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The Effects of N-ACETYLCYSTEINE as Adjuvant Therapy to Reduce TNF-A Level And Increase SPO2/FIO2 Ratio In Improving Hypoxemia In Covid-19 Patients



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Fax: +62315992445

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Research Article

Characteristics Environmental and *Anopheles* Larva Species In High And Low Clinical Malaria Cases In The Landak District of West Kalimantan Province

Khairul Bariyah¹, Budi Utomo², Sri Subekti^{3,7}, Florentina Sustini², Juniastuti^{4,7}, Fathmawati Fathmawati⁵, Heny Arwati⁶

¹Master Program in Tropical Medicine, Faculty of Medicine, Universitas Airlangga, Surabaya Indonesia

²Department of Public Health, Faculty of Medicine, Universitas Airlangga, Surabaya Indonesia

³Faculty of Fisheries and Marine, Universitas Airlangga, Surabaya Indonesia

⁴Faculty of Medicine, Universitas Airlangga, Surabaya Indonesia

⁵Department of Environmental Health, Polytechnique of Health Ministry Pontianak

⁶Department of Parasitology, Faculty of Medicine, Universitas Airlangga, Surabaya Indonesia

⁷Institute of Tropical Disease (ITD) of Universitas Airlangga, Surabaya Indonesia

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ABSTRACT

Malaria remains a health problem in Indonesia. West Kalimantan is a malaria endemic area with high and low incidence. Landak District is one of the malaria endemic area. Malaria cases were found in the areas around illegal gold mining and oil palm plantations. The aims of this study were to describe the characteristics of the breeding sites and species of *Anopheles* larvae found in high malaria cases area, namely Amboyo Utara Village and low clinical malaria cases, area namely Mandor Village. This research is a descriptive research with cross sectional design. The samples were *Anopheles* larvae collected with Accidental sampling technique in the breeding sites. Environmental characterization of breeding sites were physical characteristic including water temperature and sun exposure, chemical characteristic including water pH and salinity, and biological characteristics including water biota. The results of this study were environmental characteristics that have the potential to breed *Anopheles* mosquitoes in Amboyo Utara Village, including water temperature 26-30°C, shandy, water pH 5.0-7.6, salinity 0.2-1.0 ppt, biotas water hyacinth, grass and tadpole. The Mandor village, water temperature 29-30 °C, shandy, pH of 6.9-8.0, salinity of 0.5 ppt, water biota grass. *Anopheles* species found in Amboyo Utara village were larvae of *An. vagus* (94.30%), *An. tessellatus* (3.42%), *An. subpictus* (1.62%), *An. indefinitus* (0.81%) and *An. maculatus* (0.81%). Characteristics of breeding sites in Mandor village were larvae of *An. maculatus* (11.11%), *An. subpictus* (3.70%), and *An. vagus* (85.18%). The conclusion of this study was that different species found at breeding sites with different environmental characteristics in both high and low malaria areas in Landak District, West Kalimantan Province.

Keywords: Environment, Larvae, *Anopheles* Species

ABSTRAK

Malaria masih menjadi masalah kesehatan masyarakat malaria di Indonesia dan endemik Asia Tenggara. Kalimantan Barat termasuk daerah kasus malaria dengan insidensi rendah dan tinggi. Kabupaten Landak merupakan salah satu tempat kasus malaria yang terdapat di daerah lingkungan Pertambangan Emas Tanpa Izin (PETI), perkebunan kelapa sawit dan transmisi penularan malaria. Penelitian ini bertujuan untuk menggambarkan karakteristik tempat perindukan dan spesies larva *Anopheles* yang ditemukan di daerah kasus malaria tinggi yaitu Desa Amboyo Utara dan daerah kasus malaria klinis rendah, yaitu Desa Mandor. Metode penelitian secara deskriptif dengan rancangan cross sectional. Sampel berupa larva *Anopheles* dengan teknik Accidental sampling. Hasil penelitian ini adalah Karakteristik lingkungan yang berpotensi sebagai tempat perindukan mengandung larva *Anopheles* di Desa Amboyo Utara yaitu suhu air 26-30 °C,

teduh, pH air 5,0-7,6, salinitas 0,2-1,0 ppt, biota air eceng gondok, rumput dan kecebong. Desa Mandor yaitu suhu

