

# Uptake and Translocation of [<sup>14</sup>C-ethyl] Labeled (2-chloroethyl)methylbis(phenylmethoxy)silane [CGA-15281] in the Peach<sup>1</sup>

G. A. Couvillon, S. D. Seeley<sup>2</sup> and S. J. Kays

Dept of Horticulture, University of Georgia, Athens, GA 30602

*Additional key words.* ethylene, *Prunus persica*, growth regulator

**Abstract.** [<sup>14</sup>C-ethyl] labeled (2-chloroethyl)methylbis(phenylmethoxy)silane (CGA-15281) was applied to fruit and leaves of 4-year-old 'Bicentennial' peach trees [*Prunus persica* (L.) Batsch]. Virtually none of the parent material moved into the fruit or was taken up and transported in vegetative tissue. Of the small amount found within the vegetative tissue, there was equal distribution between acropetal and basipetal movement. The compound appears to act through the release of ethylene which penetrates the tissue rather than uptake of the parent molecule and subsequent release.

CGA-15281 has shown exceptional promise as an ethylene-releasing chemical for peach thinning (1–4, 6). This is due in part to the compound's temperature stability and ethylene-release kinetics [apparent energy of activation  $\approx 10$  Kcal mole<sup>-1</sup> (5)]. A recent study of the metabolism of CGA indicated that the degradation of the parent molecule was rapid with the majority of the metabolites dissipating into the atmosphere as hydrocarbon gases (7). Metabolites remaining on or in the fruit were principally benzyl alcohol, benzylglucoside, and benzylmethylglucoside (7).

While the effects of CGA on the abscission of peach fruits and the metabolism of the parent molecule are well-documented, the rate and extent of uptake and translocation have not been studied. We report herein the uptake and translocation of [<sup>14</sup>C-ethyl] labeled CGA in young peach fruits and in vegetative shoots.

## Materials and Methods

Four-year-old 'Bicentennial' peach trees growing in the University of Georgia experimental orchard near Athens were used in this study. Sixty uniform shoots, 48 to 64 cm long located on the periphery of 3 trees (20 shoots per tree) were used. On May 24, 1979, all fruit except the most apical on 30 shoots were removed and all fruit were removed from the remaining 30 shoots. The developing fruit's seed length varied between 15 and 18 mm on this date.

**Translocation of CGA in peach fruits.** Fruit on 30 shoots were treated with technical CGA which was formulated with Tenneco 0-500-100, and Toximul R and S adjuvants as in the formulated product and diluted to a 600  $\mu$ l/l aqueous solution. Sufficient [<sup>14</sup>C-ethyl] labeled CGA (specific activity, 15 mCi/mM) was used to give about 665,000 cpm per application. Treatment consisted of applying 50  $\mu$ l of the formulated [<sup>14</sup>C]-CGA solution to a marked 9-mm circular area on 1 cheek of each fruit. Treated

fruit were sampled at 0, 4, 8, 48, and 96 hr following application. At each sampling time, 6 treated fruit were removed from 6 shoots and cut in half along the suture dividing the fruit into 2 equal halves. A No. 6 cork-borer was used to remove a 9-mm core from directly above the treated area on the treated half of the fruit. The core was removed beginning from the cut surface of the fruit directly above the treated area. This ensured that no labeled [<sup>14</sup>C]-CGA would be transferred to the unlabeled areas of the sample by the cork-borer. The sample core was cut into 1-mm sections using a clean single edge razor blade. The cork-borer and razor blades were washed in hexane between uses. Each 1-mm sample section was placed in separate scintillation vials, capped, and cooled over ice. The seed and endocarp portions of the sample were not sectioned and were placed in separate vials as were the mesocarp samples. The cooled samples were brought to the laboratory and stored at  $-20^{\circ}\text{C}$  ( $\pm 1^{\circ}$ ) until the remaining parent material was measured.

