Potency of Salicylic Acid to Disrupt the Growth and Development of Papaya Mealybug, *Paracoccus marginatus* (Hemiptera: Pseudococcidae)

**Liza Octriana**1*, Nugroho Susetya Putra2, & Suputa2**

1) Research Institute for Tropical Fruit Plants
Jln. Raya Solok-Aripian Km.8, Solok-West Sumatera 27301 Indonesia

2) Department of Plant Protection, Faculty of Agriculture, Universitas Gadjah Mada
Jln. Flora I, Bulaksumur, Sleman, Yogyakarta 55281 Indonesia

*Corresponding author. E-mail: lizaoctriana@gmail.com

**ABSTRACT**

Mealybug is an important pest of papaya plants. Induction of plant resistance using elicitors, such as salicylic acid, might have the potency to reduce the extent of crop damage by mealybug. Therefore, a laboratory experiment was performed to determine the effect of salicylic acid on feeding preference, fecundity, oviposition period, and longevity of papaya mealybug adult, *Paracoccus marginatus*. The results showed that the application of salicylic acid increased total phenol content on papaya leaf ($r = 0.57$) hence decreased in feeding preferences and fecundity, slowed down the growth period of the nymph and pre-oviposition period, and prolonged the longevity of mealybug. The potency of using salicylic acid to control of mealybug on papaya in integrated pest management was discussed in this paper.

**Keywords:** mealybug papaya, *Paracoccus marginatus*, salicylic acid

**INTRODUCTION**

Mealybug, *Paracoccus marginatus*, is an important pest of papaya plants (Meyerdirk et al., 2004; Muniappan et al., 2008; Mastoi et al., 2011; Suartini et al., 2015). The attack of mealybug causes disruption of photosynthesis and fruit fall before ripe, thus potentially reduce plant productivity. The control technique using pesticides is relatively high, less effective, and unfriendly environmentally (Maryono & Irham, 2001; Isenring, 2010; Tanwar et al., 2010; Krishnan et al., 2016). Elicitors compound, e.g. salicylic acid, is an alternative control technique to reduce the plant damage caused by pests (Thaler et al., 2002; Qiu et al., 2009; San Vicente & Placensia, 2011; Santamaria et al., 2013).

Salicylic acid is an elicitor inducing plant resistance to many pathogens (Chen et al., 1995; Suganda, 2001; Thaler et al., 2002; Martanto et al., 2003; Suryanti et al., 2009, Sujatmiko et al., 2012; Hoerussalam et al., 2013; Thakur & Sohal, 2013). Previous studies showed that spraying salicylic acid enhanced plant resistance to sucking pests, e.g. to control population of *Nezara viridula*, *Myzus persicae*, *Empoasca lycica*, and *Tetranychus urtica* (Thaler et al., 2010; Farouk & Osman, 2011; Mahmoud, 2013; Michael et al., 2013; Afifi et al., 2015; Elhamahmy et al., 2016; Rana et al., 2016), and to decrease the fecundity and the longevity of *Bemisia tabaci* (Shi et al., 2016). However, the effect of salicylic acid on sucking pest is inconsistent. For example, Zhang et al. (2011) and Zhang et al. (2015) reported that the application of salicylic acid to *Phenacoccus solenopsis* was not reduced the growth rate of nymphs on cotton and tomato plants.

The study about the use of salicylic acid to induce the resistance of papaya plants to *P. marginatus* is needed to be done. Therefore, this study aimed to determine the effect of salicylic acid on the growth and development of *P. marginatus*. The parameters used were the feeding preference of nymph, adult fecundity, the longevity of each instar of nymphs, pre-oviposition period, oviposition period, and adult longevity. The results of this study highly expected to provide information about the use of salicylic acid to control *P. marginatus*.

**MATERIALS AND METHODS**

The study was conducted at the green house and Laboratory of Plant Pest Sciences, Faculty of Agriculture, Universitas Gadjah Mada, Yogyakarta in November 2016 to April 2017 with a daily temperature of 26–30°C, and relative humidity of 44–60%. The study used a Randomized Complete Block Design (RCBD) with
by cutting the 2 upper leaves of the papaya plant treated in the 2nd week after the application of salicylic acid. Each leaf was placed in a glass bottle (5 ml in volume) filled with water, and the stem was covered with moist cotton to keep the vigor of the leaves. Five newly hatched nymphs were fed by papaya leaves. The longevity of nymphs until adults were observed daily. Instar of the nymphs was characterized by the presence of white exuvia. When the adults emerged until an egg sac formed was recorded as the preoviposition period. The oviposition period was calculated from the formation of the egg sac until the 1st nymphs hatched. The development of nymphs and adult was observed using a binocular microscope.

