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Original Article

The Effect of Integrated Ovitrap in Reducing The Transovarial Transmission Index in DHF Endemise Areas

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ABSTRACT

Dengue hemorrhagic fever is a health problem in Indonesia. Pontianak City as the city center of West Kalimantan Province has an important role in the spread of dengue fever. An effective integrated model for vector control is needed to reduce the incidence of dengue fever in endemic areas. We carried out an intervention using the integrated CTVVC and SAMT model for 4 weeks in 100 homes as an intervention group. To determine the impact of implementing this model, blood samples were examined from the population and Aedes Spp eggs were collected into 2 groups, namely the intervention group and the control group for 4 weeks, namely 3 weeks. before the intervention and 3 weeks after. The egg samples were then colonized into adult mosquitoes with an average age of 7 days. Next, the analysis was carried out according to the immunocytochemical method of streptavidin-biotin peroxidase complex (ISBPC) and Polymerase Chain Reaction Transcription Reaction (PCR). The measurement results show that the DENV transovarial index in the intervention area is lower than in the control area, namely 40% and 50%, as well as the DENV infection index in the population shows that DENV infection is higher compared to the intervention. area, namely 26.47% and 22.50%. However, statistical analysis did not show the effect of the CTVVC-SAMT integration model on DENV transovarial transmission index and DENV infection in the population with P value: 0.66, P value: 0.109. This research succeeded in identifying the DENV-3 type.

Keywords: Integrated Ovitrap, Transovarial Transmission of DENV, Aedes aegypti.

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INTRODUCTION

Dengue Hemorrhagic Fever (DHF) is one of the health problems in Indonesia. All regions in Indonesia are at risk of contamination due to dengue fever because the transmitters, both the virus and the mosquitos, are widespread in residential housings and public facilities throughout

Indonesia. Based on today's available reports, DHF has become an endemic problem in 33 provinces, 436 regencies, 605 districts and 1800 villages. In 2014 to 2017, it reached 52.61/100,000 population with a case fatality rate (CFR) of 0.72%.¹

In 2009, the DBD CFR in West Kalimantan Province ranked 2nd in Indonesia, although the number of sufferers was only

979 cases compared to West Java with 35,453 cases, and Jakarta with 27,964 cases however, the CFR reached 3.38 %; previously was Jambi whose CFR reached 3.67%. The increase in DHF cases occurs every year. In 2017, there were 5,049 cases of DHF with 68 deaths, in five districts, including Pontianak City, which was declared as an Extraordinary Event. The West Kalimantan Province ranked 2nd in the Kalimantan Islands region after East Kalimantan, with a total of 5,762 DHF cases. 2

DHF in Pontianak City is a disease that always occurs every year (endemic), and still has the potential to cause an outbreak. During $2009 - 2016$, the DHF morbidity rate showed a fluctuating trend in which the incidence rate (IR) in 2009 was very sharp, namely 728.8 per 100,000 population; however, from 2010 to 2016, it significantly declined in 2010 (IR: 14.2), in 2011 (IR: 28.3), in 2012 (IR: 23.1), and in 2013 (IR: 17.06), and it increased in 2016 (IR: 58.85). 3

The various risk factors for DHF in Pontianak City, namely: the geographical environmental condition that is located right on the Equator, with an altitude ranging from 0.10 meters to 1.50 meters above sea level, and categorized as a lowland (less than 500 m) with high levels of mosquito populations. The temperature of Pontianak City ranges between 26.8 to 28.8 °C; such an optimal temperature along with the rainfall. This temperature is ideal (20 to 30° C) for the life of *Aedes aegypti*. The increase in the air temperature causes the incubation period of the extrinsic disease agent to be shorter, and the pattern of the Dengue virus spread has a tendency to increase over time ⁴

In general, the *Aedes aegypti* control and prevention programs in various countries, including Indonesia, still depend on fogging to extinguish the adult mosquitoes. This is costly, creating vector resistance due to inappropriate doses, and does not have a long impact since the mosquito larvae are not perished. The resistance of *Aedes aegypti* to organophosphate in Salatiga City ranges from 16.6 to 33.3 percent, while against 0.8% malathion reaches 66 to 82 percent. WHO only recommends fogging not for routine, yet only in areas that have been clearly identified. Research in Bandung shows that *Aedes aegypti* is also resistant to Allethrin, Permethrin, and Cypermethrin with a Lethal Time of 90% (LT90), ranging from 9 to 43 hours. 5

In controlling dengue fever, WHO recommends the best way to control *Aedes aegypti* mosquitoes aimed at their habitat in residential areas.⁶ One of the vector control methods for *Aedes aegypti* without insecticides that have succeeded in reducing vector density in several countries is the use of egg traps (ovitrap). This tool was developed first by Fay and, later used by the Central for Diseases Control and Prevention (CDC) in *Aedes aegypti* surveillance.