The samples were transferred to 125-ml Erlenmeyer flasks which contained a 7.5  $\times$  1-cm test tube. The vials in which the samples had been held were each rinsed with 30  $\mu$ l hexane, which was transferred to the Erlenmeyer flasks containing the sample. Care was taken to ensure that neither the sample nor the hexane wash entered the test tube that was placed within the Erlenmeyer flasks. The hexane was dried over a stream of nitrogen and the Erlenmeyer flasks were sealed with serum stoppers. Since the chloroethyl position is the most labile on the parent molecule, hydrolysis of the chloroethyl position of the remaining CGA and subsequent release of ethylene was used as a measure of the parent material at each position on the fruit and stem. Ethylene was released from the translocated parent material by injecting 6 ml of 1N NaOH through the serum caps. The released ethylene was trapped by 2 ml of mercuric acetate which had been injected into the 7.5  $\times$  1-cm test tubes. The ethylene was trapped overnight, then the mercuric acetate was transferred into scintillation vials. Twenty ml of dioxane scintillation fluid (100 gms naphthalene plus 5 gms PPO) was added to each vial and the samples counted.

**Translocation of CGA in vegetative tissue.** The upper leaf surface of the midleaf on 30 defruited shoots were treated with formulated [<sup>14</sup>C]-CGA solution (previously described) by spreading 50  $\mu$ l of solution over the leaf surface. This gave initial counts at application of about 665,000 cpm. Care was taken to prevent the label material from dripping off the leaf surface. Treated shoot samples were removed from trees at 0, 4, 8, 48, or 96 hr

<sup>1</sup>Received for publication Sept. 21, 1981. A contribution of the Georgia Agricultural Experiment Stations, College Station, Athens. This research was supported in part by State and Hatch funds allocated to the Georgia Agricultural Experiment Stations and by grant funds provided by Woolfolk Chemical Co., Fort Valley, Georgia.

The cost of publishing this paper was defrayed in part by the payment of page charges. Under postal regulations, this paper therefore must be hereby marked *advertisement* solely to indicate this fact.

<sup>2</sup>Present address: Department of Plant Sciences, Utah State University, Logan, UT 84322.

following treatment. Six shoots were removed at each sampling time and all leaves except the treated leaf were stripped from the shoot. The treated leaf was removed from the shoot and placed in a cooled scintillation vial. The shoot was then divided into acropetal and basipetal sections by cutting at the junction of the leaf petiole. Each shoot section was cut into 1-mm sections beginning from the petiole junction. The stem sections were placed in cooled scintillation vials and held over ice. The razor blades used to section the shoot were washed in hexane between each cut. The stem sections and leaves were stored in the scintillation vials at  $-20^{\circ}\text{C}$  ( $\pm 1^{\circ}$ ) until the  $[^{14}\text{C}]$ -ethylene was released. Ethylene release, trapping, and counting were as previously described.

To test the rate of breakdown of the parent molecule, 25  $\mu\text{l}$  of 600 ppm formulated CGA labeled in the chloroethyl position was applied after the fruit surface was partially wetted with 0.1% X-77 spreader, giving total cpm at 0 time of 462,000. Freshly harvested fruits (5 replications) were weighed and placed in sealed 980-ml glass jars containing a vial of 15 ml of 10% KOH with a filter paper wick and a vial of 7.5 ml of 0.25 M mercuric perchlorate. Samples were collected over an 11-hr period at  $21^{\circ}\text{C}$ . At the end of the collection period, 1-ml gas samples were removed from each container and tested for untrapped ethylene. Ethylene samples were separated on a  $1.5\text{ m} \times 3.2\text{-mm}$  column of activated alumina (70/100 mesh) and measured with a Beckman 72-5 gas chromatograph equipped with a flame ionization detector. The concentration of untrapped ethylene was found to be negligible and as a consequence, was measured for only the first 4 days.

