ELSEVIER

Food Chemistry 72 (2001) 473–477

Food Chemistry

www.elsevier.com/locate/foodchem

Degradation of pyrazophos and methidathion in fortified red and white wine under conditions of light and darkness

Panagiotis Stavropoulos, Panagiotis E. Athanasopoulos *, Nikolaos B. Kyriakidis

Department of Food Science and Technology, Agricultural University of Athens, Iera Odos 75, Votanikos, Athens 118 55, Greece

Received 14 June 2000; received in revised form 11 September 2000; accepted 11 September 2000

Abstract

The decomposition of the pesticides methidathion and pyrazophos in red and white wine stored under different conditions was studied. Samples of red and white wine were fortified with 3.25 mg/kg of each of the pesticides. One lot was stored under diffuse daylight and the other in a dark closet. Both lots were stored at the same temperature. Their degradation was followed for 80 days. Half-lives of the pesticide methidathion in white and red wine stored in diffuse daylight were 20.1 and 20.0 days, respectively, and 21.1 and 24.2 days for those stored in conditions of darkness. Corresponding half-lives for pyrazophos were 29.7 and 29.1 days for storage in daylight conditions and 31.1 and 34.5 days in darkness conditions. Photodegradation was not found to play any important role in the decomposition of these pesticides under the conditions used.

1. Introduction

A number of pesticides are used in vines (*Vitis Vinifera* L.). Among the most popular pesticides there are the insecticide methidathion and the fungicide pyrazophos. Methidathion (S-2,3-dihydro-5-methoxy-2-oxo- 1,3,4,-thiadiazol-3-ylmethyl-0,0-dimethylphosphoro dithioate) (Kidd and James, 1991) is a non-systemic insecticide and acaricide with stomach and contact action. The compound is used to control a variety of insects and mites, in many crops such as fruits, vegetables, alfalfa, and sunflower, cultivated in the open and in greenhouses. It works by inhibiting cholinesterase activity in the target pests (Gallo and Lawryk, 1991). It is commonly used in Greece, in vineyards, to control insects such as Pentatomidae (*Dolycoris baccarum* L.), Lepidoptera (*Clysia ambiquella*, Hb. and *Polychrosis botrana*, Schiff) and Coleoptera (*Rhynchites bacchus,* L. and *Hoplia minuta*, Panz.). Methidathion is a highly toxic compound listed by US EPA as a class I toxic chemical (US EPA, 1991). A number of fungicides are used to control mold growth. The major pre- and post-harvest decay is due to *Aspertgillus niger*. Penicillium spp., *Botrytis cinerea*, *Rhizopus oryzae*, and *Rizolus stolonifer* (Nair, Emmett & Parker 1987). Pre-harvest application of fungicides leads to better grape storability. The efficiency of fungicide applications depends on correct timing (Nair et al., 1987) and on the use of dissimilar chemicals as a strategy for delaying the appearance of insensitive fungal strains. Methidathion is rapidly metabolized in mammals as well in plants (The Pesticide Manual, 1997).

Pyrazophos (ethyl 2-diethoxythiophosphoryloxy-5-methylpyrazolo [1,5-α] pyrimidin-6-carboxyl ester) is a systemic fungicide, widely used in vineyards to control the disease powdery mildew, caused by the fungus *Uncinula necator*. Vines can suffer serious damage, even in dry weather. Although powdery mildew is essentially a field disease, late season infection of berries can result in a lake-like light brown colour of the skin, which may be encountered in stored grapes. Pyrazophos is used especially for the protection of wine varieties and table grapes of late crop; part of that is stored in refrigerated rooms.

Photodegradation is considered to be an important parameter affecting organophosphorus pesticide degradation. The objective of this work was to determine the rate of degradation of the pesticides pyrazophos and methidathion in fortified red and white wine and also to evaluate any effect of diffuse light on the degradation rate of these important pesticides in both products.

* Corresponding author.

0308-8146/01/$ - see front matter
PII: S0308-8146(00)00264-8

2. Materials and methods

2.1. Sampling and storage procedures

Red and white wine was bought from a local supermarket in 5-kg bottles. Both types of wine were fortified with standard solutions of methidathion and pyrazophos to obtain a final concentration of 3.25 mg/kg of active ingredient (ai) and packed into 125-ml glass bottles (24 bottles for each type of wine). The bottles were made from clear glass (thickness 0.6 cm) transparent to light at wavelength above 370 nm. Fortification was adjusted to conform to the maximum expected level of pesticides that could be transferred from the grapes to wines. Twelve bottles of each wine type were stored on a shelf in our laboratory in diffuse daylight at room temperature. Equal numbers of bottles were packed in a carton box and stored in a closet, far from the daylight. Both lots of samples were kept at the same temperature throughout the experimental period, being 20±3°C. Samples were removed and analyzed at 7-day intervals up to 80 days after pesticide addition.

