Conventional Detection of Resistance of *Aedes aegypti* Larvae as DHF Vector in Kediri District Against Temephos

Demes Nurmayanti¹, Marlik¹, Nurhaidah¹

¹Health Polytechnic of Surabaya, Indonesia

Abstract

Dengue Haemorraghic Fever (DHF) becomes health problem in the world. The most number of DHF sufferers in Kediri District in 2016 were in Pare Subdistrict, Ngasem Subdistrict, and Kunjang. However, controlling technique of DHF vector is such as fogging by using active substance of melathion for adult stadium of mosquito and larvasidation by using active substance of temephos for larvae stadium of mosquito. Moreover, resistance of vector against insecticide is global phenomenon, particularly for program management of infectious disease controlling vector and as a singular barrier in the success of vector control chemically. The vector resistance detection can be conducted by using Detection Conventionally through WHO standard method of Susceptibility test in impregnated paper. This research aimed at analyzing detection conventionally the resistance of *Aedes aegypti* as DHF vector in Kediri District against Malathion and Themepos. This research was True Experiment research and temephos was scattered to the larvae with concentration of 0.01 mg/l, 0.02 mg/l, 0.03 mg/l, and 0.04 mg/l in contact time of 15, 30, 45, and 60 minutes. Data analysis included determination of resistance status by referring to category standard from WHO and analyzing the difference of biota test mortality by using statistical different test of Anova. The result of this research was *Aedes aegypti* larvae in Kediri District was resistant against temephos with concentration in 0.01 mg/l, 0.02 mg/l, 0.03 mg/l, 0.04 mg/l and there was a significant influence of contact time against the death of *Aedes aegypti* larvae. Furthermore, suggestion for Health Office in Kediri District was the use of temephos as larvacide was needed in concentration of more than 0.04ml/l. Besides, it was also needed resistance test of *Aedes aegypti* larvae with concentration of temephos in more than 0.04 mg/l.

**Keywords:** resistance, *Aedes aegypti*, temephos

Introduction

DHF is infectious disease that is caused by dengue virus from *Falvivirus* genus. Until nowadays, DHF still becomes main health problem in the world. Geographical distribution of DHF case is found to be scattered in either tropical area or subtropical area. Based on¹, since 1968 until 2009, World Health Organisation (WHO) noted that Indonesia was the highest country with DHF case in Southeast Asia. DHF case in Surabaya in 1968 was 58 people who were infected and the 24 people died. Afterwards, DHF disease scattered to all areas in Indonesia and attacked all people, particularly children, as well as for DHF case in Kediri District.

In 2016, Kediri District had been occurred DHF case with 993 people (IR = 64.19/ 100,000 population) with total of mortality in 18 people (CFR = 1.8%). If it was compared with the case total in 2015, it had been occurred a tremendous increase in which total of DHF case was 702 people with total of mortality in 7 people. Meanwhile, the case total in 2014 was 221 people, but there was no mortality case. Case total in 2013 was 832 people with total of mortality in 11 people and in 2012 was 492 people with total of mortality in 7 people. Among 38 Districts/Cities in East Java, Kediri District was one of the districts that was categorized as Outbreak area of DHF because in 2015, it was occurred more rather than in 2014. The most number of distribution of DHF sufferers’s domicile in Kediri District in 2016 was in Pare Subdistrict with sufferer total in 107 sufferers. Then, it was in Ngasem Subdistrict with 90 sufferers.
and it was followed by Kunjang Subdistrict with 74 sufferers.(2)

Efforts that had been done in preventing and controlling DHF in Kediri District in 2015 were such as: socializing how to prevent and control DHF; monitoring and training the group of control worker (Pokjanal) in eradication of Mosquito Nest (PSN-DBD) at Subdistrict level, countryside level, and school; training for the mosquito larvae monitor for conducting periodic check; fogging focus if it was known that there was transmission of DHF case around sufferer’s house, PSN 3M plus, fishization and larvasidation.(2)

Technique to control DHF vector was such as: fogging by using active substance of malathion for adult stadium of mosquito and larvasidation by using active substance of temephos for larvae stadium of mosquito. Both of the active substances were organophosphate insecticide. Furthermore, this insecticide was used in Indonesia since 1970, but until nowadays, DHF case was still more reported. The condition made question whether it had been occurred the resistance of *Aedes aegypti* as DHF vector against Malathion and Temephos or not, particularly in Kediri District which was an endemic area of DHF.

