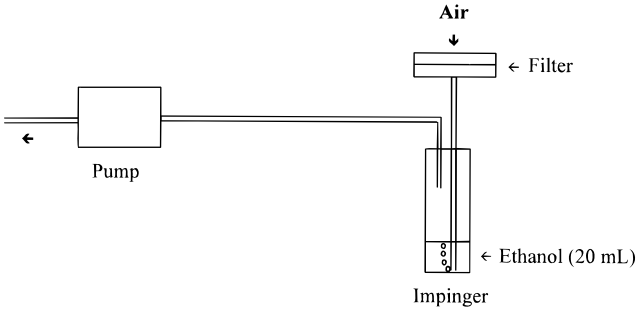

Figure 1. Air-sampling equipment: a Teflon filter (Millipore 0.8 μm) and an impinger with ethanol in series connected to a pump.

Air sampling was performed with flow rates of 1.5 L/min in the preliminary experiment (A) and 3.0 L/min in experiment B. The ethanol volume in the impingers was 20 mL. After complete sampling, the filters were extracted with ethanol and the ethanol solutions were treated in the same way as the ethanol solutions from the surface sampling.

Surface sampling was performed as described previously (Papantoni et al., 1995). The leaf was placed on the sampling device, the cylinder equiped with an O-ring was placed on the leaf, ethanol was added through the cylinder opening, and after 20 s the ethanol solution containing the dissolved fungicide was swept up with a piece of cloth. As described in Papantoni et al. (1995), when using ethanol the extraction time of 20 s is sufficient to dissolve the fungicide from the leaf surface and we assume that the risk of extracting it from the interior of the leaf is negligible. Thus, the risk for overestimating the amount of fungicide accessible for dermal exposure is judged to be small. Normally 20 or 40 cloths were collected in a specially designed steel tube and extracted with excess ethanol. After sampling, the ethanol solution was transferred to a test tube and sealed with a screw cap equipped with PFTE septa. The tubes were wrapped in aluminum foil for light protection during transport and stored in a refrigerator (8 °C) until final analysis, which normally was performed within 24 h. The time span between sampling and final analysis was less than 5 days. Storing samples in ethanol for up to 2 weeks did not noticeably change the analyte concentration.

Only the upperside of the leaves was sampled except in experiment D, when the difference in fungicide concentration between the upper- and the undersides was studied.

Sampling on hard surfaces was performed by placing a metal cylinder, equipped with an O-ring, on the surface and then using the same procedure as described above. Tightening against the surface was assured by means of the weight of the cylinder.

The ethanol solutions obtained from both air and surface sampling were evaporated to dryness, and 1-chlorobutane (0.5 mL) was added to the residue. Analysis was performed with HPLC in accordance with the procedure presented by Papantoni et al. (1995). The column used for the analysis was a silica column (Spherisorb S10W 10 μm, 20 cm length, 4.6 mm i.d., Hichrom, Reading, U.K.) operating at a flow rate of 0.5 mL/min, and the injection volume was 20 μL. Mobile phases used for the analysis of vinclozolin and triadimefon were 1-chlorobutane and 1-chlorobutane containing 1% (v/v) of ethanol, respectively. UV detection was used for the determination of vinclozolin (240 nm) and triadimefon (275 nm).

Greenhouse Experiments. In the first experiments vinclozolin spraying was applied in tomato cultivations. In subsequent experiments Ronilan FL was sprayed in cucumber cultivations. In the time span between the first and second investigations, the use of Ronilan FL in tomato cultivations was prohibited by the Swedish National Food Administration, since residues of vinclozolin had been found in tomatoes. During experiments A−C climate factors such as air temperature, air humidity, and solar radiation were measured both inside and outside the greenhouse. During experiment D climate factors were measured only inside the greenhouse. The greenhouse ground area was 528 m^2 (16 × 33 m) in experi-