The Sticky Autocidal Mosquito Trap (SAMT) is a device designed to effectively trap mosquitoes (Figure 5). The results of the research of using the SAMT tool showed that it was effective in reducing the density of *Aedes sp* mosquitoes in endemic areas, but had not been able to prevent the transovarial spread of the dengue virus from the mother mosquito to the next generation. This is because the tool immediately kills adult mosquitoes and larvae, but it cannot confirm and calculate the presence of the virus in trapped mosquitoes or larvae. Besides, the spread pattern of viruses that live and reproduce in addition to the *Aedes sp* mosquito is also in humans. A person infected with the dengue virus is a very effective source of transmission, entering from and outside the region dynamically, even becoming the most dangerous source of transmission, especially in people with the virus who do not show symptoms or only show symptoms of low-grade fever so they are late in seeking treatment, and they can go anywhere. ⁷ Patients generally come to the hospital after 4 to 5 days since someone contracting the virus in their bloodstream. During that time there has also been an escalation of horizontal virus transmission which is similar to a measurement sequence: one to two, two to four, and so on. This is also a cause of delay in carrying out focus fogging.

WHO recommends that the best control of dengue vectors be carried out in their habitat, $\frac{6}{9}$ Previous studies have intervened using the SAMT method, the results were effective in reducing the density of Aedes larvae, but not effective in reducing the DENV transovarial transmission index, because the study did not involve the community. Community is one of the two important factors in the emergence of dengue

fever, namely humans and DENV⁸. Therefore, this study combines two methods into one, combining the SAMT method and the Community-based Vector Control (CBTVC) method which aims to reduce mosquito density and also prevent vertical transmission of the virus (Transovarial DENV), and reduce DENV transmission in the population. in endemic areas

METHOD

The design in this research using a quasi-experimental design. This research uses an Interrupted time-series design with a nonequivalent no-treatment control group time series ⁹. This time-series study is usually for field research, which generally has a lot of external variables and cannot be controlled so that the power of selecting independent variables and measuring repeatedly in series before and after treatment can maintain consistency/validity and reliability of measurement effects, and minimize the effects of external variables. In this research design, the effect of the treatment was deduced from the comparison of measurements made several times before and after treatment. The intervention (X_1) in this research used The integrated Community-Based Total Vector Control And Sticky Autocidal Mosquito Trap model in the treatment area located inside and outside the home, while in the control area (X_0) there was no intervention. The research design scheme can be described as follows:

Treatment group (the integrated model of CBTVC and SAMT):

 $O_1\rightarrow O_2\rightarrow O_3\rightarrow X_1\rightarrow O_4\rightarrow O_5\rightarrow O_6$ Control Group: $O_1\rightarrow O_2\rightarrow O_3\rightarrow X_0\rightarrow O_4\rightarrow O_5\rightarrow O_6$

The sample was determined by purposive sampling based on 5 (five) criteria, namely (1) one of the high-endemic areas of DHF for 4 consecutive years had DHF cases. (2) equal characteristics of the area (settlement, vegetation, and topography), (3) there were fatal cases of DHF in the last 2 years, (4) there were new cases of more than 1 (one) case in the last 3 months before the research, and (5) has a distance of about> 5

km between the study locations (between the treatment location and the comparison location). The number of samples needed in this study were 200 people (heads of families) in 200 houses in 2 research locations, each location was taken 100 people.¹⁰ The selection of 2 locations for the study area was based on the above 5 criteria, namely: Sungai Jawi Dalam Village, West Pontianak Sub-District, and Batu Layang Village, North Pontianak Sub-District. The intervention and control areas were determined randomly, the Sungai Jawi Village as the control area and the Batu Layang Village as the intervention area. Treatment implementation technique: application of an integrated CBTVC-SAMT model (Figure 6) to the intervention area was carried out for 4 weeks. The sampling technique for population blood is based on the general clinical symptoms of DHF, namely: Fever, Headache, Nausea and Vomiting and Fatigue¹¹¹². The data collection technique calculated the effect of the CBTVC-SAMT integrated model on the transovarial transmission index of DENV in Aedes aegypti and DENV infection in the population 6 times consecutively, namely 3 times before and 3 times after treatment, with an interval of 1 (one) week, both in intervention and control areas.

