

## Reliability of CT Products in Varieties of Perishable Commodities Fumigated with Methyl Bromide

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**Abstract:** To evaluate the reliability of the CT product as an indicator of insecticidal efficacy of methyl bromide (MB), the laboratory scale test was conducted with different varieties of apples ('Alps Otome' and 'Mutsu') and nectarines ('Fantasia' and 'Shuhou') infested with egg and larval stages of the peach fruit moth, *Carposina sasakii*. The result showed that (1) the difference of the LC<sub>50</sub>'s was smaller than that of the LD<sub>50</sub>'s in two varieties of apples and nectarines when compared the concentration-mortality data with the dose-mortality data. The data suggested that if gas concentration (CT product) was the same, the same efficacy would be obtained between varieties, while even if the same dose was applied, there would be a possibility of difference in efficacy between varieties, and that (2) no significant difference was observed on eggs' response between varieties of 'Alps Otome' and 'Mutsu' apples and of 'Fantasia' and 'Shuhou' nectarines in both the LC<sub>50</sub>'s and the LD<sub>50</sub>'s, respectively. However, 95% confidential limits between apple varieties were slightly overlapped with a slight difference each other in the LD<sub>50</sub>'s. The data indicated that there might be significant difference in efficacy between two apple varieties if larger difference was obtained between confidential limits in the LD<sub>50</sub>'s, and that (3) no difference was observed on the mortality of the larval stage inside different sizes of 'Mutsu' (the weight ratio is approx 1:2.3), while the mortality in extremely small size of 'Alps Otome' was always higher than that in extremely large size of 'Mutsu' with the difference of 6.8~8.9% (the weight ratio is approx 1:9.2) and there is a possibility with the difference in efficacy in extremely large size of fruit because of influence of gas penetration.

**Key words:** methyl bromide, fumigation, *Carposina sasakii*, LC value, LD value, varieties, apples, nectarines

### Introduction

MONRO (1969) and FAO (1983) described that insecticidal efficacy of a fumigant such as methyl bromide (MB) against a given species of the pest is determined by the amount of gas in air and its penetrating power into the interstices of commodities. A most important factor affecting the action of MB is the phenomenon known as sorption with physical and chemical process. The sorptive amount of gas varies according to the com-

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modities and the amount can be identified by calculating the CT product which is known as an indicator of insecticidal efficacy in routine fumigation.

YOKOYAMA *et al.* (1987) reported that the LD<sub>50</sub>'s of the codling moth eggs, *Cydia pomonella* on 'Summer Grand' variety of nectarines were significantly more susceptible than the eggs on other varieties 'May Grand', 'Firebrite', 'Red Diamond', 'Spring Red', 'Fantasia', and that many flaws such as very fine skin cracks on 'Summer Grand' may contribute to the difference in the egg mortality by MB/fruit interaction. MAINDONALD *et al.* (1992) found differences in sorption of MB among cherry varieties. KAWAKAMI *et al.* (1998) reported that the maximum variation of the CT product between varieties was 7% in nectarines and 10% in apples, respectively when compared the CT product among 3 varieties of nectarines and 13 varieties of apples fumigated with MB.

These reports clearly suggest that the sorptive amount of MB varies according to varietal characteristics of commodities and reduction of the gas concentration during fumigation causes lower mortality of the pest, and that use of the CT product is also a possible method for the evaluation of the susceptibility of the pest in different varieties of commodities. Furthermore, there is no reports of a possibility of difference in efficacy against larval stages inside fruits because of gas penetration power.

Our objects were to examine the reliability of the CT product between varieties of fresh fruits affecting insecticidal efficacy of MB against egg and larval stages of the pest which exist on/in various varieties and sizes of fruits in the dose-response test with the peach fruit moth, *Carposina sasakii*.

## Materials and Methods

### Test Fruits

Apple varieties of 'Alps Otome' produced in Nagano Prefecture and of 'Mutsu' produced in Aomori Prefecture and nectarine varieties of 'Fantasia' and 'Shuhou' produced in Fukushima Prefecture were obtained from local packing houses, respectively. These fruits were stored at 1°C at the Research Division, Yokohama Plant Protection Station, Ministry of Agriculture, Forestry and Fisheries (MAFF) before testing. These varieties were chosen based on the previous test data shown the maximum variation of the CT product between varieties of apples and nectarines (KAWAKAMI *et al.*, 1998).

