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Research Article

Effect of Vapor Heat Treatment on the Mortality of *Bactrocera dorsalis* (Diptera: Tephritidae) and the Quality of Mango cv. Arumanis

Pengaruh Perlakuan Uap Panas terhadap Mortalitas Bactrocera dorsalis *(Diptera: Tephritidae) dan Kualitas Buah Mangga Varietas Arumanis*

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ABSTRACT

Arumanis is a superior export variety mango from Indonesia. One inhibiting factor on the production of this fruit variety is the infestation of *Bactrocera dorsalis* (Diptera: Tephritidae) fruit fly. Vapor heat treatment was recommended by ISPM No. 28 of 2007 as an effective treatment in eradicating fruit flies. This research was aimed to find out the optimum temperature and the duration of vapor heat treatment on the mortality of egg and larvae of *B. dorsalis*. The experiment was conducted in the Laboratory of Vapor Heat Treatment, BBPOPT, Jatisari, from October 2016 to January 2017. The observed parameters were temperature, duration of treatment, mortality of egg and larvae of fruit fly, and fruit quality. The results showed that vapor heat treatment at 47°C for 40 minutes (min) was effective to reduce the number of eggs and larvae of *B. dorsalis* and had no negative impact on the fruit quality.

Keywords: *Bactrocera dorsalis*, fruit fly mortality, mango cv. Arumanis, quarantine, vapor heat

INTISARI

Buah mangga varietas Arumanis merupakan varietas mangga ekspor unggulan Indonesia. Salah satu faktor pembatas produksi buah mangga varietas Arumanis adalah lalat buah B. dorsalis *(Diptera: Tephritidae). Perlakuan uap panas direkomendasikan oleh ISPM Nomor 28 tahun 2007 sebagai tindakan perlakuan yang efektif dalam mengeradikasi lalat buah. Penelitian ini bertujuan untuk mengetahui suhu dan waktu optimum perlakuan uap panas terhadap mortalitas telur dan larva* B. dorsalis *pada buah mangga varietas Arumanis tanpa merusak kualitas buah. Penelitian dilaksanakan di Laboratorium* Vapor Heat Treatment, *BBPOPT, Jatisari, pada Oktober 2016 sampai dengan Januari 2017. Parameter yang diamati adalah suhu, lamanya waktu perlakuan, mortalitas telur dan larva lalat buah, dan kualitas buah. Hasil penelitian menunjukkan bahwa perlakuan uap panas pada suhu 47°C selama 40 menit terbukti efektif membunuh telur dan larva B. dorsalis dan tidak berdampak negatif terhadap kualitas buah.*

Kata kunci: Bactrocera dorsalis, *karantina, mangga varietas arumanis, mortalitas lalat buah, uap panas*

INTRODUCTION

Indonesia is one of the largest mango producers in the world. Mango productions in Indonesia on 2011, 2012, 2013, 2014 and 2015 were about 2,131,000, 2,376,000, 2,192,000, 2,431,000, dan 2,178,000 ton respectively (BPS, 2016). Arumanis was recognized as one of export mango variety which has a good potential market (Warji & Rokhani, 2008). However, its production was frequently disturbed by the infestation of pest such as fruit fly (Purnama *et al.*, 2014). Its onset resulted in low production and poor fruit quality. Most fruit flies in Indonesia was identified as genus *Bactrocera* and one of species was *B. dorsalis* (Suputa *et al.*, 2010). The losses caused by this pest could reach 70 % (Manurung *et al.*, 2012).

International Standard for Phytosanitary Measures (ISPM) no. 28 recommended vapor heat treatment (VHT) as one of potential treatment measures for eliminating fruit flies (IPPC, 2008). VHT on fruit using water vapor ca. 40−50°C, could be carried out with the objective of eliminating insect`s eggs and adult stages, as well as serving as quarantine treatment prior to shipment to destination countries (APHIS, 1985; Le *et al.*, 2010; APHIS, 2011).