* Corresponding Author:

air 29-30°C dan teduh, pH 6,9-8,0, salinitas 0,5 ppt, biota ria_merdeka@yahoo.com

air tanaman rumput. Spesies *Anopheles* yang ditemukan di desa Amboyo Utara yaitu larva *An. vagus* (94,30%), *An. tessellatus* (3,42%), *An. subpictus* (1,62%), *An. indefinites* (0,81%) dan *An. maculatus* (0,81%). Desa Mandor ditemukan yaitu larva *An. maculatus* (11,11%), *An. subpictus* (3,70%), dan *An. vagus* (85,18%). Kesimpulan penelitian ini adalah spesies yang berbeda ditemukan di tempat perindukan dengan karakteristik lingkungan yang berbeda pula baik di daerah kasus malaria tinggi dan rendah di Kabupaten Landak, Provinsi Kalimantan Barat.

Kata kunci: Lingkungan, larvae, Spesies *Anopheles*

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INTRODUCTION

Malaria remains a public health problem in Indonesia and endemic in Southeast Asia. Malaria is transmitted by the vector. The number of malaria patients in the world in 2015 is 216 million cases each year. A total of 655,000 people died from malaria.¹ Globally, in 2016 malaria in Indonesia caused the death of 445 thousand people. The decline in the last four years occurred in endemic districts in Kalimantan and Sulawesi.²

West Kalimantan Province is one of malaria endemic areas in Indonesia. The Equator line is crossed the province precisely in Pontianak City. The spread of the *Anopheles* mosquito from the equator where mosquitoes and mosquito breeding places adapt to the equatorial climate and environment (Depkes, 2009). Landak District West Kalimantan Province with three Sub-Districts which remain endemic to malaria were Ngabang sub-districts, Menjalin sub-districts and Air Besar Sub-District. Data on malaria cases in Landak District the last three years 2014, showed that 2,742 clinical malaria cases and 18 microscopic positive malaria cases. In 2015 there were 3,566 clinical malaria cases and 41 microscopic positive malaria cases. In 2016 there were 4,409 clinical malaria cases and 15 microscopic positive malaria cases. The Annual Parasites Incidence (API) of Landak District in 2016 was 0.04 per 1,000 population, and in 2017 was 0.03 per 1,000 population (Landak Sub-District Health Office, 2017). The breeding places for *Anopheles* mosquitoes in this area includes puddles, ponds, and water gutters found in forests and oil palm plantations, rubber plantations,

illegal gold mining, irrigation channels in rice fields, wells used for bathing, washing and other kinds of breeding places located near residential areas.

Amboyo Utara village that belongs to Ngabang Sub-District is area with high malaria cases in 2016 were 58 cases of clinical malaria and 15 cases of microscopic positive malaria microscopically. In 2017 the number of clinical malaria cases was 30 cases and 6 cases of microscopic positive malaria microscopically.³ Low malaria cases in Mandor Village that belongs to Mandor Sub-district, in 2016 the number of clinical malaria cases were 47 cases and in 2017 there were 24 cases and there were no microscopic positive malaria cases (zero).⁴

Amboyo Utara Village consists of rice field, vegetables and oil palm farming. Mandor Village consists of low land areas, most of which are farming, rubber plantation, and illegal gold mining. Some population activities have an impact on the health of the surrounding community, especially the onset of malaria disease. Forests logging that disrupt the natural habitat of *Anopheles* mosquitoes.⁵

Anopheles mosquitoes in Indonesia are found around 430 species, but 30-40 species of mosquitoes can potentially be a vector for malaria transmission in humans (Soedarto, 2011). *Anopheles* mosquito species that have been identified in West Kalimantan are *An. sundaicus*, *An. maculatus*, *An. letifer*, *An. balabacensis*.⁶ Mosquito vector in the work area of Sekura Public Health Center, Sambas District Puskesmas Sekura, West Kalimantan is *An. campestris* with

breeding places in the lowlands, *An. nigerrimus* with a breeding place in the swamp and pond, *An. balabacensis* with a breeding place in a pond, a pool of water, a ditch, and *An. maculatus* with clear water and exposed to sunlight.⁷

Anopheles mosquitoes require a breeding place with certain physical, chemical and biological environmental characteristics. A research in Iran, reported that there is a relationship between habitat characteristics of *Anopheles* larvae and habitat environment with larval density. The program in vector control carried out in Iran is for malaria elimination.⁸ Species of larvae have similar breeding sites in varying environmental characteristics with different locations of the study.⁸

Thes aims of this study were to describe characteristics of breeding sites in Mandor

and Amboyo Utara villages associated with the species of *Anopheles* larvae found in both areas. This research was conducted in an effort to understand the nature of *Anopheles* larvae in various breeding sites, in contributing to the efforts to control malaria vectors to prevent malaria transmission.

