

FINAL REPORT

Project Title: Maintenance of Guthion Registrations on Pome Fruits: Fruit Residue Reduction through Spray Timing Optimization

PI: Allan S. Felsot

Organization: Food & Environmental Quality Lab, Department of Entomology, WSU, Tri-Cities

Co-PI(s): Vince Hebert, FEQL, Department of Entomology, WSU-TC
Doug Walsh, FEQL, Department of Entomology, WSU-Prosser

Cooperators: Keith Klingele, Klingele Orchards, Prosser, WA
Keith Oliver, Olsen Brothers Ranches,, Prosser, WA

Objectives:

1. Measure decline of Guthion (azinphos-methyl) apple residues in an orchard when applied one, two, or three times during the growing season.
2. Measure Guthion apple residues in composited and single-serving fruit samples at harvest following one, two, or three applications.
3. Measure Sevin (carbaryl) residues in composited and single-serving fruit samples at harvest.
4. Develop an empirical phenological (degree-day) based fruit pesticide residue decline model that predicts the best application timing window to give the lowest possible residue concentration.
5. Validate the fruit pesticide residue decline model by measuring Guthion residues in fruit harvested from commercial orchards receiving variable number of applications. Sevin residues would also be measured.
6. Determine distribution of residues in pulp and on peel.
7. Analytical Methods/GLPs/Quality Assurance development.

This project was initially funded for two years outside of the normal funding cycle in conjunction with a proposal submitted to the Washington State Commission for Pesticide Registration (WSCPR). In December 2000, a formal proposal was submitted to the WTFRC for consideration of continued funding, and a second proposal was also submitted to the WSCPR. Our goal has been to delineate how application practices affect the residues of insecticides on harvested apples. Furthermore, we sought to discover how application practices affect the magnitude of residues in individual apples, which are known as “single serving” samples. Ultimately, we wanted to build a kinetic model to predict residues at harvest given any application date throughout the growing season.

Because we were not on the normal funding cycle, we did not start the project until June 2000; we continued the project during the 2001 season. The project is somewhat unique in that the magnitude of residue studies (corresponding to objectives 1, 2, and 3) are being conducted under Good Laboratory Practice Standards, thereby requiring outside auditing of all of the procedures and data. We will be finishing all analyses of Guthion and Sevin residues on collected apples prior to June 2002. Work is ongoing for all listed objectives.

Significant Findings:

- Residues on apples collected within 24 hours of spraying did not significantly differ among spray treatments, which included one, two, or three Guthion cover sprays. Thus, for all practical purposes initial residues are the same when one-month intervals occur between sprays.
- Estimated half-lives (time to 50% loss of any given amount of residue) ranged from 17.8 to 19.7 days, but were not significantly related to number of sprays.

- Expression of residue data on a surface area basis (rather than a proportional weight basis, such as ppm or $\mu\text{g/g}$) only affected the relative magnitude of the residue after the first spray when the surface area:weight ratio ranged to 1.6.
- Average residues at harvest on composited apples and single serving apples were not significantly different. Furthermore, residues on single serving apples were at most three-fold above the average residues. However, only 7-8 apples out of 27 analyzed for each spray treatment were above the composite average.
- All residues, including those on the single serving apples were significantly below the Guthion tolerance of 1.5 ppm.

Methods:

During crop year 2000, a protocol for the field work was written for the conduct of the study consistent with GLPs. One, two or three applications of Guthion 50W (1 lb AI/acre) were made to a block of Rome apple trees on the Klingele Orchard (near Prosser). Sevin was applied by the grower. The experimental design was a random block with three replications consisting of 15 trees in each replicate plot. Apples from each of three randomly selected trees within a replicate plot were sampled frequently after each Guthion application. The apples from individual trees were bulked but were analyzed separately from apples collected on any other tree. The intention of this sampling technique was to determine the variability in residues between trees as well as to reduce the variability in residues between replicate units.