2.2. Analytical procedure

All samples were analyzed by a general method suitable for nitrogen-containing compounds (Ministry of Welfare, Health and Cultural Affairs, 1988) properly modified. According to the method, 50 g of the homogenized sample was mixed with 100 ml of ethyl acetate and 50 g of anhydrous sodium sulfate. The mixture was blended for 3 min and the extract filtered through a Whatman No. 1 filter paper containing 3 g anhydrous sodium sulfate, in a volumetric flask. During filtration, all parts were kept under crushed ice to avoid undue evaporation of the solvent. The clear filtrate was used for injection.

2.3. Gas chromatographic determination

A Hewlett-Packard, model 5890 series II, gas chromatograph was used. The machine was equipped with a splitless injector, an NPD, and a 30 m×0.5 mm i.d.×0.88 μm film thickness glass capillary column (Hewlett-Packard) coated with 5% phenyl methyl silicone. The injection port temperature and the detector temperature were 250 and 290°C, respectively. The column temperature was programmed as follows: the initial temperature of 120°C was increased at a rate of 20°C/min to 210°C with a residence time of 2 min. From 210 to 270°C a rate of 10°C/min was used with a residence time of 2 min, and from 270 to 285°C a rate of 13°C/min was used with a residence time of 5 min at the final temperature. Helium carrier gas at a flow rate of 7 ml/min was used. Samples (in triplicate) of 2 μl of the extract were injected, and quantitation of the pesticide was performed by automatic integration of the pick areas. Certified standards of the pesticides were used for external calibration.

2.4. Degradation kinetics

To determine degradation kinetics, plots of concentration against time were made for each data set, and the maximum squares of correlation coefficients found were used to determine the equations of best fit curves. For all eight cases studied, exponential relations were found to apply, corresponding to first-order rate equations. Confirmations of the first-order rate kinetics were further made graphically from the linearity of the plots of $\ln C$ against time.

The rate constant k, was calculated from the first order rate equation:

$$C_t = C_o e^{-kt} \quad (1)$$

Where C_t represents the concentration of pesticide at time t, C_0 represents the initial concentration and k is the rate constant in days^{-1}. The half-life ($t_{1/2}$) was determined from the k value for each experiment, being, $t_{1/2} = \ln 2/k$.

3. Results and discussion

3.1. Determination and recovery

The method of analysis was simple and fast. Quantitation of the pesticide in the examined samples was made by comparing the detector response for the sample to that measured before and after each injection with calibration standards.

The response of the detector for pyrazophos was linear in the studied range of 0.1–5 mg/kg. The efficiency of the method was evaluated by spiking eight control samples with pyrazophos at various concentrations from 0.1 up to 5.0 mg/kg. Average recoveries were from 89 to 108%. Relative standard deviations (RSD) for this pesticide were from 2.2 to 11.2%, values being within the accepted range for pesticide residues (Greve, 1984). The response of the detector for methidathion was linear in the studied range of 0.1–5.0 mg/kg. The efficiency of the method was evaluated by spiking control samples with methidathion at various concentration levels. Average recoveries were from 95 to 108%. Relative standard deviations for this pesticide were from 1.8 to 8.6%, values being within the accepted range for pesticide residues (Greve, 1984).

3.2. Degradation of methidathion and pyrazophos in wines

From experimental data, best fitting curves, regression equations and decomposition half-lives were

calculated according to the highest correlation coefficients (Table 1). Decomposition of both pesticides follows pseudo first order reaction kinetics (Fig. 1–4). Half-life values of degradation of methidathion in the white wine during storage in the daylight and in the dark, were 20.1 and 21.8 days, respectively, and those for the red wine were 20.0 and 24.1 days, respectively. Half-life of methidathion decomposition in white wine found 8.4% higher in the dark than that in the light, while for pyrazophos the corresponding increase was 4.7%. However, both these differences were smaller than the RSD and, accordingly, could not be taken into consideration. Half-life for red wine during storage in the dark, compared to light, was 21.0% higher for methidathion and 18.6% for pyrazophos. It can be seen that, for the red and the white wine stored under conditions of light, half-lives of each pesticide were almost the same being, accordingly, 20.0 and 20.1 days for methidathion and 29.1 and 29.7 days for pyrazophos. If photodegradation was playing a major role in the decomposition of these pesticides in the red wine, then half-lives of these pesticides (a) should be lower for the red wine compared to that for the white wine in the light and (b) decomposition in the dark should be almost equal. Accordingly, photodegradation seems not to play any important role in the degradation of these two pesticides, under the experimental conditions used.

