OBSERVATION ON THE FLOWER-VISITING INSECTS OF ROSE (ROSA X DAMASCENA) IN AN AGRICULTURAL LANDSCAPE

HIDAYAH NOR MAHMOD AND MOHAMED NOR ZALIPAH*

Faculty of Science and Marine Environment, Universiti Malaysia Terengganu, 21030 Kuala Nerus, Terengganu.

*Corresponding author: zalipah@umt.edu.my

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Abstract: Many ornamental plants are aseasonal, providing continuous food supply which leads to survival of many flower-visiting insects. Although the flower-visiting insects are always perceived as pollinators, not all visitation actually resulted in pollination of the flowers. Thus, this study aimed to identify the flower-visiting insects of a common ornamental plant, Damask rose, and to determine their potential role as pollinators from their foraging behaviour at the flowers. Data collection was conducted in a residential area located in an agricultural landscape of Jasin, Melaka, between January and April 2021. A camera was set up approximately 0.5 meter from the full bloom flower, throughout its anthesis period. From the video footage, the parameters recorded were visitation frequency, visitation type (legitimate and illegitimate visitation) and the time spent at the flower. As a result, 253 visitations were recorded by six insect taxa; four from the order Hymenoptera and one each from the order Lepidoptera and Diptera. A hymenopteran bee, Halictidae 1 was found to show the highest visitation frequency. Only three species; Halictidae 1, Heterotrigona itama and Amegilla sp. showed legitimate visits. For the time spent (mean \pm SE), Halictidae 1 (30.08 \pm 4.87 minute) recorded significantly longer time as compared to other visitors (H = 13.78, df = 4, p < 0.01). Based on the visitation rate, time spent, and visitation type. Halictidae 1 showed the highest potential as pollinator for the rose flower, followed by H. itama and Amegilla sp. It is important to understand the contributions of ornamental plants such as the rose as food source to these insects for conservation of these pollinating agents. Not only these flowers help in the survival of pollinating agents throughout the years, these pollinating agents also contribute to the survival of the flowering plants by facilitating the pollination process of the flowers they visited.

Keywords: Visitation frequency, legitimate visit, time spent, pollinators, Hymenoptera.

Introduction

Many animals visit the flowers to forage for food. As these flower-visiting animals feed on the floral resources such as flower nectar and pollen grains, they carried and transferred the pollen grains of the flowers they visited thus becoming pollinators (Willmer, 2011). The most effective pollinators can be determined from their foraging behavior such as regular visitations to the flower and effective deposition of pollen to the stigma while foraging at the flowers. Instead, some flower-visiting animals are just flower predators and nectar thieves. Flower predators resulted in destruction to the reproductive parts or to the whole flowers, while nectar thieves usually did not destruct the flowers, but neither contribute to the pollination of the flowers (Inouye, 1980).

During the enforcement of Movement Control Order (MCO) in Malaysia starting from March 2020, interest in gardening and farming in residential areas has increased among the public to occupy their time (Chung, 2020). Horticultural flowering plants, could help to extend the flowering season for the flower-visiting animals, when the flowers produced by the native plants is scarce (Mach & Potter, 2018). Hence, the residential areas have become the refugee for these animals particularly the insects, indirectly contribute to conservation of these important animal group. In the agroecosystem landscape, appropriate management strategies of these beneficiary plants in semi-natural habitat such as the residential compound could be beneficial to the neighbouring crops (Barbir, 2016).

Universiti Malaysia Terengganu Journal of Undergraduate Research Volume 5 Number 2, April 2023: 18-27

Rosa spp. is the foremost common ornamental plants and being cultivated for many millennia (Debener & Linde, 2009). The genus *Rosa* (Family Rosaceae) is native to the northern side of the equator, with over 1,302 recognised taxa (Zlasek *et al.*, 2017). Many taxa from this genus has been cultivated for its showy petal, which is also a key factor to attract the flowervisiting insects (Balfour & Ratnieks, 2023). The purposes of this study were to identify the flower-visiting insects of the cultivated rose hybrid, *Rosa x damascena*, and to observe the behaviour of these insect visitors while visiting the flowers. Hence, this study aimed to identify the insects utilising the flowers for food source and to determine which insects are potential pollinators while foraging at the flowers.

Materials and Methods

Study Site and Target Species

This study was conducted between January and April 2021 at Kampung Selandar, Jasin (2° 19' 0" N 102° 26' 0" E), located at the south of Malacca, Peninsular Malaysia (Figure 1). The study site was a private residential compound surrounded by agriculture areas such as the oil palm and rubber plantations, as well as fruit orchards.



