CHAPTER TWO

The contributions and achievements on malaria control and forthcoming elimination in China over the past 70 years by NIPD-CTDR

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Abstract

Although the past decades have seen a remarkable decrease in malaria-caused mortality and morbidity, the infection remains a significant challenge to global health. In the battle against malaria, China has gained notable feat and achievement since the 1940s through the efforts of several generations. Notably, China has not recorded a single indigenous malaria case since August 2016. The National Institute of Parasitic Diseases of the Chinese Center for Disease Control and Prevention (NIPD), as the only specialized institution for parasitic disease at the national level, has played a significant role in the malaria control, prevention, and elimination in China in the different historical periods. In order to transfer Chinese experiences on malaria control and elimination to other Low and Middle Income Countries (LMICs) and to improve global health collaboration, we have summarized and reviewed the contributions and achievements by the NIPD over the past 70 years, covering the epidemic situation; field investigation and laboratory experimental research on both parasite and vector; research and development on diagnostics, drugs, and insecticides; surveillance and response; technical and international. Support and cooperation. In addition, we also focus in particular on malaria retransmission risk, strategies on management of imported malaria cases and mobile populations, surveillance and response capacity to be maintained in post-elimination stage, challenges on diagnosis, drug resistance, and insecticide resistance as future concerns.

1. Introduction

Malaria is one of most serious vector-borne diseases transmitted by the *Anopheles* mosquito and is one of the major infectious diseases that has historically affected social development in China (Qian and Tang, 2000). Malaria has been prevalent in most parts of China, except for Gansu, Ningxia, Qinghai, Jilin, Inner Mongolia, and some other provinces,

in which malaria cases were occasionally reported or were malaria-free areas. In the early days after the founding of the People's Republic of China, malaria was epidemic in nearly 80% of the counties in the country, and 70% of the population was under threat (Tang, 2000). In 1954, according to an incomplete statistical estimate, there were 6.97 million malaria patients nationwide (Taiwan not included), with an annual incidence rate of 12.29 per 1000. Malaria was most serious in the southwestern border areas, such as Hainan Island and Yunnan, and was mainly falciparum malaria. Even in the 1970s, there were still more than 24 million cases. By 2010, after several decades of active prevention and control, only 7855 cases of malaria were reported nationwide, and the incidence rate dropped to 0.06 per one million (Yin et al., 2014). In 2010, in response to the global malaria eradication initiative proposed by the United Nations (UN) high-level meeting on the Millennium Development Goals, 13 ministries and commissions, including the Ministry of Health, jointly issued the China Malaria Elimination Action Plan (2010–20), which was proposed to eliminate malaria in the majority of the country by 2015, with the goal of complete malaria elimination by 2020 (Diouf et al., 2014; Zhou et al., 2013). Most strikingly, in 2017, China reported no indigenous malaria case reports for the first time (Feng et al., 2018).

China's malaria prevention and treatment work has achieved great progress in the past decades, resulting in effective control of the malaria epidemic and ensuring the health of its people along with the promotion of social and economic development (Shou-Pai, 1983). The National Institute of Parasitic Diseases of the Chinese Center for Disease Control and Prevention (NIPD), the national parasitic disease prevention and control work guidance centre, has played a central role in the prevention and treatment of malaria in China. In 2017, the State Commission Office of Public Sectors Reform formally approved a new title of the "Chinese Center for Tropical Diseases Research" and affiliated it with the NIPD, China CDC (NIPD-CTDR).

In the past 70 years, the NIPD-CTDR has played an important role in the demonstration, on-site prevention, organization, and implementation of parasitic disease control programmes and handling of emerging diseases outbreaks. The NIPD-CTDR has established a national malaria diagnostic reference laboratory network, parasitic disease prevention, and research base, as well as support platforms for monitoring and early warning, diagnostic testing, media control, and information services. National malaria surveillance, prevention and control of malaria epidemic within the central

region, and prevention and control of non-malaria cases have been carried out under the guidance of the NIPD-CTDR. The research field of NIPD involves global disciplines, applied epidemiology, pathogenic and vector biology, immunology, molecular biology, pharmacy, and other multi-disciplinary research methods and modern technologies, and the institution also analyzes popular epidemic characteristics and transmission of important parasitic diseases and tropical diseases. The NIPD-CTDR has consistently focused on scientific research based on disease control and prevention, including laboratory work and field needs, and has cooperated closely with disease prevention and control institutions at all levels across the country to fight side-by-side, ensuring the completion of prevention and research tasks. The NIPD-CTDR has led the national parasitic disease control and prevention programme and achieved a number of scientific and technological achievements (Table 1). This review discusses the key role of the NIPD-CTDR in the control of parasitic disease epidemics within China, progress of parasite biology, vector research, prevention and control strategies, case management monitoring, and related technical support for the national malaria control and elimination programme.

2. Epidemic situation of malaria in China

Since the founding of the People's Republic of China, malaria has presented with different epidemiological characteristics during different historical periods.