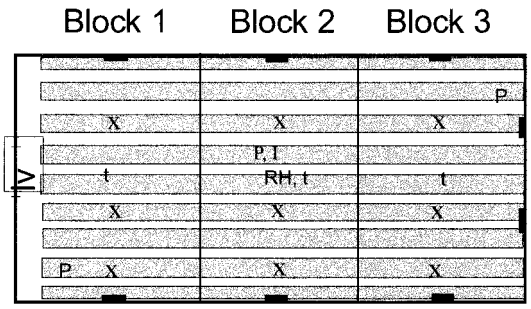

Figure 2. Positions of sampling equipment and climate measuring points in the greenhouse: t, temperature; I, pyrometer; RH, hygrometer; P, air sampling equipment. Plastic surfaces placed on the walls are marked with rectangular symbols and on the floor with x. Leaf samples were taken from the three blocks marked in the figure at a height of 1.5 m from the floor. Low-volume sprayer position is marked lv in a rectangle. The conditions for the experiments are given in Table 1.

ments A:1, A:2, B:1, and C:1 and 360 m^2 (12 × 30 m) in experiments B:2, C:2, and D. The low-volume sprayer used in the larger greenhouse was placed according to Figure 2. The experimental setups in the other greenhouses were similar.

Air Sampling. The low vapor pressures of vinclozolin and triadimefon were anticipated not to cause losses during the sampling procedure. However, to ensure that the losses were negligible, we used a combination of the filter and the impinger techniques in experiment A (see Figure 1). Since the results in these first measurements showed no detectable amount of fungicide in the impinger solutions, we used only filters in the subsequent experiment B.

In experiment A air samples were collected with stationary pump equipment during the first 24 h after application. When harvesting began, the third day after vinclozolin application, one harvester carried air-sampling equipment during 4 h (sampled air volume 360 L).

In experiment B, air sampling with stationary pump equipment was performed directly after application and air samples were collected until harvest (84 h after application).

In both experiments air sampling was performed at a height of 1.5 m (approximate breathing zone) with the sampling locations given in Figure 2. The air flow was checked with a rotameter prior to and after sampling. After sampling, the ethanol solution was transferred to a test tube and sealed with a screw cap equipped with PFTE septa and then treated in the same way as the leaf samples discussed above.

Surface Sampling. In the greenhouse experiments leaf samples were taken from three different blocks marked 1, 2, and 3 as shown in Figure 2.

In experiment A one sample was taken from each block per sampling day. In the other experiments three samples were taken from each block, in total nine samples per sampling day. All samples in experiment A consisted of 40 leaf pieces, while the samples in the other experiments (B−D) consisted of 20 leaf pieces.

In experiment A sampling was also performed on plastic sheets (acrylate on the walls and polyethene on the floor) 1, 3, and 6 days after application and on leaves 1, 2, 3, 4, 6, and 7 days after application. In experiment B sampling was performed on leaves and on plastic sheets (acrylate and polyethene) on the floor 1, 4, 7, 14, 21, and 28 days after application. In experiment C only leaf samples were taken 1, 4, 7, 14, 21, and 28 days after application, and in experiment D, 1 day after spraying, sampling was performed on both upper- and undersides of the leaves.

Climate Chamber Experiment. In the climate chamber experiments the climate factors as well as the application of fungicide could be controlled. The type of chamber (biotron) used was an artificial light chamber for which the ranges in capabilities were in terms of temperature 5−40 °C, relative humidity 30−90%, and light intensity 45−550 $\mu mol/m^2$ s. The climate chamber lights were continuously on with an intensity of 10.000 lux (140 $\mu mol/m^2$ s) and originated from Sylvania cold white fluorescent tubes. The concentration of CO_2 was approximately 350 ppm with no artificial addition. The chambers were 2.5 × 3 m in area, with a height of 2 m.

Table 2. Conditions in Climate Chamber Experiments

expt	amt of vinclozolin added for each leaf sample (μg)	temp (°C)	vapor pressure deficit[a] (kPa)	rel humidity (%)
P1	9	18	0.41	80
P2	5.5	22	0.41	84
P3	10	26	0.41	88
P4	10	22	0.79	70
P5	10	22	0.26	90

[a] Vapor pressure deficit refers to the difference in vapor pressure between present vapor pressure and the saturation vapor pressure (at the dewpoint) at the given temperature.