RESULTS

1. Transovarial Transmission Index of DENV in Aedes aegypti

Dengue virus examination on *Aedes aegypti* mosquitoes using the IHC method from eggs. An egg sample attached to the filter paper was sent to the UGM Parasitology Laboratory. Eggs were identified microscopically, enlargement of 400 and 600 times to see the condition and presence of the eggs done before the rearing process (hatching).

The mosquitoes used were *Aedes aegypti* mosquitoes with a mean age of 7 days, full of 10% sugar water solution. Each glass preparation contains 10 head squash preparations. Especially for positive and negative control mosquitoes were taken from the Laboratory of Parasitology, FK UGM mosquitoes. Following the immunocytochemical method of streptavidinbiotin peroxidase complex (ISBPC) which has been prepared and standardized by Umniyati.

DENV detection was carried out starting from material preparation, staining and microscopic examination with a magnification of 40x, 100x, 400x, and 1000x.¹³

Figure 1 shows that at a positive infection rate (+), at 400x magnification, brownish grains of sand are seen scattered among the brain tissue, but almost no cells show a brown color in the cytoplasm. On positive (++) the grains of sand were more spread out and found 1-10 cells showing brown color in their cytoplasm per field of view at 400x magnification. On positive (+++) the distribution of sand grains is more widespread and 10-100 cells are found showing a brown color on the cytoplasm so that the infection can be seen at a magnification of 100x. The picture of positive infection rates $(+++)$, $(+)$, and $(+)$ can be found on preparations in the control group, whereas in the treatment group only a positive infection rate (+) was obtained.

Figure 1. Photomicrograph of head squash with a magnification of 100x10 which shows positive Dengue antigen in the form of brownish hexagonal granules that spread on the mosquito brain tissue of the treatment group (figure C) and the control group (figure D). Figure A is a negative control of non-*Aedes aegypti* **mosquito preparations and Figure B is a positive control for an antigen from mosquitoes infected with the Dengue virus with an incubation period of 7 days.**

The results of the analysis in Table 1 show that the transovarial transmission index of DENV in Aedes aegypti in the intervention area was smaller, namely 40%, compared to the control area, which was 50%.

Table 1. Distribution of Aedes aegypti Head squash samples in the Study Area.

			Intervention Area		Control Area		
PRE/ POST	Weeek	Head sguash	$^{(+)}$ VirDen	Transovarial $DENV(\%)$	Head squash	$^{(+)}$ VirDen	Transovarial DENV $(%)$
Pre		100	15	15	100	10	10
	П	100	0	0	100	10	10
	Ш	100	10	10	100	15	15
Post	IV	100	5	5	100	0	0
	V	100	10	10	100	5	5
	VI	100			100	10	10
	Total	600	40	40	600	50	50

The results of the analysis in Figure 2 show the comparison between before and after treatment there is a tendency to decrease, from 10% to 5% at week 4, compared to the control area there is an increase at week 4, namely from 5% to 15%

Figure 2 Comparison of transovarial transmission index of DENV in Aedes aegypti before and after treatment.

2. The DENV infection in the population

The results of the analysis in Table 2 show that the The DENV infection in the intervention area is smaller, namely 25%, compared to the comparison area, which is 55%.

Figure 3 Comparison of the level of DENV infected

The results of the analysis in Figure 3 show that the comparison before and after treatment there was no tendency to decline in the DENV infection index, both in the intervention and control areas.

The results of the examination of the *Aedes aegypti* mosquito using the Polymerase Chain Reaction Transcription Reaction (RT-PCR) method found DEN3. More details can be seen in Figure 4 below.

Figure 4. DNA marker, negative control, positive control, sample numbers 1, 2, 3 and 4.

Examination of the dengue virus types in *Aedes aegypti* mosquitoes using the RT-PCR method. Based on the results of observations and documentation of the results of electrophoresis with Gel Doc as shown in Figure 4, it shows in sample number 3 positive DENV-3 from *Aedes aegypti* was taken in Batu Layang Sub-District, North Pontianak Sub-District.

Figure 5. The Design and Workings Of The SAMT Ovitrap Which Has Been Applied To The Treatment Group.