To simulate harvest, five apples were collected from each of nine trees in a replicate plot. Nine apples from the harvest day collections were composited in a food processor (called composite samples); magnitude of residue studies usually rely on composite samples. An additional 27 individual apples from each spray treatment (i.e., nine from each of the three replicates associated with a spray treatment) were processed as single servings. These apples represent exposures estimated for acute dietary risk assessments.

During crop year 2001, a GLP-compliant protocol was developed and approved for studying the effect of application number on Sevin residues. The study site was a block of Gala apples at the Olsen Brothers Ranches (near Prosser). Treatments were one spray during bloom, one spray after petal fall, and one spray three days prior to harvest. The treatments were arranged in a random block design with three blocks, and number of spray treatments were assigned randomly to each block. Apples were collected at various time intervals after application, and then they were collected at the manager's recommended harvest time.

In late September 2001, apples were collected along a transect of 50 trees in 11 commercial orchards. Apples were separated into 5 bags of 10 apples each, which were stored at -20°C . Apples from four of the bags will be processed as composites, and apples from the remaining bag will be analyzed as single servings.

Analytical methods were developed and modified for Guthion residues in single serving samples and composites. Furthermore, analytical methods were developed and validated for distinguishing Guthion residues on peels and in pulp. Methods for Sevin analysis are currently under development. All field and analytical procedures were conducted under a GLP protocol, and field phases of the Guthion and Sevin studies were audited by an independent auditor.

Results and Discussion:

Residue Decline of Guthion After One, Two, or Three Applications. Residues recovered within 24 hours after Guthion application ranged from 1.36-1.47 parts per million (ppm or microgram per gram, $\mu\text{g/g}$) but did not differ significantly among treatments (Table 1). On a surface area basis, residues varied from $0.85 \mu\text{g/cm}^2$ (one spray) to $1.31 \mu\text{g/cm}^2$ (three sprays). This comparatively larger difference between initial residues on a surface area basis is due to the surface area to volume ratio differences as the fruit matures (Figure 1). The perspective of surface area coverage, as opposed to a

weight based residue concentration, has implications for insect control because the codling moths and newly hatched larvae would be absorbing surface deposits. The lethal residue to 95% (LR95) of codling moth adults ranges from 0.31 to 0.72 $\mu\text{g}/\text{cm}^2$ (Howell and Maitlen 1987) for 96 hour and 48 hour exposures, respectively. Thus, average surface area deposits in our study were close to the baseline for efficient control. By 28 days after the first spray, residues on a surface area basis had declined to 0.19 $\mu\text{g}/\text{cm}^2$, far below the level needed for adequate control. The residues recovered would not have been sufficient to control 95% of the codling moth larvae (LR95 = 1.17 $\mu\text{g}/\text{cm}^2$, Maitlen et al. 1985).

Table 1. Effect of number of sprays and pre-harvest interval on initial residues of Guthion, residues at harvest, and dissipation half-life.

Treatment	24-h Post Spray Residue (ppm)	Pre-Harvest Interval (Days)	Residue at Harvest (ppm)	Residue Dissipation Half-Life (days)
One Spray	1.362	99	0.033	17.8
Two Spray	1.573	71	0.110	18.5
Three Spray	1.469	41	0.360	19.7