In a similar study on the degradation of methidathion on vines and in wine in Italy, results were a bit different. In Italy, methidathion showed a modest persistence on the vines having a half-life of 1.5 days (Cabras, Garau, Pirisi, Cubeddu, Cabitza & Spanedda, 1995). In Greece, relevant half-life values for methidathion decomposition were relatively higher, being 5 days for decomposition

Table 1
Kinetic parameters for the degradation of methidathion and pyrazophos in red and white wine stored in light and dark

Pesticide degradation	Equation	Correlelation coefficcient R^2	Rate constant K (days^{-1})	Half life $t_{1/2}$ (days)
Methidathion				
In white wine in the light	$C = 2.791e^{-0.0344t}$	0.9681	0.0344	20.1
In white wine in the dark	$C = 3.047e^{-0.0318t}$	0.9781	0.0318	21.8
In red wine in the light	$C = 2.836e^{-0.0346t}$	0.9679	0.0346	20.0
In red wine in the dark	$C = 2.663e^{-0.0286t}$	0.9568	0.0286	24.2
Pyrazophos				
In white wine in the light	$C = 3.239e^{-0.0233t}$	0.9471	0.0233	29.7
In white wine in the dark	$C = 3.213e^{-0.0223t}$	0.9629	0.0223	31.1
In red wine in the light	$C = 3.314e^{-0.0238t}$	0.9822	0.0238	29.1
In red wine in the dark	$C = 3.160e^{-0.0201t}$	0.9352	0.0201	34.5

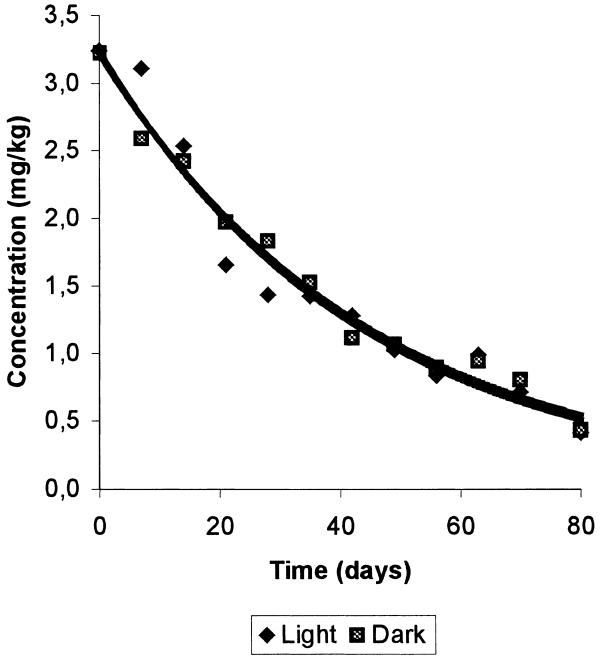

Fig. 1. Degradation of pyrazophos in white wine under conditions of light and darkness.

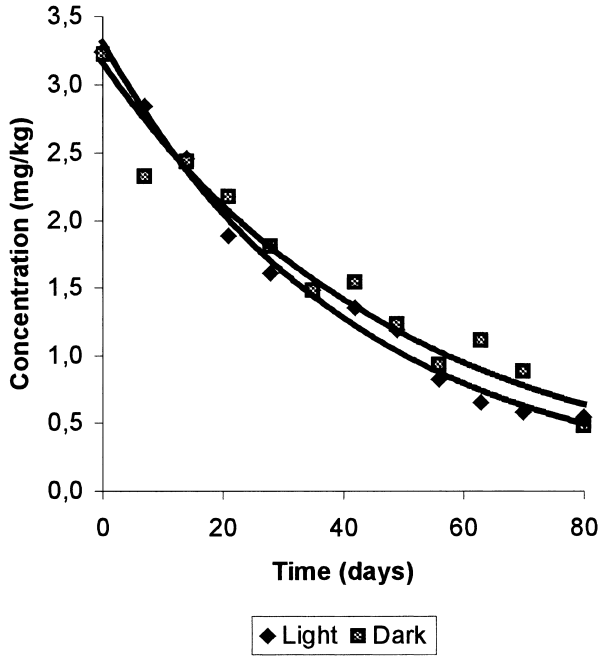

Fig. 2. Degradation of pyrazophos in red wine under conditions of light and darkness.

