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Sub lethal and Lethal Effects of Crude Leaves Extract of Gamal (*Gliricidia maculata*) on Red Ants (*Solenopsis* sp.)

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Abstract: *Our previous studies showed that crude water extract of gamal (Gliricidia maculata) leaves powder contains flavonoids that are toxic to mealybugs. Our investigation shows that one of the mutualistic symbionts of mealybugs is red ants (Solenopsis sp.). This study is intended to determine whether gamal extract is also toxic to the symbionic ants of the mealybugs. Gamal leaves extract used in this study is crude water extract at the concentration of 0.11%, the LC50 of the extract on mealybug. For comparison, we use a synthetic insecticide, Regent 50 SC at a concentration of 0,1/200ml as positive control and distilled water as normal control. Two experiments were conducted, sub-lethal and lethal test. For sub-lethal test 30 individuals of red ants were grouped into three (10 ants each). The three groups of ants were exposed to rice that has mixed with distilled water, gamal extract, and synthetic insecticide respectively. The relative distance and position of ants to food was observed every 30 minutes for 12 hours. For mortality test, red ants (n=30) were also grouped into three (10 ants each). The three groups of ants were successively sprayed with distilled water, gamal extract, and synthetic insecticide respectively. Sub-lethal test results showed that gamal leaves extract showed anti-feedant effect on red ants. Mortality test results showed that gamal leaves extract has a lethal effect similar to the synthetic insecticide.*

Keywords: *gamal, Gliricidia maculata, mealybug, red ant, Solenopsis sp.*

INTRODUCTION

Gamal (*Gliricidia maculata*) can be used as an insecticide. Gamal leaves are known to contain flavonoid compounds are effective for controlling sucking insects caterpillars and fruit (Sudarmo, 2005). Flavonoids are secondary metabolites of the compounds derived from green plants with structure polyphenols (Tapas, et al., 2008).

Based on the research results Nukmal et al. (2017) known to thin-layer chromatography analysis of water and methanol extract of leaves of gamal produces a row and seven of the flavonoid fraction. Both types of extract affects the level of insect mortality by level of concentration, but based on the value of LC_{50} and LT_{50} water fraction showed higher efficacy than methanol.

Couples research results (2018) Unknown rough water extract powder KLU Gliricidia leaves (cultivar North Lampung) is more effective in shutting down *Planococcus minor* pest on cocoa crops than pure extract water, because the crude extract water Gliricidia leaf powder has a value smaller than the clock LC_{50} pure water leaf powder extract gliricidia (0,11%: 0,27%). In addition, according to Nukmal et al. (2019) leaf powder extract water crude gamal of four cultivars LU (North Lampung), PW (Pringsewu), LB (Lampung Barat) and BL

(Bandar Lampung) can be shut off with a papaya mealybug pest LC_{50} consecutive hours 0,033 %, 0,090%, 0,184% and 1,818%. gamal leaves crude extract powder known exhibits a nematicide, an antimicrobial and larvicidal activity in mosquitoes (Krishnaveni, et al., 2015; Nazli, et al., 2011).
Fitrisia research results (2017) note that there is a polar extract effect Gliricidia leaves against a symbiotic ants with mealybug on coffee plants, cocoa and soursop ($p < 0{,}01$). The average mortality ant synthetic insecticide treated more than 1,07 to 1,42 times and 7.55 to 11.07 times the botanical insecticides when compared to controls.
Based on the above background research on the effects of crude extract water gamal leaf powder as an insecticide plant have been carried out. However, to nontarget organisms such as ants still little is researched. Therefore, this study was conducted to determine the effect of crude extract water gamal leaf powder to red ant (*Solenopsis* sp.).