**Analysis of Leaf Phenol Content**

Analysis of leaf phenol content was carried out using the method by Senter et al. (1989) and conducted at the Laboratory of Chemistry and Biochemistry of Food and Agricultural Products, Department of Food and Agricultural Products Technology, Faculty of Agricultural Technology, Universitas Gadjah Mada, Yogyakarta. One ml of papaya leaf extract was diluted to 100 ml, then 1 ml of the dilution was taken and diluted again with 10 ml hence the total dilution became 1000× (df = 1000×). One ml of dilution was added with 5 ml of alkaline Na₂CO₃ 2%, and incubated for 10 minutes, then added 0.5 ml of Folin Ciocalteau reagent, vortexed, and incubated for 30 minutes. Absorptions were measured at a wavelength of 750 nm. The phenolic concentration was calculated based on the standard curve obtained from pure phenol solution using the following formula:

\[
\text{% Phenol} = \frac{x \cdot df \cdot 100\%}{\text{sample (mg)}}
\]

Note: df = dilution factor

Each treatment concentration has for replications. Leaf samples for analysis were harvested on the 12th day after application of salicylic acid.

**Data Analysis**

The effect of treatments on fecundity, the longevity of the newly hatched nymphs, 2nd instar, 3rd instar, preoviposition period, oviposition period, the longevity of adults and leaf phenol levels were tested by Variance Analysis (ANOVA). The post-hoc analysis using the LSD test was carried out at 5% when there
Salicylic acid in papaya plants affects the biology of P. margaratus and increased the leaf phenol level thus reduce the feeding preference of 3rd instar nymphs. Although salicylic acid prolonged the longevity of the 2nd and 3rd instar nymphs, shorten the longevity of the 1st instar nymph was shorter. Therefore, the use of salicylic acid to papaya plants has the potential to reduce P. marginatus population. The results also showed that papaya leaves treated with salicylic acid had higher phenol level than untreated leaves (Table 1; P < 0.05). Nevertheless, the leaf phenol level was not significantly different from the concentrations of salicylic acid 50, 100 and 200 mg/l.

Salicylic acid, o-hydroxybenzoic acid, is a phenolic compound naturally exists in plants in low concentrations (Kawano et al., 2004, Janda et al., 2014, War et al., 2015). The function of salicylic acid is a phytohormone and has a role in physiological processes, for example in the management of resistance to pests. The synthesis of salicylic acid in plants is a complex mechanism. Plants synthesize salicylic acid from cinnamic acid produced by phenylalanine ammonia lyase with benzoic acid as an intermediate precursor, which takes place in the cytosol. Salicylic acid is also synthesized in chloroplasts from isochorismate acid produced from chorismic acid (Kawano et al., 2004, Hayat et al., 2010). Previous study stated that salicylic acid has a role in the plant resistance to diseases (Dempsey & Klessig, 2017). Lennon et al. (1997) also reported that salicylic acid is an inducer to increase the resistance of tobacco plants to Tobacco Mosaic Virus (TMV) through an increase in alternative oxidase proteins. Furthermore, War et al. (2012) stated that salicylic acid has a role in plant resistance to herbivorous insects, both piercing-chewing and biting-sucking pests.

This study showed that salicylic acid was applied to papaya increased leaf phenol levels hence decreased feeding preference. The test showed that the number of 3rd instar nymphs fed on papaya leaves exposed to salicylic acid was smaller than those fed on untreated leaves (controls) (Figure 1). Other studies by War et al. (2011), Farouk & Osman (2011), and Damodaram et al. (2015) also stated that the application of salicylic acid can increase phenol levels. In the other hand, the study of Elhamahmy et al. (2016) showed that the use of salicylic acid did not increase phenol levels in canola leaves (Brassica napus). In general, the role of phenol compounds in developing plant resistance to herbivorous insects has also been studied in several types of herbivorous insects.

Other studies showed that salicylic acid is a deterrent for aphids and B. tabaci (Mahmoud & Mahfouz, 2015; Shi et al., 2016), hence those population in plants was lower than in control treatments. These studies proved that salicylic acid inhibited the feeding preference of piercing-sucking pests. Another study by Ollerstam and Larsson (2003) showed that salicylic acid has an important role in the resistance of Salix viminalis to gall midge (Dasineura marginetorumquens). However, Sekido and Sogawas (1976) reported that salicylic acid is a feeding stimulant on Nilaparvata lugens, but it reduced the feeding preference of Laodelphax striatellus.