### Test Insects

The peach fruit moth were obtained from the Aomori Apple Experiment Station in June 1998 and 1999. They were reared by using immature apples as described by KAWASHIMA (1991) in the rearing room at 25°C, 70% R. H. with a 16L:8D photoperiod at the Research Division, Yokohama Plant Protection Station, MAFF.

Twenty-five to thirty mated females were released and allowed to oviposit on apple and nectarine fruits for 24 hours in the cage. In order to facilitate the counting of the number of eggs on fruit, the stalk cavity and the calyx end of apples and nectarines were filled with melted wax. The egg on a piece of filter paper was also prepared for the test with nectarine varieties because nectarine fruits are decay easily during the test and it is

difficult to evaluate the number of the test insect. These infested fruits and filter papers were placed in the rearing room for one overnight and then stored at 15°C for apples and at 20°C for nectarines before fumigation. The stage thus reared was considered as two-day-old eggs which were the most resistant to MB fumigation of all stages of the peach fruit moth (KAWAKAMI *et al.*, 1994).

Test fruits infested with larval stages were prepared as follows; in the test in 1998, 4th instar larvae reared in immature apples were placed on the stem of 'Mutsu' (15-20 larvae/fruit) and then stored for 48 hours at 22°C until fumigation, while in the test conducted in 1999, a piece of paper with eggs were placed on 'Alps Otome' and 'Mutsu' and then stored for 18 days under rearing conditions until 3rd to 4th instar larvae were obtained.

### Fumigation

Fumigation for the egg stage was conducted in a 29.5l fiber-glass fumigation box (26.0 cm × 28.0 cm × 41.0 cm in size) equipped with a circulation fan, ventilation apparatus, and ports for gas application and sampling, a manometer and temperature probes. Each variety of the fruit infested with the peach fruit moth eggs were weighed by a top-loading digital balance (LIBROR EB-3200D, Shimadzu) with special attention to be adjusted fruit loading of 0.16kg/l for apples and of 0.14kg/l for nectarines and then placed in the fumigation box in the fumigation room maintained at 15°C for apples and at 20°C for nectarines. Six difference doses (12-36g/m<sup>3</sup> for apples and 6-24g/m<sup>3</sup> for nectarines) of MB were applied to determine the mortality in the dose-response test. MB was introduced with a gas-tight syringe and then fumigated for 2 hours. The circulation fan was kept on during fumigation.

Fumigation for the larval stage inside 'Mutsu' in 1998 was conducted with the same manner as fumigation of the egg stage. Fumigation for 'Alps Otome' and 'Mutsu' in 1999 was conducted in a 0.52m<sup>3</sup> stainless steel fumigation chamber equipped with a circulation fan, ventilation apparatus, and ports for gas application and sampling and a manometer and temperature probes. Ninety-five of 'Alps Otome' and 25 of 'Mutsu' were placed in four plastic bins with many vents on four sides (0.062m<sup>3</sup>/bin in capacity) and then placed in the fumigation chamber at the same time with filler fruits for supporting 50% loading. MB enclosed in an ampul was applied by using a built-in ampul breaker. Each apple varieties was fumigated at doses of 15.6g/m<sup>3</sup> and 16.9g/m<sup>3</sup> for 2 hours at 15°C. The built-in circulation apparatus was kept on during fumigation.

Gas concentrations were measured with a gas chromatography (GC-8AIF with FID) at time intervals of 10, 30, 60, 90 and 120 minutes after the commencement of fumigation. Temperatures of fruit pulp and air in the fumigation box were also monitored with a multi-channel automatic temperature recorder (Hybrid Recorder: AH, Chino) during fumigation. Fumigation was followed by 2 hours of exhausting using the ventilation apparatus.

### Determination of Mortality

Fumigated fruits and pieces of filter papers with eggs were removed from the fumigation boxes and then placed for 9 days in the room maintained at 25°C, 70% R. H. until

confirmation of their hatching. Apple fruit infested with larvae were stored for 3 days and then assessed the mortality.

### Data Analysis.

The CT product was calculated similarly to the method described by MONRO (1969). Data for egg responses to LD<sub>50</sub> and LC<sub>50</sub> were analyzed by the computer program of the Probit analysis (POLO-PC; LeOra Software, 1987). Linearity of regression lines obtained from the statistical analysis were tested by the Chi-square test. The LD<sub>50</sub>'s and LC<sub>50</sub>'s were considered to be significantly different when their 95% confidence limits did not overlap.