2.1 Key investigation and research stage (1949–59)

Before the founding of the People's Republic of China in 1949, historical documents on malaria were very scarce. Endemic instances of malaria were only reported and recorded in a few provinces, such as Yunnan, Guangxi, Guangdong, and Guizhou. From 1935 to 1938, Lanzhou Feng published "China's Malaria Epidemiology" and other scientific papers, which outlined the regional and seasonal distribution of malaria within China, as well as the types of malaria parasites and vectors, and summarized the main characteristics of the malaria epidemics within China (Yin et al., 2014). From 1947 to 1949, the former Central Health Laboratory conducted a dichloro-diphenyl-tricgloroethane (DDT) indoor residual spraying and mosquito killing experiment in Dongshan Town, Nanjing. From 1951 to 1952, the Central Health Research Institute Huadong Branch worked on *Anopheles sinensis* in Pengxingzhou and Nanjing South Suburb of Dangtu County,

Table 1 Primary research focus of the NIPD during different time periods.

Period (year)	Research area of NIPD		
	Parasite	**Vector**	**Control and elimination strategies**
1949–59	• Plasmodium species investigation • Establishment of animal models of malaria • Development of new antimalarial drugs	• Investigations on malaria vector distribution • Investigations on malaria vector species • Investigation on vector competence	• Investigations on malaria epidemic situation • Classification on epidemic areas • Pilot study on malaria control prevention • Guidelines and comprehensive control strategies
1960–79	• In vitro culture of *Plasmodium falciparum* • Establishment and improvement of anti-malaria drug sensitivity • Multi-drug resistance testing • Indirect fluorescent antibody test (IFAT) • Enzyme-linked immunosorbent assay	• Investigations on malaria vector distribution • Research on *Anopheles sinensis* • Investigations on insecticide resistance	• Radical treatment on vivax malaria • Malaria vector control • Investigation on the degree and geographical distribution of drug resistance to chloroquine
1980–99	• The establishment of a single epitope monoclonal antibody against P. falciparum • Biological research on *Plasmodium vivax*	• Investigations on insecticide resistance • Research on vector competence of *An. sinensis* and other species	• Epidemiological investigation and surveillance strategies • Control strategies on malaria outbreak • Vector control
2000–09	• Identification of Plasmodium species by PCR method • Drug resistance surveillance	• Research on *Anopheles pseudovespa* in Tibet • Susceptibility study on different vector species	• Malaria control strategies • Optimization and adjustment on surveillance and control strategies
2010–20	• Molecular marker for drug resistance • Development of new antimalarial drugs • Drug resistance surveillance and related mechanism research • Development of novel antimalarial drugs	• Surveillance on insecticide resistance • Molecular studies on main malaria vector developmental immunity • Development of novel vector control strategy	• Research on malaria elimination strategy • Development on the 1-3-7 strategy • Malaria surveillance post-elimination • Risk prediction and assessment post-elimination • Pilot study in African countries

Anhui Province, performing some minor antimalarial test interventions, such as residual spraying or larval killing of mosquitoes in wintering places. At that time, 239 people were surveyed in the urban area of Quanjiao County, Anhui Province, and were found to have an infection rate of 41.4% (of which *Plasmodium falciparum* accounted for 74.7%) and a splenomegaly rate of 74.8% (Tang, 2009; Zu-Jie, 1981).

The number of malaria cases was the highest of all infectious diseases at the beginning of the founding of the People's Republic of China. Malaria was prevalent in 1829 counties (cities), accounting for 70%–80% of the counties at that time. In 1954, the Ministry of Health conducted surveys in Yunnan, Guizhou, Guangdong, Guangxi, Hubei, Sichuan, Jiangxi, Henan, and other provinces and found that the number of malaria cases accounted for about 60% of 19 infectious diseases surveyed. Malaria was more prevalent in Yunnan, Guizhou, Guangdong, and Guangxi than in other provinces, especially in the southwestern border areas of Hainan Island and Yunnan. There were local outbreaks in some places in north and central China, such as Yongzhou Hunan, Xinyang Henan, Quanjiao Anhui, and Xingcheng Liaoning. The epidemic pattern of malaria seriously jeopardized the health of the people. Anaemia and hepatosplenomegaly caused by malaria was very common in these areas. South of 25° north latitude, malaria was highly endemic in most areas and only a few areas were moderately endemic. Generally, plains areas were low or moderately endemic with an unstable pattern, while mountainous areas were highly prevalent with a relatively stable pattern. From 25 to 32° north latitude, malaria was very prevalent and mainly caused by vivax malaria with high population density where the epidemic was unstable. There were often low or moderate malaria outbreaks in this region. The malaria situation in the mountainous area was more serious than in the plains area. To the north of 32° latitude, the malaria epidemic was relatively mild. The transmission period was between 3 and 6 months, and the splenomegaly rate was generally below 10%. Malaria was mostly distributed in the low-lying areas along rivers and lakes. The immunity of the residents was very low, which often led to outbreaks. In the northwestern region, annual malaria cases ranged from 9000 to 18,000, and the annual incidence rate was 179–364/100,000. The incidence rate north of Xinjiang was higher than that south of Xinjiang. The epidemic areas were mainly distributed in swamps, low-lying areas, and some rice-growing areas on both sides of the river. Malaria maintained a low endemic profile near the Kashgar, Yarkant, and Tianhe rivers in southern Xinjiang. After 1958, malaria prevention and treatment was also