Material and Methods

Research conducted at the Laboratory of Zoology II, Department of Biology, Faculty of Mathematics and Natural Sciences, University of Lampung, making ants from Kedaton, Bandar Lampung. The study was conducted in December 2018 until January 2019.
The tools used are fabric tricot to cover the top of the jar and the Petri dish, rubber bands to bind the fabric tricot, jam jars and the petri dish as the test insects treated, aluminum foil place put food (rice), markers to give limit on a petri dish, syringes for spraying insecticides on ant when observed mortality, a pipette to drip feed ant insecticide at the time of behavioral observation, a measuring cup to measure the volume of distilled water and synthetic insecticides, as well as stationery to record observations. While the materials used include red ant (*Solenopsis* sp.) As the test insects, water crude extract powder Gliricidia leaves North Lampung cultivars with LC_{50}= 0.11% were obtained from previous studies,
This research was a randomized block design (RAK) in two ways, namely mortality and behavioral observations of red ants. In observation mortality red ants, the first treatment without spraying botanical and synthetic insecticides (control). The second treatment with insecticide spraying water crude extract powder gamal leaves that have the potential to kill the pest mealybugs with LC_{50} = 0,11% and the third treatment with a synthetic insecticide spraying (Regent 50 SC) according to the recommended dose (0,1 ml / 200 ml distilled water), then do three repetitions. Red ant mortality was observed at 1, 3, 6, 12, 24 and 48 hours after treatment. Ant experiencing mortality data were analyzed using the Anara and a further test BNT level of 5% SPSS version 15.0 when there are differences among the treatments. While on the red ants behavioral observations, the first treatment without feeding on ants (control). Then the second treatment to feed rice without insecticide compound, the third treatment to feed rice mixed with botanical insecticide powder in the form of crude extract water Gliricidia leaves that have the potential to kill the pest mealybugs with LC_{50} = 0,11% and the fourth treatment with feed and rice mixed with synthetic insecticides (Regent 50 SC) in accordance with the recommended dose (0,1 ml / 200 ml of distilled water). Fourth treatment do ten repetitions. The effect of treatment on test symbiotic ants with mealybug observed during the 12 hours from 08:00 am - 19:00 pm every 30 minutes at each time of observation. Red ant behavior change that approach and stay away from food were analyzed descriptively.

Results and Discussion

On the results of analysis of variance (Table 1) is known that treatment without spraying insecticides (control), insecticide spraying botanical and synthetic insecticides, the interaction between the time of observation and treatment, as well as the observation time effect on the mortality of red ants (*Solenopsis* sp.) To the cocoa crop $p \leq 0{,}05$ and groups were not significantly different ($p = 0{,}964$).

Table 1. Results of analysis of variance mortality red ant (*Solenopsis* sp.) in the treatment and observation time is different

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	780,630(a)	19	41,086	80,764	,000
Intercept	474,074	1	474,074	931,906	,000
Group	,037	2	,019	,036	,964
Treatment	301,037	2	150,519	295,880	,000
Time	299,704	5	59,941	117,828	,000
Treatment * Time	179,852	10	17,985	35,354	,000
Error	17,296	34	,509		
Total	1272,000	54			
Corrected Total	797,926	53			

Note: *) The interaction of treatment and Time

Average mortality of red ants were given three treatments (Table 2) were significantly different mortality is indicated by red ants on synthetic insecticides to 1,86 times (5,78: 3,11) more than the ants by insecticide treatment. This is because the toxic compounds contained in synthetic insecticides to work faster than the toxic compounds contained in plant-based insecticide. This opinion is supported by Hendayana (2006) working power plant insecticide slower compared to synthetic insecticides and can not directly kill the target's body.

Table 2. The LSD influence the treatment of the three red ants (*Solenopsis* sp.)

Treatment	Mortality Ants (Average± SD)
Non Treatment	0,00 ± 0,000[a]
Spraying Botanical Insecticide	3,11 ± 3,479[b]
Spraying Synthetic Insecticide	5,78 ± 4,138[c]

Description: The average value followed by the same lowercase not significantly different at the level of α = 5%

On the results of further test BNT influence of observation time is different to the average mortality of red ants in Table 3, discovered that the average mortality of red ants at 1-3 hours after treatment has yet to show a real difference, and this is because the mortality rate of red ants still low. Meanwhile, on the observation of 6-48 hours after treatment was significantly different with increasing time. This happens because the longer the exposure time, the more insecticides toxic compounds that enter into the ant's body. This opinion is supported by Lu (1994), which states that if a lot of toxic substances into the body and undergo biotransformation will cause a lot of energy is needed to neutralize the toxic compound causing insects run out of energy that can stop the process of metabolism in the body. In addition, the toxic compounds that work in the ant's body lead to weak nervous system and inhibit the mechanisms of energy in the mitochondria that disrupts the electron transport system and causes the ants are not able to perform activities including communications (Raymond et al., 2005; Muta'ali, et al., 2015).

Table 3. The LSD influence of different observation times against average mortality of red ants (*Solenopsis* sp.)

Time (Hours)	Mortality Ants (Average± SD)
1 hour	0,11± 0,333[a]

3 hours	0,33± 0,707[a]
6 Hours	1,89± 2,472[b]
12 hours	3,89± 3,723[c]
24 hours	5,33± 4,387[d]
48 Hours	6,22± 4,738[e]

Description: The average value followed by the same lowercase not significantly different at the level of α = 5%

On the results of LSD test the interaction effect of treatment and observation time is different to the average mortality of red ants (Table 4), it is known that in 1-3 hours of observation have not shown a real difference is due to insecticide exposure time on the red ants are still short. At 6 hours after treatment, the average mortality ant insecticide treated significantly different synthetic insecticide treatment and non treatmen. However, the average mortality by botanical insecticide treatments were not significantly different with the non treatmen, this situation caused the death rate of ants in botanical insecticide is still low, and due to the power toxic synthetic insecticides faster work than botanical insecticide.

