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Assessment of Safe Harvesting After Methyl Parathion Application in Peaches

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Methyl parathion is a widely used potent anticholinergic compound found as active ingredient in several plant protection products. Commercially available formulations of methyl parathion are usually in the form of dust, emulsifiable concentrates (EC) and microcapsules. Up to now there are 25 different formulations registered in Greece (Gianopolitis 2000). Most of them (22) are emulsifiable concentrates (EC), classified as poisonous to humans and toxic to the aquatic organisms. Two of them are microencapsulated formulations and classified as highly toxic to aquatic organisms but are not classified for humans. Only one is granular (GR) and is classified as toxic to humans and toxic to aquatic organisms. Methyl parathion is rapidly absorbed into the bloodstream through dermal, oral and inhalation exposure. Methyl parathion does not accumulate in the body, and is almost completely excreted via kidneys in the urine within 24 hours, as phenolic metabolites (Department of Transportation 1984; Wayland 1991). In man, the major metabolites are 4-nitrophenol and dimethyl phosphate. In case of poisoning the recovery of patients was not only associated with blood concentration of poison on admission, but also with a variety of factors dealing with patient 's prior status and emergency treatment employed, including cathertic measures and the administration of antidotes (Tsatsakis et al. 1995). The major metabolites formed in plants are p-nitrophenol, p-nitrophenyl glucopyranoside and P-S demethyl parathion methyl. Very important is the fact that this substance used close to the harvest time. The objective of this study was to investigate the dissipation of methyl parathion on peaches under local climatic conditions.

MATERIALS AND METHODS

The field study was done according a defined protocol. All the procedures were done according to the Good Laboratory Practice (GPL) applied to field studies. A motorized back pack sprayer was used for the application after a proper calibration. The test field was divided in three plots, T1, T2 and C. T1 was the plot of single application, T2 was the plot of double application and C was the control plot. The calculation of spraying solution are shown in table 1. During application the temperature was 21°C, the relative hydration 75%, soil conditions were fine and foliage was dry. The calculation of spraying time was

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done according to the results of calibration. Both treated and non-treated plots were labeled on trees and in front of each plot. The operator did both the application and the mixing loading. After the application and when the fruits were dry we did the first sampling. Temperature (min max), number of fruits in each sample and diameter of the selected fruits were registered each sampling day. Samples were taken every second day for 16 days. Each sample was packed separately and labeled. The total weight of each sample was 1 Kg. The samples were chopped and blended before deep freezing without any prior processing. A specific protocol based on the directions of the FDA was used for the sampling procedure (Clifton 1996). An imaginary grid divided into approximately 100 areas was superimposed on the plot. Furthermore we extended this grid at height until the higher part of the trees. The sampling was done by chance at any level. The sample places of each sampling were marked in proper sheets. Then the fruits were packed intact and each sample was numbered. A different pair of gloves was used for each plot only once.

For the determination of methyl parathion in the fruit samples, a multi-residue method was applied (Krijgsman et al. 1976). The analytical determination of methyl parathion was performed by gas chromatography in a modified Varian 3700 instrument with an NPD detector at 270 °C, injector at 250°C and oven temperature at 100-260 °C with a rate of 5 °C /min. The column used was RTX – 1701(crossbond 14% cyanopropylphenyl-86% dimethyl polysiloxane) 30meter, 0.53mm ID. The solvent used for the extraction of methyl parathion from the samples was ethyl acetate (pestanal) and sodium sulfate was added in the homogenates in order to remove moisture. The samples were chopped and blended before deep freezing. The determination of methyl parathion concentration was done in 50 gr of the blended pulp in triplicate. The 50 gr sub-sample was further homogenized in 100 ml of ethyl acetate for 1 min, sodium sulphate was added and further homogenization followed. The homogenate was left for 30 min and then filtered through filter paper. The solvent extract was evaporated under nitrogen to 5 ml, which was used for GC.

The calibration plot was established at 5 concentration levels i.e. 1, 0.5, 0.1, 0.05, 0.02 ppm with five replicates per level. The respective equation is $y = (56071.25 \pm 1731.87)x - (150.119 \pm 870)$, $R=0.996836$ and CL = 95%. The respective values, for the limit of detection (LOD) and the limit of quantification (LOQ) are 0.0013 ppm and 0.0036 ppm. The recovery of methyl parathion from the fruits was studied at three fortification levels adding 12, 40, and 200 mg of methyl parathion at 200 gr peach tissue with 3 replicates per level. The whole procedure was done using peach tissue extract (with ethyl acetate). From the stock solution of 10, 100, and 1000 ppm we used 0.2, 0.4 and 1.2 ml respectively, in order to infect the peach tissue. Then we implemented the method. The results are shown on Table 2.