affected by the "the Great Cultural Revolution," which derailed the situation at that time. In addition, after severe natural disasters in 1959, malaria prevention and control work was stopped and stagnated for many areas. In 1960, malaria outbreaks occurred in many areas. The incidence rate in 1960 was 1.8 to 600 times higher than that in 1959 in Hebei, Shandong, Henan, Jiangsu, and Anhui provinces alone, and the number of malaria cases was as high as 9.53 million.

2.2 Control of the pandemic transmission stage (1960–79)

From 1960 to 1979, comprehensive prevention and control strategies were planned and developed based on the investigation, prevention, and control of malaria in China during the 1950s. However, the malaria prevention and control work during this period was disrupted by the impact of natural disasters, the country's economic difficulties, shortage of funds for prevention and treatment, tight supply of antimalarial drugs, and the impact of social unrest, resulting in repeated epidemics and poor control effects (Zu-Jie, 1981). In the early 1960s and 1970s, vivax malaria was prevalent in the Huanghuai Plain and Jianghan Plain within the central region. According to reported data, this period had the most serious malaria epidemic in China after the founding of the People's Republic of China. In the 20 years from 1960 to 1979, more than 180 million malaria cases were reported nationwide, and the highest case number was reported in 1970 at about 24 million cases, with an incidence rate of 296 per million. The highest number of mortalities was 2,049 in 1963. During this time, the number of malaria cases in the country accounted for 12.40%–63.15% of the total number of infectious diseases reported in the epidemic, indicating that malaria was the most serious infectious disease in China. According to statistics of counties (cities, districts), the prevalence in some regions was very high. For example, during 1973–75, 46.92%–63.85% of the counties (cities, districts) in the central region, including Jiangsu, Henan, Anhui, Hubei, and Shandong provinces, had an annual incidence rate of more than 1% (Kai-Jie et al., 2016). Moreover, the incidence in 14.39%–26.61% of the counties (cities, districts) was above 10%. By 1979, many historical endemic areas still had an incidence rate above 1% (Fig. 1A and B).

2.3 Intensive prevention and control stage (1980–99)

Since the implementation of comprehensive prevention and control programmes starting in 1971, the epidemic situation of malaria in the