MATERIALS AND METHODS

Location

The area with high clinical malaria cases was Amboyo Utara village with located in Ngabang Sub-District, while area with low clinical malaria cases was Mandor village. Which located in Mandor Sub-District, both villages were located in Landak District, West Kalimantan Province.

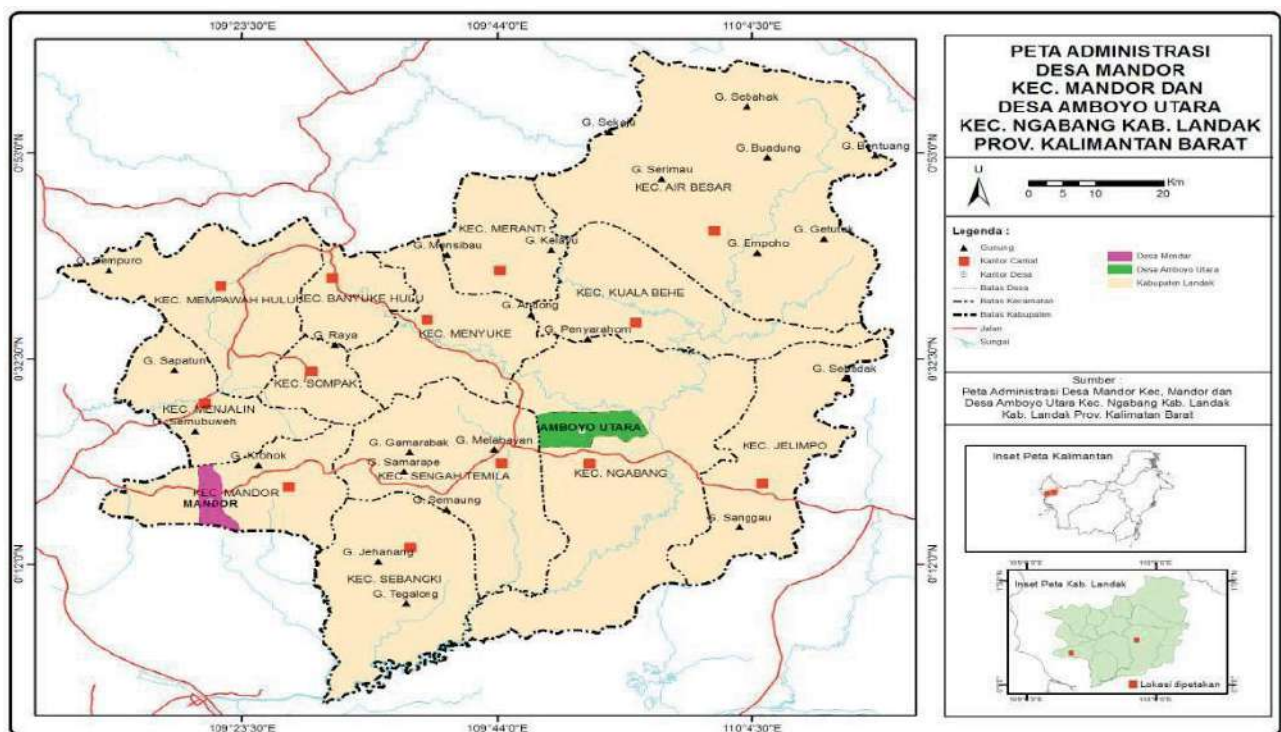


Figure 1. Amboyo Utara Villages is located in Ngabang Sub-District (green color) and Mandor Village is located in Mandor Sub-District (pink color). Both are located in Landak District, West Kalimantan Province.

Research Subject

The subjects of this research were *Anopheles* larvae and their breeding sites found in Amboyo Utara village and Mandor village.