RESULTS AND DISCUSSION

Our results provided us detailed information about the relation of residues of

Table 1. Calculation for the spray solution

Name		Unit	Formula	Digits	T T1, T2
Plot size	PS	m^2	-	1	125.0
Number of replicates	Rep	m^2	-	-	1
Total plot size to be treated	TPS	m^2	PS*Rep	1	125.0
Plant / plot	PP	./plot	-	-	5
Plants / hectare	PH	...ha	-	-	400
Total plants to be treated	TPP	m^2	PP*Rep		5
Factor	F	-	TPP/PH	7	0.0125000
Compound / hectare in gr or ml /ha	Cha	.../ha	-	-	2000
Spray volume / ha	Wha	l	-	-	2000
Compound/ plot or treatment in ml (if liquid)	Cp	ml	Cha*F	4	25.0000
Spray volume / plot or treatment	Wp	ml	Wha*F*1000	-	25000
Safety factor absolute	Sfabs	ml	-	-	6250
Safety factor relative	SF%		Sfabs/Wp+1	4	1.2500
Amound / volume of compound to be prepared in gr or ml	Cp-sf	.....	Cp*SF	2	31.25
Volume of water to be prepared	Wp-sf	ml	Wp*SF	-	31250
Tolerance (Min Max)	T	....%	Wp*(1± T/100)	-	1250

Table 2. Fortification levels and recovery rates of methyl parathion from peaches

Replicate	Injection volume µL	Level of fortification ppm	Recovery rates %	SD	Average of recoveries
A	4		72.86		
B	4	1	69.27	3.25	69.31
C	2		65.81		
A	4		80.70		
B	4	0.2	75.11	15.73	68.96
C	4		51.08		
A	4		73.60		
B	4	0.06	93.03	10.53	80.95
C	4		76.14		

active ingredient versus time and applied doses. Our research was a part of study dealing with the system of integrated management in agricultural production as a part of the management of rural environment. Figure 1 shows the results of concentration of methyl parathion in peaches versus time. The degradation curves of methyl parathion show that the levels of the active ingredient, which remain initially on the fruits, are different in the two application scenaria. This difference, which is 0.083 ppm, was due to the double quantity of spraying solution sprayed on the foliage and double spraying time. Also the hair of the fruit are of some importance in relation with this difference because hair form a mesh which prevents spray droplets from running off, and so allows the active ingredient to be deposited. The main reasons for the degradation are solar radiation and heat. The greater difference of residues between two sequential samples occurred between the 2^{nd} and 4^{th} day. This was due to the increase of temperature from 29 to 33.5 C^{o} between these days. During the trial period there was no rain so there was no run off of active ingredient from the fruit surface. Maturation also affects the degradation progress. As we know, the variation in the size of the fruit as a result of the ripening process affects the quantity of residues. This is due to the increase of weight and the increase of the surface of the fruit. At this stage the effect is small because 70% of the fruits had already reached the final size and the only alteration was the change of the color and of the percentage of brix. Table 3 shows the number of fruits, the range of diameter and the mean of diameter of peaches collected for 16 consequently days. The degradation rate of methyl parathion between the day of application and the 6^{th} day at the second scenario is faster than the first. The respective values are 0.041 mg/Kg/day and 0.054 mg/kg /day.

As far as we could see the double spraying procedure did not contribute to the plant protection significantly. Furthermore it increased the production cost and the pollution because most of the spraying solution ended up in the soil and a small fraction moved through the air (drift). On the 4^{th} day the residue levels on fruits were lower than the MRLs (maximum residue limits) in both application scenarios. This fact is very important because we could have a safe harvest after this day. After 8^{th} day there were no detectable residues with the method that we used. Also very important is the fact that temperatures are normal for the season of trial. Figure 2 shows the higher day and night temperatures at sampling days.

In a 4-week study, for chronic toxicity with volunteers given 22, 24, 26, 28 or 30 mg / person / day, mild cholinesterase inhibition appeared in some individuals in the 24, 26 and 28 mg dosage groups (Wayland 1991). In the 30 mg / person / day (about 0.43 mg / Kg / day) group, red blood cholinesterase activity was depressed by 37%. Also other studies with human volunteers have found that doses of 1 to 22 mg / person /day have no effect on cholinesterase activity. The 24 mg / person / day correspond to 0.342 mg / kg /day, for a person weighing 70 Kg. Samples after the application give us for single and double application scenarios 0.266 mg/Kg and 0.35 mg/Kg respectively. That means for the dose of 0.342 mg / Kg / day correspond for the single application to 0.342/0.266 = 1.29 kg peach tissue. According to these results a human of 70 Kg should consume about 1.29 * 70 = 90.3 Kg of peach tissue per day. With the same calculations for the double