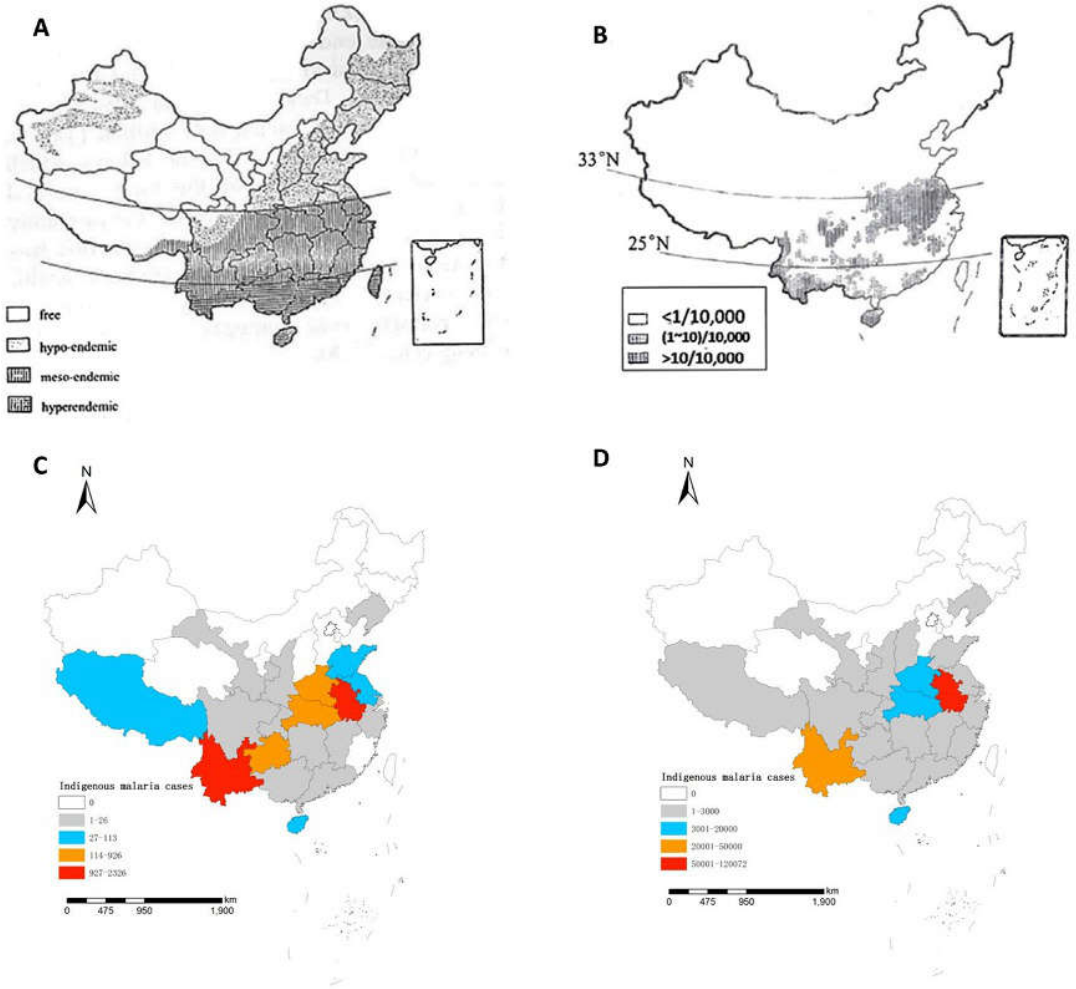

Fig. 1 The incidence and prevalence of malaria in China during different periods. (A) Malaria epidemics during the 1970s. (B) Malaria incidence rates in the 1980s. (C) Malaria epidemics in China from 2002 to 2009. (D) Malaria epidemics in China during 2010–16. Since the beginning of 2017, there have been no local malaria cases reported in China.

country has changed significantly (Huang, 1980). The severe epidemics that raged during the early 1970s are now effectively under control. The incidence of malaria in the country has dropped to its lowest level (257.54/100,000), except for the rate in 1958 and 1959. Between 1980 and 1999, the malaria epidemic in the country was in a stable downward trend, and some areas also experienced spot-like or patchy outbreaks (Goodman, 1986). Except for the rebound in 1989 and 1994 (Sleigh et al., 1998), the efforts of continuous implementation of various prevention and control measures has resulted in the gradual decline in the incidence of malaria in the country every year, as compared to the previous year. After 1980, the prevention and control work was strengthened throughout the country. Through continuous

prevention and control, some counties (cities) gradually eliminated malaria, and the incidence of malaria still continued to decline every year in other counties (cities). Due to the strengthening of the prevention and treatment of falciparum malaria, the number of endemic provinces (districts) in China have been decreasing every year. After 1990, falciparum malaria was prevalent in only Hainan and Yunnan provinces, and the morbidity and mortality rates reported in those regions were significantly reduced (Xiao et al., 2012). In 2010, there were only 17 counties in Yunnan Province with falciparum malaria. In 1980, China was at the height of a 10-year cycle of malaria epidemics. According to the epidemic report, the annual national incidence rate was 337.87/100,000, which was 31.17% higher than the previous year.

From 1980 to 1999, the prevalence of malaria in the country was gradually reduced, and the counties (cities, districts) with high incidence rates have continued to decrease every year, while those with low or previously malaria-free areas have slowly reported increased malaria cases. However, the public health system was established under the planned economic system during this stage. The grassroots health organizations have been greatly affected by the formation of the market economic system. The under-reporting rate for malaria might have been high according to the monitoring and special surveys at the province level. As such, the actual number of malaria cases in the country should have been greater, and the actual prevalence might be higher than the situation reflected in the epidemic report.

2.4 Consolidation of the achievements of prevention and control (2000–09)

After 50 years of prevention and control, the malaria epidemic was controlled in most parts of the country, and the malaria cases were mainly concentrated in areas where episodes of malaria were typically more common. Since 2000, although the incidence of morbidity has increased in some areas, the overall epidemic has been stable and maintained at a low level. According to the epidemic report, the national incidence was in a dynamic trend from 2000 to 2009 (Fig. 1) (Expert Advisory Committee on Malaria, Ministry of Health, 2001). The epidemic situation was mainly caused by the malaria outbreak in the Huaibei District of Anhui Province. The prevalence of malaria was still relatively stable in other parts of the country. From 2000 to 2009, the prevalence of malaria in most areas of