Sample Collection

The study was conducted in March 2018. The retrieval of *Anopheles* mosquito larvae was based on *Accidental sampling* techniques.¹⁰ Arrest of

larvae is carried out at 07.00 - 17.00 WIB. *Anopheles* larvae in all breeding places locations in the two study villages. Larvae capture was carried out in accordance with WHO standards using a dipper device with a capacity of 320 ml. Identification of *Anopheles* larvae by using the keys of O'connor and Soepanto.⁹

Analysis data

Data were analyzed descriptively with a *cross sectional* approach.

RESULTS AND DISCUSSION

The survey results of breeding place types in Mandor village along with the characteristics of the environment in Amboyo Utara village in Table 1 and Mandor village in Table 2. The breeding place in North Amboyo village in Ngabang Subdistrict was found breeding sites containing *Anopheles* larvae, were water ditches, swamp, Excavation wells, rice fields, and drill wells. Mandor villages are found breeding places for illegal mining gold and Excavation wells.

The results of distribution of breeding characteristics in the environment that contained *Anopheles* larvae based on malaria cases were low and high in Table 1 and Table 2.

Table 1 showed that based on nine positive breeding sites of *Anopheles* larvae in North Amboyo Village, the characteristics of breeding sites in the physical environment with sun

exposure of shady, temperatures of 25-30°C and shade in the village of North Amboyo.²³ temperature in the range 25-30°C the highest level of larval density is at 27°C in Karossa District, Mamuju Regency, Central West Sulawesi Province. Chemical environment at pH 5.0-7.6 and salinity 0.1-0.5 ppt in North Amboyo village. This study is in accordance with Mardiana and Perwitasari that 0 per mile of salt is found in the breeding place in the form of rice fields and ditches. Species found with a pH of 7-8 at potential breeding sites *An. vagus* positive in Sub-District Labuan, Pandeglang Sub-District, Banten Province.^{11, 12, 15}

Biological environmental characteristics were found consisting of water hyacinth, tadpole and grass at the *Anopheles* larva breeding site in North Amboyo village. The presence of aquatic plants and leaves that fall in the breeding can be used by larvae to hide and obtain nutrients that contain nutrients that make larvae survive longer.²³ Tadpole (*Rana sp*) can be a predator but is a competitor to larvae. Some theories also say tadpole animals are aquatic animals that are herbivores that eat algae and other plants. However, there are also several types of tadpoles eating everything that can be eaten including larvae or animals.¹⁸ The presence of animals and predators also influences the development and density of larvae, such as larva-eating fish, namely tin-head fish, tilapia and others.¹²

Table 1. Results of characteristics of breeding sites in the positive environment of *Anopheles* larvae in Amboyo Utara village, Ngabang District, Landak Regency.

No	Type of breeding placse	Amount of places breeding	Physical		Chemical		Water Biota	
			Sun Exposure	Temperature (°C)	pH	Salinity (ppt)	Animal	Plants
1.	Water ditch	1	Shady	26	7.3	0.5	-	Water hyacinth
2.	Excavation wells	3	Shady	25-29	5.0-7.5	0.1-0.4	-	-
3.	Swamp	3	Shady	26-30	5.7-6.6	0.1-1.0	-	Grass
4.	Rice fields	1	Shady	28	7.0	0.5	Tadpole	-
5.	Drill wells	1	Shady	26	7.6	1.0	-	-
Amount Average		9	Shady	25-30	5.0-7.6	0.1-0.5	Tadpole	Hyacinth&grass

Table 2. Results of environmental characteristics with positive *Anopheles* larvae at breeding sites in Mandor village, Mandor Sub-district, Landak Regency.

No	Type of breeding place	Amount of places breeding	Physical		Chemical		Water Biota	
			Sun Exposure	Temperature (°C)	pH	Salinity (ppt)	Animal	Plants
1.	Gold mining wells	1	Shady	30	6.9	0.5	-	grass
2.	Excavation wells	1	Shady	29	8.0	0.5	-	-
Amount Average		2	Shady	29-30	6.9-8.0	0.5	-	grass

Table 2 showed that based on two positive breeding sites of *Anopheles* larvae in Mandor village illegal gold mining wells and excavation wells in a physical environment with water temperatures of 29-30°C and shady. The water temperature at the larval breeding site that is good for larval development is 28°C.¹³ The optimum average temperature for mosquito development is 25-27°C.¹⁴