Figure 1: The location of study site at Jasin, Malacca, in West Coast of Peninsular Malaysia (left). The sampling was conducted in a private residential area in the agriculture landscape (right)

In this study, the rose (genus *Rosa*) was selected as 30 rose plants of several cultivars and hybrids were planted in the compound of approximately one acre. The genus *Rosa* consists of woody perennial plants that can grow flowers and produce seeds for many years. Predominant species from this genus are native to Asia, in

which cultivars and hybrids are widely grown for its large, showy, and fragrance flowers. In this study, Damask rose flowers (*Rosa x damascena*) were selected for observations (Figure 2). It is a rose hybrid derived from *Rosa gallica*, *R. moschata* and *R. fedtschenskoana* (Huxley, 1992; Harkness, 2003). The flowers ranges from light to moderate pink to light red in colour.



Figure 2: The pink petal of Damask rose (*Rosa x damascena*) flower selected for observations of the flowervisiting insects. In the photo with the arrows indicating the reproductive organs of the flower namely anthers (A) and stigma (B)

Observations of the Flower-visiting Insects

In this study, determination of flower stage was conducted following Ranu et al. (2009) for Rosa spp. (Figure 3). A total of 20 flowers at stage 1 from 4 plants (five flowers each plant) were selected and marked for further observations. When the flower reached stage 4 (at progressive stages of opening), a unit of 4K Ultra HD (16 mega pixels) sports action camera (1920 x 1080p video capture resolution) was installed in facing position to the flower. The camera was set up at a distance of approximately 0.5 m to observe the flower-visiting insects. The camera was set to record the insect visitors starting from 0700 hr until 1800 hr every day. Observations of the flowers were ceased once the petals start to fall off indicating its end of anthesis phase. In total, the recordings were conducted for a total of 744 hours.

The video footage was then transferred into a computer for reviewing using Windows Media Player application. For each hour however, observations of the visitors were conducted for only the first 15 minutes. The times of observations on insect pollinators through digital video recordings varied with goals and approaches (Gilpin et al., 2017; Steen, 2017; Bonelli et al., 2020; Pegoraro et al., 2020). In the present study, the footage from only the first 15 minutes of every hour was analysed to ease detailed observations. Thus, overall observations of the visitors conducted in this study were 186 hours. From the video footages, the identity of the visitors, the visitation frequency, the type of visits (either legitimate or illegitimate), and the time spent at the flowers were recorded. The image of each visitor was used for its identification to the lowest taxonomic level possible following McGavin (2002) and Gullan and Cranston (2014). A visit was counted when the visitor was in contact with the flower, until it flies away. The visit was considered legitimate only if the visitor contacting the anthers or stigma of the flower (Inouye, 1980). The time spent at the flower by each visitor was recorded from the time stamp of the video footage.



Figure 3: The flower stage for *Rosa* spp. as described by Ranu *et al.* (2009). Stage 1 is the flower bud, stage 2 is the partially opened bud, stage 3 is the complete opened bud, stage 4-6 are the flower at progressive stage of opening, stage 7 is the opened flower and stage 8 is the full opened flower in which after this stage the petals start to wilted and fall off

Data Analysis

As the data was not normally distributed, the Kruskal-Wallis test was used to compare the mean visitation rate (visit/hour) and time spent (in minute) between the visitors with more than a single visit. Other than that, Mann-Whitney test was used to compare the visitation rate (visit/hour) between the two visitation types (legitimate and illegitimate visits), conducted only for visitors which showed both visitation types. Data was analysed using IBM Statistical Package for Social Science (SPSS) version 26.

Results and Discussion

Flower Visitors of Rosa x damascene.

In this study, a total of 6 insect taxa were recorded visiting the rose flowers (Figure 4). Four of the insect taxa were from the order Hymenoptera, while a taxon was recorded for the order Lepidoptera and Diptera each (Table 1). From the 6 taxa, a single taxon from the order Hymenoptera and Diptera each were identified to Family and Order level only. Hence, named as Halictidae 1 and Diptera 1 respectively. A total of 253 visitations were observed by these visitors. Between the 6 taxa, Halictidae 1 recorded the highest visitation rate with 0.758 visit/hour, while the small branded swift (Pelopidas mathias) recorded only a single visitation to the flowers. For the 5 taxa with more than a single visitation, the Kruskal-Wallis test showed significant difference in the visitation rate between the flower visitors (H = 14.887, df = 4, p < 0.01). Multiple comparison further indicated that Halictidae 1 was with the highest visitation rate, followed by Heterotrigona itama (Malaysian stingless bees). Meanwhile, the other 3 taxa were with the lowest visitation rates (Figure 5).