## Results and Discussion

### Reliability of CT Products as an Indicator of Mortality Efficacy

Table 1 shows estimated LC<sub>50</sub>'s from the concentration-mortality data and LD<sub>50</sub>'s from the dose-mortality data for the peach fruit moth eggs on two apple varieties in the dose-response test. The LD<sub>50</sub>'s for 'Alps Otome' and 'Mutsu' were 18.1 and 16.5mg/l, while the LC<sub>50</sub>'s for 'Alps Otome' and 'Mutsu' were 19.1 and 19.0mg/l. Table 2 also shows estimated LC<sub>50</sub>'s and LD<sub>50</sub>'s for the peach fruit moth eggs on two nectarine varieties. The LD<sub>50</sub>'s for 'Fantasia' and 'Shuhou' were 20.8 and 22.4mg/l, while the LC<sub>50</sub>'s for 'Fantasia' and 'Shuhou' were also 17.7 and 17.9mg/l.

The difference of the LC<sub>50</sub>'s was smaller than that of the LD<sub>50</sub>'s in two varieties of apples and nectarines when compared the concentration-mortality data with the LD<sub>50</sub>'s from the dose-mortality data. The data indicated that even if the same dose was applied, there would be a possibility of difference in efficacy between varieties of commodities, while if gas concentration (CT product) was the same, the same efficacy would be obtained between varieties, and that if the susceptibility (LD<sub>50</sub>'s) of the pest by using one variety of

**Table 1.** Estimated LC<sub>50</sub> and LD<sub>50</sub> values for the peach fruit moth eggs on apple varieties fumigated with methyl bromide at 6 doses for 2 hours at 15°C with 0.16kg/l loading.

Variety	Replicate	Number tested	LC <sub>50</sub> (mg/l) (95%CL)	LD <sub>50</sub> (mg/l) (95%CL)
Alps Otome	3	15,256	19.1 (18.6 - 19.5)	18.1 (17.5 - 18.5)
Mutsu	3	24,323	19.0 (17.4 - 20.3)	16.5 (14.9 - 17.6)

**Table 2.** Estimated LC<sub>50</sub> and LD<sub>50</sub> values for the peach fruit moth eggs\* on nectarine varieties fumigated with methyl bromide at 6 doses for 2 hours at 20°C with 0.14kg/l loading.

Variety	Replicate	Number tested	LC <sub>50</sub> (mg/l) (95%CL)	LD <sub>50</sub> (mg/l) (95%CL)
Fantasia	3	5,320	17.7 (16.6 - 19.0)	20.8 (19.5 - 22.3)
Shuhou	2	5,220	17.9 (16.9 - 19.3)	22.4 (21.2 - 24.1)

\* Mortality data from eggs on filter paper were used for evaluation due to rot infested fruits before determination of mortality.

the commodity was determined in the dose-response test, the difference in efficacy for different varieties could be determined by measuring the CT product in each variety of commodities.

No significant difference was observed on eggs' response between varieties of 'Alps Otome' and 'Mutsu' apples and of 'Fantasia' and 'Shuhou' nectarines in both  $LC_{50}$ 's and  $LD_{50}$ ', respectively. However, 95% confidential limits between 'Alps Otome' and 'Mutsu' apples were slightly overlapped each other in the  $LD_{50}$ 's. The data suggested that there might be significant difference in efficacy between two apple varieties if larger difference was obtained between confidential limits in the  $LD_{50}$ 's.

### CT Product of Mortality Efficacy for Insects inside Fruits

Table 3 shows the CT product and the mortality ratio of the peach fruit moth larvae inside different sizes of 'Mutsu' apples (the weight ratio is approx 1:2.3). The difference of the mortality between sizes was 3.1% at  $14g/m^3$  and 0.8% at  $20g/m^3$ . Although the mortality ratio of the larvae inside small size fruit was higher than that inside large size fruit, no difference was observed on the mortality between sizes in 'Mutsu'. Table 4 also shows the mortality ratio of larvae inside 'Mutsu' of extremely large size and 'Alps Otome' of extremely small size of apples (the weight ratio is approx 1:9.2). The mortality at  $15.6g/m^3$  ( $28.1 g \cdot h/m^3$ )- $16.9g/m^3$  ( $31.9 \sim 32.2g \cdot h/m^3$ ) was 70.0~77.1% in 'Mutsu' variety and

**Table 3.** Mortalities of the peach fruit moth larvae inside different size of 'Mutsu' apples fumigated with methyl bromide at 14 and  $20g/m^3$  for 2 hours at  $15^\circ C$  with  $0.17kg/l$  loading.