Efficacy of vapor heat treatment against fruit flies has been reported by some researchers. Hasbullah *et al.* (2013), reported that VHT at 46.5°C for 20−30 minutes (min) was effective in eradicating the eggs of *B. carambolae* on the start fruit and no significant changes in the fruit weight loss, moisture content, hardness, color, total soluble solid and vitamin C. Report of Indonesia-Japan Economic Partnership Agreement (IJ-EPA) research revealed that vapor heat treatment at 47°C for 30 min was effective in eliminating the eggs and larvae of *B. papayae* on mango variety of Gedong Gincu without any effect on quality (IJ-EPA Unpublished, 2013). Until now, the information pertaining to the effect of VHT against *B. dorsalis* on mango variety of Arumanis has not been available yet. This research was aimed to find out the optimum temperature and the duration of VHT on mortality of eggs and larvae of *B. dorsalis* on mango variety of Arumanis and its impact on fruit quality.

MATERIALS AND METHODS

The experiment was conducted in the Laboratory of Vapor Heat Treatment (VHT), Pest Forecasting Institute (PFI), Ministry of Agriculture, Karawang, West Java, from October 2016 to January 2017.

Insects Rearing

Bactrocera dorsalis was formely known as *B. papayae* (Schutze *et al*., 2014) and for this research the they were originally obtained from the colony which was reared in Laboratory of VHT, Balai Besar Peramalan Organisme Penggangu Tumbuhan (BBPOPT), Ministry of Agriculture. Maintaining and rearing of fruit fly were performed in biotron (STH-19PS SBT-10-JL-1; Sanshu Sangyou Co. Ltd.) ca. 28°C temperature, and ca. 65−70% relative humidity (RH). Imagoes were fed with artificial sugar and autolise yeast (4:1 ratio) and larvae were fed with artificial diet (wheat bran, granulated sugar, Mau-ripan, HCL 3.5%, Sodium benzoate, tissue, and water) formulated by BBPOPT. Eggs and larvae were collected from 14-days old female fruit flies by placing egg-laying bottles (8 cm in diameter and 13 cm in height) in which their walls were proliferated with as much as 240 holes with diameter ca. 0.5 mm. Egg-laying bottle was placed for an hour. Mango juice was put in its surface for egg laying.

Preparation of Mango Fruits

The Arumanis mango was originally obtained from Majalengka, West Java with ca. 400−500 g in weight, ca. 70−80% maturity level, and were harvested from 6−7 years old trees.

Effect Vapor Heat Treatment on the Mortality of Eggs and Larvae of B. dorsalis

The objective of this assay was to find the optimum temperature which is capable for eliminating eggs and larvae of *B. dorsalis*. Four insects from each stage were inoculated on mango fruits with three replications (4 insects × 3 replications × 7 temperature treatments). Egg inoculation containers were 3.4 cm in length and 2.2 cm in width, with 5 mm in diameter of hole. Afterwards, the container was covered with water and heat-resistant materials (heat-resistant cellotype tape). The inoculated fruits were kept in incubator at 28°C until the vapor heat treatment was started.

The VHT machine (Model: EHK-1000D, Sanshu Sangyo EHK-1000) was used in this research. Heat temperatures were set up at 45, 45.5, 46, 46.5, 47, 47.5°C, and control (without any vapor heat treatment). Inoculated fruits with eggs and larvae of *B. dorsalis* were put into machine at 48.5°C VHT and 95% RH. During treatment, core temperature of the treated fruit was measured with three censors which was displayed on monitor. After treatment, fruits were immediately cooled by tapping in the water for 10 min. And then fan dried for 30 min. To bring them into normal condition. The observations on the mortality of eggs and first instar larvae were conducted at 4-days after treatment while the mortality of second and third larvae was observed at 3-days after treatment. Fruits were then observed the number of unhatched eggs and dead larvae were counted. This experiment was designed with Completely Randomized Design with three replications.