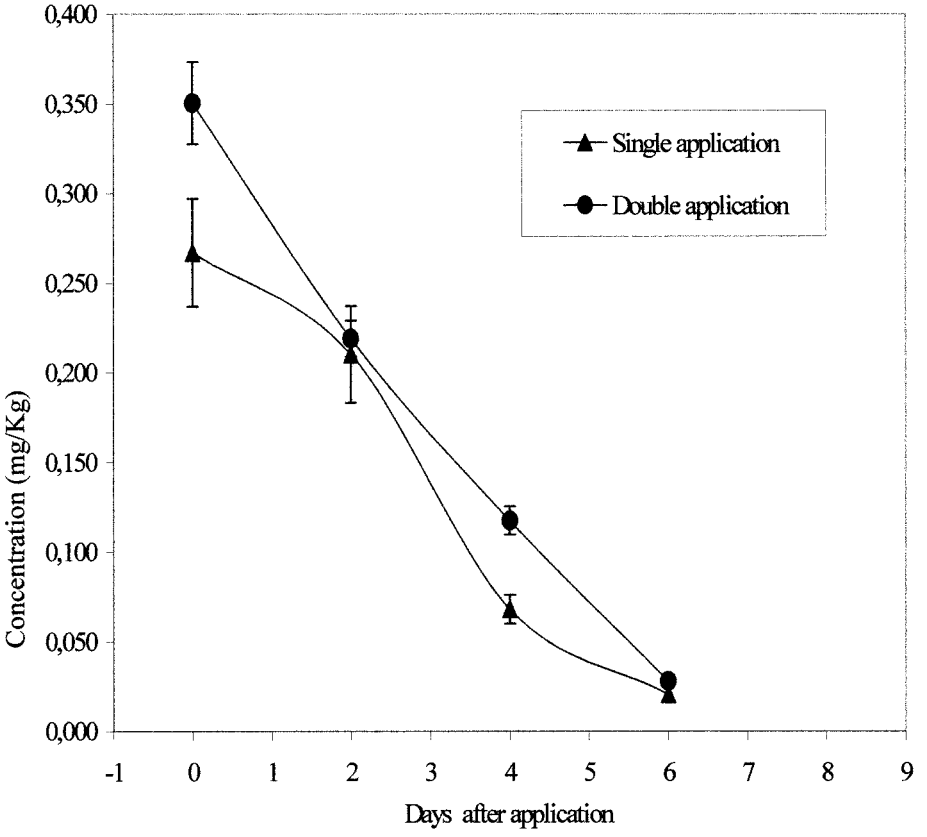

Figure 1. Methyl parathion concentration (mg /Kg) in peaches versus time

application scenario the 0.342 mg / Kg / day correspond to 0.342 / 0.35 = 0.97 Kg and a human of 70 Kg should consume about 0.97 * 70 = 67.9 Kg of peach tissue per day.

The results of this trial should take in account during the planing of plant protection at the Integrated Crop Management Systems both in normal and crisis circumstances. At the normal plan the scientist should consider: the residues levels of the active ingredient which remain on the fruits after the application, the cost of the plan and also the potential pollution of the environment in order to achieve a satisfactory fight of the insects. Furthermore the decisions, for example, which substance should be used, in which concentration, with which application method, at which growth stage, are easy to be taken. But dealing with a crisis as, for example, an insect infestation very close to harvesting is very complicate, because if the application performed before harvesting, there is a possibility the the residue levels on the fruits to be found above the MRLs. Also of great importance for the effectual resultant of the trial are the Standard Operated Procedures (SOPs). Any staff involved in a field study should be trained in the use

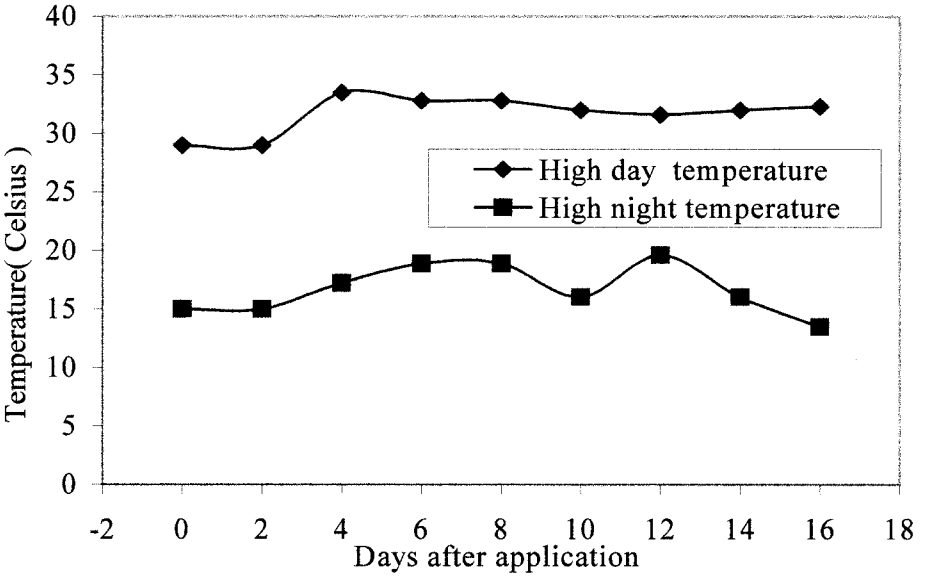

Figure 2. High day and night temperatures at sampling days

Table 3. Number of fruits (N), range of diameter (R) cm, mean of diameter (±SD) for sixteen consequently days of sampling after single and double application of methyl parathion in peaches

	N	R	Mean (±SD)
Single application	108	3	0.51
Double application	108	3	0.57

of Standard Operated Procedures. Conclusively Good Laboratory Practice (GLP) Principles applied to field studies ensure the credibility of the results.

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