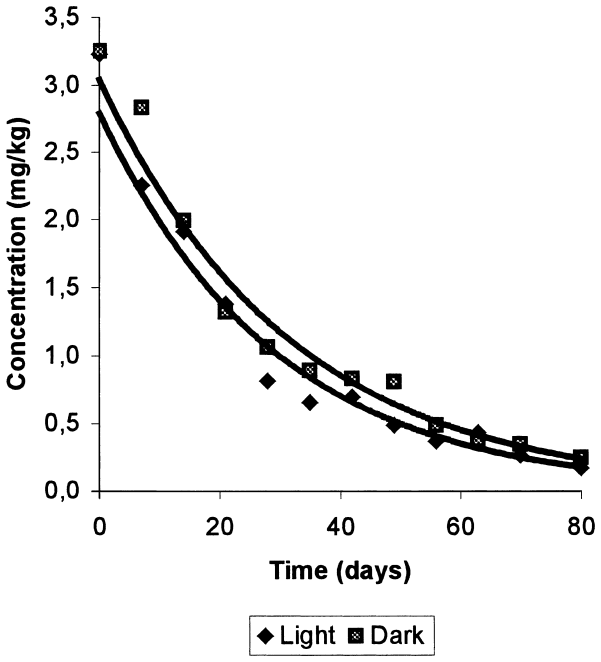

Fig. 3. Degradation of methidathion in white wine under conditions of light and darkness.

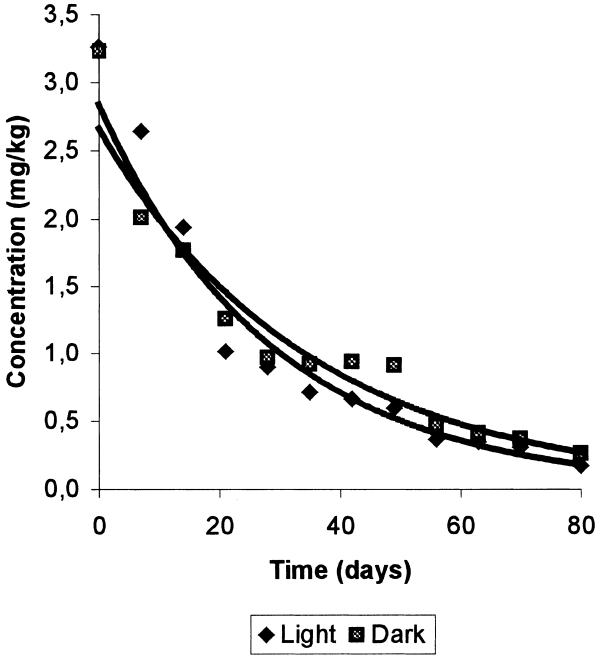

Fig. 4. Degradation of methidathion in white wine under conditions of light and darkness.

on grapes from uncovered vines and 7 days for decomposition on grapes from covered vines (Kyriakidis, Athanasopoulos, Thanos, Pappas & Yialitaki, 2000).

Cabras, Garau, Pirisi et al. (1995) reported that, during the wine making process, almost half of the initial residue was transferred to the liquid phase (must) and did not undergo further reduction during the 15-day fermentation process for wine production. In our case, we had a considerable (40.5%) decrease of methidathion during a 15-day period in wine. Our results are similar to that by Farris et al. (1992) who found a 50% methidathion rate during processing for table olive production. Half-life found for degradation of methidathion in olive oil was 10.5 days (Cabras et al., 1997). In orange fruits, still on the trees, methidathion showed high persistence, after 69 days being still above the legal limit (Cabras, Garau, Melis et al. 1995).

Concerning pyrazophos degradation (Table 1), half-lives found, were 29.7 and 31.1 days for white wine stored in the light and in the dark, respectively, and 29.1 and 34.5 days, respectively, for red wine. In this case, as also for methidathion, it can be seen that, in the white wine, light does not exhibit any effect on the decomposition rate of pyrazophos (difference was less than the SD). The red wine stored in diffuse daylight exhibited a decrease of half-life rather similar to methidathion (15.6%), compared to wine stored in the dark. Degradation rates of pyrazophos in wines were almost 40–50% higher than corresponding rates of methidathion.

