ANTI-MALARIAL DRUG RESISTANCE

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Abstrak

Tujuan studi ini adalah untuk menjelaskan mekanisme resistensi parasit malaria dan usaha-usaha yang dapat dilakukan untuk menghadapi munculnya strain parasit yang resisten terhadap artemisinin. Metode yang digunakan adalah studi kepustakaan. Resistensi *P.falciparum* terhadap obat-obat anti malaria disebabkan oleh perubahan spontan yang terjadi pada beberapa gen seperti *P.falciparum multi drug resistance1* (Pfmdr1), *P.falciparum chloroquine transporter* (Pfcrt), *P.falciparum dihydropteroate synthase* (Pfdhps), *P.falciparum dihydrofolate reductase* (Pfdhfr), and *P.falciparum multidrug resistance-associated proteins* (Pfmrp). Penyebaran resistensi tersebut dipengaruhi oleh tingkat transmisi di sebuah wilayah. WHO telah menjalankan usaha untuk menanggulangi penyebaran resistensi tersebut misalnya dengan merekomendasikan penghentian monoterapi artemisinin, dan pemberian anti malaria setelah konfirmasi laboratorium. Selain itu, perlu adanya penggunaan obat kombinasi, produksi rejimen dosis tetap, dan pengembangan obat anti malaria baru. Kesimpulan dari hasil studi ini ialah munculnya malaria resisten terhadap artemisinin akan menghambat usaha eradikasi malaria karena itu diperlukan usaha-usaha untuk menanggulanginya.

Kata kunci: resistensi obat anti-malaria, artemisinin-combination therapy (ACT), malaria resisten artemisinin, *Plasmodium falciparum*

Abstract

The objective of this study was to describe the development of anti-malarial drug resistance of the parasites and the efforts taken to contain the emergence of artemisinin resistant malaria. This was a literature study. The development of resistance to anti-malarial drugs are due to spontaneous changes in certain genes such as of P.falciparum multi drug resistance1 (Pfmdr1), P.falciparum chloroquine resistance transporter (Pfcrt), P.falciparum dihydropteroate synthase (Pfdhps), P.falciparum dihydrofolate reductase (Pfdhfr), and P.falciparum multidrug resistance-associated proteins (Pfmrp). The spread of the resistance depends on the transmission rate within each area. WHO has established a global plan to contain the spread of this resistance, such as recommendation to withdraw artemisinin-based monotherapies and administration of treatment after laboratory confirmation. In addition, administration of anti-malarial drug combination, production of fixed dose regimen and development of new drugs are necessary. The Conclusion is emergence of artemisinin resistant malaria will threaten malaria eradication thus some efforts are necessarily needed to contain it.

Keywords: anti-malarial drug resistance, artemisinin-combination therapy (ACT), artemisinin resistant malaria, Plasmodium falciparum

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INTRODUCTION

Malaria is still one of major infectious diseases nowadays, especially in Sub Saharan Africa where most deaths of the disease occur. It is caused by the parasite *Plasmodium spp* and transmitted by female *Anopheles* mosquitoes. There are currently five species known to cause malaria in human; *Plasmodium falciparum, Plasmodium vivax, Plasmodium ovale, Plasmodium malariae* and *Plasmodium knowlesi.*

Development of anti-malarial drugs to treat the disease has reduced morbidity and mortality due to malaria. WHO has achieved a very good progress in eliminating malaria, as 43 countries reported a more than 50 % reduction in the number of malaria cases in 10 years, between 2010 and 2000.[1] Globally, the number of malaria cases has reduced 17% in 2010 since 2000. These achievements are due to comprehensive ongoing efforts undertaken by WHO in eliminating the disease through several projects. However, the ability of the parasites to adapt with the drugs and eventually develop resistance will threaten WHO malaria eradication program.