China remained stable at an annual incidence rate of about 1/100,000 (Feng et al., 2015a). However, due to the rebound or outbreak of malaria in the Huaibei area of Anhui Province, the annual incidence rate was over 10/10,000. The number of high incidence rates in counties (cities, districts) increased (Fig. 1C). At this stage, the under-reporting rate of malaria in primary health care institutions was still serious, especially in high-endemic and unstable transmission areas. In 2002, the Global Fund-China Malaria Project verified the actual malaria incidence in 10 project provinces (Wang et al., 2014a). The results showed that the actual number of malaria cases in each province in 2002 was 3.95–100.09 times the number in epidemic reports, and the actual number of cases in 10 provinces was 20.55 times higher than the number reported in epidemic reports. In 2003, the Malaria Expert Advisory Committee of the Ministry of Health conducted a serious discussion and analysis on the actual incidence of malaria in the country. According to the survey results, the experts estimated that the actual number of malaria cases in the country in 2002 was about 740,000, which was 20.96 times the number of epidemic reports (35,300). The under-reporting of cases was mainly concentrated in areas where social and economic development was relatively lagging, grassroots health organizations were weak, and malaria was more common as an infectious disease. By contrast, the incidence of epidemic situation in most regions with prevalence of counties (cities, districts) below 1/10,000 was comparable to the actual incidence in these regions and areas, basically consistent with the situation reflected in the epidemic report. However, the actual prevalence of the county (city, district) with an incidence rate above 10/10,000 was much higher than that in epidemic report.

2.5 Approaching the malaria elimination stage (2010–20)

In order to construct more efficient malaria elimination strategies in China, 2858 counties in China were divided into four categories according to the local malaria epidemic situation in 2006–08, with 75 counties in tier 1, 678 counties in tier 2, 1432 in tier 3, and 664 in tier 4. These counties were divided as such to help explore and accelerate the goal of eliminating malaria, cope with the challenge of imported cases, and promote work in all areas by drawing upon the experience gained on key points (Sheng et al., 2016). In 2010, the national malaria elimination programme (NMEP) decided to choose malaria endemic counties and selected a tier of counties (districts)

as pilot counties and initiated pilot work to eliminate malaria in six provinces (cities), including Shanghai, Zhejiang, Fujian, Shandong, Hebei, and Guangdong. Since the launch of the action plan, the national malaria epidemic has declined overall. The number of cases and prevalence of indigenous infections have continued to decrease (Hu et al., 2016). In 2016, only three indigenous cases were reported in Yingjiang County of Yunnan Province and Chayu County of Tibet District (Fig. 1D), which was a 99.9% decrease from 2011 (4262 cases) (Li et al., 2016). Furthermore, the distribution range also decreased from 155 counties (districts) in 13 provinces (municipalities and autonomous regions) in 2011 to 2 counties (districts) in 2 provinces (autonomous regions) in 2016. In terms of parasite species classification, 10,589 cases (31.4%) of vivax malaria, 16,211 cases (48.1%) of falciparum malaria, 1669 cases (4.9%) were oval malaria, 383 cases (1.1%) were malaria cases, 396 cases were of mixed species (1.2%), 2 cases were *Plasmodium knowlesi* infection, and 503 cases (1.5%) were not classified. There were also 3884 clinically diagnosed cases (11.5%). It is particularly noteworthy that in 2017, for the first time, China did not report any indigenous cases of malaria (Fig. 2) (Feng et al., 2018).

Fig. 2 National report on the incidence of malaria during 1993–2018.

3. Monitoring and information management

In terms of the current malaria status and malaria epidemic characteristics and trends in China, it is necessary to carry out planned, continuous, and systematic monitoring of malaria cases, vector, and drug resistance, including control, elimination, and post-elimination (Fig. 3) (Feng et al., 2014b). The monitoring objectives, monitoring priorities, and blood tests of fever patients are different at each stage. According to the National Malaria Elimination Surveillance Programme (2015 Edition), malaria surveillance includes an elimination and post-elimination phase. Counties (cities, districts) that have not yet passed the provincial malaria elimination assessment work should follow the monitoring requirements of the elimination phase. Counties (cities, districts) that have passed the provincial malaria elimination and non-malaria endemic counties (cities, districts) should carry out work in accordance with the post-elimination monitoring requirements. Elimination phase monitoring includes: case discovery and reporting, case review and epidemiological case investigation, epidemic investigation and classification, active screening of returnees, sentinel hospital monitoring,

