Vitis 20, 329—334 (1981)

Benaki Phytopathological Institute, Kiphissia, Athens
Vine Institute, Lycovrissi-Amaroussion, Athens, Greece

# Residues of triazine herbicides in a vineyard after a long-term application[1])

by

M. E. Damanakis and B. T. Daris

**Herbicides triazine résiduaires dans un vignoble après application à long terme**

Résumé. — Dans un vignoble expérimental de la région d'Attique, où depuis 1965 on emploie régulièrement les herbicides atrazine et simazine à une dose annuelle de 5 kg m.a./ha, nous avons mesuré les résidus dans le sol au moyen d'un test biologique.

Ces mesures ont montré qu'après 6, 9 et 14 ans d'application continue l'accumulation de l'atrazine ou de la simazine est faible dans les couches de sol de 0—10 cm et de 10—20 cm.

La quantité de l'herbicide décelée ne dépassait pas les 2 kg m.a./ha ou les 40 % de la dose annuelle appliquée.

Nous avons étudié aussi l'influence de l'humidité et de la température sur la dégradation de l'atrazine; nous avons observé qu'elle est faible dans les cas où seulement l'un des deux facteurs est favorable.

## Introduction

The migration of peasants to urban and industrial centers of Greece has created a shortage in labour hands needed in vineyards and so has facilitated the introduction of herbicides for the control of weeds.

Chemical weed control in Greek vineyards is based mainly on the application of residual herbicides of the triazine group (simazine or atrazine) or of urea substitutes (diuron). The annually applied dose keeps the soil of the vineyards free of annual weeds for one season and a smaller dose is needed on the following years.

Repeated application of residual herbicides in vineyards and orchards over many years has created the necessity to investigate the possibility of residue accumulation in soil (Holly and Roberts 1963). Research conducted in apple orchards (Clay 1978) and raspberries (Clay and Yvens 1966) in Great Britain as well as in vineyards in U.S.A. (Dawson *et al.* 1968), France (Agulhon *et al.* 1969, Agulhon 1971) and Bulgaria (Nikov *et al.* 1977) has shown that no herbicide residues had accumulated in the soil.

Degradation of residual herbicides can only be accomplished if both soil temperature and moisture are favourable (Holly and Roberts 1963, Tarbert and Fletchall 1964). Since most Greek vineyards are not irrigated and summer rainfall is quite limited, it was of primary importance to examine the possible danger from repeated application of residual herbicides.

[1]) Summary of this work was presented in the Hellenic Agricultural Research Conference, Chalkidiki, Greece, 5—8 May 1981.

To study this, a non irrigated vineyard was selected in Attica, a typical viticultural region, where annual rainfall is 476 mm, only 20.7 mm of which fall during the 3 summer months (June, July, August).

## Materials and methods

The vineyard in which the level of triazine herbicide residues was determined is situated in the experimental farm of the Vine Institute, at Lycovrissi near Athens.

In this vineyard a long-term experiment, designed in split plot with 3 replications, was established in 1965. The herbicides tested were atrazine and simazine applied at 5 kg a.i./ha early in February each year from 1965 to 1979. The experimental plots were of 105 $m^2$.

The soil of the vineyard is a clay-loam with a pH 7.8 and a total calcium content of 36.2 %. The organic matter content was 1.54 % in the simazine plots and 1.66 % in the atrazine plots during the experiment (PIPPAS and DARIS 1978).

The soil samples were taken from two depths (0—10 cm and 10—20 cm) with a sampler of 5 cm in diameter, passed through a 3 mm sieve, air-dried and then kept in plastic bags at room temperature. Sampling was performed after the herbicide had been used in each plot for 6, 9 and 14 years.

Measurement of herbicide residues in the soil was conducted with a sensitive bioassay using lettuce *(Lactuca sativa)* as a test plant. The lettuce seedlings were produced by germinating seeds on wet filter paper in plastic transparent bags and kept in an illuminated chamber at 25 °C for 4 d.