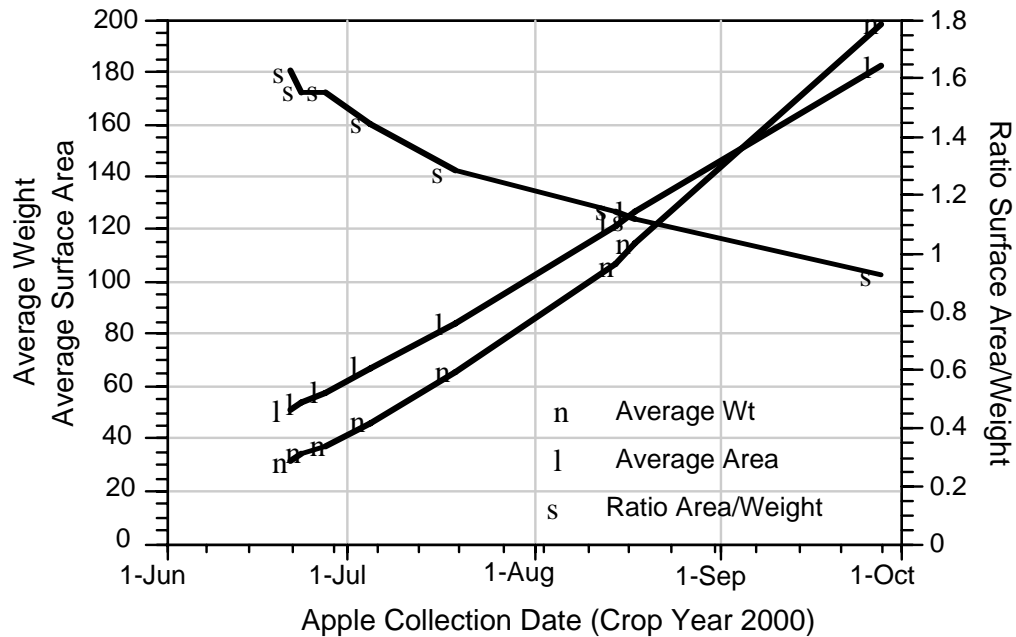


Figure 1. Relationship between average apple weight (grams) and surface area (cm^2) for apples collected on selected dates.

Half-life of Guthion was not affected by the number of sprays (Table 1), but was prolonged relative to the average 10 days reported for foliage (Maitlen et al. 1985).

Measure Guthion apple residues in composited and single-serving fruit samples at harvest following one, two, or three applications. Residues on apples at harvest following the three-spray treatment were at least five fold below the tolerance of 1.5 ppm. A reduction of the pre-harvest interval by about 28 days was associated with approximately a three-fold increase in residues.

Average residues from single serving samples (n=27) were not significantly different than average residues from composited apples (n=9) (Table 2), but variability was greater as evidenced from a comparison of standard deviations. However, the residue concentrations ranged at most three-fold above the average residues, which is on the low side of the range of 2-13 reported for several other single serving studies (Hamey and Harris 1999) (Figures 2-4).

Determine distribution of residues in pulp and on peel. Analysis of samples collected during September 2000 showed that nearly all of the residues reside on the peel. Diffusion of residues into the pulp seemed insignificant by comparison.

Table 2. Comparison of average residues for individually analyzed apples (“single serving apples”) and composited apples collected at harvest.

Treatment	Single Serving Apples	Composite Apples
One Spray	0.03 ± 0.03	0.03 ± 0.01
Two Sprays	0.08 ± 0.07	0.11 ± 0.06
Three Sprays	0.25 ± 0.19	0.36 ± 0.10

Develop an empirical phenological (degree-day) based fruit pesticide residue decline model. The first steps toward model development have been completed with first-order kinetic analysis of the residue data collected thus far. The next step is to further refine the kinetic analysis with all of the residue data and then to tie the model to accumulation of degree days. Apples were collected from 11 grower orchards that represent variable management practices. These are in the process of analysis to determine if the kinetic model is predictive of the residue at harvest.

Measure Sevin residues in composited and single-serving fruit samples at harvest. Apples collected during crop year 2001 at the Olsen Brothers Ranch are being handled consistent with a GLP protocol. After method validation, both composites and single serving apples will be analyzed, similarly to the analysis conducted for Guthion. Also, apples that were collected several times throughout the growing season will be analyzed to determine half-life of Sevin residues.

