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Investigating Malaria Susceptibility in Central Maluku District: A Focus on *Anopheles Mosquito Habitats*



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Abstract:

Malaria remains a formidable challenge to global public health, with an estimated 241 million cases reported across 85 endemic countries in 2020. Within this context, Indonesia, and particularly the Central Maluku Regency, has reported a notable burden of the disease, evidenced by 102 confirmed cases in 2022 as per the annual parasite incidence (API) data, highlighting indigenous transmissions in specific locales. This research was conducted to assess the susceptibility to malaria within the operational area of the Hila Perawatan Primary Healthcare Centre (Puskesmas), situated in the Leihitu sub-district of Ambon Island, through an examination of Anopheles mosquito breeding sites, larval densities, and habitat indices. Employing a descriptive research design, this cross-sectional observational study was carried out on October 26-27, 2023, to meticulously document the ecological footprint of the Anopheles mosquito, particularly Anopheles farauti. Findings reveal a habitat index (HI) of 33% in Kaitetu village with a larval density of 20%, indicating a significant presence of Anopheles farauti larvae. These findings suggest that environmental and behavioral factors within households, such as the use of gauze and ceilings, nocturnal activities, application of mosquito repellents, wearing of long-sleeved clothing, and utilization of mosquito nets, are pivotal in influencing malaria transmission dynamics. This study underscores the imperative of integrating environmental management with community engagement strategies to mitigate malaria transmission in endemic regions. The results not only provide a nuanced understanding of the Anopheles mosquito's breeding patterns and its implications for malaria transmission but also offer a foundational basis for tailoring targeted interventions aimed at reducing the malaria burden in the Central Maluku District.

Keywords: Malaria; *Anopheles*; *Plasmodium*; Home environment; Community behavior; Habitat index; Larval density

1. Introduction

Malaria remains an important public health problem in many countries around the world, including Indonesia. In 2020, 241 million cases of malaria were reported in 85 endemic countries, an increase from 227 million in 2019. Especially significant increases occurred in countries included in the World Health Organization (WHO), such as Africa, South Asia, and Southeast Asia regions (World Health Organization, 2021). This disease affects the high mortality rate of infants, toddlers, and pregnant women. Every year, more than 500 million people worldwide are infected with malaria, and more than 1,000,000 people die. Malaria is still a problem in Indonesia. According to

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the Director General of Disease Prevention and Control (P2P) in 2019, out of 514 districts/cities in Indonesia, 300 (58%) are malaria-free, and 214 are still endemic areas. Of the 214 endemic districts, 160 (31% of total districts) are low endemic, 31 (6% of total districts) are medium endemic, and 23 (4% of total districts) are high endemic. Based on this data, it appears that 77% of Indonesia's population already lives in malaria-free areas and around 23% of Indonesia's population still lives in malaria-endemic areas. 89% of districts/cities in Indonesia have achieved an API of < 1 per 1000 population and 58% have achieved malaria elimination. Based on the trend of positive malaria cases and API, there was a significant decrease in cases from 2010-2014 but tended to stagnate from 2014-2019. The stagnant trend in cases can be attributed to the stagnant and increasing trend of malaria cases in Papua Province. Overall, there was almost a decrease in malaria cases in all provinces in Indonesia from 2015 to 2019. Malaria is an infectious disease caused by blood protozoan parasites belonging to the genus *Plasmodium spp.* Insects transmit these parasites to various vertebrates. It is estimated that about 250 different *Plasmodium parasites* infect various animal species, including birds, reptiles, snakes, and mammals. Among them, 27 *Plasmodium* species have been identified to infect a diverse range of non-human primates globally, including macaques, siamangs, and New and Old World monkeys (World Health Organization, 2019; World Health Organization, 2020).

Human infection with various primate malaria strains, such as *Plasmodium knowlesi*, *P. cynomolgi*, and *P. inui*, has been documented in several locations in Malaysia. Similar phenomena likely occur in areas of Indonesia that are natural habitats for non-human primates. Malaria with a clinical diagnosis without parasites found is categorized under clinical codification B54. If parasites are found, *Plasmodium falciparum* is classified under B50; *Plasmodium vivax* (B51); *Plasmodium malariae* (B52); and other parasites (B53).