Optimalization Period of Vapor Heat Treatment

The following steps were the disinfestation process of eggs on fruits under 47°C for 0, 5, 10, 15, 20, 25, 30, 35, 40 min, and control (without any vapor heat treatment). The objective of this assay was to find the optimum duration of vapor heat treatment at 47°C. Twenty seven mango fruits were inoculated with 150 eggs, placed into machine at temperature of 48°C and RH 95%. After treatment, fruits were immediately cooled with tapping water for 10 min and then fan dried for 30 min to bring them into normal condition. The observation on the mortality of eggs was conducted at 4-days after treatment. Fruits were then cleaved to count the number of unhatched eggs. This experiment was designed with Completely Randomized Design with three replications.

Effect of Vapor Heat Treatment on Fruit Quality

The assay was carried out on 30 mango fruits at 47°C for 40 min and other 30 mango fruits as control. Treated fruits were kept at room temperature of 28°C. The quality was observed on 5 and 10 days after treatment. The quality parameter were weight loss, hardness, change of peel color, content of sugar, vitamin C, and taste.

Data Analysis

For egg and larval mortality test, the corrected mortality was calculated using the following formula of Abbott's (Abbott, 1925; Toshiyuki *et al*., 2014):

$$\text{Corrected mortality (\%)} = \frac{\text{mortality (treatment)} - \text{mortality (control)}}{100 - \text{mortality (control)}} \times 100\ \%$$

The obtained corrected mortality was analyzed with analysis of variance (ANOVA) to view the effect of optimum temperature and time duration of vapor heat treatment against mortality of eggs and larvae and proceeded with Tukey test ($\alpha = 5\%$).

For fruit quality test, which was conducted before and after treatment, data were then analyzed using t-test ($\alpha = 5\%$) of SPSS software, while data from taste simulation test were analyzed using chi-square test ($\alpha < 5\ \%$).

RESULTS AND DISCUSSIONS

Effect of Vapor Heat Treatment on the Mortality of Eggs and Larvae of B. dorsalis

The results showed that vapor heat treatment and *B. dorsalis* mortality had positif correlation as show on Table 1. Vapor heat treatment was effective in eradicating eggs and larvae of *B. dorsalis* on mango fruit. IJ-EPA (Unpublished, 2013) reported that vapor heat treatment was effective in diminishing eggs and larvae of *B. carambolae* and *B. dorsalis* on mango fruit variety of Gedong Gincu.

Data analysis showed that mortality of eggs and larvae of *B. dorsalis* under vapor heat treatment was significantly different with those in control. This assay demonstrated that mortality of eggs and larvae increased along with the increase of treatment temperature (Table 1). Wigglesworth (1977) described that the incapability of insects in surviving at high temperature was caused by the imbalance of metabolism rate happening in their bodies so that the insects produced some secondary metabolites which would be secreted from their bodies and make the food reserves in them exhausted and the insects then would be dead.

Table 1 also showed that egg was the most resistant stadia. Mortality of eggs which was lower than larvae. Emekci *et al*. (2001) explained that egg respiration was lower than larval respiration of some insects. This finding was in accordance with the research of Heard *et al*. (1992) which showed that egg was the most heat-resistant stage compared to other stadia under vapor heat treatment by using *Queensland fruit fly*. Armstrong *et al.* (2009) also reported that egg stage was the most heat-resistant on some species of genus *Bactrocera*.

Heat temperature will quicken and can even kill the larvae. Temperature also affects insect growth enzymes. Insect growth enzymes will be in normal action at room temperature but will increase if the temperature was raised, which causes accumulation of metabolites and can not spread rapidly throughout the body (Wigglesworth, 1977).

Moss and Jang (1991) reported that the lack of oxygen was the cause of mortality of Mediterranean fruit flies under hot water treatment. Neven (1998) also noted that the elevated temperature under heat treatment affected the respiration of larvae. Oxygen was an important and most required substance for respiration process.

