Note

Transference of Organophosphorus Pesticides to Wine from Fruits during the Process of Making Fruit Wine

(Received February 14, 1997)

Toshihiro NAGAYAMA

(The Tokyo Metropolitan Research Laboratory of Public Health: 3-24-1,
Hyakunin-cho, Shinjuku-ku, Tokyo 169, Japan)

Fruit wines were produced using fruits which contained residual organophosphorus pesticides. Some of the dialifos and prothiofos contained in strawberries and some ethion in grapefruit was transferred into the fruit wines (1 to 7%, respectively). The content of pesticides in the wines was maximum after soaking for one day, and then gradually decreased.

The rate of this decrease was well described by the following first-order equation.

$$\log (Tr) = A - 0.4 \times \log (D)$$

where Tr is the content ratio of pesticide transferred from the fruit to the wine (%), A is the content transferred to the wine after one day of soaking, and D is the number of days of soaking.

Key words: organophosphorus pesticide; fruit; fruit wine; translocation; simple equation; regression expression

Introduction

Pesticides which are applied to crops may leave residues by adhering to the surface of the crop, combining with components of the crop or translocating into the crop tissue. The form of such residues depends on the nature of the pesticide, on the time elapsed since the pesticide was applied, and on the conditions of crop storage. The decrease in the amount of pesticide present during crop processing also depends on the character of the pesticide and the reactivities of the various materials in the crop with the pesticide. A further decrease may be caused by pre-consumption treatment, such as cooking.

The preparation of fruit wine is accomplished by prolonged soaking of fruit without heating. Residual pesticide in the fresh material may be transferred into the wine. Several studies have been done on the loss of pesticides in crops during cooking¹⁻⁸⁾. In these studies, however, the dissipation was examined using experimentally treated samples¹⁻⁷⁾, and the cooking involved a heating process. There is one study

dealing with the translocation of pesticides in Chinese cabbage when the cabbages were pickled in vinegar and soaked in dilute alcohol for several days⁹⁾. Translocation has also been examined using experimentally treated samples, and the correlations between behavior during short-term treatments (within three days) and the physicochemical properties (such as solubility in water) of pesticides have been described. Few studies have been conducted on the behavior of pesticides in fruits during cooking or on the loss of pesticides during prolonged immersion.

This paper deals with the translocation of three pesticides (dialifos, prothiofos and ethion) in commercial fruit (strawberries and grapefruit) during soaking for fruit wine preparation.

Materials and Methods*Materials**1. Samples*

(1) Strawberries: Commercial strawberries containing 0.28 and 0.52 ppm of dialifos and prothiofos, respectively, were obtained at a local

market in Tokyo.

(2) Grapefruits: Commercial grapefruits containing 0.22 ppm of ethion in the pericarp were obtained at a local market in Tokyo.

2. Analytical standards and reagents

(1) Analytical standards: All of the pesticides used as analytical standards were provided by Wako Pure Chemical Industries (Osaka, Japan).

(2) Reagent: Organic solvents were of pesticide analysis grade, provided by Wako. Other reagents were of pesticide analysis grade.

3. Apparatus

(1) Blender: Excel Auto Homogenizer (Nihonseiki Kaisha Ltd., Tokyo, Japan).

(2) Gas chromatograph: Shimadzu Model GC-9A (Shimadzu, Kyoto, Japan) with a flame photometric detector (FPD) and a flame thermionic detector (FTD) interfaced with a Shimadzu Chromatopac C-R2AX. The following gas chromatographic conditions were used: a column of 1.5 m × 3.2 mm i.d. glass tubing, packed with 2% Silicone DCQF-1 on 80–100 mesh Chromosorb G AW-DMCS for FPD, and 1.0 m × 3.2 mm i.d. glass tubing, packed with 3% Silicone OV-101 on 80–100 mesh Chromosorb W AW-DMCS for FTD; column oven at 180°C to 250°C depending on the pesticide; injector and detector at 250°C; helium carrier gas at 50–60 mL/min; hydrogen at 60

(FPD) or 3.5 (FTD) mL/min; air at 60 (FPD) or 150 (FTD) mL/min.

4. Processing

(1) Strawberry wine: De-calyxed strawberries (dialifos; 400 or prothiofos; 320 g) were rinsed with 1,000 mL of distilled water and immersed in 720 or 580 mL of liquor with 100 or 80 g of lemon mesocarp and 40 or 32 g of sugar (Scheme 1). After standing for 60 days at ambient temperature in the dark, the strawberries, lemon mesocarp and wine were separated.

(2) Grapefruit wine: Grapefruits (450 g) were peeled and separated into the pericarp and mesocarp. The pericarp (125 g) was prepared from grapefruits which contained ethion, and the

Scheme 1. Strawberry wine preparation procedure

Scheme 2. Grapefruit wine preparation procedure

Fig. 1. The content of dialifos during the fruit-immersion step of strawberry wine-making

●: in strawberries, □: in lemon, ○: in wine

Dialifos content was the average of 2 trials.