Malaria is an infectious disease caused by blood protozoan parasites belonging to the genus *Plasmodium spp*. These parasites are transmitted to various vertebrates via insects. It is estimated that about 250 different *Plasmodium* parasites infect various animal species, including birds, reptiles, snakes, and mammals (Garnham, 1966; Poinar, 2009; Valkiunas, 2004). Among them, 27 *Plasmodium* species have been identified to infect a diverse range of non-human primates globally, including macaques, gibbons, and New and Old World monkeys (Martinelli & Culleton, 2016). In contrast, humans can contract several *Plasmodium* species, such as *P. falciparum*, *P. vivax*, *P. ovale*, and *P. malariae*. In the past, certain malaria parasites from monkeys were deliberately transmitted to humans through mosquito bites in experiments. These include *P. cynomolgi* (Coatney et al., 1961; Eyles et al., 1960), *P. knowlesi* (Chin et al., 1968; Coatney et al. 1966), and *P. inui* (Coatney et al., 1963) from Old World monkeys, as well as *P. brasilianum* (Contacos et al., 1963) and *P. simium* (Contacos et al., 1963; Deane et al., 1966) from New World monkeys. In addition, *P. schwetzi*, previously considered a distinct species, is now recognized as a parasite similar to *P. vivax* or *P. ovale*, and was originally identified in chimpanzees (Contacos et al., 1970).

In addition to *Plasmodium spp*. parasites, mosquitoes can act as vectors (intermediate animals) if they fulfill several requirements, such as malaria. Central to these requirements is the engagement between mosquitoes and human populations, quantifiably assessed through the man-biting rate (MBR). Mosquito species are more dominant than other species because they generally have a fairly long lifespan. In other locations, the species have been confirmed as a vector (Lu et al., 2023). *Anopheles sp.* mosquitoes are one of the mosquitoes that like places close to the ground. They also like rice fields that have stagnant water as a breeding place, in addition to bushes and gardens (Wibowo, 2021).

Central Maluku District is on the verge of being eliminated. According to Central Maluku District API data in 2022, there were 102 positive malaria cases, with indigenous cases in several Hila Perawatan primary health working areas. This has increased when compared to 2021, with 56 cases, and 2020, with 43 cases. Central Maluku Regency may have been selected because it is known as a malaria-endemic area. Endemic areas are characterized by sustained transmission of malaria parasites, making them critical locations for studying the epidemiology, transmission dynamics, and control strategies of this disease. The geographical conditions of Central Maluku Regency, such as climate, topography, and proximity to water bodies, can influence mosquito breeding habitats and malaria transmission intensity. These factors contribute to the suitability of the area for malaria research and intervention implementation. Central Maluku Regency can be a priority area for malaria control and elimination efforts due to the disease burden it poses on public health. Understanding the local malaria situation and the factors contributing to its transmission is crucial for designing effective control measures tailored to the needs of the area.

"Malaria receptivity" refers to the inherent suitability of an area for the transmission and establishment of malaria parasites within a population. In the context of Central Maluku, understanding malaria receptivity involves assessing various environmental and ecological factors. Environmental conditions, such as temperature, humidity, and rainfall, influence malaria receptivity by affecting the breeding and survival of mosquitoes and the vectors responsible for malaria parasite transmission. The tropical climate in Central Maluku, characterized by warm temperatures and high humidity, creates favorable conditions for mosquito breeding, thus increasing the area's receptivity to malaria transmission. The presence of suitable mosquito breeding habitats, such as stagnant water bodies, vegetation, and land use patterns, plays a significant role in malaria receptivity. The diverse landscape of Central Maluku, including forests, coastal areas, and urban settlements, provides ample breeding grounds for

malaria vectors, contributing to the region's receptivity to malaria. The term "receptivity" is a critical component of any malaria elimination or reintroduction prevention program, enabling the planning and stratification of program areas. While important for this purpose, there is a lack of clear guidance for programs and policymakers on how to understand and measure "receptivity". This study therefore explored malaria receptivity based on *Anopheles* breeding sites, larval density, and HI in the Hila Perawatan primary health working area.

2. Method

This investigation employed a descriptive research design, focusing on the delineation of *Anopheles* mosquito breeding sites, larval density, and HI within the operational area of Hila Perawatan primary health, situated in the Central Maluku Regency of Maluku Province. The methodological approach adopted was observational, characterized by a cross-sectional data collection framework executed over the duration of October 26-27, 2023. Various factors, such as weather patterns, historical malaria data, and logistical considerations, influence the decision to conduct research in October. In some areas, October may coincide with the peak malaria transmission season, while in others, it may not. Therefore, understanding the local context is crucial for determining whether October falls within the peak malaria transmission season in Central Maluku. October falls within the transition period between the wet and dry seasons in many tropical regions, including Central Maluku. During this time, there are fluctuations in temperature, humidity, and rainfall, which can affect mosquito breeding and malaria transmission dynamics. By conducting research in October, potential changes in malaria transmission patterns associated with these weather variations can be captured.