First line therapy for malaria has shifted several times due to antimalarial drug resistance development of the parasites. From 1632 until 1900, the only drug used to treat malaria is quinine, which was derived from the bark of the cinchona tree and used to treat fever in Peru.[2,3] Nowadays, it is still the drug of choice to treat severe falciparum malaria.[4]

In 1934 chloroquine, a derivative of quinine, was produced and then became first line therapy of malaria for years.[3] Unfortunately, since 1957, resistant parasites to the drug started to emerge from four different foci, namely The Thai-Cambodian border region in South-east Asia, Venezuela, Magdalena valley in Colombia, and Port Moresby in Papua New Guinea (PNG), which gradually spread throughout the world.[3,5] Although resistant parasites were also found in Africa in 1978, the strain was actually similar to that in South-East Asia, leading to the conclusion that the parasite resistance spread from South-east Asia to Africa.[3] Due to the emergence of chloroquine resistant parasites, some countries considered to replace its deployment as the first line therapy of malaria. Thailand initiated this step in 1973, followed by some other countries, until 1988 when African countries also started to replace chloroquine.[3]

After the era of chloroquine deployment, a combination of two anti-folate drugs, sulphadoxine-pyrimethamine, known as Fansidar®, became the drug of choice for malaria treatment in some regions.[6] Besides, proguanil, another anti-folate drug, became the most potent prophylaxis. In contrast with the development of resistance to chloroquine which took more than 20 years, the resistance to proguanil and pyrimethamine developed promptly after its deployment in some areas.[3] Pyrimethamine resistance was firstly reported in 1967 in Thailand, soon after it was introduced in the same year.

On the other hand, some areas in Thailand started to use mefloquine replacing chloroquine in 1973, and the government officially recommended it as the drug of choice in 1984. Unfortunately, in 1990, the resistance to this drug started to emerge.[7]

In November 1999, a new venture called The Medicines for Malaria Venture (MMV) was established by WHO and some pharmaceutical industries in order to develop new anti-malarial drugs, including artemisinin (qinghaosu) derivatives which were isolated from the Chinese wormwood, *Artemisia annua*.[8] There are three different compounds have been developed, namely artemisinin, artesunate, and artemether. Currently artemisinin and its derivatives are considered as the most potent drugs to treat malaria and is used as the first line therapy as recommended by WHO.[1] Yet, some recent studies conducted in the Thai-Cambodia border region suggested that artemisinin resistant strain of *P. falciparum* has emerged in that area.[9]

Considering the occurence of resistance to anti malarial drugs as described above, it is important to understand the mechanism of the development of drug resistance of the parasites, especially *P. falciparum* which can cause the most severe form of malaria known as cerebral malaria, and to identify the factors that attribute to increase the risk of the emergence of resistant strains. Thus, the aim of this paper is to describe those mechanism and the key attributes of the resistance development. In addition, some efforts which are necessary to contain the spread or the emergence of artemisinin resistance parasites will be discussed.

METHODS

This essay is done by searching and reviewing several studies regarding anti-malarial drug resistance of *P. falciparum*.

RESULTS AND DISCUSSION

It is suggested that a spontaneous genetic change, either a point mutation or multiplication of certain genes in the parasite is the major source of resistance to anti-malarial drugs.[7]The number of mutated genes involved in the mechanism of resistance and the half-life of drugs affect the time period needed to the emergence of a resistant strain.[3] Whether a single or multiple mutations required for the development of the resistance depends on the administered drugs.[10] The resistance to anti-malarial drugs are more commonly observed in *P.falciparum* than the other *Plasmodium* species.[3]

Chloroquine resistance developed after several mutations in the parasite genes.[5] A number of genes involved in the mechanism of resistance to this drug, namely *P.falciparum* chloroquine transporter (*Pcrt*); *P. falciparum* multidrug resistance 1 (*Pfmdr1*), *P. falciparum* multidrug resistance-associated proteins (*Pfmrp*), and *P. falciparum* Na^+/H^+ exchanger (*Pfnhe*).[11]

Pfcrt which is located in the chromosome 7 encodes a trans-membrane protein in the parasite digestive vacuole (DV).[11] In the DV, the haemoglobin digestion products are polymerized by chloroquine, producing toxic haem which kills the parasite.[8] It was found that parasites with a lysine to threonine mutation at codon 76 (K76T) in *Pfcrt* accumulate less chloroquine in the DV compared to those without such mutation.[11] In the resistant strains, less accumulation of chloroquine is due to drug efflux or reduced uptake.[8]It is because mutant parasites undergo a change in the DV membrane from being positively charged to being uncharged, allowing the di-protonated drugs being actively transported out from the vacuole.[11]