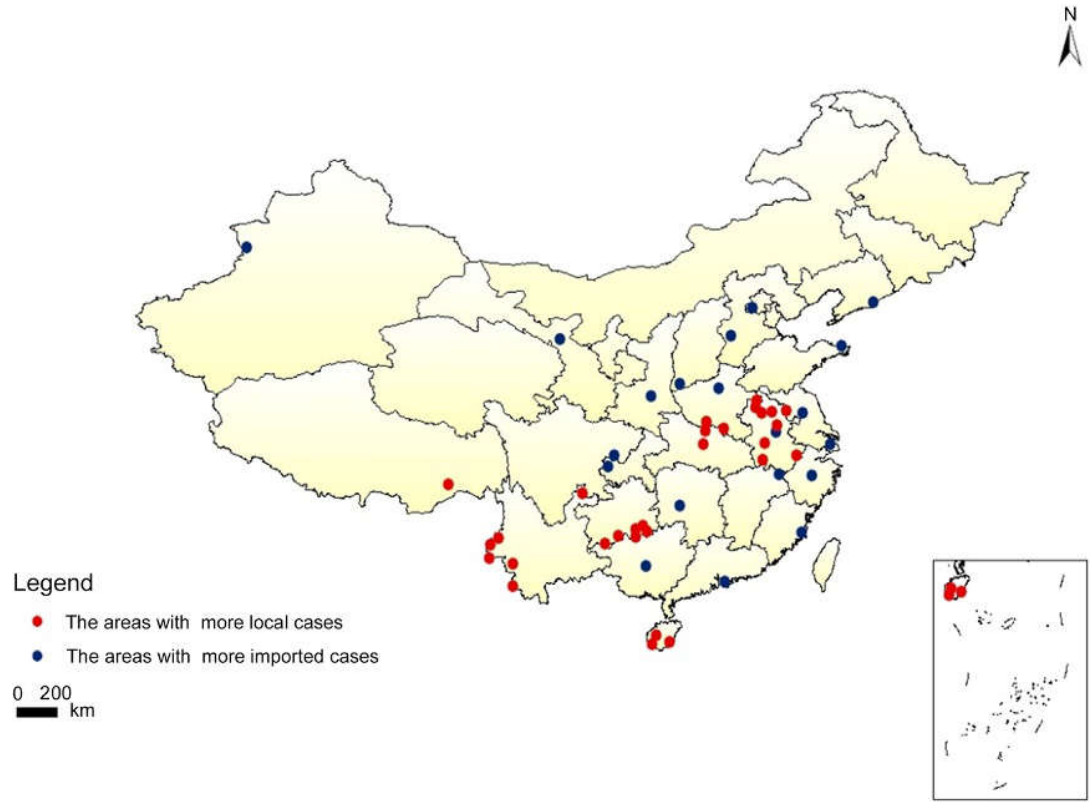

Fig. 3 Distribution map of the national malaria surveillance sites in 2014.

antimalarial drug sensitivity monitoring, and insecticide sensitivity. Post-elimination monitoring should include case monitoring, vector monitoring, and anti-malaria drug resistance.

The malaria control stage aimed to understand the current status of malaria transmission and its influencing factors and to master the prevalence and trends of malaria in order to provide a scientific basis for evaluating the prevention, treatment, and development of malaria control strategies. In the early 1960s, research at the NIPD regarding malaria epidemiology and its prevention and control in China mainly focused on two tasks. First, it was about radically curing vivax malaria. Most of China's mainland was in the northern temperate zone, where the spread of malaria began in the spring and stopped in the winter. This was conducive to the implementation of a radical treatment for vivax malaria. Therefore, it was proposed to carry out anti-relapse treatment during the malaria transmission period as one of the key measures to eliminate vivax malaria and to formulate a technical plan for malaria control. Monitoring during the elimination stage mainly focused on the timely removal of infection sources and blocking possible transmission via the timely detection of each source of infection and possible epidemic of transmission. Post-elimination monitoring is a key measure to maintain work capacity, consolidate and eliminate outcomes, and prevent secondary transmission caused by imported malaria.

The malaria control stage focuses on suspected cases, clinically diagnosed cases, and confirmed cases. The NIPD has tried to identify malaria outbreaks likely to occur, but laboratory detection and diagnosis were not emphasized in this stage. Monitoring during the elimination phase is based on the improvement of overall malaria prevention and control ability, focusing on confirmed cases, strengthening the importance of laboratory diagnosis, reducing clinical diagnosis cases and suspected cases through the establishment of malaria diagnostic reference laboratories, and improving the laboratory detection rate and proportion of confirmed cases. The monitoring in post-elimination stage focuses on the risk assessment of reintroduction, emphasizing the definition of imported and local infections and assessing transmission risk through case screening and vector investigations. For example, in 2013–18, a total of 18,658 malaria cases were reported nationwide, with a 24-h case report rate of 100.0%; 18,525 cases were detected in laboratory with a detection rate of 99.3%; and 18,336 cases were confirmed in laboratory with a coincidence rate of 98.3%. The epidemiological investigation was conducted among 17,773 cases with rate of 95.3%, and 12,509 related foci were disposed within seven days.

During the malaria control stage, the cases with suspected malaria and unexplained fever were included in the blood test. The annual blood test rate should be not less than 5% in the area with high incidence (1/1000 or more, the township as the unit), while the annual blood test rate should be not less than 2% in the unstable area (1/1000 to 10,000). In the areas with lower incidence (less than 1/10,000), the annual blood test rate should be not less than 1%. The blood test for fever patients should be carried out throughout the year. In the elimination stage, patients with fever for unknown reasons are included in the blood test. In first and second class counties (the township as the unit), the number of blood tests per year should be not less than 1% of the total population of the jurisdiction. In the third class of counties, the number of blood tests per year should be not less than 0.5‰ of the total population of the jurisdiction. The number of patients with unexplained fever during the spread season should be not less than 80% of the total number of blood tests. In the post-elimination stage, surveillance is conducted in sentinel hospitals in areas with high retransmission risk rates for patients with unexplained fever.