Cucumbers were sown December 5, 1992, and the plants were moved to the climate chambers on January 26, 1993. For later experiments cucumbers were sown January 14, 1993, and the plants were moved to the climate chamber on March 18, 1993. The fungicide decline was studied with vinclozolin as model substance during two experimental periods. During the first period (February 15−March 17, 1993) experiments P1, P2, and P3 were performed, and during the second period (April 12−May 12, 1993) experiments P4 and P5 were performed. Conditions for climate chamber experiments are given in Table 2.

An experiment in which the temperature was kept at 22 °C and the vapor pressure deficit was 1.32 kPa, resulting in a relative humidity of 50%, was also performed. These conditions led to brittle leaves, which made sampling difficult as the ethanol liquid penetrated the leaves more quickly. Consequently, no sampling was performed during these conditions.

Leaf samples were taken in accordance with a randomized plan. One sample consisted of 20 leaf pieces. Leaf surfaces to be sampled were cut to match the sampling device, in a way that the extracting solution did not come into contact with cut edges.

RESULTS AND DISCUSSION

Greenhouse Experiments. *Air Sampling.* In preliminary experiments (A) the concentration of triadimefon was, at a sampling period of 0−2 h after application, below the detection limit (3 $\mu g/m^3$). Hence, air exposure most likely is of no concern with respect to human health.

The morning after spraying, air concentrations of vinclozolin were found to be low in the greenhouse. When low-volume spraying was used, the concentrations in experiment A were 8.5 $\mu g/m^3$ (an average value for the exposure) between 1.5 and 3.8 h after application, 74 $\mu g/m^3$ 8.5−11.5 h after application, and 37 $\mu g/m^3$ 15.5−18.5 h after application. When using high-volume spraying, the concentrations meaured were 6.5 $\mu g/m^3$ 0.25−3.25 h after application and 25 $\mu g/m^3$ 13.5−16.5 h after application. These low concentrations ought to be of no concern regarding the health of workers in the greenhouse. An experiment performed with carried air-sampling equipment during harvesting, 3 days after fungicide application and with a sampling period of 4 h, did not result in detectable amounts of vinclozolin (detection limit is 0.4 $\mu g/m^3$).

No alterations were made regarding work procedures in the greenhouse during air sampling. Variation in fungicide air concentrations from time to time and place to place is thus expected.

The results of long-term air sampling of vinclozolin performed in experiment B are given in Table 3. Here we can see that after low-volume spraying the air concentration is high directly after application but declines rapidly to a very low level.

Table 3. Air Sampling of Vinclozolin in Experiment B

time after appln (h)	concn after low-volume spray (SD)[a] (μg/m^3)	concn after high-volume spray (SD)[a] (μg/m^3)
0−2	445 (253)	5.0
2−4	8.4 (3.6)	<0.4
4−7	10.9 (4.8)	0.9
8−11	4.1 (0.6)	2.5
12−15	1.3 (1.9)	3.7
16−19	−[b]	7.4
24−27	2	−[b]
32−35	1.4	1.3
48−51	1.1	1.0
84−87	1.0	1.1

[a] The values within parentheses represent standard deviations (SD) based on three samples. When no value is given within parentheses, three samples were merged to one, due to low concentrations. [b] No final analysis of these two sample was made because of problems with one of the pumps.

The concentration after low-volume spraying decreased rapidly from 445 (0−2 h sampling interval after spraying) to 8.4 μg/m^3 (2−4 h sampling interval). The last value matches the one in experiment A after 2−4 h sampling in low-volume-sprayed cultivation.

In high-volume-sprayed greenhouses the air concentration of vinclozolin was small from the start. Vinclozolin concentration was only 5 μg/m^3 1 h after application. Between 24 and 87 h after application, the concentration in air varied between 1 and 2 μg/m^3 for both spraying techniques. The risk of respiratory fungicide exposure should be negligible if the greenhouse is properly ventilated the morning after application.

Since the air concentration of vinclozolin declined rapidly, we decided not to perform air exposure measurements in experiments C and D.