Fig. 2. The content of prothiofos during the fruit-immersion step of strawberry wine-making

●: in strawberries, □: in lemon, ○: in wine

Prothiofos content was the average of 2 trials.

mesocarp (300 g) was prepared from grapefruits in which ethion was not detected. Both pericarp and mesocarp were immersed in 850 mL of liquor with 150 g of lemon mesocarp and 50 g of sugar (Scheme 2). After standing for 7 days at ambient temperature in the dark, the grapefruit pericarp was removed. The remainder was left to stand for another 53 days, then the juices of the grapefruit and lemon mesocarp were squeezed into the mixture.

Analytical method

For the assays of organophosphorus pesticides in the crops and soak garbage from the wine preparation, each solid sample (20 g) was extracted with 200 mL of acetone. Immediately after the addition of sodium chloride to 5%, the wine (100 or 150 mL) was extracted with dichloromethane.

The extracts were evaporated under reduced pressure at below 40°C. The residue was diluted with 100 mL of 5% sodium chloride solution, and the mixture was extracted with 100 and 50 mL of dichloromethane twice. The dichloromethane layers were collected, dried over anhydrous sodium sulfate and evaporated to dryness under reduced pressure at below 40°C. For the grapefruit peel sample, the residue was partitioned with acetonitrile and *n*-hexane. The acetonitrile layer was evaporated to dryness under reduced pressure at below 40°C. The small amount of solvent remaining was vaporized by purging with nitrogen gas. The residue was

dissolved in 1.0 mL of *n*-hexane.

An aliquot of 5 μL of the solution was directly subjected to gas chromatography.

Results and Discussion

Translocation of organophosphorus pesticides into liquor

(1) Strawberry wine

The pesticide content in the strawberries after immersion, and the amounts in the soaked lemon and soaking liquor are shown in Figs. 1 and 2.

The pesticides (dialifos and prothiofos) in the strawberries were translocated into the liquor. The pesticide content in the strawberries decreased rapidly in the first 1–3 days, and continued to decrease with the increasing immersion time. The pesticide content in the liquor was maximum after one day of soaking, and decreased gradually after that. The translocation ratio of dialifos from the strawberries into the liquor (the ratio of the pesticide amount in the liquor to the residue amount in the fresh strawberries) was a little higher than that of prothiofos. After 60 days of soaking, the translocation ratios into the liquor of dialifos and prothiofos were about 7 and 1%, respectively. The pesticide content in the lemon mesocarp immersed with the strawberries increased with soaking time, and about 7% of the dialifos and 3% of the prothiofos in the strawberries were translocated to the lemon through the liquor

Fig. 3. The content of ethion during the fruit-immersion step of grapefruit wine-making
 ●: in the peel, ■: in the flesh, □: in lemon, ○: in wine
 Ethion content was the average of 2 trials.

after 60 days of soaking.

(2) Grapefruit wine

The concentration of the ethion remaining in the grapefruit was very high in the pericarp, and scarcely detectable in the mesocarp⁹. However, the weight of grapefruit mesocarp is about four times that of the pericarp, and even though the level of ethion residue in the mesocarp in the present experiment was below the detection limit (0.001 ppm), there might be a significant amount of ethion therein. For the investigation of pesticide translocation, the mesocarp of grapefruit in which ethion was not detected was used.

The ethion content in the grapefruit pericarp rapidly decreased during the first three days of soaking (Fig. 3). The ethion content in the liquor was about 7% of that in the fresh pericarp after one day of soaking, and gradually decreased thereafter. After 60 days of soaking, the translocation ratio of ethion into the liquor was about 1%. Conversely, the amounts in the mesocarp of the grapefruit and the lemon immersed at the same time gradually increased (Fig. 3). The behavior of the pesticides was virtually the same in the strawberry wine and grapefruit wine preparation processes.

Decrease of pesticide content after translocation into the fruit wine

The translocation ratio from the fresh materials to the liquor was the highest after one day of

Fig. 4. The relationship between the translocation ratio of pesticides and the number of days of soaking (after the first day) in strawberry and grapefruit wine preparation

●: dialifos in strawberry wine; ○: prothiofos in strawberry wine; ■: ethion in grapefruit wine

soaking, and gradually decreased thereafter. The pesticide remaining in the wine was suggested to be decomposed by the fruit components or combined with some materials of the fruit. The relation between the translocation ratio and the number of soaking days after the first day of soaking was examined (Fig. 4). The pesticide decrease with soaking time after the first day was related to the logarithm of the number of soaking days, and the regression expressions are:

$$\log (Tr) = 1.57 - 0.40 \times \log (D) \quad r = 0.996$$

for dialifos in strawberry wine preparation,

$$\log (Tr) = 0.87 - 0.44 \times \log (D) \quad r = 0.984$$

for prothiofos in strawberry wine preparation, and

$$\log (Tr) = 0.84 - 0.43 \times \log (D) \quad r = 0.995$$

for ethion in grapefruit wine preparation, where Tr is the percent of pesticide translocated into the wine from the fresh fruit and D is the number of days of soaking. The regression lines obtained were essentially the same; the rates of decrease of the organophosphorus pesticides translocated into the wine were consequently similar.