Temephos is one of larvasidations from organophosphate class that is more used by Health Office or non-governmental organization for controlling population of *Aedes aegypti* larvae. The use of temephos was began since the government proclaimed abatitation in 1980 for determining DHF transmission chain by distributing abate powder to the society. This program was conducted continuously throughout the year without any larvicidal rotation by proposing to avoid the epidemic. Resistance of insect against an insecticide would be occurred if it was used intensively for 2 until 20 years and continuously throughout the year.(3)

The resistance of temephos and malathion in 0.8% had been occurred to *Aedes aegypti* in West Jakarta, East Jakarta, and South Jakarta.(4) *Aedes aegypti* larvae was resistant against temephos in 0.02 mg/l at Plasokerep Village, Sumobito Subdistrict, Jombang District, East Java-Indonesia.(5)

Concerning with the use of temephos had been used in Kediri City for more than 20 years as an effort of preventative and chain breaker of DHF transmission when it was occurred the outbreak or infectious disease epidemic of dengue. Hence, it was needed to be conducted a determination of the susceptibility status of *Aedes aegypti* larvae against temephos at three Subdistricts of endemic DHF.

This research aimed at detecting conventionally the resistance of *Aedes aegypti* larvae as DHF vector in Kediri District against temephos.

**Methods**

This research was true experiment research and the sample was 3rd descent (F3) of *Aedes aegypti* larvae stadium that was bred in laboratory of Department of Environmental Health, Health Polytechnic of Surabaya, from parental that came from trapped egg in ovitrap in DHF endemic area in Pare, Ngasem, Kandat, and Kunjang, Kediri District, East Java Province, Indonesia. Temephos was scattered to larvae with concentration in 0.01 mg/l, 0.02 mg/l, 0.03 mg/l, 0.04 mg/l with contact time in 15, 30, 45, and 60 minutes. Data analysis included determination of resistance status in Kediri District by referring to category standard from WHO and analyzing the difference of biota test mortality by using statistical different test of Anova.

**Findings**

Result of Conventional Resistance test which scattering 20 *Aedes aegypti* larvae that came from Kediri District for 60 minutes by using temephos with either concentration in 0.01; 0.02; 0.03; or 0.04 mg/l, for control group, it used etanol 1 cc. Every 15 minutes, it was conducted an observation. Result that was obtained in either first 15 minutes, second 15 minutes, third 15 minutes, or fourth 15 minutes was there was no dead *Aedes aegypti* larva, except for concentration in 0.03 mg/l, the percentage average of *Aedes aegypti* larva death was in 0.83%. However, *Aedes aegypti* larva was dead that was caused by residual temephos with concentration in 0.01; 0.02; 0.03; and 0.04 mg/l and also etanol 1 cc for 24 hours (1440 minutes) was in 1.25%; 0.42%; 1.25%; 0.42%; and 2.08%.