Surface Sampling of Triadimefon. The recovery values for leaves and acrylic sheets obtained previously (Papantoni et al., 1995) were used to correct for losses during the analysis procedure.

Triadimefon residues in experiment A were low. After low-volume spraying the concentrations on leaves were 0.002−0.004 μg/cm^2, which is close to the detection limit (0.002 μg/cm^2). One day after high-volume spraying, the residue concentration on leaves was on average 0.009 μg/cm^2. One week after spraying, no changes in concentration on the surfaces were observed.

On floor surfaces the triadimefon concentrations were on average 0.010 μg/cm^2 after low-volume spraying and 0.013 μg/cm^2 after high-volume spraying. On walls the triadimefon concentrations were on average 0.009 μg/cm^2 after low-volume spraying and 0.011 μg/cm^2 after high-volume spraying. Six days after low-volume spraying, the concentration of triadimefon was below the detection limit (0.002 μg/cm^2), and after high-volume spraying, the concentration of triadimefon was estimated to be 0.004 μg/cm^2.

Surface Sampling of Vinclozolin. The recovery values for leaves and acrylic sheets obtained previously (Papantoni et al., 1995) were used to correct for losses during the analysis procedure.

The first experiment (A) was performed during a relatively short period. This makes an estimation of the decline rate too uncertain, since the amount of surficial fungicide residues at the end of the sampling period still was high.

In experiment A, 1 day after spraying, the amounts of vinclozolin on acrylate sheets placed on the greenhouse floor were 0.92 and 2.9 μg/cm^2 after low- and high-volume spraying, respectively. These values are considerably higher than the values obtained on leaves after low-volume (0.25 μg/cm^2) and high-volume spraying (0.68 μg/cm^2). After 6 days, the amount of vinclozolin residue on the acrylate sheets was 0.39 μg/cm^2 after low-volume spraying, and after high-volume spraying, the detected amount was still the same as 1 day after application (no leaf sampling was performed during the sixth day). We expected a lower value 6 days after application, but this might be explained by variations of the amount of fungicide applied. We know from measurements on leaves in different blocks in the greenhouse that the variation of applied amounts in different areas is large.

Table 4. Concentrations of Vinclozolin on the Surface of Plastic Sheets Placed on the Floor in Experiment B

	acrylic sheets (μg/cm^2)		polyethene sheets (μg/cm^2)	
days after spraying	high volume	low volume	high volume	low volume
1	1.59	1.81	1.40	1.59
4	1.53	1.52	1.02	1.23
7	1.02	0.89	0.79	0.08
14	0.48	0.60	0.45	0.11
21	0.01	0.08	0.04	0.03
28	a	0.04	a	0.02

[a] No sampling performed.

The results from experiment B on the greenhouse floor are shown in Table 4. Polyethene sheets are frequently used to protect greenhouse floors, and this study was intended to give an idea of the amount of dislodgeable fungicide on the polyethene sheets accessible for dermal exposure.

The presence of high amounts of fungicide on the floor (the average concentration of vinclozolin is 1.60 μg/cm^2) 1 day after spraying underscores the importance of wearing clothes that also protect feet and legs from exposure when working in treated greenhouses.

The decline rate of vinclozolin was calculated according to the decay equation

$$C = C_0 e^{-kt}$$

where C = concentration (mg/m^2), C_0 = initial concentration value (mg/m^2), t = time elapsed since spraying (days), k = time constant (days^{-1}).

The time constants on acrylic plates were 0.11 and 0.10 using low-volume and high-volume spraying techniques, respectively. Laboratory studies showed a faster decline rate on acrylate sheets with a time constant of 0.23. This may depend on the difference in initial concentration application of the fungicide in the laboratory (ca. 0.3 μg/cm^2), compared to the average value obtained on the acrylate sheets in the greenhouse (ca. 1.7 μg/cm^2) 1 day after fungicide application.

Results concerning the decline of the fungicide vinclozolin on leaves in experiments B and C are shown in Figure 3.