Relatively similar half-lives were found for the decomposition of pyrazophos on grapes in vines, being 17 days for pyrazophos decomposition on uncovered trees and 32 days for decomposition on covered trees (Athanasopoulos, Kyriakidis, Thanos & Pappas, 2000). Rather similar half-lives, in the order of 31.4 days, were also found for decomposition of pyrazophos on greenhouse tomatoes (Miliadis, Aplada-Sarlis & Liapis 1994).

From a comparison of half-lives of pyrazophos and methidathion during storage it can be seen that half-lives of the two pesticides were shorter under conditions of light than in darkness. Photodegradation of pesticides is a well-known phenomenon (Hogenboom, Niessen & Brinkman, 1999; Sanz-Asencio, Plaza-Medina, Martinez-Soria & Perez-Clavijo 1999) but pyrazophos and methidathion have not been studied up to now.

In conclusion, half-life values for both pesticides were very similar to those found during decomposition of these pesticides on fruits and trees. It also seems that photodegradation does not play any important role in the decomposition rate of these two pesticides in red and white wine.

References

Athanasopoulos, P. E., Kyriakidis, N. B., Thanos, A., & Pappas, Ch., (2000). Decomposition of the pesticide pyrazophos on trees and during storage of Greek grapes Sultanina. *Italian J. Food Sci.* (in press).

Cabras, P., Angioni, A., Garau, V. L., Melis, M., Pirisi, F. M., Karim, M., & Minelli, E. V. (1997). Persistence of insecticide residues in olives and olive oil. *Journal of Agricultural and Food Chemistry, 45*, 2244–2247.

Cabras, P., Garau, V. L., Melis, M., Pirisi, F. M., Spanedda, L., Cabitza, F., & Cubeddu, M. (1995). Persistence of some organophosphorus insecticides in orange fruit. *Italian J. Food Sci., 7*, 291–298.

Cabras, P., Garau, V. L., Pirisi, F. M., Cubeddu, M., Cabitza, F., & Spanedda, L. (1995). Fate of some insecticides from vine to wine. *Journal of Agricultural and Food Chemistry, 43*, 2613–2615.

Farris, G. A., Cabras, P., & Spanedda, L. (1992). Pesticide residues in food processing. *Italian J. Food Sci., 4*, 149–169.

Gallo, M. A., & Lawryk, N. J. (1991). Organic phosphorus pesticides. In W. J. Hayes, & E. R. Laws, *Handbook of pesticides toxicology*. (pp. 5–3) New York, NY: Academic Press.

Greve, P. A. (1984). Good laboratory practice in pesticide residue analysis. In A. Ambrus, & R. Greenhalgh, *Pesticide residue analysis*. Proceedings of a FAO/WHO Course (p. 281). Rome: FAO/WHO.

Hogenboom, A. C., Niessen, W. M. A., & Brinkman, U. A. Th. (1999). On-line solid-phase extraction-short-column liquid chromatography combined with various tandem mass spectrometric scanning strategies for the rapid study of transformation of pesticides in surface water. *Journal of Chromatography A, 841*, 33–44.

Kidd, H., & James, D. R. (1991). *The agrochemical handbook*, 3rd ed. Cambridge, UK: Royal Society of Chemistry, Information Services.

Kyriakidis, B. N., Athanasopoulos, E. P., Thanos, A., Pappas, Ch., & Yialitaki, M. (2000). Decomposition of methidathion on Greek Soultanina grapes on the vine trees and during storage. *Journal of Agricultural and Food Chemistry*, (in press).

Miliadis, G. E., Aplada-Sarlis, P., & Liapis, K. S. (1994). Dissipation of pyrazophos residues in greenhouse tomatoes. *Bulletin of Environmental Contamination and Toxicology, 53*, 883–887.

Ministry of Welfare Health and Cultural Affairs. (1988). Analytical methods for residues of pesticides in foodstuffs (part 1, p. 3). P. A. Greve, Rijswijk, The Netherlands.

Nair, N. G., Emmett, R. W., & Parker, F. E. (1987). Programming applications of dicarboximates to control bunch rot of grapes caused by *Botrytis cinerea*. *Plant Pathology, 36*, 175–179.

Sanz-Asensio, J., Plaza-Medina, M., Martinez-Soria, M. T., & Perez-Cavijo, M. (1999). Study of photodegradation of the pesticide ethiofencarb in aqueous and non-aqueous media, by gas chromatography–mass spectrometry. *Journal of Chromatography A, 840*, 235–247.

British Crop Protection Council, The pesticide manual, 11th ed. Farnham, Surrey, UK: C.D.S. Tomlin.

US Environmental Protection Agency. (1991). Memorandum from the Office of Pesticides and Toxic Substances to Office of Pesticides Programs Division Director, Washington, DC.