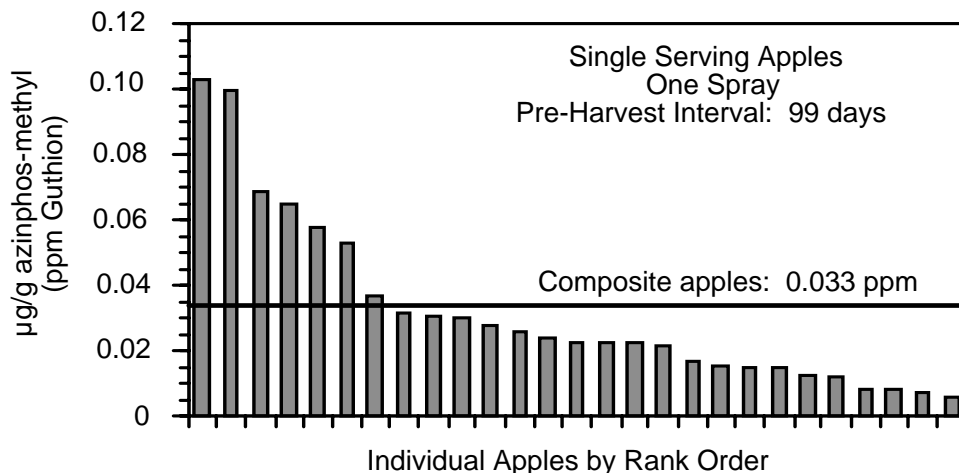


Figure 2. Rank order distribution of Guthion residues on individual apples collected 99 days after a single spray. The solid horizontal line represents the residue recovered from a composite of nine apples.

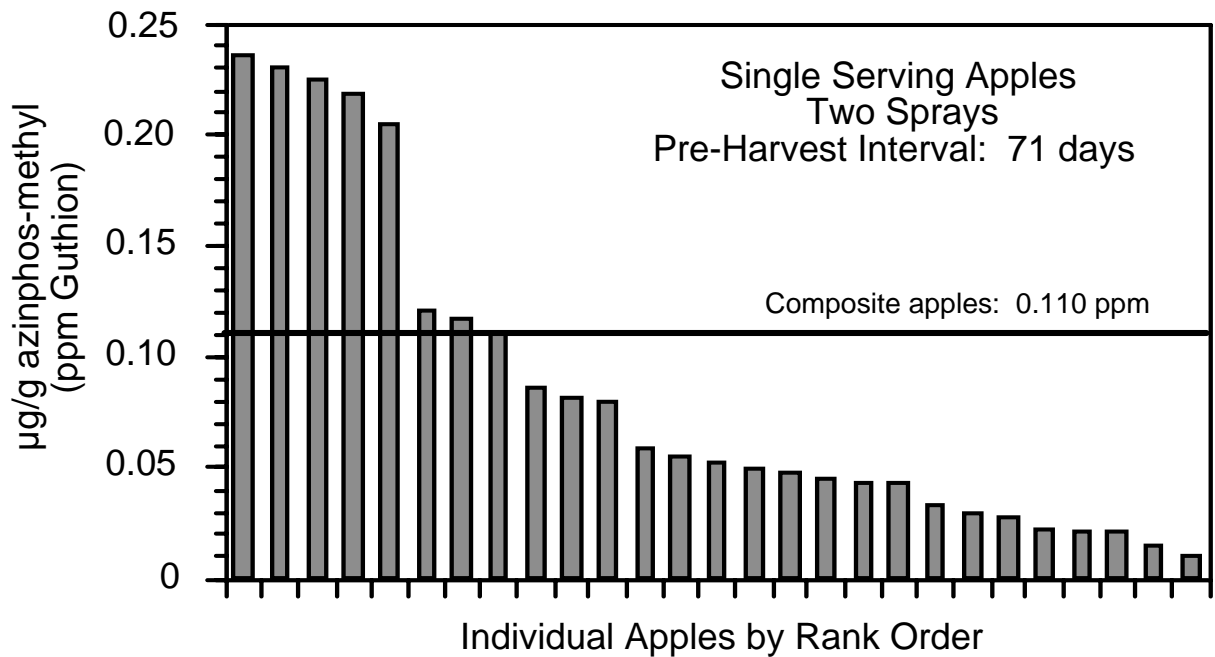


Figure 3. Rank order distribution of Guthion residues on individual apples 71 days after a second spray. The solid horizontal line represents the residue recovered from a composite of nine apples.