*Aedes aegypti* larva in Kediri District was resistant against temephos with concentration in 0.01 mg/l, 0.02 mg/l, 0.03 mg/l, and 0.04 mg/l (Table 1).
Table 1. Resistance Status Against Temephos in Kediri District 2018

| Contact Time (minute) | Concentration of Temephos |
|-----------------------|---------------------------|
|                       | 0.01 mg/l | 0.02 mg/l | 0.03 mg/l | 0.04 mg/l |
|                       | %        | Status    | %        | Status    | %        | Status    | %        | Status    |
| 15                    | 0        | Resistant | 0        | Resistant | 0        | Resistant | 0        | Resistant |
| 30                    | 0        | Resistant | 0        | Resistant | 0        | Resistant | 0        | Resistant |
| 45                    | 0        | Resistant | 0        | Resistant | 0        | Resistant | 0        | Resistant |
| 60                    | 0        | Resistant | 0        | Resistant | 0        | Resistant | 0        | Resistant |
| 1440 (24 hours)       | 1.25     | Resistant | 0.42     | Resistant | 1.25     | Resistant | 0.33     | Resistant |

Discussion

*Aedes aegypti* larva from Kediri District that was scattered by using temephos with concentration in 0.01; 0.02; 0.03; and 0.04 mg/l until 60th minutes, there was no dead *Aedes aegypti* larvae. In certain concentration, *Aedes aegypti* larvae died because as the effect of residue from temephos with concentration 0.03 mg/l. As we know that temephos is the substance of organophosphate insecticide which is effective to be used in water and to be able to kill mosquito larvae. This substance is substance from *abate* that is often used by the society. How to use this temephos insecticide is by hindering enzyme in the insect nervous system, thus, it is not able to have normal function. Larvae of *Aedes aegypti* mosquito in Kediri District which was almost all in varied concentration did not undergo death and this was caused by the concentration that was used was perhaps too small. Similar research was conducted by Merty, et al.\(^6\) in Proceedings of Medical Education who researched resistance of Malathion in 0.8% and Temephos in 1% to adult *Aedes aegypti* mosquito and larvae in Bandung City. Observation was conducted during these two weeks for *Aedes aegypti* larvae that was still sensitive with insecticide of temephos 1% with percentage average of death in 100%. However, the difference with recent conducted research was the concentration that was used in this research was in mg/l, meanwhile, in conducted research by Merty et al.\(^6\) used percent. Concentration and similar units which were conducted by Ridha, et al.\(^7\) who researched by using insecticide of temephos 0.02 mg/l to *Aedes aegypti* larvae in Generation F1 in Banjarbaru City, that the average percentage of death for larvae in the area was in 95%.

Insecticide that was used by society commonly was *abate*, which another name of *abate* was temephos. Dosage of *abate* use in society was 10 gram/100 liter, thus, dosage of *abate* that was used by society was 10%. In this research, the substance of *abate* that was used was temephos with concentration in 0.02 mg/l. Result of the research was all larva of *Aedes aegypti* mosquito in Kediri District was resistant in concentration 0.01; 0.02; 0.03; and 0.04 mg/l. This was caused by toxicity that was given to larva was too small.

Similar research was conducted by Handayani\(^8\) who used temephos with concentration in mg/l. Her research by using concentration 0.625; 0.31; 0.15; 0.078; and 0.039 mg/l concluded that larvae in perimetry area was in tolerant category with mortality rate in 96%, meanwhile, in buffer area was in resistant category with mortality rate in 68%. Another conducted research by Sinaga, et al.\(^9\) used concentration 0.0025 mg/l; 0.005 mg/l; 0.01 mg/l; 0.02 mg/l; 0.04 mg/l; and 0.08 mg/l. Mortality percentage of *Aedes aegypti* larvae reached 100% in concentration 0.08 mg/l.

All in all, the use of *abate* continuously will make larvae to be able to adapt with the environment around, hence, it causes larvae will be resistant with chemical substance. This condition is proved from several areas where have endemic for DHF and always use *abate* as one of mosquito controls. Giving *abate* with concentration...
0.02 mg/l that is conducted continously causes larvae is resistant, thus, it needs higher concentration. Controlling by using abate must be noticed more. Furthermore, abate cannot be given continously. It can be conducted periodically in order to avoid resistant mosquito larvae against chemical substance. However, the main mosquito control is through controlling mosquito population in the environment. One of the most effective ways in controlling the mosquito is through draining, closing, and burying, which in Indonesian.