The calibration lines for simazine and atrazine were produced as described below:

A quantity of 150 g of soil from untreated plots was added in small plastic pots along with 20 ml of a known herbicide solution and left for 1 h for equilibration. After this, the soil was mixed thoroughly and the pots were subirrigated in trays.

6 lettuce seedlings with a shoot of 1—2 cm were placed on the soil surface of each pot, which was previously slightly pressed, taking care for the rootlets to be oriented towards the center of the pot. The rootlets of the seedlings were then covered with a small quantity of washed sand.

The pots with the seedlings were placed in a growth chamber (25 °C and 16 h photoperiod of 25,000 lx). Fresh weights of shoots were taken after a 3-week period of growth.

The inhibition of growth expressed as a percentage of the untreated controls was plotted against the herbicide concentration on a log-probability graph paper and the calibration line was drawn (Fig. 1).

A similar technique was used for the determination of residues in the soil samples taken from the treated plots. From preliminary tests it was found that the soil from the treated plots had to be diluted down with untreated soil so that the total concentration falls within the limits of the calibration lines (0.01—0.08 ppm). A quantity of 150 g of diluted soil was placed in each pot along with 20 ml distilled water and the previously described technique was followed. The concentration of the herbicide in each pot was found from the calibration line using the percentage inhibition of fresh weight. This concentration was multiplied by the dilution factor to find the concentration in the soil sample.

It must be noted that the sensitivity of the bioassay used (0.01 ppm) is approximately 100 times higher than the sensitivity of gas chromatography method.

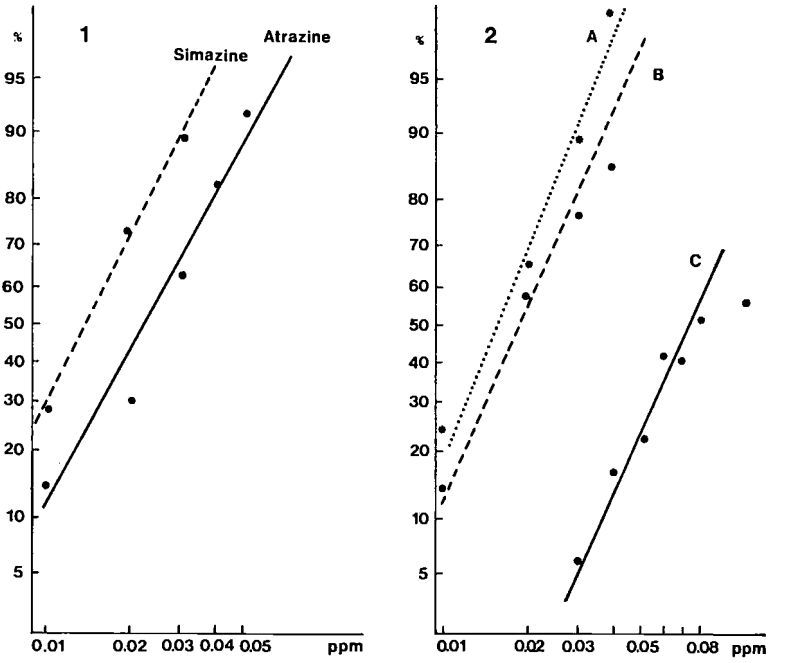


Fig. 1: Calibration lines for simazine and atrazine in soil bioassays. Horizontal axis: Herbicide concentration. Perpendicular axis: Reduction of fresh weight.

Fig. 2: Influence of temperature and moisture on atrazine degradation. Bioassays after 4 month incubation on the following conditions: 25 °C and dry soil (A), 5 °C and wet soil (B), 25 °C and wet soil (C). Horizontal axis: Initial concentration of atrazine in soil. Perpendicular axis: Reduction of fresh weight.

Fig. 1: Courbes de référence pour la simazine et l'atrazine aux tests biologiques. Axe horizontal: Concentration de l'herbicide. Axe perpendiculaire: Réduction du poids de matière fraîche.