Labeled ethylene was measured by placing 2 ml of the 0.25 mercuric perchlorate trapping solution in the bottom of a 125-ml flask containing a test tube with 3 ml of 0.1 M mercuric acetate. Ethylene was released by injecting 2 ml of 10 M LiCl into the base of the flask containing the mercuric perchlorate

and trapping overnight. The mercuric acetate was then transferred to a scintillation vial and counted with a dioxane "cocktail" (100 g naphthalene + 5 g PPO/liter dioxane).

## Results

The percentage of initial radioactivity of peach fruits decreased 9% by 4 hr following treatment and was only 57% of the total after 8 hr (data not shown). The loss of radioactivity remained stable for 48 hr following treatment but by the 96-hr sampling time there was only 41% of the initial radioactivity remaining. A loss of the label CGA continued at a rapid rate following the 4th day. About 75% of the radioactivity was lost within the first 10 days following application after which there was a decrease in the rate of label loss (Fig. 1). Twenty-one days following application, only about 13% of the total label applied remained and about 9% remained at harvest which occurred 35 days following application (Fig. 1).

There was very little, if any,  $[^{14}\text{C}]$ -CGA penetration into the peach fruit. The 0- and 96-hr sampling time showed similar penetration characteristics (Fig. 2). Over 99% of the radioactivity on or within the fruit was located in the first mm of mesocarp and epidermal tissue. There was essentially no radioactivity in mesocarp samples removed from the other depths. The rapid loss of radioactivity during the first 8 days following treatment probably represents degradation of the  $[^{14}\text{C}]$ -CGA located on the surface of the fruit.

Very little CGA was translocated from the leaf into the shoots (Fig. 3). Less than 0.7% of the total label applied was found in shoots that were sampled 4 hr following treatment. The greatest amount of label was found in the 3- to 8-mm sections from the acropetal portions of the shoots. At 96-hr post-treatment, the acropetal 8- to 10-mm sections contained the greatest amount of label although the cpm were about 60% less than that found in the 3- to 8-mm acropetal sections 4 hr following treatment.