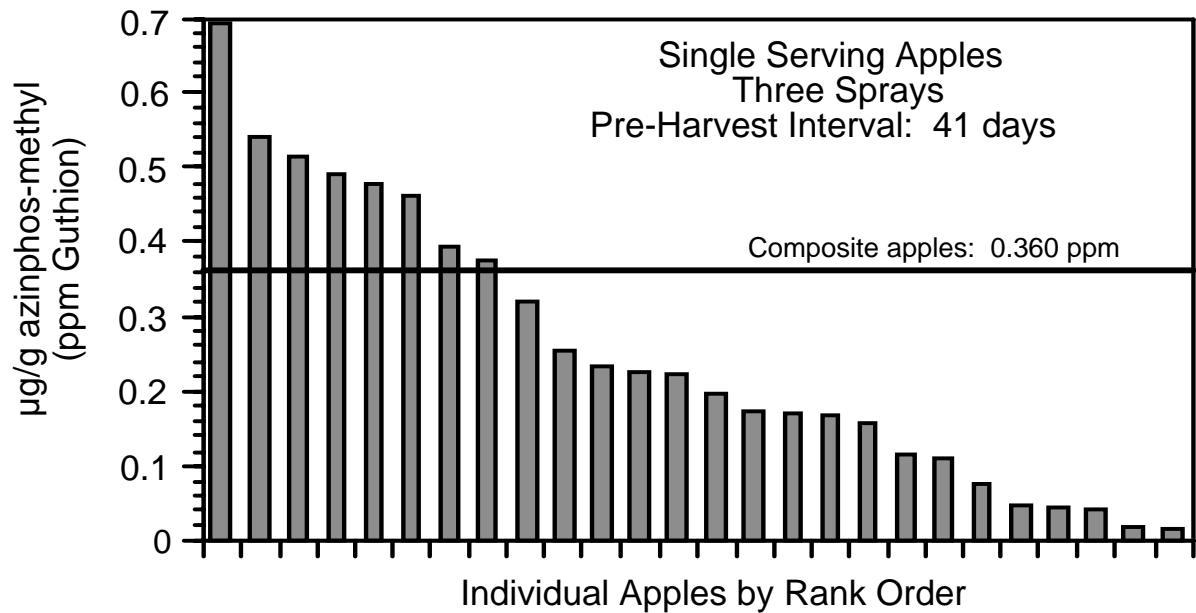


Figure 4. Rank order distribution of Guthion residues on individual apples 41 days after a third spray. The solid horizontal line represents the residue recovered from a composite of nine apples.

References Cited:

- Hamey, P. Y. and C. A. Harris. 1999. The variation of pesticide residues in fruits and vegetables and the associated assessment of risk. *Regulatory Toxicology and Pharmacology* 30:S34-S41.
- Howell, J. F. and Maitlen. 1987. Accelerated decay of residual azinphosmethyl and phosmet by sprinkler irrigation above tree and its effect on control of codling moth based on laboratory bioassays as estimated by laboratory simulation of insecticide deposits. *J. Agric. Entomol.* 4(4):281-288.
- Maitlen, J. C., A. Cantu, and A. Palmer. 1985. Efficiency of pesticide application: azinphosmethyl on apples. Report to Cooperators 1985: USDA-ARS, Yakima Agricultural Research Laboratory 14 pp.

Budget:**Maintenance of Guthion Registrations on Pome Fruits: Fruit Residue Reduction through Spray Timing Optimization**

Allan S. Felsot

Project Duration: 2 years (2000-2002)

Year	Year 1 (2000-2001)	Year 2 (2001-2002)
Total	\$39,393	\$32,572

Budget Breakdown

Expenditure	FY 2001		FY 2002	
	WTFRC	WSCP	WTFRC	WSCP
Salaries (Research Aide II, 0.75 FTE)	16,440	8,220	17,098	8,549
Employee Benefits for Aide	4,603	2,302	4,787	2,394
Technical Assistant I (Non-classified)	5,850	2,925	0	0
Travel (to Orchards)	375	188	500	0
Equipment (GC supplies)	2,750		1,000	5,000
Other (Sample Analysis Cost)	8,375	4,188	9,188	3,063
Other (Study Audit)	1,000	1,875	0	2,000
Total	\$39,393	\$19,697	\$32,572	\$21,005

Project total: \$71,965