Fig. 2: Influence de la température et de l'humidité sur la dégradation de l'atrazine. Test biologique après une incubation de 4 mois dans les conditions suivantes: 25 °C et sol sec (A), 5 °C et sol humide (B), 25 °C et sol humide (C). Axe horizontal: Concentration initiale de l'atrazine dans le sol. Axe perpendiculaire: Réduction du poids de matière fraîche.

In order to study the influence of temperature and humidity on the degradation of atrazine in the soil the following technique was used:

In 3 series of pots (A, B and C) containing 150 g of soil from untreated plots, 8 concentrations of atrazine, within the limits of the calibration line (0.01—0.08 ppm), as well as a control were made.

After the addition and mixing of the herbicide solution in series A, the soil was air-dried, placed again in the pots and kept in a dark chamber at a constant temperature of 25 °C (favourable factor for degradation only temperature).

The pots of series B and C, after addition of the herbicide solution and mixing, were closed in plastic bags to retain their humidity. Then the pots of series B were placed in a refrigerator at 5 °C and of series C in a dark chamber at 25 °C.

Pots of both series B and C were weighed every week and losses of moisture were replaced.

After 4 months, an equilibration of moisture was made in all pots and the described bioassay of lettuce was followed.

## Results and discussion

### Residue level in the soil

Residues in the soil of atrazine and simazine applied at 5 kg a.i./ ha for 6, 9 and 14 years are given in the table.

The total quantity of both herbicides varied between 1.05 and 1.75 kg a.i./ha and was never higher than 2 kg/ha, which corresponds to 40 % of the annually applied dose.

In the surface soil layer of 0—10 cm the residues were higher than in the lower layer of 10—20 cm.

These findings support the results of workers from other countries: In an irrigated vineyard in the U. S. A., after a 6-year application of simazine, 86—100 % of the residues were kept in the superficial 10 cm of the soil (Dawson *et al.* 1968). In southern France in a vineyard on clay-sandy soil, simazine residues were found mainly in the first 10 cm of soil after 6 years of continuous application (Agulhon *et al.* 1969). Finally, in a vineyard on a clay-sandy soil in southern Bulgaria, which was under continuous application of simazine for 12 years, simazine residues were found mostly in the upper 10 cm of soil but after 5—6 months the degradation of an annual dose to nontoxic products was almost complete (Nikov *et al.* 1977).

### Influence of temperature and moisture on the degradation

Influence of temperature and moisture on degradation of atrazine is given in Fig. 2. It can be observed that the 0.03 ppm concentration of atrazine after 4 months of incubation caused a growth inhibition of lettuce which was 88 % in series A (25 °C and dry soil), 75 % in series B (5 °C and moist soil) and only 6 % in series C (25 °C and moist soil). These results confirm previous findings by Holly and Roberts (1963) and Tarbert and Fletchall (1964) that both factors (temperature and moisture) must be simultaneously favourable for a quick degradation of a triazine herbicide.

Although the region in which this trial was conducted is dry during the 3 summer months (June, July, August) the results of this study show that degradation of atrazine and simazine is rather quick.

Another observation is that the detected residues of triazines are rather equal after 6, 9 or 14 years of continuous application.

No dangerous accumulation of these herbicides was therefore observed in this vineyard after a long-term application. Taking into account that a grafted vine has a productive life of 26—30 years, application of atrazine or simazine once a year for 14 years has not been proved dangerous on the vines and the soil. A decrease in organic matter of the superficial soil layer has been observed however (Pippas and Daris 1978).