When harvesting began the third day after fungicide application, vinclozolin residues on leaves were approximately 80% of the applied amount regarding both spraying techniques (Figure 3). After 1 week, approximately 50% remained on the surfaces, and after 4 weeks the amount of fungicide decreased to close to zero (detection limit was 0.001 μg/cm^2).

The decline rate of vinclozolin was calculated on leaf surfaces using the equation mentioned above. The constants calculated for the two spraying techniques were, after low-volume spraying, 0.235 (SD = 0.045)

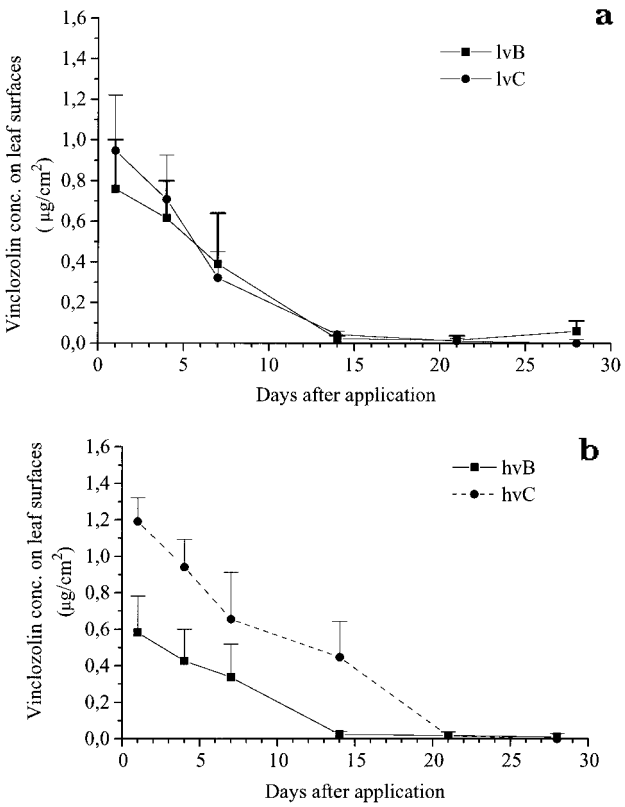

Figure 3. Decline of vinclozolin on leaves (μg/cm^2) in greenhouse after low-volume (a) and high-volume (b) application in experiments B and C.

and, after high-volume spraying, 0.167 (SD = 0.059), respectively. The decline rate is significantly faster after low-volume spraying.

After low-volume spraying in both experiments B and C and after high-volume spraying in experiment B, there was a significantly higher initial concentration at the 95% confidence level (using a *t*-test) in block 1 closest to the sprayer compared to that in block 3 farthest away from the sprayer.

Differences between blocks after high-volume spraying in experiment B can be explained by the variation in application due to manual distribution of the liquid sprayed. Examination of the results regarding the whole experimental period shows the differences within the blocks as well as between the blocks to be considerable.

With the dosage of 1.15 kg of vinclozolin/ha applied in cucumber greenhouse cultivation, the concentrations on leaves were on average 0.87 μg/cm^2 the first day after application and on average 0.005 μg/cm^2 (close to the detection limit = 0.001 μg/cm^2) 28 days after application. In outdoor cultivation Zweig et al. (1985) found 31 days after application concentrations of 0.0089 μg/cm^2, 32 days after application 0.0054 μg/cm^2, and 33 days after application 0.0052 μg/cm^2, using a dosage of 1.12 kg of vinclozolin/ha in strawberries. These values are approximately at the same level as the values obtained after 28 days in our investigation (0.005 μg/cm^2 on average).

Climate Chamber Experiment. The results from climate chamber experiments are shown in Table 5 and Figure 4. Our hypothesis was that higher biological activity present at higher temperature would increase the decline rate. The higher vapor pressure of the fungicide at elevated temperature should also increase the decline rate. We also assumed that a higher vapor pressure deficit or a lower relative humidity would lead to a slower decline since possible hydrolysis reactions would be slower. To test these assumptions, time constants were calculated from the values in Table 5. These constants are given in Table 6.

