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EFFECTIVENESS OF ENVIRONMENTALLY FRIENDLY MOSQUITO TRAP CONTAINED SUGAR YEAST SOLUTION

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Abstract

Environmentally friendly mosquitoes trap using common daily materials used by community may give hopes in reducing mosquitoes density. This study aims to determine the effectiveness of environmentally friendly mosquitoes trap made from bottle contained sugar yeast solution for reducing the number of trapped mosquitoes. This study consist of two phases and quasi experimental design was used. Mann Whitney test was used to determine the differences the number of trapped mosquitoes indoor and outdoor. The result showed *p* value 0,000 which was lower than α value (0,05), so there was significant difference between the number of indoor and outdoor trapped mosquitoes. The average Rank score of outdoor mosquitoes trap (42,75) was more than indoor mosquitoes trap (18,25). We concluded that the mosquitoes trapped which contained of sugar-yeast solution was effective for trapping the mosquitoes especially outdoor .

Introduction

Mosquitoes are vectors of many diseases and various effort to control adult mosquito have been done including the use of insecticides (Nuryanti, 2003). However, the active ingredients or synthetic chemical compounds that are used as insecticides likely to cause resistance. Based on this phenomenon, it is necessary to develop an environmentally friendly mosquito trap, sustainable, involve participation of the community and effectively reduce mosquito density.

This study used the principle of attractant, which is used materials that attract mosquitoes to come to the host, such as carbon dioxide (CO_2). The ability to detect chemical compounds released by host (particularly vertebrates) is one of the important mosquito behavior to find a host. CO_2 also play a role to increase mosquitoes interest toward odors released by humans (Smallegange, 2010).

To date, the source of CO_2 to attract mosquitoes came from dry ice, pressurized gas cylinder and propane-powered trap. However, these sources of CO_2 is not always easy to obtain. Dry ice is a valuable CO_2 producer which relatively inexpensive, but for daily use by the community as a source of CO_2 to attract mosquitoes into mosquitoes trap would be relatively expensive. The flow of the CO_2

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produced by the dry ice has a high variability and the longer its use will decrease the concentration of CO_2 produced. CO_2 generated by a propane powered mosquito trap does have the advantage in producing CO_2 with a steady stream, but this source is burdensome and expensive, therefore can't be used for mosquito trap used by the community in the long term as an effort to lower density of mosquitoes. Limitations of each source of CO_2 raise the idea of using other materials as a source of CO_2, which is relatively inexpensive, easy to make and the sustainability of its use as a mosquitoes attractant can be maintained for a long period.

The process of sugar fermentation produces bioethanol and CO_2, while sugar is a daily groceries being used by every household as a sweetener in food and beverages. The use of daily used materials by community is expected to improve the sustainability.

Based on this background, we want to test the effectiveness of mosquito trap made from used plastic bottles containing sugar-fermenting yeast in trapping mosquitoes. Preceded by determining the type of sugar solution that is most effective to be mosquito trap which by comparing the sugar-yeast solutions, brown sugar-yeast solution, and well water as a negative control. The research conducted in Pabean Pekalongan, north part of Pekalongan city, Central Java Province, which is an endemic area of *Filariasis bancrofti*. One risk factor of *Filariasis bancrofti* infection is high density of mosquito vectors, the large number bites of infective filariasis vector mosquito (Ramadhani, 2010).

Method

This study was a descriptive study using a quasi-experimental approach with post test only design. The study consisted of two phases, the first phase aimed determine the most effective sugar-yeast solution as an attractant and the second phase to determine how the study can be implemented in the community.