Levels of atrazine and simazine residues in a vineyard after the application of 5 kg a.i./ha annually for 14 consecutive years

Niveaux des résidues de l'atrazine et de la simazine dans un vignoble après un traitement annuel de 5 kg m. a./ha pendant 14 années consécutives

| Herbicide | Soil layer (cm) | Years of herbicide application | Date of sampling | Residues in the soil[1]) (ppm) | Range of residues (ppm) | Residues expressed in kg a.i./ha |
|---|---|---|---|---|---|---|
| Atrazine | 0—10 | 6 | 19. 5.71 | 1.12 | 0.64—1.55 | 1.230 |
| | 10—20 | 6 | 19. 5.71 | 0.47 | 0.21—0.90 | 0.520 |
| Simazine | 0—10 | 6 | 19. 5.71 | 0.81 | 0.51—1.25 | 0.890 |
| | 10—20 | 6 | 19. 5.71 | 0.28 | 0.08—0.94 | 0.310 |
| Atrazine | 0—10 | 9 | 21.11.74 | 1.05 | 0.21—1.52 | 1.150 |
| | 10—20 | 9 | 21.11.74 | 0.17 | 0.08—0.37 | 0.190 |
| Simazine | 0—10 | 9 | 21.11.74 | 0.80 | 0.22—1.92 | 0.880 |
| | 10—20 | 9 | 21.11.74 | 0.18 | 0.06—0.36 | 0.200 |
| Atrazine | 0—10 | 14 | 19. 6.79 | 1.09 | 0.88—1.23 | 1.190 |
| | 10—20 | 14 | 19. 6.79 | 0.39 | 0.19—0.81 | 0.430 |
| Simazine | 0—10 | 14 | 19. 6.79 | 0.61 | 0.19—1.15 | 0.670 |
| | 10—20 | 14 | 19. 6.79 | 0.35 | 0.09—0.89 | 0.380 |

[1]) Average of 6 replications. The annual dose of 5 kg a.i./ha is equivalent to a concentration of 4.54 ppm assuming herbicide distribution to a depth of 10 cm.


## Summary

In an experimental vineyard of Attica, where triazine herbicides (atrazine and simazine) were applied yearly since 1965 in annual doses of 5 kg a.i./ha, determinations of herbicide residues in the soil were made with a sensitive bioassay.

Determinations after 6, 9 and 14 years of application have shown that no significant accumulation of atrazine or simazine is observed in the soil layers of 0—10 cm and 10—20 cm.

The detected quantity of herbicides has never overpassed the 2 kg a.i./ha which is 40 % of the annually applied dose.

The influence of temperature and moisture on the degradation of herbicide was also studied. It was found that, when only one of the two factors is favourable, the degradation of atrazine is slow.


## Literature cited


1. Agulhon, R., 1971: La pollution du vignoble par les techniques viticoles modernes. Vignes et Vins (200), 14—16.
2. — — , Gagne, R., Rozier, J. P. et Vergnes, A., 1969: Etude des résidus d'herbicides préventifs dans le sol des vignes. Progr. Agric. Vitic. 86, 184—189.
3. Allot, D. J., 1969: The persistence of simazine applied annually in the prolonged absence of soil cultivation. Weed Res. 9, 279—287.

4. Clay, D. V., 1978: Residues of simazine in soil following repeated application to fruit, ornamental and forestry crops. ADAS Exp. Hort., 30, 46—55.
5. — — and Yvens, G. W., 1966: An interim report on a soil management study in raspberries. Proc. 8th Br. Weed Control Conf. 85—91.
6. Dawson, J. H., Bruns, V. F. and Clore, W. J., 1968: Residual monuron, diuron and simazine in a vineyard soil. Weed Sci. 16, 63—65.
7. Holly, K. and Roberts, H. A., 1963: Persistence of phytotoxic residues of triazine herbicides in the soil. Weed Sci. 3, 1—10.
8. Nikov, M., Koltcheva, B. et Atanassova, D., 1977: Etude des résidus de simazine dans les vignobles et leur action sur la vigne. Connaiss. Vigne Vin 11, 93—103.
9. Pippas, B. and Daris, B. T., 1978: Influence of a long term weed control on the organic matter of the vineyards soil. Agricult. Res. 11, 5—17.
10. Tarbert, R. E. and Fletchall, O. H., 1964: Inactivation of simazine and atrazine in the field. Weeds 12, 33—37.

*Eingegangen am 3. 7. 1981*

Dr. B. T. Daris
Vine Institute
Lycovrissi-Amaroussion
Athens
Greece