Meanwhile, *Pfmdr1* which is located in the chromosome 5 encodes an ATP-binding cassette (ABC) protein which, similar to *Pfcrt*, also exists in the DV membrane.[11] Mutations in several amino acid positions in this gene decrease the accumulation of some anti-malarial drugs such as chloroquine, quinine, mefloquine, artemisinin, lumefantrine, and halofantrine, and increase the drug efflux.[11] Some studies have identified the positions of mutations in the gene that might confer resistance in the parasite, such as asparagine to tyrosine mutation at codon 86 (N86Y), serine to cysteine at codon 1034 (S1034C), asparagine to aspartic acid at codon 1042 (N1042D), and aspartic acid to tyrosine at codon 1246 (D1246Y).[12]

Besides, *Pfmrp*, which is located on chromosome 1, encodes a protein that is located in the plasma membrane and membrane-bound vesicles of the erythrocytic stages of the parasite.[11] In contrast with mutations of *Pfcrt* and *Pfmdr1* which cause decrease susceptibility of the parasite to anti-malarial drugs, mutation in *Pfmrp* results in an increase anti-malarial susceptibility of the parasites.[11]

On the other hand, the parasite might confer resistance to anti-folate drugs such as sulphadoxine-pyrimethamine when point mutations occur in some genes involved in folate pathway, namely*P. falciparum* DHPS (*Pfdhps)* and *P. falciparum* (*Pfdhfr*),because anti-folate drugs act by inhibiting the enzymes used in the folate synthesis such as dihydropteroate synthase (DHPS) and dihydrofolatereductase (HFR).[10],[11]DHPS is inhibited by sulfonamide, while DHFR is inhibited by pyrimethamine.[10]

Finally, even though it has been suggested that artemisinin resistance parasites might have emerged, the mechanism of the resistance to this drug is still unclear.[1] Some studies have been conducted focusing on several mutations that occur in the genes that involved in chloroquine resistance, but none has proposed the true mechanism of artemisinin resistance.[11]

In addition to spontaneous mutations, it was proposed that the resistance to certain anti-malarial drugs are influenced by several other factors, such as mutation rate, high parasite load, the strength of the drug selection, and the treatment compliance. Exposure to sub-optimal drug concentration due to inappropriate dose, fake or counterfeit drugs, and reinfection during anti-malarial elimination phase, which commonly occur in high-transmission areas, also attribute to the emergence of resistance.[11]

Geographical difference in the emergence of resistant parasites

As mentioned before, choloroquine resistant parasites emerged from four different foci, 2 in south America, 1 in South East Asia, and 1 in Papua New Guinea.[3,5] This was concluded from the result of molecular analysis of the parasite resistant strains in those foci.[5] Although most malaria deaths occur in Africa, there is no original chloroquine resistance from this region.[5]

Parasites that confer mutations do not always survive, depending on the local epidemiology of the disease, such as the transmission rate of mutated parasites and their selective advantage.[5] In addition, the risk of increased drug pressure depends on the treatment seeking behavior of the infected patients which is influenced by their immunity to the infecting agents.[7] Hence, the difference in the intensity of parasite transmissions causing different immune responses of population in each area results in different geographical distribution of anti-malarial resistant parasites.[7]In a low transmission area, such as South East Asia, selective mutations leading to resistance to anti-malarial drugs are more likely to occur and disseminate since most of the population are non-immune, and consequently most infections are symptomatic and require treatments.[5] On the contrary, in the areas with high transmission rate such as in Africa, most of the population have developed immunity, resulting in asymptomatic disease and less drug deployment.[5,7]

However, in high transmission areas, drug pressure might be higher when infected patients take anti-malarial drugs with long half-lives, which can remain in sub-therapeutic level in the blood for weeks.[7] Those people are more likely to get a re-infection, thus the newly infecting parasite is more likely to be exposed to the remaining low level of drugs, increasing the drug pressure. In addition, transmission of mutant parasites may occur more easily in high transmission areas because when an anti-malarial drug is administered, sensitive parasites will be killed but the resistant ones are not.[7] Therefore, instead of transmitting the sensitive strains, mosquitoes will only transmit the mutant parasites.

Pharmacological development to contain anti-malarial resistant parasites

It is very important to identify the efforts that can be undertaken to contain the resistance to anti-malarial drugs. Some strategies that have been done are reducing monotherapies, deploying drug combination therapies, administration of antimalarial drugs after laboratory confirmations, and developing some new drugs.