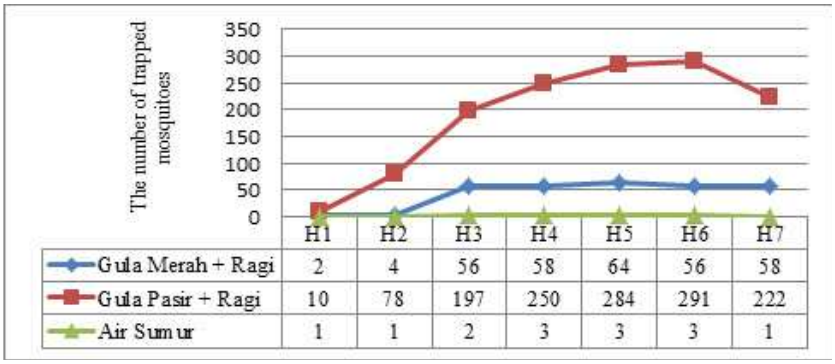
The first phase was mosquitoes traps placement in 30 houses, each house was assembled with 6 bottles of mosquito trap consist of 3 bottles inside the house and 3 bottles outside the house. Each bottle placement consisted of bottles contain sugar-yeast solution, brown sugar-yeast and well water as a negative control. Bottle of mosquito traps placed at every 5:00 pm and counted the number of mosquitoes trapped at 09.00 am until the number of mosquitoes trapped didn't increase.

The second phase was installing bottle of mosquito trap contained the most effective solution from the result of the first phase. Trap placement mosquitoes carried out since September 2015 until January 2016. Each house was fitted with 4 pieces of mosquito traps, 2 traps placed in the house and 2 traps placed outside the home. Placement of the traps in the house is in one of the rooms often occupied by residents of the house such as the living room and bedroom. Placement of the traps outside of the house is in the outside area of the walls of the house until out of the roof.

The steps to make mosquito trap were as follow: (1) plastic bottles of soft drinks packaging volume 1.5 L cut at the 1/3 upper part ; (2) 1/3 upper part of the bottles inserted into remained pieces of the bottle upside down ; (3) both pieces of the bottle sealed using tape; (4) the outside covered with black plastic. While, the steps to make mosquito attractant were as follow: (1) 50 g of sugar dissolved in to 200 ml of hot water; (2) let the solution until the temperature reach warm; (3) 1 g of yeast inserted into the bottle without stirring; (4) bottle's mouth closed using plastic for 24 hours; (5) then the plastic lid opened when a mosquito trap will be installed. At the time of the mosquito traps placed, around the bottle trap gave a circular line using chalk (lime ants) to prevents ants entering the bottle. If the contents of the bottle spilled then it will immediately replaced with fresh solution.

Mosquitoes trap installment started at 05:00 pm and the number of mosquitoes trapped counted at 09.00 am seven days later by the same person. Subsequently, sugar-yeast solution was replaced with a new sugar-yeast solution, and so on. Temperature, humidity and rainfall were recorded every week. The number of mosquitoes trapped counted by opening the black plastic covering the bottle and poured the contents of the bottle into a container equipped with a filter. Mosquitoes trapped counted one by one regardless the species and blood-sucking or not.

Figure 1. Total Mosquitoes Trapped During 7-Day Observation



	H1	H2	H3	H4	H5	H6	H7
Gula Merah + Ragi	2	4	56	58	64	56	58
Gula Pasir + Ragi	10	78	197	250	284	291	222
Air Sumur	1	1	2	3	3	3	1

Source : Primary Data

Results and Discussion

Result of Mann Whitney test showed the p value was 0.117, greater than α value 0.05, which means no differences were found between the number of mosquitoes trapped in brown sugar-yeast solution and sugar-yeast solution. However, there were significant differences in the number of mosquitoes caught in traps contained brown sugar-yeast solution and sugar-yeast solution compared to trap contained well water, which indicated with *p*-value of 0.00 (less than the α value 0.05).