Figure 6. The Integrated CBTVC-SAMT Model.

3. The effect between mean transovarial transmission index of DENV in *Aedes aegypti* **before and after the intervention.**

Table. 3 Analysis of the effect between transovarial transmission index (TTI) of DENV in *Aedes aegypti* and DENV infection in the population

* Pairs samples t-test

**Wilcoxon Signed Ranks Test

4. The effect between mean DENV infection in the population before and after the intervention.

Table 4. Analysis of the effect DENV infection in the population

Transovarial	Research Area				
Transmission Index (Mean + Standard Deviation)		D			
(TTI)	Intervention	Control Area			
TTI Pre	$8.33 + 7.63$	$11.60 + 2.88$	$0.66*$		
TTI Post	$5.00 + 5.00$	$5.00 + 5.00$	$0.65***$		

* Pairs samples t-test

**Wilcoxon Signed Ranks Test

DISCUSSION

The results of this research show that the percentage of transovarial transmission index of DENV with the Integrated Community-Based Total Vector Control and Sticky Autocidal Mosquito Trap model was 40% lower than that of the control area, namely 50%. This condition is still lower when compared to previous studies in endemic areas of Pontianak City, namely the transovarial transovarial transmission index of DENV in 2012 of 76.6%.¹⁴

Although the results of the study using the CBTVC-SAMT integrated model do not have a significant effect on the transovaria transmission index. However, it can be seen that the results of examining the mosquito samples by PCR in Figure 1 show that the positive (+) infection rate is lower in the intervention area compared to the control area, namely the positive infection rate $(+ +$ +). This mechanism can occur because a number of Aedes mosquitoes are trapped in the SAMT in the intervention area, causing damage to mosquito morphology, and affecting the autophagy process of Aedes aegypti and disruption of virus proliferation in mosquito cells. This can be sure that DENV as an intracellular parasite Obligate is very dependent on the condition of the host cell, if the cell is damaged it can interfere with the virus growth process, even DENV will die. Based on the results of virological research, it shows that the specific conditions that cause virus propagation in different organs during the embryogenesis process or at the final stage of mosquito life can vary.15121617,18

Table 3 shows that there is no effect in the results of the Integrated CBTVC-
SAMT intervention model on the SAMT intervention model on the Transovarial Transmission index, both in the intervention and control areas. This shows that this model does not directly affect the transovarial transmission index, but has a direct effect on larvae density, as previous studies by Saepudin M, et al. Showed that SAMT had an effect on Aedes larvae density and had no effect on the Transovarial Transmission Index. [9] This happens because of several factors that directly have a tendency to increase the DENV Taransovarial Transmission Index, namely the habit of biting, the presence of patients in the environment/family, spatial (environmental conditions), as well as residents.

The existence of environmental factors as confounding variables affects the intervention, namely population density, humidity, temperature, and the presence of predators in the two regions. High population density affects the occurrence of faster transmission because the distance from one house to another is close together. Apart from that, the existence of a house that is close to each other can increase the temperature and humidity which is more optimal for both vector reproduction and the extrinsic incubation period for the dengue virus is shorter. The optimum temperature for the development of *Aedes spp* is 25-27°C and the optimum humidity is at 70–90%, this is closely related to the extrinsic incubation period of dengue virus in mosquitoes from 11–14 days to 7–12 days, the virus will move throughout the body, including the salivary glands of mosquitoes, with an increase in temperature of 2°C The extrinsic incubation period for DENV will be shortened and more mosquitoes will be infected available for a longer period of time. In addition, mosquitoes will bite more often because dehydrated and thus further increasing human-mosquito contact.

Tidal environmental conditions can worsen sanitation and increase new breeding places for mosquitoes especially during high tide after heavy rains, water quickly fills natural reservoirs such as used cans, used buckets, and used glasses of mineral water. Based on the results of the virtual index calculation in the two regions, "medium to high" shows that these areas are at high risk as a breeding ground for the *Aedes* mosquito. Thus the intervention carried out within 4 weeks has not succeeded in reducing transovarial transmission index of DENV, because it cannot detect eggs in natural water reservoirs.

