

HOST PREFERENCE OF THE PUMPKIN FRUIT FLY, *ZEUGODACUS TAU* (WALKER) UNDER LABORATORY CONDITIONS

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Abstract

The pumpkin fruit fly (*Zeugodacus tau*) is an important agricultural pest in many regions of the world. It has more than 91 economic host-plant species, particularly in the family Cucurbitaceae. The infestation of this fruit fly species has caused damage to quality and quantity of agricultural products. The host preference of the pumpkin fruit fly with choice and no choice tests using cucumber, pumpkin and bitter gourd as host-plants were performed under laboratory conditions. The results demonstrated that the number of pupae and adult pumpkin fruit fly found in the three cucurbit host-plants were significantly different ($P < 0.05$). The highest number of pupae and emerged adults for the choice test were found in pumpkin, followed by cucumber and bitter gourd, respectively. For the no choice test, the highest number of pupae and emerged adults were observed in cucumber, followed by pumpkin and bitter gourd, respectively. The host preference of the pumpkin fruit fly in this study indicated that female fruit flies use vision and olfactory cues to locate the host-plant. They were attracted by cucumber odor and yellow color of pumpkin. In addition, the nutrient compositions of the host-plant also affect fruit fly performance. Tephritid fruit flies preferred host with high concentration of water, carbohydrate and fat. In conclusion, the study on host preference of the pumpkin fruit fly revealed factors affecting host-plant selection that can be used as information for pest management for the imported and exported cucurbit products.

Keywords: Pumpkin fruit fly, Host-plant, Host preference, Cucurbitaceae

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Introduction

The pumpkin fruit fly, *Zeugodacus tau* (Walker) (Diptera: Tephritidae) is a significant polyphagous pest of more than 91 economic cultivated species in family Cucurbitaceae and other families (Singh et al., 2010; Shi et al., 2014; Jaleel et al., 2018). It was first described in the People's Republic of China, however, currently it has been spread all over South Asia, East Asia, Southeast Asia and the South Pacific region (Jaleel et al., 2018). In Thailand, this fruit fly species can be found in all geographical regions throughout the year (Baimai et al., 2000; Saelee et al., 2006; Kitthawee & Julsirikul, 2019). The pumpkin fruit fly infests fruits and vegetables by punctures with its ovipositor and lay eggs. The hatched larvae of pumpkin fruit fly feed on host fruit tissues lead to the decomposition and premature dropping of the fruits. In addition, a secondary infestation by other herbivorous insects and plant pathogens is occurred at puncture marks. These result in a reduction in yield and quality of agricultural products and also losses in export opportunity because of strict pest quarantine regulations imposed by many countries (Vargas et al., 2015; EFSA Panel on Plant Health, 2020). The pumpkin fruit fly is a polyphagous insect, thus it has ability to move among and infest various host-plants (Kennedy & Storer, 2000). Furthermore, the pumpkin fruit fly has long dispersal distance (Shi et al., 2014) with high reproductive potential (Singh et al., 2010) and very adaptable to environmental change (Huang et al., 2020). Therefore, these factors are keys that allow populations of the pumpkin fruit fly to persist at any time of the year and cause seriously damage on agricultural crops.

The Cucurbitaceae or cucurbits are a plant family, consist of more than 800 species and cultivated around the world. In addition, they are regarded as an important food and medicinal plant in many countries (Rolnik & Olas, 2020). In Thailand, cucurbits are one of the most important economic crops which grown for domestic consumption and exportation, such as, watermelon, cucumber, pumpkin, gourd and cantaloupe (DOA, 2013). However, the infestation of cucurbit fruit fly is the huge threat to cucurbit cultivations. Accordingly, the pre-harvest and post-harvest managements are required to control pests without causing chemical residues and environmental problems (Verghese et al., 2006; Prange, 2012). Thus, various research studies involving in biology, ecology, genetics and behavior of insect-pests, including host preference of the

polyphagous insect are essential (Aluja & Mangan, 2008). Host preference of the polyphagous insect is associated with several factors, such as, oviposition strategy of the female, characteristic and quality of the host-plant, as well as coevolution between insect-pest and host-plant (Kennedy & Storer, 2000). In Thailand, there are only few researches that studied on host-plant preference of tephritid fruit fly including *Bactrocera papayae* (Petlamul et al., 2009), *Bactrocera dorsalis* (Rattanapun et al., 2009) and *Bactrocera latifrons* (Rattanapun et al., 2021). Therefore, the present study aimed to determine the host preference of the pumpkin fruit fly on three commercial cucurbit host-plants. The results will provide information about host-plant selection by the fruit fly that may be required to the pest management program and facilitate the economic crops exportation of Thailand.