The chemical environment with a pH of 6.9-8.0 pH 7 is an ideal condition for the development of larvae and a mean salinity of 0.5 ppt because there is a low difference in salinity in the breeding sites of gold mining wells

and excavated wells due to the water environment contaminated by mercury. Biological environment found grass at the larvae breeding sites in Mandor village. Larvae take shelter in aquatic plants with many aquatic plants, moss grass that is submerged in water.¹⁷

Table 3 showed that there were nine positive breeding sites containing *Anopheles* larvae as many as 123 in the Amboyo Utara village. Most species of *Anopheles* larvae were identified in the breeding sites in the dug fields *An. tessellatus* larvae 2.43%, *An. maculatus* larvae 0.81% and *An. indefinitus* larvae 0.81%, and *An. vagus* larvae

Table 3: Results of identification of *Anopheles* species at breeding sites in North Amboyo Village, Ngabang District, Landak Regency.

No	Types of breeding places	Amount of breeding places	Species	Amount of larvae (tail)	%
1.	water ditch	1	<i>An. indefinitus</i>	1	0.81
			<i>An. vagus</i>	2	1.62
2.	Excavation wells	3	<i>An. vagus</i>	19	15.45
			<i>An. tessellatus</i>	1	0.81
			<i>An. maculatus</i>	1	0.81
3.	Swamp	3	<i>An. vagus</i>	23	18.70
			<i>An. tessellatus</i>	1	0.81
			<i>An. subpictus</i>	2	1.62
4.	Rice fields	1	<i>An. tessellatus</i>	1	0.81
			<i>An. vagus</i>	64	52.03
5.	Driil wells	1	<i>An. vagus</i>	8	6.50
Amount Average		9		123	100

52.03%, according to Mading (2014) in Selong Belanak Village, Central Lombok Regency. *Anopheles* larvae were found, including *An. vagus*, *An. subpictus*, *An. sundaicus*, *An. maculatus*, *An. aconitus*, and *An. anullaris*, in this research on the bioecological aspects. This contains *Plasmodium vivax* sporozoites which are

thought to be malaria vectors to be able to ensure the need for surgery for mosquito salivary glands of *Anopheles* species.¹⁷

The results of the recapitulation of the species in the village of Amboyo Utara was *An. maculatus* larvae 0.81%, the number of these species that were found little larvae was

caused by the weather of the rainy season at the time of research in the village of North Amboyo. Density *An. maculatus* is high in the dry season, while in the rainy season the vector is somewhat reduced because the breeding places are affected by rain so that the larvae are brought in by water when flooded.¹⁷

An. indefinitus 0.81% was found in the ditch, according to stated for *An. indefinitus* has habitat and lives breed in irrigation channels and rice fields that are close to the forest. There for this type of species is the least number of larvae found in breeding sites.¹⁸

Larva *An. tessellatus* larvae 2.43% was found in the Amboyo Utara of village. *An. tessellatus* is a malaria vector in Sri Lanka and has the potential as a vector in Kalimantan (Munif and Imron, 2010) and in Donggala, Central Sulawesi. *An. tessellatus* Larvae can be found in habitats^{19,20} protected from sunlight, fresh water that flows slowly, although relatively high salinity has been reported.⁸ *An. subpictus* larvae was found 1.62% of breeding sites in North Amboyo village. In *Anopheles* mosquitoes, some of the ecology on the beach which tends to have brackish water is also a breeding ground for *An. subpictus*.¹⁵

An. vagus larvae 94.30% are found most in each breeding place in the village of Amboyo Utara. *An. vagus* is found to rest more in livestock cages compared to people's homes. In Vietnam, is found *An. vagus* with brackish water habitat and *P. falciparum* cannot develop in mosquitoes *An. vagus*.¹⁶ Different research in Thailand, at *An. vagus* found *P. falciparum* and *P. vivax* able to live in freshwater habitats through salivary gland surgery and ELISA. This is a difference in strains or species due to ecological differences in geographical distribution.