The presence of predators *Micronecta*, known as water insects, will increase along with the availability of food in the water reservoir. The presence of a *Micronecta* predator is believed to have preyed on the infected larvae vertically. According to Mardihusodo et al., (2007), every infected adult mosquito will have a transovarial transmission in the next generation, the dengue virus will develop following the mosquito life cycle. As explained by Mardihusodo et al., a gravid female *Aedes aegypti* mosquito infected with Dengue virus acts as a host to the ovum (egg) in the mosquito's uterus, which eventually propagates in the embryo in the egg. Furthermore, the Dengue virus uses larvae to image as a living medium for its propagation. Humans can be infected with the dengue virus when the mosquitoes that emerge from their puppies in the water first bite and suck blood.

This is also related to the optimal humidity which has a direct effect on increasing the population of female gravid mosquitoes to lay their eggs in water reservoirs, then after hatching they become larvae instar 1 and 2 as a source of food for predators. Thus the increase and decrease in transovarial transmission index of DENV are not determined by interventions using standardized Integrated CBTVC-SAMT model or Ovitrap modifications, but due to the presence of predators in water reservoirs in the study area.

The biting habit of mosquitoes is influenced by the presence of a virus in the mosquito's body, resulting in behavioral changes that lead to an increase in vector competence or the ability of mosquitoes to spread viruses, where mosquitoes become less reliable at sucking blood, even though they repeatedly thrust the proboscis but fail to suck blood, so the mosquitoes move from one person to another, as a result, the risk of virus transmission becomes even greater. The *Aedes* mosquito gets the virus when it sucks the blood of people whose blood contains the virus or gets it by transovarial transmission, where since the egg, the mosquito has contained the virus passed down by its mother.

The entry of infected *Aedes* mosquitoes from outside the study area cannot be limited and controlled, because of the large ecological area, all areas in open conditions have access roads to other areas. This directly affected the spread of the dengue virus during the intervention, so the increase and decrease in transovarial transmission index of DENV can be determined by other than the intervention.

Another factor is the technical examination of the virus is only taken from *Aedes spp* eggs, breeding is carried out in the laboratory until it reaches an adult mosquito

aged 7 days for head squash examination, so it does not describe the real transovarial transmission index of DENV . According to the results of the treatment in the Ovitrap index (AOI) area before and after treatment, it shows a small number both in the intervention and control areas, namely 12% in the control area and 5% in the treatment area.

The results of this research showed that the percentage of the level of DENV infecteted with the Integrated model of CBTVC-SAMT was 22.50% lower than that of the control area, which was 26.47%. The results of the study using the Integrated model of CBTVC-SAMT showed no statistically significant effect in the level of DENV infecteted. This is in line with the number of mosquitoes and larvae trapped in the intervention area which affects the population, it also affects the number of bites of mosquitoes that are infused in people around the patient.

Table 4 shows that there is no effect in the results of the Integrated model of CBTVC-SAMT intervention on the incidence of DENV infection in both the intervention and control areas. This shows that this model does not have a direct effect on the incidence of DENV infection in the population, but has a direct effect on larval density, as Saepudin M, at al, previous research showed that SAMT had an effect on Aedes larvae density and had no effect on the rate of DENV infection.⁷ The existence of compliance with CBTVC to increase community participation also does not have a significant effect on the incidence of infection in the population.²⁰²¹ This is followed by a change in behavior that takes a long time. besides the incidence of DENV infection is directly influenced by the habit of biting vectors, vector density, vector movement from one place to another; as well as the presence of sufferers in the environment/family, mobilization and exposure to mosquitoes, age and sex; also influenced by environmental conditions: rainfall, temperature, sanitation, and population density. ²² Another thing that has a direct effect on the occurrence of DHF incidence is the number of viruses that attack the population of an area, and is highly correlated with the increase in cases both in quantity and quality, namely severe dengue, dengue / DSS.

Virulence of dengue virus is also one of the factors that determine the occurrence of infection in hosts, among the most virulent strains is the *Dengue* strain 3 as evidenced by the results of the research found DENV-3 as the main virus strain which is the most virulent strain. This virulence was shown by the discovery of different fatal cases before the intervention in the two intervention areas, CFR din Batu Layang Village was higher, namely 7.14% compared to the Sungai Jawi Dalam Exit 4.44% ²³ Thus, the effect in virulence in these two locations has an effect on the increased incidence of dengue transovarially over time.