Loganathan *et al*. (2011) stated that insect resistance against heat was determined by species, its position within fruit, combination of temperature and time exposure, as well as the development of insect stages. Meanwhile Neven (2000) explained that physiochemical factors also determined insect mortality under heat treatment such as change of metabolism, decline of respiration, nerve distortion, endocrinal system, and production of heat shock protein. According to Hosking (2007), the death of the insect was indicated with the breakage of plasma

Table 1. Mortality of eggs and larvae of *Bactrocera dorsalis* after VHT at temperature of 28−47.5°C

Temperature (°C)	Corrected Mortality (%)*			
	Egg	1st Instar	2nd Instar	3rd Instar
Control (28.0)	0.00 c	0.00 c	2.51 b	3.90 b
45.0	58.67 b	79.70 b	97.23 a	100.00 a
45.5	62.23 b	83.60 ab	99.17 a	100.00 a
46.0	63.33 b	88.90 ab	100.00 a	100.00 a
46.5	69.57 ab	87.50 ab	100.00 a	100.00 a
47.0	99.13 a	100.00 a	100.00 a	100.00 a
47.5	100.00 a	100.00 a	100.00 a	100.00 a

*The numbers which was followed by the same letter on the same column is not significantly different based on Tukey-test ($\alpha = 5\%$).

membrane and protein denaturation and then resulting in enzymatic interference in DNA.

Duration of Vapor Heat Treatment on Mortality of B. dorsalis *Eggs*

Test of vapor heat treatment under various temperatures revealed that 100% of eggs mortality happens at 47.5°C (Table 1). However, it was assumed that fruits kept in VHT machine had not reached desired temperature under those treatments, so in this test a 47°C temperature with varying duration was used. According to Singh *et al*. (2001), there would be different core temperature of fruit between fruits located on middle, bottom and upper racks during VHT process. The alternative solution was by adding time of treatment in order to get similiar target of core temperature. The previous research of IJ-EPA (Unpublished, 2013) reported that vapor heat treatment at 47°C for 30 min was effective in eliminating eggs and larvae of *B. dorsalis* on mango fruit of Gedong Gincu variety. Jacobi and Giles (1997) also recorded that quarantine treatment on mango using VHT or HWT methods, i.e. in temperature range of 46−47°C depending on fruit size and variety.

The result showed that the longer the time of treatment, the higher the mortality of eggs, which means that vapor heat treatment at 47°C for 40 min was effective in eradicating eggs of *B. dorsalis* (Table 2). It also indicated the difference with the finding of Indonesia-Japan Economic Partnership Agreement (IJ-EPA) which showed that vapor heat treatment at 47°C for 30 min could kill 100% of *B. dorsalis* eggs on mango fruit of Gedong Gincu variety. It was supposed due to difference in fruit weight and shape between those two varieties which could influence the mortality of fruit fly eggs after treated with vapor heat. Other supporting factors were the thickness of the pulp of Arumanis mango,

Table 2. Effect of the duration of VHT at temperature of 47°C on mortality of *Bactrocera dorsalis*

Treatment (minute)	Corrected Mortality (%)*
	Egg
Control	13.11 c
0	96.17 b
5	98.43 a
10	99.77 a
15	99.77 a
20	100.00 a
25	100.00 a
30	100.00 a
35	100.00 a
40	100.00 a

*The numbers which was followed by the same letter on the same column is not significantly different based on Tukey-test ($\alpha = 5\%$).

thus requiring a considerable long time to kill eggs of *B. dorsalis*. Omura *et al*. (2014) reported that fruit weight and shape affected the mortality of eggs under vapor heat treatment. A similar result was also reported by Yoshinaga *et al*. (2009) who concluded that fruit weight influenced the increase of inner pulp temperature. Fruit thickness caused the significant difference in of inner pulp temperature's fluctuation.