Located in the northern part of Ambon Island, Leihitu Sub-district has a district government, a working area composed of 11 Negeri and a total population of 47,96 people. Located at 3,250-3,400 South latitude and 126,500-127,300 East longitude, the sub-district is bordered to the west by the West Leihitu Sub-district, to the east by the Salahutu Sub-district, to the north by the West Seram Regency, and to the south by Ambon City. The study population consisted of all breeding sites, residents, and houses in the survey area. The sample in the activity is the breeding site shown by the accompanying officer/Puskesmas. Residents and houses around the breeding sites were sampled.

3. Results

Observations and surveys conducted in Hila Perawatan primary health resulted in three villages serving as breeding grounds. The observations included village names and habitat types. Measurements were taken to calculate HI, larval density and water salinity. The complete results are presented in Table 1.

Total larvae of Water Quality **Points** Village Type of Habitat Classified Anopheles НІ (Salinity ppt) (per Dipper) 25 Kaitetu Receptive 1-10 Swamps Hila Swamps Unreceptive 0 24 33% 25 Waitomu Swamps Unreceptive 0

Table 1. Characteristics of breeding sites

Point 1 is located in Kaitetu village with an area of 60 m² and 10 m² away from residential houses; point 2 has an area of 50 m² and is 50 m² away from Hila village; and point 3 has an area of 50 m² located in Waitomu subvillage and is 10 m² away from residential houses. Table 2 describes the larvae identified as *Anopheles*.

Table 2. Identification of *Anopheles* larvae

Points	Date of Sampling	District	Sub-District	Total Flick	Total Anopheles Larvae	Percentage (%)
1	26 October 2023	Leihitu	Kaitetu	50	10 An. farauti dan koliensis	20
2	26 October 2023	Leihitu	Hila	20	0	0
3	27 October 2023	Leihitu	Waitomu	15	0	0

Table 1 shows that the positive breeding place was in Kaitetu Village, with 10 *Anopheles* larvae found from three breeding places. While the breeding place in Hila village did not have any *Anopheles* larvae, HI of *Anopheles* sp. larvae was at 33%, with one of the three habitats observed positive for *Anopheles*. This still exceeds the required limit, which must be below 1%. The observation results (Table 2) show that of the three breeding places, one of them found *Anopheles* larvae (20%), referring to the Minister of Health Regulation No. 2 of 2023 concerning Environmental Health Quality Standards, i.e., the HI < 1. Data obtained from the Hila Perawatan primary health, malaria cases from January to December 2022 were four cases, Hila Village with two malaria cases and Kaitetu

Village Kalauli Hamlet with two cases. Malaria cases at the Hila Perawatan primary health show a decrease in malaria cases. Different *Anopheles* species exhibit varying degrees of tolerance to salinity levels. Some species, known as freshwater species, prefer breeding in habitats with low salinity. In contrast, other species, known as brackish-water species, can thrive in environments with higher salinity levels. Understanding the salinity tolerance of specific *Anopheles* species found in larval habitats is crucial for assessing the suitability of those habitats for mosquito breeding.

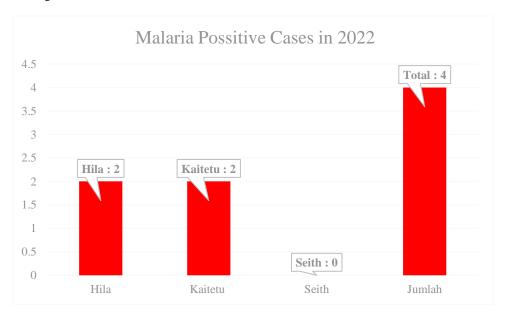


Figure 1. Data on positive cases of malaria in 2022 based on health facility location

Based on observations, malaria occurs (Figure 1) due to home environment factors and population behavior. Table 3 describes the community's home environment, such as the use of gauze and ceilings. Table 4 describes the behavior of the community, such as the habit of going out at night, the use of mosquito repellent, long-sleeved clothing, and mosquito nets when sleeping.

Table 3. Home environment against malaria risk factors in 2023

No.	Home Environment	Yes	No
1	Use of gauze	0	28
2	Ceilings	8	20

Table 4. Behavior of the community against malaria risk factors in 2023

No.	Community Behavior		No
1	Habit of going out at night	28	0
2	Use of mosquito repellent	0	28
3	Use of long-sleeved clothing	0	28
4	Use of mosquito nets when sleeping	0	28

Table 3 shows that 28 (100%) of the houses in Hila Village, with 8 (28.6%) having a ceiling and 20 (71.4%) without one, do not use gauze for ventilation. As shown in Table 4, 28 (100%) respondents have the habit of leaving the house at night, and all of them do not use mosquito repellent, long sleeves when going out at night, or mosquito nets when sleeping.