The decrease in feeding preferences by mealybug on papaya leaves treated with salicylic acid may effect the growth and development of the longevity of nymphs and adults. The results showed that the use of salicylic acid prolonged the longevity of 2nd and 3rd instar nymphs, although shorten the longevity of 1st instar nymph (Table 2). In use to that, the exposure of salicylic acid had an effect on preoviposition and longevity of adults. In general, the application of salicylic acid in higher concentrations tends to prolong the preoviposition period and the longevity of adults. Meanwhile, the use of salicylic acid did not affect the oviposition period (Table 3; P > 0.05).

<table>
<thead>
<tr>
<th>Concentration (mg/l)</th>
<th>Phenol (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>0.320 ± 0.04b</td>
</tr>
<tr>
<td>50</td>
<td>0.385 ± 0.03a</td>
</tr>
<tr>
<td>100</td>
<td>0.345 ± 0.01ab</td>
</tr>
<tr>
<td>200</td>
<td>0.381 ± 0.04a</td>
</tr>
</tbody>
</table>

Remarks: Means followed by the same letter were not significantly different according to BNT test (p = 0.05).
Table 4 showed that the use of salicylic acid to papaya leaves decreased fecundity of *P. marginatus* compared to controls (*P* < 0.05). Nonetheless, the concentration of 50, 100, and 200 mg/l of salicylic acid to papaya leaves did not significantly affect the fecundity. Damodaram *et al.* (2015) reported that the application of salicylic acid increased plant phenol levels, thereby reducing the preference of *Bactrocera dorsalis* for laying eggs. Meanwhile, the application of elicitor compounds, such as salicylic acid, increased the levels of toxic compounds, e.g. phenol, peroxidase enzymes, polyphenol oxidase, and hydrogen peroxide (War *et al.*, 2011; Damodaram *et al.*, 2015), which potentially decrease the growth of larvae or nymphs, and fecundity.

The exposure of salicylic acid induces chemical compounds that affect the chemical and mechanical characteristics of plant tissue to insect growth (Cipollini *et al.*, 2004; War *et al.*, 2013). Phenol is a signaling molecule (precursor) to activate the resistance genes of plants to pests and diseases (War *et al.*, 2012). Oxidation of phenol compounds is catalyzed by polyphenol oxidase and peroxidase produce quinone which is toxic to insects or inhibits insect appetite (War *et al.*, 2012; Shoorooei *et al.*, 2013), thus effecting insect growth and development.
This study showed the potency of an elicitor compound, salicylic acid, to manage herbivorous insect populations by affecting the chemical or mechanico-physic characteristics of plants (War et al., 2013; Damodaram et al., 2015). Nevertheless, several studies also explained that the production of elicitor compounds (e.g. phenol) for plant resistance was influenced by abiotic factors, such as light and soil nutrients (Dudt & Shure, 1994). In the other study, plants grow in different types of soil will have different phenol content (Doyle et al., 1978). Therefore, the plants would generally contain more defense compounds in soils with lower nutrient.

In this study, observations of the growth and development of *Paracoccus marginatus* were employed on leaves was not picked on fresh plants. This might affect the growth of *P. marginatus*. However, because all treatments, both controls and those applied by salicylic acid were carried out by the same method, the result of the effect of salicylic acid on the growth and development of *P. marginatus* was considered reliable. Moreover, this study will give additional information about pest management using a resistance induction technique. The use of inducing compounds, such as salicylic acid, has the potency to reduce the growth and development of *P. marginatus*. However, the concentration of elicitor compounds induced to plants and the influence of external factors, such as temperature, light, and humidity, is important to be considered.

**CONCLUSION**

The use of salicylic acid potentially can control the population of *P. marginatus*, through a mechanism of emphasis on feeding preference of nymphs and decreased fecundity of *P. marginatus*.

**ACKNOWLEDGEMENTS**

We thank the Agricultural Research and Development Institute for funding this research. This paper is a part of the thesis for graduating from the Department of Plant Pest Science.

**LITERATURE CITED**


---

**Table 4. Effect of salicylic acid on fecundity of *Paracoccus marginatus***

<table>
<thead>
<tr>
<th>Concentration (mg/l)</th>
<th>Fecundity</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>236.32 ± 24.63a</td>
</tr>
<tr>
<td>50</td>
<td>151.50 ± 12.48b</td>
</tr>
<tr>
<td>100</td>
<td>112.97 ± 8.02b</td>
</tr>
<tr>
<td>200</td>
<td>151.13 ± 16.03b</td>
</tr>
</tbody>
</table>

Remarks: Means followed by the same letter were not significantly different according to BNT test (p = 0.05)