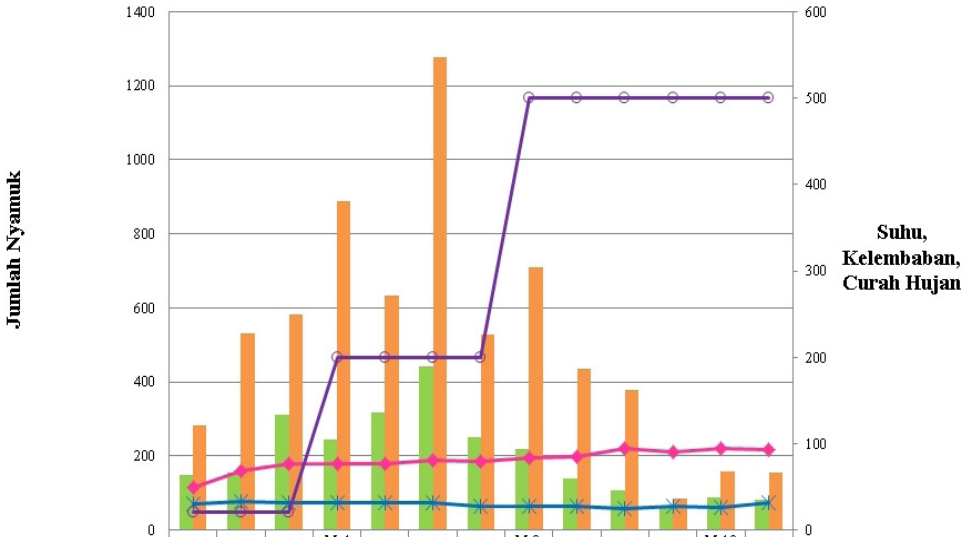
The results of this study indicated the number of mosquitoes caught in traps that contained brown sugar-yeast solution and sugar-yeast solution did not differ significantly (Figure. 1). In the process of fermentation, sugars serve as substrate. Coconout sugar, palm sugar or brown sugar, similar to sugar, have a carbohydrate called sucrose, a disaccharide which digested glucose and fructose.

Among the number of mosquitoes trapped, 25% were blood fed mosquitoes. The vertebrate blood-sucking mosquitoes groups need blood for their eggs maturation. According to the life cycle of mosquitoes in general, mosquitoes will always look for the water to lay their eggs until become a pupae (Lahondere, 2012). Therefore, this study used bottles of mosquito trap contained only well water as negative control. The Man Whitney test showed significant differences between the number of mosquitoes trapped on the bottle contained brown sugar-yeast solution, sugar-yeast solution compared to the bottles that contained only well water

Glucose can be fermented by yeast cells into alcohol by releasing CO_2 gas. Fermentation of glucose into ethanol and carbon dioxide by yeast (*Saccharomyces cerevisiae*) occurred via fructose diphosphate pathway. *Saccharomyces cerevisiae* may live well in sufficient oxygen as well as lack of oxygen conditions. In sufficient oxygen condition, *Saccharomyces cerevisiae* will conduct regular respiration. But, in the lack of oxygen environment, *Saccharomyces cerevisiae* will perform fermentation. In the anaerobic state, pyruvic acid produced by glycolysis is converted to acetic acid and CO_2 (Polakowski, 2008). Regarding to the conditions above, in this study the bottles of mosquito traps covered with plastic sheets for 24 hours to induced an anaerobic atmosphere and plastic cover will be opened when a bottle of mosquito traps will be used.

Figure 1 showed that the number of mosquitoes caught in bottle of mosquito traps contained solution of sugar-yeast and brown sugar-yeast solution did not increase again after 7th day. These results differ from Astuti (2011) results which showed that the use of sugar-yeast solution as an attractant was effective trapping mosquitoes until the 18th day. This difference results due to research conducted by Astuti (2011) carried out in the laboratory with controlled environment while this study was conducted in the field. The wind may affected the levels of CO_2 in trapping mosquitoes in the field. However, it was consistent with research conducted by Guidobaldi (2013) showed that a decrease in the number of *Aedes aegypti* mosquitoes caught in traps by attractant palm sugar and 30% chilli solution started in the 4th day of installments of mosquitoes traps which is caused by evaporation of the attractants. Smallegange (2010), stated that

Figure 2. The Differences of Total Mosquitoes Trapped Inside and Outside The House along with Temperature, Humidity, Rainfall Rate in Pabean area in Pekalongan City in 2015-2016



	M-1 (Okt)	M-2 (Okt)	M-3 (Okt)	M-4 (Okt-Nov)	M-5 (Nov)	M-6 (Nov)	M-7 (Nov)	M-8 (Nov-Des)	M-9 (Des)	M-10 (Des)	M-11 (Des)	M-12 (Des-Jan)	M-13 (Jan)
Jumlah nyamuk terperangkap di dalam rumah	149	156	311	244	319	441	251	220	140	107	58	89	82
Jumlah nyamuk terperangkap di luar rumah	284	530	583	888	633	1277	527	709	436	377	84	159	156
Suhu (°C)	30	33	32	31	32	31	28	27	27	25	27	26	31
Kelembaban (%)	49	69	77	76	76	81	79	84	85	95	90	95	93
Curah Hujan (mm)	20	20	20	200	200	200	200	500	500	500	500	500	500

Source : Primary Data

the CO_2 produced by the yeast will decrease its attractant properties between 38-48 hours after the preparation of the solution.The average flow of CO_2 decreased from 60 ml/min to 0 ml/min after 30 hours.