Fruit size (g/fruit)	Replicate	$14g/m^3$			$20g/m^3$		
		Larvae tested	CT product ( $g \cdot h/m^3$ )	Mortality (%)	Larvae tested	CT product ( $g \cdot h/m^3$ )	Mortality (%)
LL 623	3	358	$28.4 \pm 0.2$	$86.0 \pm 1.3$	329	$40.2 \pm 0.4$	$98.7 \pm 0.7$
S 276	3	472	$27.3 \pm 0.1$	$82.9 \pm 2.9$	493	$38.8 \pm 0.2$	$97.9 \pm 0.9$

1. Survival ratios in untreated control lots; 95.1-100%.

2. The test was conducted in 1998.

**Table 4.** Mortalities of the peach fruit moth larvae inside 'Alps Otome' (extreme small size variety) and 'Mutsu' (extreme large size variety) apples fumigated with

Dose ( $g/m^3$ )	CT product ( $g \cdot h/m^3$ )	Mutsu			Alps Otome			Difference of mortality (%)
		Size (g/fruit)	Larvae tested	Mortality (%)	Size (g/fruit)	Larvae tested	Mortality (%)	
16.9	31.9	416	363	77.1	45	407	86.0	8.9
16.9	32.2	416	349	76.8	45	275	83.6	6.8
15.6	28.1	416	350	70.0	45	309	78.6	8.6

1. Fumigation schedules:  $15.6$  and  $16.9 g/m^3$  of MB for 2 hours at  $15^\circ C$  with  $0.16kg/l$  loading.

2. Survival ratios in untreated control lots; 95.1-100%.

3. The test was conducted in 1999.

78.6-86.0% in 'Alps Otome', respectively. Namely, the mortality ratios in 'Alps Otome' were always higher than those in 'Mutsu' with the difference of 6.8~8.9% under the same CT products of MB.

These data suggest that there is a possibility with the difference in fruit size affecting gas penetration in the case of 'Alps Otome' and 'Mutsu' which have extremely different size each other, while the difference in mortality dose not take place between fruit with normal size, *i.e.* the weight ratio between fruit is as large as 3 time.

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## 和 文 摘 要

臭化メチルくん蒸における  
果実品種間の CT 値の信頼性

溝渕 三必・土屋 芳夫・土肥野利幸・三角 隆・内藤 浩光  
高野 利達・丹野 昌浩・相馬 幸博・松本 信弘<sup>1)</sup>・齋藤 学<sup>2)</sup>  
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横浜植物防疫所調査研究部消毒技術開発担当

リンゴ2品種(“アルプス乙女”及び“陸奥”)及びネクタリン2品種(“ファンタジア”及び“秀峰”)に寄生したモモシנקイガ, *Carposina sasakii* の卵及び幼虫態を臭化メチルくん蒸し、殺虫効果の指標としての CT 値の信頼性を評価した。その結果、(1) 薬量-致死率のデータから得られた LD<sub>50</sub>値と濃度-致死率データから得られた LC<sub>50</sub>値を比較すると、リンゴ及びネクタリンともに LC<sub>50</sub>値の方が LD<sub>50</sub>値よりも両品種の差が小さかった。これは、品種間における殺虫効果はガス濃度(CT 値)が同じであれば同じ効果が得られるが、逆に、薬量が同じであっても濃度(CT 値)異なれば殺虫効果が異なることを示している。(2) リンゴ及びネクタリンの各品種間に寄生し

た卵の感受性は、LD<sub>50</sub>値及び LC<sub>50</sub>値の95%信頼限界値に基づけば有意差が認められなかった。しかし、“アルプス乙女”と“陸奥”の間の LD<sub>50</sub>値の信頼限界値はわずかに交差しているにすぎず、これらの差がわずかでも拡大すれば殺虫効果に有意差が生じることが判明した。(3) 異なるサイズの果実(“陸奥”;重量比において約2.3倍及び“アルプス乙女”と“陸奥”;重量比において約9.2倍)の内部に寄生した幼虫態の殺虫率は、“陸奥”のサイズ間では差が認められなかったが、“アルプス乙女”と“陸奥”の間では、アルプス乙女の方が常に殺虫率が6.9~8.8%高く、サイズが大きく異なる果実ではガス浸透力が殺虫効果に影響を及ぼしていることが判明した。

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