Analysis result showed that there was a significant influence between contact time and larva death, but concentration of temephos and interaction of contact time with temephos did not have any significant influence against death of Aedes aegypti larvae. It meant that larvae will die with longer contact time. The influence of several dosages of bacillus thuringiensis var israelensis Serotype H14 against Aedes aegypti larvae in West Kalimantan was a conducted research by Perwitasari\(^{10}\). In her research, she stated that the use of insecticide in Bacillus thuringiensis type with dosage based on WHO that was 0.02 ml/l could kill Aedes aegypti larva of LT50 with contact time in 5,046 hours, meanwhile, in contact time of 24.68 hours, larva population was dead (95%). Furthermore, temephos is insecticide in organophosphate which is insecticide category from organic substance that is added by phosphate substance. Phosphate is non-toxic acid type. Characteristic of this acid will impede growth process from mosquito larvae. The larvae can be alive in pH 5.8-8.8. If pH of the water under 5.8, larva will die. However, controlling larvae through insecticide must be kept and controlled well. Safe use for killing larvae is by using insecticide concentration from organophosphate, which is in small concentration in quite long exposure time. Giving insecticide is not allowed continously, but it can be given periodically and insecticide substance, is always evaluated.

**Conclusion**

* Aedes aegypti Larvae in Kediri District was resistant against temephos with concentration in 0.01 mg/l, 0.02 mg/l, 0.03 mg/l, 0.04 mg/l and there was a significant influence between contact time and larva death.

The use of temephos as larvacide was needed with concentration in more than 0.04 mg/l. Giving abate was needed to be done periodically and also evaluated every time. Moreover, it needed resistance test for larvae by using other insecticide substance besides from organophosphate type. In addition, it needed to be conducted monitoring of vector resistance periodically in different area.

**Conflict of Interest-** No

**Source of Funding-** Authors

**Ethical Clearance-** Yes

**References**

1. MoH-RI. Window Epidemiology Bulletin (Bul. Jen.Epi). Jakarta: Jakarta: MoH-RI; 2010.
2. Health Office of Kediri District. Health Profile of Kediri District in 2016. Kediri: Health Office of Kediri District; 2016.
3. Georghio GP, Melon R. Pest Resistance to Pesticides, in Georghio GP, Sito T. (editors). New York: Plenum Press;1998.
4. Prasetyowati H, Hendri J, Wahono T. The Status of Resistance of Aedes aegypti to Organophosphates in the Three Municipalities of DKI Jakarta. BALABA. 2016.
5. Fitriani, Yunita E. Resistance Test of Aedes aegypti Larvae Against Larvasida Temephos. Surabaya; 2017.
6. Merty DK, Rusmartini T, Purbaningsih W. Resistance of 0.8% Malathion and 1% Temephos in Adult and Larva Aedes aegypti Mosquitoes in The City Buah Batu, Bandung. Proceedings. 2017.
7. Ridha MR, Nisa K. Aedes aegypti Larvae is Tolerant to Temepos in Banjarbaru City, South Kalimantan. Jur.Vektora. 2017;3(2).
8. Handayani N. Resistance Status of Larvae Aedes aegypti Against Temephos in the Region of the Perimeter and Buffer Port of Tanjung Emas Semarang. 2015.
9. Sinaga L, Martini S, Saraswati LD. Status of Resistance of Aedes aegypti (Linnaeus) Larvae to Temephos (Study in Jatiasih Sub-District Jatiasih District, Bekasi City, West Java Province. Jur.Kes. Mas. 2016;4(1).
10. Perwitasari D, Musadad DA. Sahat P Manalul, Munif A. The Influence of Some Doses of Bacillus Thuringiensis Var Israelensis Serotype H14 Against Aedes aegypti Larvae in West Kalimantan. Jur.Eko. Kes. 2015;14(3).