Population mobility affects increasing horizontal Dengue virus transmission. A person who has dengue virus dynamically enters and exits the research area becomes a very effective source of transmission, the most dangerous source of transmission is people with the virus who are asymptomatic or only showing symptoms of low-grade fever because they can go anywhere. High mobility in urban areas plays an important role in the transmission of DENV than the mobility of *Aedes aegypti* itself.24–26

This condition is exacerbated by delays in going to the hospital or health services, which will become a mobile and effective source of transmitting the virus horizontally or vertically. Sufferers generally come to the hospital after 4 to 5 days since a person has the virus in their bloodstream. During that time there has also been an escalation of horizontal virus transmission which is similar to a measurement sequence: one to two, two to four, and so on. This is also a cause of delay in carrying out focus fogging. Along with population mobility and movement, a transovarial transmission has become more widespread throughout the world, for example in Malaysia, Thailand, and Singapore, and Indonesia. Such transmission also occurs in several DHF endemic subdistricts in Yogyakarta City, in several DHF endemic districts in Central Java, and also in Sampit, Kotawaringin Timur Regency, Central Kalimantan. This will likely continue to expand to other areas in Indonesia.²⁷¹⁹

Some of the limitations of this research include the difficulty in controlling the entry of *Aedes* mosquitoes from outside the study area. The entry of *Aedes* mosquitoes from outside the area may exist because the

scope of the study is only limited to a radius of 100 houses around the patient's house (+ 500 meters). At the research location, this was difficult to control because almost all areas were in an open condition (having access roads to other areas). Efforts to create a buffer area will reduce the number of existing housing population. The existence of climate change and the physical environment which directly affect the breeding pattern of *Aedes spp* mosquitoes in the area. Also, the intervention activities for the application of Integrated CBTVC-SAMT model was carried out in two different regions and did not compare with other types of ovitrap, so it was not possible to determine the effectiveness of Integrated CBTVC-SAMT model as a trap for *Aedes Aegypti*.

The entry of predators into SAMT was also inevitable, with worms, ants, and lizards trapped, and frogs attracted to mosquitoes trapped on ovitrap. For control purposes, this is beneficial because it is supportive. However, this condition can affect the ability to modify SAMT and also the results of research, especially the number of mosquitoes that died before being detected, so the results may be smaller than the actual condition.

Increasing the application of the integrated SBTVC-SAMT model will have a positive impact, namely having an impact on improving better environmental management, as well as on awareness and community participation in DHF control. To see this, further research is needed. The use of used goods to make this model makes it possible to increase the price value of these used goods and benefit the local community, such as used plastic glasses and all kinds of used goods that can hold water. This also helps government programs, especially in waste management using the 4R principle; Reduce (reduce), Reuse (reuse), Recycle (recycle), and Replace (replace). This CBTVC-SAMT integrated model is very appropriate as a local-based vector control solution in dengue endemic areas in urban areas in developing countries such as Indonesia.²⁸

The integrated use model of CBTVC-SAMT in the long term can also be one of the ways to control the dengue vector in an integrated manner with other vector control programs. The CBTVC-SAMT utilization model that is integrated in the long term can also be a way of controlling the dengue vector in an integrated manner with other vector control programs. in the future, innovative programs are needed, so that allows fast detection of cases and proper clinical management can reduce mortality from severe dengue fever.²⁶

This model is the most complete modification, compared to the ovitrap designed by several previous researchers. This model can function 3 in one, namely functioning first as a killer for adult benthic mosquitoes and new young mosquitoes that hatch from Pupa, this second model as an alternative in monitoring trapped mosquito species, the third as a trigger for increased community participation in prevention and control. DHF that is easy and cheap takes advantage of local wisdom in the region. Hopefully in the future government policies, in this case, the Ministry of Health of the Republic of Indonesia, in an integrated *Aedes aegypti* vector control activity with the application of this the Integrated CBTVC-SAMT model and Dengue Hemorrhagic Fever vector surveillance activities are not only focused on entomological indicators such as Larva Free Rate (LFR), but are further enhanced on virus surveillance at the vector, as an effort of the Early Warning System (EWS) to prevent Dengue Hemorrhagic Fever Outbreak.

CONCLUSION

The application of the BCTVC-SAMT integrated model, although statistically showed no significant influence on either the DENV infection index or transovarial transmission index, nevertheless there was a tendency for a decrease in the DENV infection index and transovarial transmission index in the intervention group compared to the control group. Examination using the Polymerase Chain Reaction Transcription Reaction (RT-PCR) method succeeded in identifying the DENV-3 virus type as the main cause of Dengue Hemorrhagic Fever cases in endemic areas.

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CONFLICT OF INTEREST

All of us researchers hereby declare that there is no conflict of interest in writing this manuscript,

either for any individual or company**.**

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