4. Technical support to the malaria control and elimination programmes

The control and elimination of malaria is not only an improvement of disease prevention and control goals, but also a major shift in disease prevention and control strategies. The transformation from the traditional "comprehensive prevention" mode to the new "eliminate foci and infection source" model was based on new theories, techniques and methods, and innovations and breakthroughs in key technologies. Over the years, at the technical level, in order to ensure the effective implementation of the elimination of malaria strategy measures and the timely realization of the elimination of targets, the NIPD has fully guaranteed the planning documents, funding, human resources investment, supervision and inspection, and international cooperation.

In order to strengthen the technical guidance for national malaria research and prevention work, the Ministry of Health established the Malaria Special Committee, consisting of the experts and secretaries of the NIPD, in March 1981. Since the establishment of the special committee, substantial work has been done in coordinating national malaria research projects, organizing key scientific research projects, conducting prevention and treatment technical guidance, and formulating national malaria prevention and control plans,

regulations, and regulations. Among them, in the early 1960s, the NIPD's research on malaria epidemiology and its prevention and treatment in China mainly focused on some key points. For instance, it aimed to eliminate vivax malaria in most of China's mainland since most of the areas were located in the northern temperate zone, which created the opportunity to block the spread of malaria in the winter and spring and was conducive to the implementation of elimination. Therefore, it was proposed to carry out anti-relapse treatment outside the malaria transmission period as one of the measures to cure and eliminate vivax malaria. This finally formulated a technical plan for malaria control. In the early 1960s, tens of millions of mass prevention campaigns were also promoted by the technical plan. The "eight-day therapy" programme was first proposed. In 1963, the "four-day therapy" with less adverse reactions and easier application was proposed. In the same period, technical groups from the NIPD were sent to the four provinces to conduct on-the-spot investigations, write survey reports, and select Jiangsu Binhai County to track the investigation of outbreak trends.

From 1991 to 1994, the NIPD led a collaboration group with 23 counties (cities) in 11 provinces (cities), such as Henan and Hunan, and conducted malaria prevention and control in nearly 15 million population areas (*An. sinensis* as main vector) to study new features of epidemiology and to monitor programme research (Liu et al., 1996). A multidisciplinary approach, such as parasitology, serology, vector biology, and health economics, was used to investigate the new epidemiological characteristics of malaria prevention and control in this area and to explore economic and feasible simplified monitoring strategies. The transmission potential of *An. sinensis* was assessed by vector capacity, and the transmission intensity of malaria transmission index was calculated by the ratio of secondary cases induced by imported cases to imported cases. The cost and effect of the monitoring programmes were evaluated by a comprehensive effect index. The study found that in the later stage of malaria control in China, the malaria transmission was low, and the epidemic situation was stable. The main cases were imported, but rarely spread. New features of epidemiology included the epidemic foci, which were scattered, and the proportion of typical clinical symptoms of patients, which increased. The human blood index and vector competence of *An. sinensis* were significantly reduced, indicating that the potential of malaria transmission in this region has been at a low level. According to the results of the implementation of the experimental area and the new characteristics of epidemiology, the monitoring measures in this area are proposed as the blood test for fever patients with

malaria and suspected malaria. The annual blood test rate is 0.3%, which can simplify the management of malaria in the mobile population and monitor the programmes for epidemic treatment, thereby saving manpower and anti-malaria funds. The research results won third prize for the National Science and Technology Progress Award in 1998.

After the national malaria elimination programme launched in 2010, more intensive work on the surveillance and response in all 24 malaria endemic areas. With strengthening surveillance and response system, particularly to identify the local and imported malaria cases, the implementational works on surveillance and response were standardized as "1-3-7" surveillance approach, which means case reporting within one day, case investigation within three days and focus investigation and action within seven days (Cao et al., 2014). In recent years, in order to ensure the quality of the "1-3-7" work, the National Anti-Malaria Technical Expert Group conducts an audit analysis of each reported case every quarter (2017) or semi-annual (from 2018) to ensure that each case is confirmed, and the information is accurate and reliable. Also, the National Critical Malaria Rescue Expert Group conducts an annual review of each death case to find out the cause of death and improve the ability of each province to diagnose and reduce the number of death. In 2017, the expert group reviewed 2651 cases from 24 malaria endemic provinces, of which 58.2% were qualified in the initial examination, and 92.5% passed the expert review. In 2018, the expert group reviewed 2546 cases from 24 malaria epidemic provinces, of which the rate of the preliminary examination was 74.27%, and the confirmation rate of the expert review was 98.08%. From 2016 to 2018, the NIPD trained 85,016 laboratory inspectors in the nationwide institutions. In addition, 256,694 clinical doctors were trained, including 44,977 ICU doctors, 20,489 doctors from communicable diseases department, and 191,228 doctors from other departments. At the same time, the National Health and Health Committee holds the National Parasitic Disease Prevention and Control Technology Competition every year. Through selection and competition, it has received good results in promoting competition. The NIPD was responsible for training and providing technical support and guidance to provincial laboratories and regularly assessing the microscopic examination and genetic testing capabilities of provincial laboratories. The NIPD also carried out parallel detection and confirmation of specimens from malaria cases with controversial results in provincial laboratories, established a national malaria case information database for the exchange and sharing of relevant information, and researched new technologies and