The average mortality of red ants at 12-48 hours after being given treatment (Table 4), it is known that treatment with synthetic insecticides significantly different with botanical insecticides and non treatmen. This situation is caused by the compound on a synthetic insecticide fipronil as a contact poison that is quicker to react in the body of an ant compared flavonoid compounds derived from plant-based insecticide. This opinion is supported by Djojosumarto (2008) insecticide that works as a contact poison will kill insects when exposed directly to the body parts of insects and insecticides into the insect's body through the skin (cuticle) will be translocated into the insect body parts where the insecticide active work. Also according to zhao et al.

Table 4. Hasi LSD treatment interaction effect and time of observation different to the average ± SD mortality red ants (*Solenopsis* sp.)

Time (Hours)	Non Treatmen	Spraying Insecticide Plant	Spraying Insecticide Synthetic
1	0,00 ± 0,000[a]	0,00 ± 0,000[a]	0,33 ± 0,577[a]
3	0,00 ± 0,000[a]	0,00 ± 0,000[a]	1,00 ± 1,000[a]
6	0,00 ± 0,000[a]	0,67 ± 1,155[a]	5,00 ± 1,000[c]
12	0,00 ± 0,000[a]	3,33 ± 1,528[b]	8,33 ± 0,577[d]
24	0,00 ± 0,000[a]	6,00 ± 1,000[c]	10,00 ± 0,000[e]
48	0,00 ± 0,000[a]	8,67 ± 1,155[d]	10,00 ± 0,000[e]

Description: The average value followed by the same small letters on the same line are not significantly different at the level α = 5%.

In Figure 1 unknown percentage of red ants at least approached the food on treatment with synthetic insecticides and most of the rice without insecticide treatment exceeds 85%. Meanwhile, if the comparison between the mixture of rice with botanical insecticides and synthetic insecticides, both types of ants more approaching food laced with insecticide compared to synthetic insecticides. This is due to the absence of insecticides that are toxic content in rice without insecticide treatment, causing the ants more closer to the food. As for the treatment of rice mixed with botanical insecticides are flavonoid compounds that are toxic and rice mixed with a synthetic insecticide fipronil-containing compounds are higher than the toxic level of flavonoid compounds in based insecticide. This opinion is supported by Petrosida Gresik (2016) which stated that the active ingredient fipronil contained in synthetic insecticides used are systemic work in contact and stomach and can affect the nervous system stimulation and causing organisms convulsions and can lead to death. Also according to Morello & Rejessus (1983); Prijono (1994) that the content of toxic compounds contained in insecticides is antifeedant and repellent which works as an insect repellent to eat by reduced insect appetite, caused insects to die of starvation and inhibit development of insects.

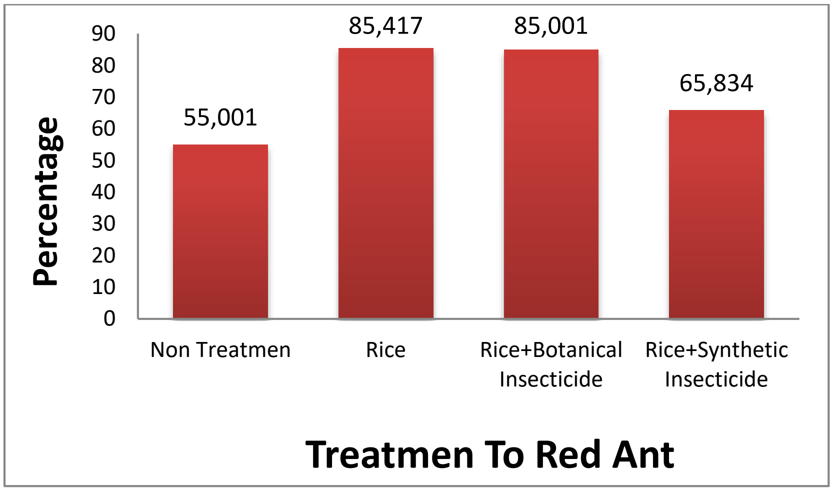

Figure 1. Percentage tendency red ant (*Solenopsis* sp.) approached the food.

Conclusion

The crude extract of gamal leaf powder (*Gliricidia maculata*) Cultivars North Lampung effect to mortality and behavioral approach and stay away from food on a red ant (*Solenopsis* sp.). However, the degree of influence of botanical insecticide not exceed the influence of synthetic insecticides.

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