Figure 4: The flower-visiting insects of *Rosa x damascena* flowers in Jasin, Melaka.
(A) Halictidae 1, (B) *Heterotrigona itama*, (C) *Amegilla* sp., (D) *Xylocopa confusa*, (E) *Pelopidas mathias*, (F) Diptera 1

Table 1: The number of visitations recorded by each flower visitors of Rosa x damascena in Jasin, Melaka

Order	Family	Taxa	Vernacular Name	Total Visitation	Visitation Rate (visit/hour)
Hymenoptera	Halictidae	Halictidae 1 Halictid bee		141	0.758
	Apidae	Heterotrigona itama	Malaysian stingless bee	77	0.414
		Amegilla sp.	Blue-banded bee	23	0.124
		Xylocopa confusa	Yellow carpenter bee	5	0.003
Lepidoptera	Hesperidae	Pelopidas mathias	Small branded swift	1	0.001
Diptera	-	Diptera 1	Fly	6	0.003
			Total	253	1.360



Figure 5: The boxplot of visitation rate (visit/hour) for the five flower-visiting insects with more than a single visit to *Rosa x damascena* flowers in Jasin, Melaka. Hs = Halictidae 1., Hi = *Heterotrigona itama*, As = *Amegilla* sp., Xc = *Xylocopa confusa*, Ds = Diptera 1. The box represents inter-quartile range, the horizontal line indicates the median value, while the whiskers outside the box represent the minimum and maximum values. Same small letter indicates no significant difference in visitation rate between the taxa from multiple comparison (step-down method) following significant result of the Kruskal-Wallis test (*p*<0.05)

Universiti Malaysia Terengganu Journal of Undergraduate Research Volume 5 Number 2, April 2023: 18-27

The hymenopteran bees were the most common visitors observed to the flowers of Rosa x damascena, with a total of 4 taxa, and with a cumulative of 98% of the total visitations recorded to the flowers in this study. The interaction between the bees and flowers is widely known, in which bees rely heavily on both pollen and flower nectar as food source, both during their adult and larvae stage (Willmer, 2011). In agricultural areas, this interaction is economically important, in which the hymenopteran bees is one of the major pollinator groups for the agricultural crops other than the dipteran flies (Wojcik, 2021). Although urbanisation generally resulted in biodioversity lost including in pollinator decline (Koh et al., 2016), the hymenopteran bees nevertheless found to thrive in the urban settings, hence become crucial pollinators in the area (Theodorou et al., 2020).

Between the hymenopteran bees, Halictidae 1 was found to register the highest visitations to the flowers, followed by *H. itama* and *Amegilla* sp. A study in the United States of America on *Rosa multiflora* (Multiflora rose) indicated the Syrphid flies (Order Diptera) as its common flower-visiting insects (Jesse *et al.*, 2006). This dipteran flies, together with the hymenopteran bees; the Western honey bee (Apis mellifera) and bumblebees (Bombus spp.) were reported as pollinators of this rose species elsewhere (Stougaard, 1983; Lee et al., 1995). The only dipteran fly recorded in present study (Diptera 1) however, registered comparatively low visitation rate (0.003 visit/hour) as compared to the hymenopteran bees. The lepidopteran taxon, P. mathias on the other hand, was the only taxon which recorded a single visit. Butterflies are known to prefer the flowers with red, yellow, blue, and purple in colour, than the white and pink flowers, with tubular shaped as compared to the open flowers (Tiple et al., 1984) such as the pink rose observed.

Visitation Behaviour of the Flower-visiting Insects

From the total of 6 insect taxa recorded, only 3 taxa showed both legitimate and illegitimate visitations to the flowers, while the other three taxa recorded only the illegitimate visits (Table 2). Between these three species, only Halictidae 1 showed significant visitation rate (Mean \pm SE) between the legitimate (0.52 \pm 0.19) and illegitimate (0.24 \pm 0.03) visits (Mann-Whitney test, U = 0.500, p < 0.05).