At the second phase of study, we used bottle mosquito traps containing sugar-yeast solution because it was a daily ingredients which the cost was cheaper than brown sugar. The costs that needed to made a mosquito traps for 1 week was Rp.352 for brown sugar and Rp.346 for granulated sugar. This means the cost was cheaper than use a mosquito coils which price Rp 800 per day and Rp.1392 for insect repellent spray. Moreover, mosquito coil and insect repellent spray may have side effects from its chemical compounds. In addition, sugar was a daily ingredients more frequently use than brown sugar, so ensure the sustainability of its use.

Mosquito attractant is a compound that chemically or visually attract the insect. This attractant could be CO_2, lactic acid, fatty acids and octenol. Fatty acid that produced by a normal flora is supposed to be an effective attractant (Kline, 2012).

The mosquito attractant that used on this research was carbon dioxide (CO_2) that produced from fermentation of sugar and yeast. Glucose could be fermented by yeast cells converted to alcohol and CO_2 gas. Many studies showed that the use of CO_2 to increase the mosquitoes trapped because CO_2 is a substance which responsible for activation and guide those mosquitoes to reach vertebrates host (Carde, 2010; Mweresa et al, 2014; Lahondere & Lazzari 2012). Therefore, CO_2 is the most frequently use as mosquito traps (Jawara, 2011). This day, CO_2 often added to artificial odors to attract mosquitoes for various interests and objectives.

Mosquito has a simple sensory organ or a structural unit from composite sensory organ called sensillum (plural: sensilla). Olfactory sensilla of mosquito is formed by multiporous hair and peg sesilla on antenna that form flagella and multiporous peg sensilla on palpus maxillary. There are 4 compound types that easily evaporate and responded by sensila i.e. (i) Carbon dioxide (CO_2); (ii) fatty acid; (Iii) group of essential oil, which is derived from plants, including geraniol, eugenol, citral, α-pinene and thujone; (iv) compound group known as attractants for oviposition such as 4-methylphenol and 4-methylcyclohexanol and

(v) pheromones was known for attract *Culex pipiens* to laid their eggs called 6-acetoxy-5-hexadecanolide.

Single-walled multiporous peg sensilla on mosquito palpus maxillary contain sensory neurons that able to respond to CO_2 concentration alteration, although the change is small. Mostly, single-walled peg sensilla multiporous in mosquitoes contain three big sensory neurons. These neuron capable of producing enormous action potential but only one neuron that sensitive to CO_2. It consist of components which provide respond to female mosquitoes behavior against CO_2 alteration. These neurons sensitive to CO2 concentration at the range from 0.015% to 0.03% ,which is almost the same with the concentration of CO_2 in the atmosphere (0.02% to 0.04%). These neurons are CO_2 detector and its sensitivity were not reduced by continuous exposure of CO_2. Besides that, its sensitivity are not affected by the previously CO_2 concentration. A slightly increased in CO_2 concentration able to increase the activation of these neuron significantly. Increased CO_2 concentration at 0.01% above threshold of neuron sensitivity are able to increase the response time 2-4 times. These neurons give response to CO_2 surrounding the vertebrate host.

The molecular mechanism of smelling in insects are very complex. Olfactory signals is an external stimuli that influence the mosquitoes behavior in finding a host, laying eggs and sucking food (sugar) (Cook, 2011; Guha, 2012; Mathew, 2013). Mosquitoes are classified in hematophagous group and it should able to find a suitable host for the reproductive process. These mosquitoes group require blood protein from the host to the development of the eggs (Guidobaldi, 2013).