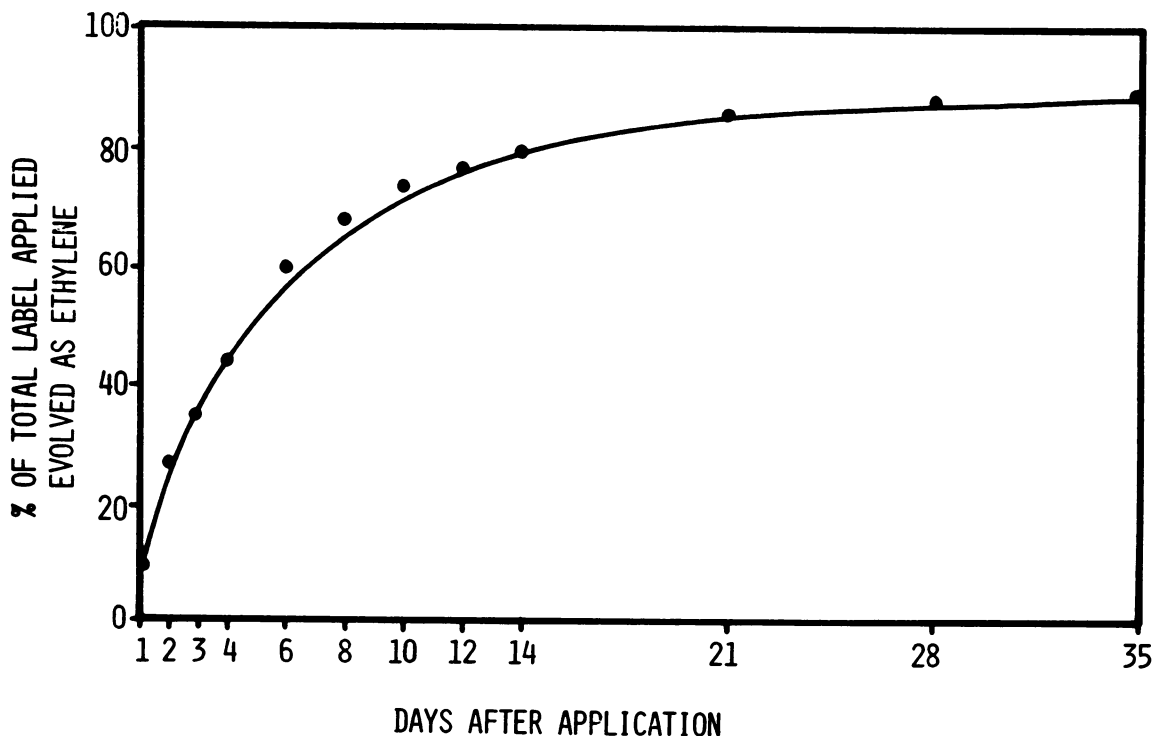


Fig. 1. Loss of radioactivity during 35 posttreatment days from peach fruit treated with  $[^{14}\text{C}$ -ethyl] labeled (2-chloroethyl)methylbis(phenyl-methoxy)silane.

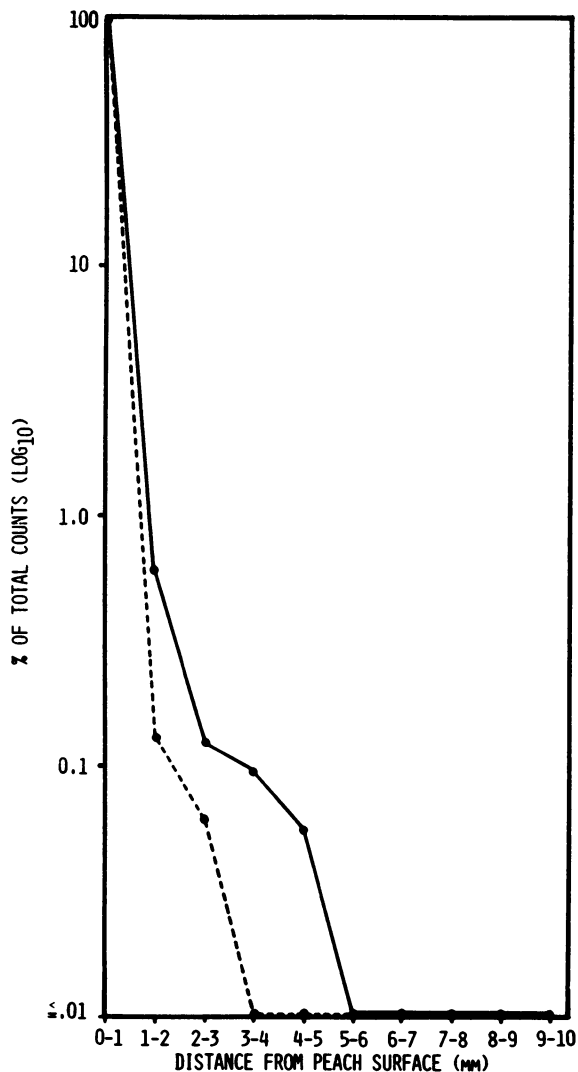


Fig. 2. Penetration of [<sup>14</sup>C-ethyl] labeled (2-chloroethyl)methylbis(phenyl methoxy)silane, in peach fruits immediately after application(---)and at 96 hr(—)following application.

Basipetal shoot samples showed a similar translocation pattern (Fig. 3). The degree of translocation of CGA into peach shoots is similar to that found in peach fruits. In both cases the amount was extremely small.

#### Discussion

We have shown that the majority of the parent CGA molecule (i.e. 99%) does not penetrate beyond the outer 1 mm of the fruit tissue. It is anticipated that much of the parent molecule which is included in the outer 1-mm section is in fact on the outer surface of the section. In addition, little CGA is taken up by the foliar tissue and translocation within the plant. Of the small fraction (<.7%) that is translocated, a discernable preference between acropetal and basipetal movement was not detected. This lack of uptake and transport of the parent molecule reflects the extremely low solubility (i.e. <10 ppm) of the molecule in water.