It has been suggested that mono-therapy may induce resistance in the parasites more rapidly compared to combined drugs, as observed in a rapid atovaquone resistance development when the drug is used as monotherapy, yet the resistance developed more slowly when it is used in combination with other drugs, such as tetracycline or proguanil.[10] Thus, one effort to contain the emergence of anti-malarial resistance is withdrawing the available monotherapy regimen, particularly artemisinin monotherapy that are still produced and marketed in some countries.[1,8]

In addition, as suggested by Professor Wallace Peters, drug combinations should be used for malaria treatment.[8] Therefore, the current first line therapy is artemisinin combination therapy (ACT) which consists of artemisinin derivatives and a partner drug with longer half-life and different pharmacodynamic.[1,13] Another alternative of anti-malarial drug combination is quinine with tetracycline or doxycycline as the second line therapy in some countries.[13]

Moreover, it is assumed that using non-fixed combination of anti-malarial drugs will reduce patient adherence to the drugs, resulting in an increased risk of resistance development of the parasites compared to using fixed-dose combinations.[7] Therefore, at present, some fixed co-formulated combinations of anti-malarial drugs are being developed, such as Co-Artem®, consisting of artemether-lumefantrine, Artekin®, consisting of dihydroartemisinin and piperaquine, and Amonate FDC®, consisting of artesunate-amodiaquine.[7,14]

Furthermore, responding to the report of the emergence of artemisinin resistance emergence in the Greater Mekong region, South East Asia, WHO has established the global plan for artemisinin resistance containment (GPRAC) in 2011. This includes

conducting drug efficacy monitoring in every malaria endemic countries every 24 months, and elimination of counterfeit or substandard anti-malarial drugs.[1]

Another very important effort is to avoid administration of anti-malarial drugs before laboratory confirmation either by microscopy examination or rapid diagnostic test (RDT).[1]The administration of anti-malarial drugs in non-infected patients may increase the risk of resistance developmen.

Finally, Medicines for Malaria Venture (MMV) has provided funding and research for new drugs development for uncomplicated malaria treatment which are efficacious to combat the resistant strains, with single dose administration and short term duration. There are three strategies used in this project, namely re-designing existing drugs, using older drugs in new ways, and targeting new specific sites in the parasite. Currently there are 9 undergoing projects which is being tested in clinical trials, and 12 projects in exploratory phase.[15]

One of the drug combinations that are currently being studied is chloroproguanil and dapsone.[8] Chloroproguanil-dapsone (Lapdap)-artesunate acts against DHFR and DHPS, thus might be effective to combat sulphadoxine-pyrimethamine resistant para-sites. Besides, combinations of artemisinin derivatives with some drugs that have been used as monotherapy for almost 20 years in China as malaria treatment such as pyro-naridine and piperaquine are currently devel-oped. Those combinations include pyronar-idine-artesunate and piperaquine-dihydroar-temisinin.

Other drugs being developed include synthetic endoperoxide based drugs, such as OZ277, which has been proven clinically effective in mice with unclear mechanism of action, pyridines, a new drug targeting the destruction of the parasite mitochondria resulting in the parasite cell death, and DB289, an analogue of pentamidine which has been proven to be effective in a phase I trial to treat malaria in a 3 day course therapy with single dose.[15]

Besides that, studies to improve the effectiveness of some drugs that have been proven to be effective such as Co-Artem and amodiaquine are also being done. These include development of a powder paediatric regimen of Co-Artem to improve treatment for infants and children and a modification of amodiaquine known as isoquine to reduce the amodiaquine toxicity, without affecting its efficacy in treating chloroquine resistant parasites.[15]

CONCLUSION

The first line anti-malarial drug has been changed several times due to the ability of malarial parasite to develop resistance to the drug. Resistance occur as a result of genetic changes in the parasite due to point mutation or multiplication of certain genes. The resistance spread and occurrence in every region could be different depending on the parasite transmission rate. Since the emergence of the resistant parasites has been a major threat to WHO malaria eliminating program, some pharmacological development are currently being undertaken, such as administration of anti-malarial drug combination, production of fixed dose anti-malarial regimen, and development of new drugs.

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