The climate chamber study showed little variation in decline rate among the different climate conditions in the experiment. The decline rate seems neither humidity nor temperature dependent, within these experimental conditions, since the time constants not differ significantly from each other, x^2 test ($P = 0.05$).

Comparison of Results in Greenhouses and Climate Chambers. In our decline studies we have measured the decrease in the amount of fungicide accessible for dermal exposure. A larger fraction of fungicide may be present in pores on old and brittle leaves compared to fresh leaves, yielding lower sampling values in the former case. Due to air movements the cucumber leaves in climate chambers seem to deteriorate more rapidly than in normal greenhouses. This might lead to an underestimation of the residue concentrations, especially for the last readings in a decline rate experiment.

A comparison of high-volume applications in greenhouses and in climate chamber experiments showed that there was no significant difference on average decline rate of the fungicide at the 95% confidence level using Student's *t*-test. The agreement between results obtained in the climate chamber and in high-volume spraying in greenhouses might be explained by the similarity in application of fungicide formulation regarding runoff and film spreading.

However, there is a significant difference on average decline rate at 95% confidence level when the values obtained in climate chamber experiments and low-volume spraying in greenhouse experiments are compared. The faster decline in low-volume application may depend on smaller droplets produced by this spraying technique. The droplet sizes from the two sprayers, measured with water as liquid with a Malvern 2600 laser instrument (Malvern Instrument Ltd., Malvern, England), resulted in a volume median diameters of 26 and 43 μm using the low-volume sprayer and the high-volume sprayer, respectively. The difference in droplet size using Ronilan FL may be even greater since the formulation contains emulsifiers that lower the surface tension (D. Mortensen, BASF Denmark, personal communication, 1995) of the sprayed liquid, which normally leads to a decrease in droplet size. For some additives, for example oils, the droplet size is instead increased (Höstgaard, 1991). The formulation concentration in low-volume spraying is 1.7% of Ronilan FL and in high-volume spraying is 0.1% of Ronilan FL. Thus, the droplet size when using the low-volume spraying technique is more influenced by the formulation than when using the high-volume spraying technique.

Sprayers. The main problem in determining average concentrations and decline rates of fungicides on different surfaces is the variation in deposition due to uneven spray distribution. This leads to a variation in leaf concentration in different blocks of the greenhouse. Thus, a large number of samples are required at each sampling event to make it possible to obtain reliable decline curves.

When using low-volume spraying, the droplets reach not only the crop but all parts of the greenhouse. This has been discussed by van Os et al. (1993), who found

Table 5. Decline of Vinclozolin in Climate Chambers during 4 Weeks

	vinclozolin concn (SD)[a] (μg/cm^2)				
expt	P1	P2	P3	P4	P5
day 0	0.53 (0.206)	0.39 (0.08)	0.39 (0.08)	0.62 (0.13)	0.50 (0.06)
day 4	*b*	*b*	0.27 (0.08)	0.4 (0.40)	0.33 (0.031)
day 7	0.22 (0.061)	0.24 (0.07)	0.15 (0.04)	0.32 (0.12)	0.24 (0.04)
day 14	0.12 (0.05)	0.16 (0.02)	0.06 (0.01)	0.13 (0.025)	0.12 (0.02)
day 21	0.13 (0.01)	0.14 (0.03)	0.05 (0.005)	0.16 (0.09)	0.10 (0.06)
day 28	0.11 (0.01)	0.09 (0.03)	0.01 (0.005)	0.06 (0.037)	0.05 (0.01)

[a] The standard deviation (SD) values within the parentheses are based on three samples. [b] Lost samples.