The results of heat progress observation in vapor heat treatment duration test on *B. dorsalis* eggs mortality showed that fruit temperature increase was slower than VHT machine temperature increase. Center temperature of 47°C was achieved by using VHT machine temperature of 48° C and RH>95%. The observation results showed that the time needed for Arumanis mangoes to reach center temperature of 47°C is 150 minutes (Figure 1) while research that has been done by the IJ-EPA (Unpublished, 2013) on Gedong Gincu mangoes small size, to reach center temperature of 47°C, 75 minutes is needed. Based on that, there are differences in the

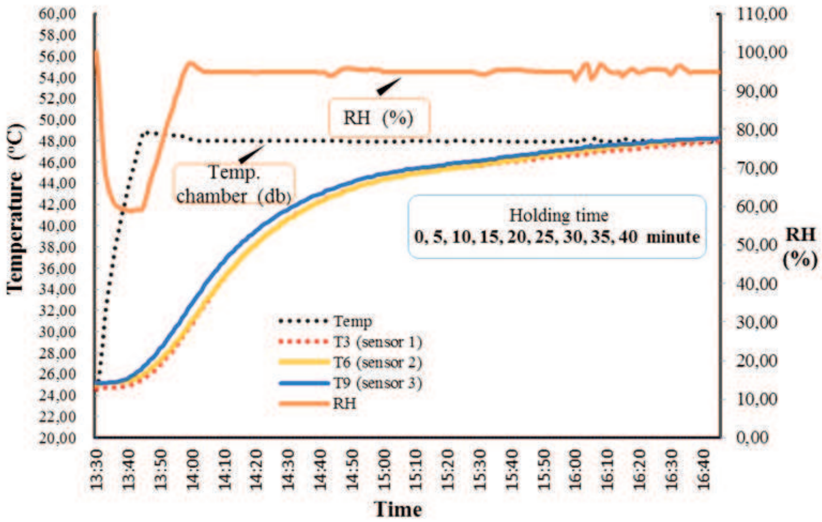

Figure 1. Changes of fruit core temperatures, chamber air temperature and relative humidity (RH) in VHT chamber during test

time needed to reach the center temperature between both varieties. It was supposed to be due to the thickness and texture of the fruit.

Effect of Vapor Heat Treatment on Fruit Quality

Weight loss. Table 3 showed that vapor heat treatment did not significantly affect weight loss. The similar result was reported by Hasbullah *et al.* (2001) who found that heat treatment could not crucially influence weight loss of mango variety of Irwin during storage period. The identical finding was also reported by Sunagawa *et al.* (1987) who recorded that VHT could not affect the weight loss of mango variety of Irwin. The given heat treatment on fruit might decrease as well as increase the weight loss. It could reduce weight loss by transforming the structure of epicuticular wax and filling up cracks of cuticle (Schirra *et al*., 2000). In addition, heat treatment could also improve the weight loss concomitant with time duration of heat treatment associated with the wider cuticle cracks because of overdoses heat treatment (D'hallewin & Schirra, 2000).

Fruit hardness. Fruit hardness was one of the most common indicators in recognizing fruit quality. The results revealed that VHT did not significantly affect the decrease of fruit hardness (Table 3). It was supposed that this happened since there were the presence of obstacle and the improvement of pectin hydrolysis activity by elevated temperature as consequence of vapor heat treatment. Shalom *et al.* (1996) stated that those phenomena were main important factors for fruit hardness as consequence of heat treatment.

Jacobi and Giles (1997) reported that treated mango variety of Kensington within 15 min was softer than control. On the contrary, Le *et al.* (2010) documented that heat treatment in terms of hot water treatment (HWT) and VHT applied to mango variety of Taiwan could maintain the fruit hardness for a week during storage, but it then declined after 3 weeks' storage.

Peel color. Peel color of fruit was measured with color reader that shown L, a, and b value. L value showed the lightness, a value showed yellow or red colour, and b value showed yellow colour. Table 3, indicated that VHT did not significantly influence the component of peel color. The significant one was reflected by value of L on 10-days after inoculation. During storage, value of a tended to encounter the improvement indicated by change of green to yellow and value of b experienced also during storage (Table 3). Hasbullah *et al.* (2001) also reported similar results, in which time duration of VHT did not crucially affect the color change of mango variety of Irwin during storage. Process of mango peel color from green to be yellow was assumed due to chlorophyll degradation. Wills *et al*. (1989) stated that the yellowing color was caused by degraded chlorophyll during maturity as consequence of carotene pigment. This pigment was stable in peel fruit, but its appearance was covered by chlorophyll.