Screens in home ventilation are typically considered a preventive measure to protect against *Anopheles* mosquitoes, which can be malaria vectors. However, damage or holes in the screen, which allow mosquitoes to pass through the barrier, can still increase the risk of *Anopheles* mosquitoes entering the house (Saili et al., 2024; Sunarsih et al., 2020; Sutriyawan et al., 2021). The chances of mosquito entry can be increased by several factors, such as the size of the holes or damage to the screen, the activity of mosquitoes at night when they search for blood sources, and the attractiveness of humans as a source of blood meal (Sulistyawati et al., 2020; Tizifa et al., 2022). In addition, environmental conditions around the house, such as stagnant water as a breeding ground for mosquitoes, can also affect the risk of *Anopheles* mosquito entry (Mulyani, 2022). Therefore, regular maintenance and upkeep of ventilation screens, along with other malaria prevention strategies, are important measures to reduce

the likelihood of *Anopheles* mosquitoes entering the home and protect the health of householders from malaria risk.

House ceilings play an important role in preventing *Anopheles* mosquitoes, which are vectors of malaria, from entering the house. The ceiling serves as an effective physical barrier to prevent mosquitoes from entering the room where humans rest (Kayendeke et al., 2023). With a ceiling in place, the likelihood of *Anopheles* mosquitoes biting the occupants of the house is significantly reduced. The importance of ceilings in *Anopheles* prevention is mainly related to the behavior of these mosquitoes. *Anopheles* mosquitoes tend to be active at night, especially at dusk and early morning, when people are usually resting indoors (Bofu et al., 2023; Pinder et al., 2016). A tight, sealed ceiling effectively blocks mosquitoes' access to beds or other areas where humans are present (Ngadjeu et al., 2020). In addition, a good ceiling also helps prevent *Anopheles* mosquitoes from laying eggs inside the house. Female *Anopheles* mosquitoes generally seek dark, sheltered places to lay their eggs, and a good ceiling prevents them from approaching such areas (Bofu et al., 2023). Good ceiling maintenance is also an important part of a malaria prevention strategy. By ensuring there are no holes or damage to the ceiling, the risk of *Anopheles* mosquitoes entering the house can be minimized. Thus, the ceiling of the house acts as a significant physical barrier in protecting residents from *Anopheles* mosquitoes, making it a key element in malaria prevention efforts.

Night outings can influence the occurrence of malaria because *Anopheles* mosquitoes, which are the vectors of the disease, tend to be active during this time. The *Plasmodium* parasite, which causes malaria and transmits it through the bite of the *Anopheles* mosquito, primarily transmits the disease at night (Kenney, 2023; Riva et al., 2020). At night, especially at dusk and early morning, female *Anopheles* mosquitoes actively search for blood sources to fulfill nutritional needs in their life cycle. The habit of going out at night or being outdoors at this time can increase the risk of being bitten by mosquitoes carrying malaria parasites. Activities outside the home at night, such as working in the garden, fishing, or traveling without adequate protection, can make a person more vulnerable to mosquito bites. Going out at night can also increase the risk of contact with areas or environments that are potential breeding grounds for *Anopheles* mosquitoes, such as stagnant water or small ponds. If one is near these locations at night, the risk of being bitten by a mosquito carrying the malaria parasite is also higher.

The use of mosquito repellents plays a crucial role in efforts to prevent malaria cases. Mosquito repellents, such as insecticides, have a direct effect on *Anopheles* mosquitoes that act as malaria vectors (Figueira et al., 2023; Ignell & Hill, 2020; Mapossa et al., 2021). Insecticides can be applied to various surfaces, such as mosquito nets, clothing, and house walls, to kill or repel mosquitoes that could potentially carry *Plasmodium* parasites (Havyarimana et al., 2023; Okyere, 2021). The use of insecticide-treated bed nets is one of the most effective prevention strategies. These nets are designed to provide a physical and chemical barrier against mosquito bites during the night when *Anopheles* mosquitoes are actively seeking blood (Afify & Potter, 2020; Dhawan et al., 2014; Luo et al., 2022). The insecticides contained in bed nets not only kill mosquitoes that come into contact with the nets but also provide longer protection against mosquito infestation in the protected area (Mapossa et al., 2019). In addition to bed nets, the use of insecticides on clothing and personal protective equipment can also be an important step in malaria prevention. The use of mosquito repellents on skin or clothing can help repel mosquitoes and reduce the risk of bites that carry the malaria parasite. Indoor residual spraying (IRS) involves applying insecticides to the walls of a house or building structure. Through this method, insecticides are sprayed on the walls or surfaces inside the house, providing additional protection against mosquitoes entering the house. This helps reduce human contact with nocturnal *Anopheles* mosquitoes.