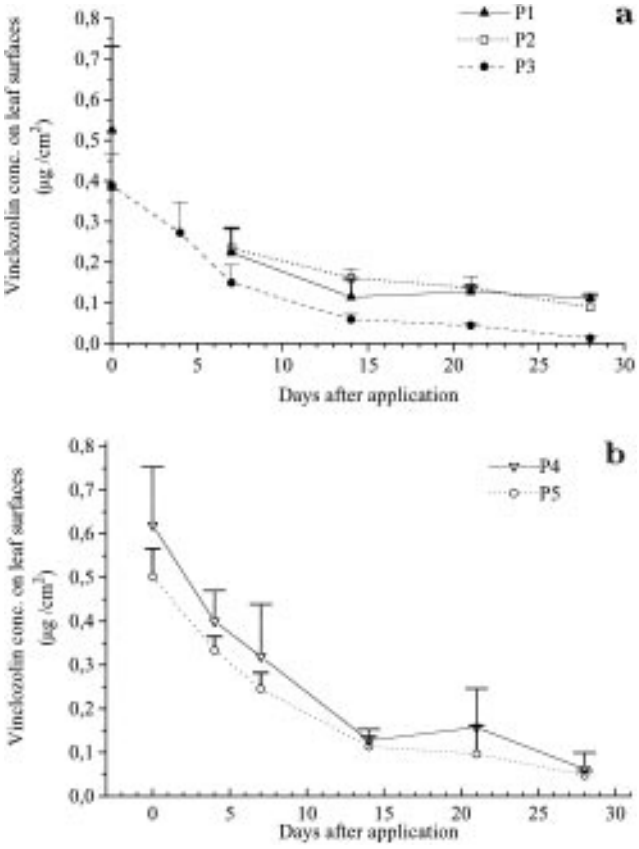

Figure 4. Decline of vinclozolin on leaves (μg/cm^2) during the first (a) and second periods (b) in climate chambers. Conditions are given in Table 3.

Table 6. Time Constants (*k*) Calculated from Values in Table 5 for Experiments P1−P5 Performed in Climate Chambers[a]

expt	time constant (*k*)	expt	time constant (*k*)
P1	0.18	P4	0.14
P2	0.08	P5	0.12
P3	0.11		

[a] C, concentration (mg/m^2); C_0, initial concentration value (mg/m^2); t = time elapsed since spraying (days); k = time constant (days^{-1}).

that the portion of the pesticide that reaches the crop is only 5−20%, while 25−50% of the pesticide escapes from the greenhouse to the outside, mostly by ventilation.

The high-volume sprayer gives a better opportunity to control the direction of the sprayed liquid by manual operation. Nevertheless, a large fraction of the applied pesticide contaminates the floor, since the volume applied is larger and the high-pressure mist is difficult to control.

In experiments A−C only the uppersides of the leaves were sampled, since most of the fungicide was assumed to be on this side. The hypothesis that a negligible fraction of the pesticide reaches the underside of the leaves was confirmed in experiment D. Using the low-volume technique, the average concentration of vinclozolin on the upperside was found to be 1.47 μg/cm^2 (SD 0.73, $n = 7$) and on the underside 0.146 μg/cm^2 (SD = 0.278, $n = 8$). These values are significantly different at the 99% confidence level using a *t*-test. Similar results have been obtained by Nielsen and Kirknel (1990) after low-volume spraying in greenhouses. In the top part of a potted plant (*Chrysanthemum*), 95% of the fungicide was found on the upperside and 5% on the underside of the leaves (Nielsen and Kirknel, 1990).

A *t*-test was performed by comparing the means of the estimated time constants for high- and low-volume application procedures. A significant difference was obtained in decline rate on leaves between the two techniques at the 95% confidence level. The decline after low-volume spraying was faster than after high-volume spraying.

After low-volume spraying, the applied substance is distributed over all surfaces in the greenhouse. This, together with the faster decline, forces the user to spray more often to achieve the same effect as after high-volume spraying. Thus, the low-volume spraying is potentially more harmful to health.

Conclusion. Results obtained in greenhouses and climate chambers indicate that the risk of dermal exposure is significant when harvesting begins. It seems possible to determine the degradation on leaves in greenhouses from the initial concentration in a single climate chamber experiment, performed at some intermediate conditions, by estimation of the decline rate of a substance. This information, along with measurements of initial concentrations on surfaces in the actual greenhouse, could then be used to predict the risk of dermal exposure. However, more investigations are needed to prove the general applicability of this approach.

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