Thus, all three pesticides in the fresh fruit were rapidly translocated into the immersion liquor, and their contents therein gradually decreased with the time elapsed, according to a simple equation. Similar straight lines were

Fig. 5. The relationship between soaking time and pesticide contents in the liquor

●: dialifos from strawberries, ○: prothiofos from strawberries, ■: ethion from grapefruits

obtained for several other pesticides^{10, 11)}. It was suggested that the relation between the amount of pesticide residue and the time depends more on the decrease in the amount of pesticides on the crops by degradation, metabolism and run-off than on the amount of the pesticides applied to the crops¹¹⁾. A similar situation seems to hold for the pesticides translocated from fresh fruit into liquor in the present study.

Since the decrease of pesticide content is linear, the residual pesticide content at “ n ” days of soaking can be estimated from that after one day of soaking. The amounts of remaining translocated pesticide at “ n ” days of soaking (T_n) was calculated from the following equation, where T_1 represents the amount of pesticide translocated at one day of soaking and C represents the rate constant of decrease:

$$\log(T_n) = \log(T_1) - C \times \log(n).$$

This equation can be transformed to:

$$\begin{aligned} T_n &= 10^{\log(T_1) - C \times \log(n)} \\ &= 10^{\log(T_1)} / (10^{\log(n)})^C \\ &= T_1 / n^C. \end{aligned}$$

From the results of the above examination (Fig. 5), it was determined that $C \approx 0.3$ in strawberry wine production, and the remaining pesticide content after 60 days of soaking is given by:

$$T_{60} = T_1 / 60^{0.3} \approx T_1 / 3.42 \approx 0.3 \times T_1$$

These findings should be applicable to the

estimation of residual pesticide amounts when producing fruit wine, pickling vegetables or the like for human consumption.

Conclusion

The organophosphorus pesticide residue in fresh fruits gradually decreased during the production of fruit wine. The amount of pesticides translocated from the fresh fruits into the liquor was the greatest after one day of soaking, and then gradually decreased with time. The regression expression of the relation between the pesticide content in the fruit wine and the immersion time is:

$$\log(P) = A - B \times \log(D)$$

where A is the logarithm of the translocated pesticide content in the wine after one day of soaking, B is the coefficient of decrease (0.4 in the wine production), P is the amount of pesticide translocated into the wine from the fresh fruit, and D is the number of days of soaking.

References

- 1) Takeda, M., Otsuki, K., Sekita, H., Tanabe, H., Okajima, S., Sakai, Y.: J. Food Hyg. Soc. Japan **14**, 142–148 (1973).
- 2) Hearnberger, A. P., Kilgore, L. T., Rogers, R. W.: J. Agric. Food Chem. **24**, 677–678 (1976).
- 3) Wen, K., Shimamoto, T., Nishihara, T., Kondo, M.: Jpn. J. Toxicol. Environ. Health **31**, 256–259 (1985).
- 4) El-Zemaity M. S.: Bull. Environ. Contam. Toxicol. **40**, 74–79 (1988).
- 5) Hori, Y., Chonan, T., Sato, M., Okada, M.: J. Food Hyg. Soc. Japan **33**, 144–149 (1992).
- 6) Tsumura-Hasegawa, Y., Tonogai, Y., Nakamura, Y., Ito, Y.: *ibid.* **33**, 258–266 (1992).
- 7) Nakamura, Y., Sekiguchi, Y., Hasegawa, S., Tsumura, Y., Tonogai, Y., Ito, Y.: J. Agric. Food Chem. **41**, 1,910–1,915 (1993).
- 8) Nagayama, T.: *ibid.* **44**, 2,388–2,393 (1996).
- 9) Nagayama, T., Kobayashi, M., Shioda, H., Ito, M., Tamura, Y.: J. Food Hyg. Soc. Japan **36**, 383–392 (1995).
- 10) El-Zemaity, M. S.: Bull. Environ. Contam. Toxicol. **40**, 80–85 (1988).
- 11) Yamamoto, I., Fukami, J. ed.: “Pesticide Design—Strategy and Tactics—” p. 1,128 (1979) Soft Science, Inc., Tokyo Japan.