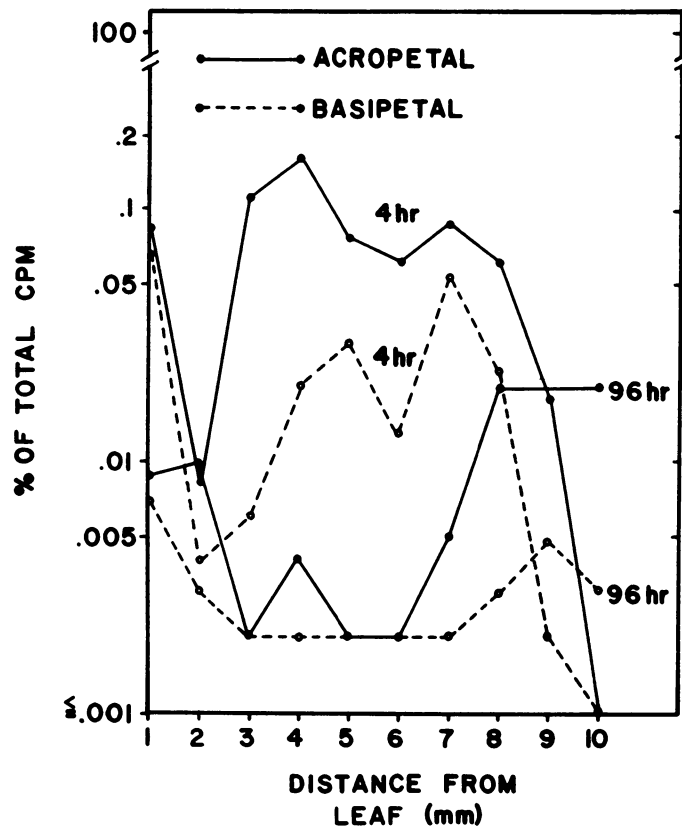


Fig. 3. Acropetal and basipetal movement of [<sup>14</sup>C-ethyl] labeled (2-chloroethyl)methylbis(phenylmethoxy)silane in vegetative tissue of the peach.

From the results of this study, it would appear that the growth regulator CGA acts through the release and subsequent penetration of ethylene into the plant tissue rather than through the uptake of the parent molecule and release of ethylene within the fruit tissue. If this is the case, then atmospheric conditions resulting in turbulence could alter the effectiveness of the growth regulator in inducing fruit abscission.

#### Literature Cited

1. Byers, R. E. 1976. Peach fruit thinning with CGA-15281. HortScience 11:324. (Abstr.)
2. Byers, R. E. 1978. Chemical thinning of peach fruits with CGA-15281 and CGA-17856. J. Amer. Soc. Hort. Sci. 103:232-236.
3. Daniell, J. W. 1978. Fruit thinning in peaches with CGA-15281. HortScience 13:345. (Abstr.)
4. Gambrell, Jr., C. E. and G. E. Stembridge. 1978. Thinning peaches with CGA-15281. HortScience 13:261. (Abstr.)
5. Olein, W. C. 1980. Ethephon-induced gummosis in sour cherry (*Prunus cerasus*): Effect of gum on xylem function and influence of temperature. PhD Thesis, Michigan State University, East Lansing.
6. Porpiglia, P. J. and J. A. Barden. 1980. Peach leaf abscission following CGA-15281 and CGA-17856 applications as affected by temperature. J. Amer. Soc. Hort. Sci. 105:227-229.
7. Seeley, S. D., G. A. Couvillon, and S. J. Kays. 1981. Metabolism of an ethylene releasing growth regulator (CGA-15281) in young peach fruit. J. Amer. Soc. Hort. Sci. 107:682-687.