The spread of larvae in the breeding site is not evenly distributed on the surface of the water, but collected in closed places such as moss, floating water plants, garbage and grass - grass on the edge of a river or ditch. Fresh water is used as a breeding ground, is open and gets direct sunlight.²¹ Larva species in Mandor Village and Amboyo Utara Villages found five types of larvae species *Anopheles* larvae that consisted of *An. vagus*, *An. subpictus*, *An. maculatus*, *An. tessellatus*, and *An. indefinitus* which is confirmed as an *Anopheles* species which acts as a transmission of malaria vectors in Indonesia.²²

The results of the distribution of *Anopheles* larvae species in the breeding place with the number of larvae based on low clinical malaria cases in the area of Mandor Village, Mandor Subdistrict, Landak District, West Kalimantan Province in Tables 4.

Table 4 showed that identification of Ngabang Sub-District, Landak Regency, West Kalimantan Province in Tables 4.

The results of the distribution of *Anopheles* larvae species in North Amboyo village, *Anopheles* species larvae showed that there were 27 tail positive breeding sites containing 27 *Anopheles* larvae in Mandor village. The larvae found are not many at the breeding site because the breeding sites contain mercury and this type of groundwater storage is partly acidic and yellow soil which affects the proliferation of *Anopheles* larvae. Residents of the settlement dig the ground to search for gold stones illegally so that gold mining well is a risk factor for malaria cases. The occurrence of malaria is an outdoor activity at

Table 4. Results of identification of *Anopheles* species at breeding sites in Mandor Village, Mandor Sub-District, Landak Regency

No.	Type of breeding place	Amount of breeding places	Species	Amount of larvae (tail)	%
1.	Gold mining wells	1	<i>An. maculatus</i>	3	11.11
			<i>An. vagus</i>	9	33.33
2.	Excavation wells	1	<i>An. subpictus</i>	1	3.70
			<i>An. vagus</i>	14	51.85
Amount Average		2		27	100

night, this is the habit of some exophagic mosquitoes at night, gold workers sleeping at gold mine sites and rubber farm workers who cut rubber. Mosquitoes endophagic like to suck human blood inside the house, including bite of active *An. maculatus* between 21.00-03.00 pm.²⁷⁻²⁸

The results of the identification of *Anopheles* larvae species found the breeding sites in the gold mining wells were found by larvae *An. maculatus* 11.11% and *An. vagus* 33.33%, while larvae of digging wells were found larvae *An. subpictus* 3.70% and *An. vagus* 51.85%.²⁸ in a study in Aceh Besar District stated that *Anopheles* mosquito species found in larval breeding sites are *An. sundaicus*, *An. vagus*, *An. barbirostris*, *An. kochi*, *An. maculatus*, and *An. tessellatus* which is thought to have potential as a malaria vector.

There were two breeding sites that contained *Anopheles* larvae in the Mandor village, namely *An. maculatus* 11.11%. Mosquito *An. maculatus* can develop well in open waters both flowing and not flowing and in the form of stones or soil.²³ Larva *An. subpictus* 3.70%, this species has been recognized as an important malaria vector in Sri Lanka, so bionomics *An. subpictus* in every type of breeding place becomes transmission in India.²⁴ Larva *An. vagus* 85.18% at the breeding site in the Mandor village. Larva *An. vagus* which is often found in calm or slow-flowing water and brackish water.²⁵⁻²⁶ Study states that breeding sites have potential *An. vagus* is positive for sporozoites as a companion vector to transmit malaria to humans in Labuan Subdistrict, Pandegelang Regency, Banten Province.

CONCLUSION

The conclusion of this study is environmental characteristics that have the potential to breed *Anopheles* mosquitoes in Amboyo Utara Village, including water temperature 26-30°C, shandy, water pH 5.0-7.6, salinity 0.2-1.0 ppt, biotas water hyacinth, grass and tadpole. The Mandor village, water temperature 29-30 °C, shandy, pH of 6.9-8.0, salinity of 0.5 ppt, water biota grass *Anopheles* species found in Amboyo Utara village were larvae of *An. vagus* (94.30%), *An. tessellatus*

(3.42%), *An. subpictus* (1.62%), *An. indefinitus* (0.81%) and *An. maculatus* (0.81%). Characteristics of breeding sites in Mandor village were larvae of *An. maculatus* (11.11%), *An. subpictus* (3.70%), and *An. vagus* (85.18%). There is different species were found at breeding sites with different environmental characteristics in both high and low malaria cases in Landak District, West Kalimantan Province.

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CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

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