Table 3. Effect of vapor heat treatment on fruit quality of Arumanis mango

Parameter of observation*	Control	Vapor heat treatment with temperature 47°C for 40 minute
Weight Loss (%)		
5 day after treatment	10.01 a	10.17 a
10 day after treatment	12.58 a	12.71 a
Fruit Hardness (kg)		
5 day after treatment	0.25 a	0.27 a
10 day after treatment	0.07 a	0.06 a
Peel Color (L)		
before treatment	51.38 a	53.53 a
5 day after treatment	54.51 a	57.19 a
before treatment	49.36 a	49.12 a
10 day after treatment	49.10 a	52.29 b
Peel Color (a)		
before treatment	-11.43 a	-10.86 a
5 day after treatment	9.08 a	9.58 a
before treatment	-11.22 a	-11.07 a
10 day after treatment	1.33 a	3.31 a
Peel Color (b)		
before treatment	26.53 a	27.01 a
5 day after treatment	51.76 a	54.47 a
before treatment	25.65 a	24.30 a
10 day after treatment	59.03 a	59.64 a
Content of sugar (%)		
5 day after treatment	15.82 a	15.72 a
10 day after treatment	0.14 a	0.15 a
Content of vitamin C (mg/100g)		
5 day after treatment	47.04 a	43.58 a
10 day after treatment	45.86 a	42.06 a

The numbers which was followed by the same letter on the same row is not significantly different based on t-test ($\alpha = 5\%$).

Content of sugar. The results elaborated that vapor heat treatment did not significantly influence the content of fruit sugar (Table 3). It was parallel with Le *et al.* (2010) who reporting that HWT and VHT could not affect the change of sugar content on mango variety of 'Tuu Shien" for three weeks storage. It was also in line with Kim *et al.* (2009) who documented that heat treatment at 46.1°C for 70, 90, and 110 min respectively did not have significant effect against change of sugar content on mango variety of 'Tommy Atkin". Conversely, Djioua *et al.* (2009) reported that their final results showed higher sugar content after 9 days' storage after dipping treatment with hot water of 50°C for 30 min. Sugar content of mango fruit would experience the increase and decrease during storage. Helmiyesi *et al.* (2008) exhibited that the increase of sugar content was caused by the breakage process of polysaccharides into sugar (sucrose, glucose, fructose) occurring at post-harvest period, whereas decrease of sugar content was resulted by the established polysaccharide reserves run low.

Vitamin C. The results showed that vapor heat treatment did not significantly affect the content of vitamin C (Table 3). These results were parallel with the research of Hasbullah *et al.* (2013) who reporting that disinfection method using vapor heat did not has real effect on content of vitamin C in star fruit. However, Wang and Camp (2000) contrarily found that the content of vitamin C could decrease concomitant with the increase of temperature. Vitamin C was easily degraded, either by temperature, light, or surrounding air (Begum *et al.*, 2009).

Taste assay. Taste assay was carried out to find out whether there was difference in taste between mango fruit treated with vapor heat and control (without any treatment). The results revealed that there was no difference in taste of the treated and control fruits (Table 4). This finding was in line with the result obtained by Jacobi *et al.* (1993) who reported that heat treatment on mango did not influence the taste, aroma, pH, total soluble solids,

Table 4. Effect of vapor heat treatment at temperature 47°C for 40 minute on flavor of Arumanis mango

Treatment	Taste test*
5 day after treatment	2.45
10 day after treatment	1.25

Chi-square test α < 5 % (Table 5 % = 3.84)

and total of acid. The similar result was documented by Jacobi and Giles (1997) that heat treatment at 47°C for 15 min did not affect the taste of Kensington mango.

CONCLUSION

Vapor heat treatment at 47°C for 40 min was effectively proven in eradicating the eggs and larvae of *B. dorsalis* and did not affect quality of Arumanis mango variety.

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