Materials and Methods

The pumpkin fruit fly (*Zeugodacus tau*)

The pumpkin fruit fly in this study was obtained from laboratory colony at the Department of Biology, Mahidol University, Bangkok, Thailand and then, maintained at the Department of Biology, Burapha University, Chonburi, Thailand. The identification of fruit fly species as *Z. tau* was confirmed using taxonomic keys from White & Elson-Harris (1992) and Sumrandee et al. (2011). Adult flies used for the host preference experiments were mass-reared in a ventilated plastic cage (24.5 cm in diameter), provided with 10% solution of honey and yeast extract powder as food source at laboratory conditions (25 ± 2 °C, 65 ± 10 % RH, and 12-h natural daylight).

Host preference of the pumpkin fruit fly

Choice and no choice host preference experiments of the pumpkin fruit fly were conducted simultaneously under laboratory conditions with three replications of each experiment. Fruit flies used for the experiments were 60 pairs (1♀:1♂) of newly emerged adult in ventilated plastic cage and provided with food source. The adult pumpkin fruit flies were reared for 14 days until reach the oviposition period (Singh et al., 2010) before starting the experiments. The three commercial cucurbit host-plants were cucumber (*Cucumis sativas* L.), pumpkin (*Cucurbita moschata* Duchesne), and bitter gourd (*Momordica charantia* L.). All host-plants were purchased from Nongmon

market, Chonburi and soaked in 1% salt solution to reduce pesticide residues, then rinsed with tap water before used in the experiments.

1. Choice test

The cucurbit host-plants were cut to size 4x8x2 cm and placed on the petri dish (5 cm in diameter). Then put all three host-plants in the experimental cage that contain 60 pairs of the pumpkin fruit fly for 24 h. After that, the exposed cucurbit host-plants were removed and replaced with a new set of host every day for 5 days. The infested hosts were kept separately in ventilated plastic box (11x11x5.5 cm) with straw absorbent paper at the bottom for pupation. The data were collected as a total number of pupae and emerged flies in each host-plant.

2. No choice test

For the no choice test, three experimental cages were setup, each with one type of cucurbit host-plants (described above) and 60 pairs of the pumpkin fruit fly. Host-plant were exposed in the cage for 24 h, removed and replaced with new host every day for 5 days. The subsequence procedures including the data record were performed as described in the choice test.

Data analysis

Host preference for choice and no choice tests of the pumpkin fruit fly was evaluated in term of average number of pupae and emerged flies with standard deviation (SD). The percentage of adult flies emergence was also calculated. The data on average number of pupae and emerged flies were subject to analysis of variance (ANOVA) using SPSS version 23 (IBM Corporation).

Results

The number of pupae and adults obtained from three cucurbit host-plants revealed the significant difference among three host-plants, for both choice and no choice tests ($P < 0.05$). However, there was no significant difference among the egg laying date and no effect of host-plant type and egg laying date on the number of pupae and adults ($P > 0.05$) (Table 1). The Least significant difference (LSD) test between pairs of means at the 0.05% level of significance showed that the highest number of pupae, emerged adults and percentage of emergence for the choice test were observed in pumpkin, followed by cucumber and bitter gourd, respectively. While

the highest number of pupae, emerged adults and percentage of emergence for the no choice test were observed in cucumber, followed by pumpkin and bitter gourd, respectively (Table 2).

Table 1 Analysis of variance (ANOVA) for the average number of pupae and adult fruit flies collected from three cucurbit host-plants

Source	Choice test				No choice test			
	pupae		adults		pupae		adults	
	F	P value	F	P value	F	P value	F	P value
Host-plant type	320.926	0.000	285.387	0.000	382.674	0.000	108.132	0.000
Egg laying date	0.809	0.529	1.782	0.158	1.428	0.249	0.188	0.943
Interaction	1.653	0.152	2.122	0.065	1.080	0.403	1.311	0.276

Table 2 Number of pupae and adult fruit flies (average \pm SD) and adult emergence (%) in three cucurbit host-plants

Host-plant type	Choice test			No choice test		
	Pupae	Emerged adult	% emergence	Pupae	Emerged adult	% emergence
Bitter gourd	21.60 \pm 2.36 ^a	14.07 \pm 4.44 ^a	65.12	35.87 \pm 6.61 ^a	21.60 \pm 9.22 ^a	60.22
Cucumber	64.27 \pm 7.77 ^b	47.20 \pm 4.95 ^b	73.44	105.73 \pm 7.74 ^b	82.27 \pm 12.66 ^b	77.81
Pumpkin	73.40 \pm 6.68 ^c	54.60 \pm 6.63 ^c	74.39	86.20 \pm 6.85 ^c	64.53 \pm 11.21 ^c	74.86

Remark Average values of pupae and adult fruit flies in the same column followed by the different superscript letters are significantly different ($P < 0.05$)