Table 2: The mean $(\pm SE)$ visitation rate (visit/hour) of the flower-visiting insects to *Rosa x damascena* flowers according to the visitation type. N = total visitations. NC = not calculated

		Type of visits			
Taxa	Ν	Legitimate (Mean ± SE)	Illegitimate (Mean ± SE)	Mann-Whitney Test, U	
Halictidae 1	141	0.52 ± 0.10	0.24 ± 0.03	U = 0.500, p = 0.029	
Heterotrigona itama	77	0.20 ± 0.04	0.22 ± 0.03	U = 8.000, p = 1.000	
Amegilla sp.	23	0.04 ± 0.01	0.08 ± 0.03	U = 12.500, p = 0.200	
Xylocopa confusa	5	0	0.03 ± 0.02	NC	
Pelopidas mathias	1	0	NC	NC	
Diptera 1	6	0	0.03 ± 0.0	NC	

Only 3 taxa from the order Hymenoptera showed the legitimate visitation (Halictidae 1, *H. itama*, and *Amegilla* sp.), while the other taxa showed only the illegitimate visits. *Heterotrigona itama* is one of the eusocial insects that live in perennial colonies (Amano *et al.*, 2000). The workers collect pollen, nectar, and propolis for the queen for reproduction. As compared to the higher legitimate than illegitimate visitations by Halictidae 1 while

foraging at the flowers, high visitation frequency showed by *Amegilla* sp. and *H. itama* in contrast consists of equal legitimate and illegitimate visitations. As compared to honeybees, the stingless bees in general collecting more pollen grains while foraging at the flowers as compared to nectar, an inverse pattern showed by the honeybees (Heard, 1994). The stingless bees nevertheless were reported to supplement the pollination activities by the honeybees, indicating their importance as pollinators (Layek *et al.*, 2022) for the plant they forage at. For the time spent at the flowers, Halictidae 1 showed the longest time (Mean \pm SE) with 30.08 ± 4.87 minutes, while *P. mathias* recorded the shortest time spent at the flower with 2.16 minutes from the only visit recorded by this insect. The Kruskal-Wallis test conducted showed a significant difference in time spent between the other five flower-visiting insects to *Rosa x damascena* (H = 13.78, df = 4, p < 0.01). Multiple comparison further indicated that Halactidae 1 showed the longest time spent, followed by *H. itama*, while the other 3 taxa showed the shortest time spent at the flowers (Figure 6).



Figure 6: The boxplot of time spent (in minute) between the flower-visiting insect of *Rosa x damascena* in Jasin, Melaka. Hs = Halictidae 1, Hi = *Heterotrigona itama*, As = *Amegilla* sp., Xc = *Xylocopa confusa*, Ds = Diptera 1. The box represents inter-quartile range, the horizontal line indicates the median value, while the whiskers outside the box represent the minimum and maximum values. Same small letter indicates no significant difference in visitation rate between the taxa from multiple comparison (step-down method) following significant result of the Kruskal-Wallis test (p < 0.05)

Insect pollinators are known to prefer to visit plants with particular floral traits (Frund *et al.*, 2010). In Rosacea family for example, flowers horticulturally-modified varieties of the genus *Prunus* and *Rosa* with double petals of clustered flowers with showy sepals found to attract less bees as compared to the varieties with more accessible nectar and pollen (Mach & Potter, 2018). The wide landing area (petals)

in *Rosa* spp. flowers might allows the flowervisiting insects to land on the flowers, collecting the nectar at the base of the petals without contacting the anthers and stigma (illegitimate visits). This behaviour was reported for the small bodies' bee, as compared to their large size counterparts (Tangmitcharoen *et al.*, 2006). The difference in time spent between the flowervisiting insects could be due to the depleted

OBSERVATION ON THE FLOWER-VISITING INSECTS OF ROSE (ROSA X DAMASCENA) IN AN AGRICULTURAL LANDSCAPE

pollen grains in the already exploited flowers (Silva *et al.*, 2013). In present study, Halictidae 1 and *H. itama* were found to spent longer time as compared to other visitors, as these 2 insect taxa were often observed foraging at the flowers in the early morning, much earlier than the other insect visitors.

Conclusion

In total, 6 insect taxa were recorded visiting the flower of Rosa x damascena in Jasin, Melaka, which consists of four hymenopteran bees, and a single taxon for each dipteran (fly) and lepodipteran (butterfly) insects. From these, three hymenopteran taxa were determined as the most common visitors to the flowers, which were Halictidae 1, H. itama and Amegilla sp. These 3 insect taxa were found commonly contacting the reproductive part of the flowers during their visits to the flowers, as well as spending long duration to forage at the flowers. Hence, these 3 insects showed a high potential as pollinators of Rosa x damascena. Furthermore, Halictidae 1 was with the highest potential as pollinator of Rosa x damascena, followed by H. itama and Amegilla sp. However, no further observation was made to confirm the pollination success following visitations by these flower-visiting insects to the observed flowers. Furthermore, observations on subsequent visits by the same flower-visiting insects is useful to determine their role as pollinators of the flowers they visited. This is due to repeated visitations to the same flowers by the insects might resulted in self-pollination to occur on the flowers, thus reducing the pollinator effectiveness.

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