Host attractiveness are grouped into three i.e. short-range, moderate-range and distant-range attraction. Odor stimuli alone is a distant-range attraction. If the odor stimuli combined with CO_2 will be classified as moderate-range attraction; and CO2, body heat, body moisture and visual signals will be classified as short-range attraction (Mukabana, 2002, Mathew, 2013). Mosquitoes are able to use its sensitive olfactorius to select and identify the chemical compounds in the breathing air, perspiration, excretion from human skin (Verhulst, 2011).

The *p*-value at the Mann Whitney test analysis was 0.000 less than the value α 0.05 then Ho was rejected, which means that there were differences in the number of mosquitoes trapped in the mosquitoes trap inside and outside the house which observed until 13th week since the trap was set up. The average score of Rank to trap mosquitoes outside (42.75) was larger compared to mosquitoes trap inside the house (18.25). Thus, it could be concluded that a mosquito trap containing a solution of sugar-yeast more effectively used outside the house and the number of mosquitoes trapped outside more than the number of mosquitoes trapped in bottles placed inside the house.

Mosquito trap that placed inside the house where people live and stay, mosquitoes were also attracted by the other attractants materials released by humans. As known, various substances or compounds secreted by humans are attractants for mosquitoes, such as lactic acid, octenol and fatty acids. Thus, mosquitoes are more interested in human who released more kind of attractant odors than the attractant in a bottle of mosquito trap containing only CO_2 alone (Backer, 1995). Saitoh et al. (2004) stated that CO_2 attractiveness against mosquitoes increased when combined with synthetic attractant odors. Mbita, a mosquito trap products contain ammonia, L-lactic acid and 3-methyl-1-butanol.

CO_2 is a substance that able to attract mosquitoes approaching the trap. However, the findings showed that mosquitoes trapped increased if added human odors or the synthetic attractant than only CO_2 alone (Spitzen, 2008). Olanga (2010), stated that CO_2 is an effective to attract mosquitoes approaching the host. However the effect CO_2 as an attractant will be stronger if combined with lactic acid.

The capture of mosquitoes using mosquito traps which environtmentally friendly performed at temperature between 25˚C - 33˚C and humidity ranged from 49% - 95%. Simon (2015), mentioned that the rainfall and humidity associated with vector population dynamics. Rainfall provide mosquito breeding place and relative humidity for at least 50%-60% which prolong the survival life of mosquitoes. Relative humidity under 60% shorten the

mosquitoes life-span. Similar results also found in research conducted by Murty (2010), that the temperature in between 26˚C - 32˚C and the average relative of humidity 55% allowed the mosquitoes reach the highest abundance. Total abundance of mosquitoes trapped is 1.918 mosquitoes. The highest mosquitoes population trapped was *Cx. Quinquefasciatus* (1,577), followed by *Anopheles indefinitus*, *Culex vishnui, Culex tritaeniorhynchus, Aedes aegypti* and *Culex bitaeniorhynchus*. Ramadhani (2010), stated that *Cx.quinquefasciatus* caught in Pabean area in Pekalongan city has highest density in October until November. It is consistent with current research that said the most mosquitoes trapped in between the end of September until early November. At these time, there was an increase in rainfall that resulted in the emergence of puddles and could be used as mosquito breeding place. While, from the end of November, the rainfall is very high resulted in mosquito eggs washed away by high debit water on various water drainages.

Most species of mosquitoes trapped in this study was *Cx.quinquefasciatus*. Culex mosquito larvae easy to adapt and survive in almost all habitats such as household sewer, garbage plastic bottles, rob puddles, puddles in the swamp that dries (Okiwelu, 2012). The location of this research was the Pabean in north part of Pekalongan city, Central Java Province. Based on demographic data of Padukuhan Kraton Pekalongan city, Pabean area lies at a height of 1 meter above sea level, and it's the lowest area in Pekalongan, so it's a basin. These geographic conditions cause sea water runoff can not return to the sea immediately and become breeding places of mosquitoes.

Conclusion

Mosquito trap made from a plastic bottle containing a solution of sugar-yeast effectively trapped mosquitoes compared to well water as control. Mosquito trap containing a solution of sugar-yeast were placed outside the house were more effective than the mosquito trap placed inside the house.

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