Discussions

The three cucurbit host-plants used in the study of host preference by pumpkin fruit fly under laboratory conditions were cucumber, pumpkin and bitter gourd because they were important economic crops and can be found in all geographical regions of Thailand (DOA, 2013). In addition, these cucurbits have distinct morphological characteristics such as color, shape, texture and are inexpensive. Thus, they are suitable for use as materials in the experiments. The previous studies on the host preference of the pumpkin fruit fly revealed that it preferred cucurbit crops, such as sponge gourd (Khan et al., 2011) and cucumber (Yang et al., 1994; Wu et al., 2011). However, the pumpkin fruit fly has been considered as a phytophagous and polyphagous

fruit pest (Singh et al., 2010; Jaleel et al., 2018), therefore, there were reports that it can infests tomato, eggplant, papaya, guava, tangerine and banana under field and laboratory conditions (Yang et al., 1994; Khan et al., 2011; Wu et al., 2011; Boopathi et al., 2017; Shi et al., 2017). The study on host-plant selection in polyphagous insects revealed that the process of host selection by female insects depends on several factors, including host-plant and environmental factors such as temperature, wind and light conditions (Bernays & Chapman, 1994). In general, the females preferred to oviposit their eggs on the suitable host-plant that increase the survival ability of their offspring (Thompson, 1988). The host selection behavior of polyphagous insects can be divided into host finding of the females that are attracted by odor, color and shape of the host-plants. Then, in the host acceptance stage, the physical and chemical properties of the plants such as size and nutritional value are employed in decision making of female insects (Bernays & Chapman, 1994).

The host preference of pumpkin fruit fly in the present study for the no choice test was consistent with Yang et al. (1994) and Wu et al. (2011), that it preferred cucumber. Khan et al. (2011) demonstrated that cucumber odor appeared to be an attractant for egg laying of female pumpkin fruit fly because plant odors were used as a cue for host location (Quilici et al., 2014). While Devi et al. (2020) showed that the most attractive odor for the pumpkin fruit fly was hydrolysed protein since tephritid fruit fly required protein diet for female maturity. In this study, the pumpkin fruit fly was adequately provided with protein diet (yeast extract powder) during the experiment, therefore, the influence of plant odor on host selection behavior of the pumpkin fruit fly was clearly demonstrated. In addition, the female fruit fly also used visual perception in their host-plant selection. The evaluation of chromatic cues for the preference of the pumpkin fruit fly showed that yellow and yellowish green were the most attractive color (Li et al., 2016). Accordingly, the pumpkin with yellow flesh was the most preferred host for the choice test in this study and may be indicated that color has more influence than odor on host-plant selection of the pumpkin fruit fly. However, it was found that the percentages of fruit fly emergence in pumpkin and cucumber were not obviously different although pupa density was different. This may be due to the influence of nutrient content of the host-plants. Wu et al. (2011)

explained that the percentage of adult emergence in the pumpkin fruit fly was influenced by pupa health which determined by nutritional composition in the host-plants (Bernays & Chapman, 1994; Awmack & Leather, 2002). The most preferred hosts in this study were cucumber that made up of 96.73% water, 2.16% carbohydrate, 0.16% fat (Uthpala et al., 2020) and pumpkin which contains 92.24% water, 5.31% carbohydrate, 0.15% fat (Dhiman et al., 2009). High concentration of water, carbohydrate and lipid in the host-plant promote larval performance in Tephritid fruit flies (Hafsi et al., 2016) while mineral nutrition are essential for adult fecundity (Awmack & Leather, 2002). Therefore, the percentages of adult emergence in cucumber were approximate to pumpkin and higher than those of bitter gourd because cucumber and pumpkin contain greater essential nutrients for fruit fly performance than bitter gourd (Sorifa, 2018).

The study of host preference of the pumpkin fruit fly revealed the status of host-plant which generally divided into a natural host, a conditional host and a non-host (Aluja & Mangan 2008). Host status determination is the key of the strategic decision-making on the national and international trade of fruit and vegetable (Aluja & Mangan 2008). Additionally, it also revealed the factors influencing and behavior of host selection of the fruit fly. Several studies reported that cucurbit crops were natural host of the pumpkin fruit flies and they were attracted by cucumber odor (Khan et al., 2011). However, the tephritid fruit flies did not used only chemical cue but also visual cue for host-plant selection. Thus, the yellow and yellowish green conditional hosts are attracted to them as well which may finally resulted in the host expansion (Bernays & Chapman, 1994). Furthermore, the results in the present study indicated the non-significant ANOVA of egg laying date, consistent with Yang et al. (1994) that the pumpkin fruit fly had relatively constant egg production during oviposition period and declined after age of 80 days. The long oviposition period combined with the ability to expand on a new host-plant of the pumpkin fruit fly are the important factor in causing huge damage to agricultural products.

Conclusions

In conclusion, the host preference of insect pest reveals behaviors and factors affecting host-plant selection that can be used as information for pest management. For example, removing the fallen, overripe and damage fruits from the planting area prevents insect attracting odor. On the other hand, fruit odor extract combined with protein bait can be used to trap female tephritid fruit fly in the outbreak area. Moreover, it can also determine the host-plant status and the risk of insect pests that may be found in the imported and exported agricultural products. However, the host preference of the pumpkin fruit fly in this study was conducted only in three cucurbit host-plant species under laboratory conditions. Therefore, it would be better to continue the experiment on more host-plants as well as expanding to field study that will provide useful information for the designation of the pest quarantine, control and elimination methods effectively.

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