The use of long sleeves plays an important role in efforts to prevent malaria cases. Long sleeves provide an additional physical layer that can help protect the body from the bites of *Anopheles* mosquitoes that act as vectors of malaria (Arsunan et al., 2020; Mponzi et al., 2022). Directly covering the arms can be an effective barrier against mosquito access to the skin, especially on areas of the body that are prone to bites, such as the arms (Ekusai-Sebatta et al., 2021; Zou et al., 2023). Long sleeves are especially relevant when *Anopheles* mosquitoes are actively searching for blood sources, which generally occurs at night (Kazwaini & Wahyuni, 2021). When a person wears long sleeves when outdoors or in areas at high risk of malaria transmission, the risk of mosquito bites can be significantly reduced (Naserrudin et al., 2023). The importance of wearing long sleeves can also be understood in the context of malaria prevention in high-transmission areas. People living in these areas are often exposed to the risk of mosquito bites daily, and the use of clothing that protects the body, including long sleeves, can be a simple but effective step in reducing the risk of disease transmission (Duut & Alhassan, 2022). In addition to providing direct protection against mosquito bites, the use of long sleeves also contributes to a holistic approach to prevention. It can be part of a combination of strategies, such as the use of insecticide-treated bed nets, mosquito repellents, and vector control, that can help reduce exposure to Anopheles mosquitoes and minimize the risk of developing malaria.

Bed nets play a central role in malaria prevention efforts and are one of the most effective strategies for protecting individuals from the bites of *Anopheles* mosquitoes, the vectors of the disease. Insecticide-treated bed nets, specifically designed for bed use, provide physical and chemical protection during critical periods, especially at night when *Anopheles* mosquitoes are actively seeking blood sources (Lindsay et al., 2021; Pooseesod et al., 2021). Mosquito nets form an effective physical barrier, preventing direct contact between mosquitoes carrying

malaria parasites and sleeping humans (Hutchins et al., 2020; Sidiki et al., 2020). In addition, the presence of insecticides in bed nets can kill or repel approaching mosquitoes, increasing the effectiveness of protection. The insecticide contained in bed nets also provides longer protection against mosquito attacks, helping to keep sleeping spaces and surrounding areas in a safe zone. The importance of bed nets in malaria prevention becomes more significant in areas with a high risk of disease transmission. People living in these areas, particularly children and pregnant women who are vulnerable to malaria complications, benefit greatly from the use of bed nets during the night (Hamre et al., 2020; Omonijo et al., 2021; Unwin et al., 2023). Public health programs and malaria prevention campaigns often promote the use of bed nets as a key strategy. Mass-distributed insecticide-treated bed net programs in areas with high malaria prevalence have proven effective in reducing malaria case rates. In addition, government and WHO initiatives include the distribution of free bed nets to high-risk populations. The limitations of larval surveys may not provide a complete picture of the mosquito population because they only capture a small portion of the total mosquito population. Larval surveys may not capture the different larval habitats or breeding behaviors of some mosquito species, leading to an incomplete representation of mosquito population dynamics.

4. Conclusions

Field surveys revealed that swamps with both direct and indirect sunlight were the characteristics of *Anopheles* larval breeding sites. In three breeding places, Kaitetu village exhibited the highest density of *Anopheles* larvae, with one breeding place identified as positive for *Anopheles* larval presence. Conversely, in two other breeding places located within Hila village, no larvae were detected. The overall HI was calculated to be 33%. In addition, it was found that residents often went out at night and did not use mosquito repellent, mosquito nets, or gauze for ventilation, which are risk factors for the home environment and behavior of residents. It is critical to engage local communities and raise their awareness of malaria prevention and control measures, thereby sustaining elimination efforts. Community-based interventions, such as encouraging the use of insecticide-treated bed nets and promoting early treatment-seeking behavior, can help reduce malaria transmission rates and improve health outcomes.

Ethical Approval

This study adheres to strict ethical guidelines, ensuring the rights and privacy of participants. Informed consent was obtained from all participants, and personal information was protected throughout the study. The methodology and procedures of this research have been approved by the appropriate ethics committee. Participants were informed of their rights, including the right to withdraw from the study at any time. All collected data is used solely for the purpose of this research and is stored and processed in a secure and confidential manner.

Data Availability

The data used to support the research findings are available from the corresponding author upon request.

Conflicts of Interest

The authors declare no conflict of interest.

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