methods related to malaria parasite diagnosis. In addition, the NIPD actively conducts international exchanges and cooperation, introduces and promotes the application of advanced and applicable technologies, and makes full use of internationally funded projects, such as the Global Fund to support malaria elimination operations. For example, the NIPD has established a cross-border malaria prevention and control cooperation mechanism to accelerate the control and elimination of malaria in China's border areas. Since 2015, in order to promote cross-border cooperation on malaria elimination in the China-Burma border region, the China CDC and relevant departments in Myanmar have held annual seminars and invited the National Health and Health Commission, WHO, Health Poverty Alleviation, and other departments and international organizations to participate. The strategic plan for malaria elimination in the border areas of China and Myanmar was drafted, and the "same strategy in the same region" was taken as the guiding ideology to jointly promote the joint prevention and control of malaria in the border areas. The NIPD has actively contacted Southeast Asia and relevant African institutions and has launched and implemented the China-British-Tanzania Malaria on-site prevention and control project and the China-Australia- Papua New Guinea malaria prevention and control project, which has effectively reduced local malaria morbidity and mortality (Wang et al., 2019; Xia et al., 2014).

5. Experimental research in the biology of the malaria pathogen

5.1 Investigation into the *Plasmodium* species

Based on survey data from 1951 to 1961, the composition of malaria parasites in China was as follows: *Plasmodium vivax* accounted for 52.1%, *Plasmodium falciparum* accounted for 36.9%, *Plasmodium malariae* accounted for 7.5%, and the remaining 3.5% was *P. vivax* mixed infection with *P. falciparum*. The composition of the *Plasmodium* species in different latitudes also differed. There are four *Plasmodium* species distributed south of 25° north latitude. *P. falciparum* and *P. vivax* were the main species, of which *P. falciparum* accounted for 45.9%, *P. vivax* accounted for 42.0%, and *P. malariae* and mixed infection accounted for 7.4% and 4.7%, respectively. Only *P. vivax* was found north of 33° latitude. Between 25° latitude and 33° latitude, three kinds of *Plasmodium* existed, but *P. vivax* dominated and accounted for 74.3%, while *P. falciparum* accounted for 17.0%, and *P. malariae* and mixed infection accounted for 7.9% and 0.8%, respectively.

The distribution and composition of malaria parasites varied in each province and region. *P. vivax* had a very wide distribution range and was distributed in various kinds of malaria endemic areas, such as the tropical, subtropical, and temperate regions of China. The distribution of *P. falciparum* had certain limitations. It was widely distributed south of Nanling Mountains, including almost all areas in the mountains, hills, and plains. The infection rate of *P. falciparum* was around 20%, which was the dominant species in these areas. Between Nanling and Qinling, *P. falciparum* was mostly distributed in mountain basins with a high temperature and low terrain. It was difficult to find its distribution north of the Qinling Mountains. *P. malariae* was scattered within a certain range, and it was mainly distributed south of the Qinling Mountains, but it was not a dominant species. *P. ovale* was only found minimally disturbed southsouthwest of Yunnan Province, Guizhou Province, and Hainan Island (Xiao et al., 2010). After more than 60 years of active prevention and control, the geographical distribution of the four Plasmodium species changed greatly in China. In recent years, *P. ovale* has not been rediscovered in any place, and *P. malariae* has been rarely found. Only 17 counties (cities) in Yunnan Province have reported falciparum malaria. The proportion of falciparum malaria has been greatly reduced, while the number of all types of malaria cases has dropped significantly. In recent years, only vivax and falciparum malaria have been distributed within the border areas of Yunnan Province (Wang et al., 2014b).

5.2 Research on *Plasmodium falciparum*

5.2.1 *Continuous* in vitro *culture of* P. falciparum *and its application*

In 1979, the NIPD successfully established the *in vitro* culture of *P. falciparum* by using the candle cylinder method. The culture method was greatly improved. The relatively expensive cost of the foetal bovine serum in the original method was changed to rabbit serum, and the serum ratio in the medium was reduced from 10%–12% to 8%–10%. Thus, the improved method facilitated the continuous culture of *P. falciparum in vitro* and at the same time, reduced the expenditure. In addition, some progress with the *in vitro* culture of *P. falciparum* gametophytes was also made. This progress created conditions for the screening of antimalarials *in vitro*, testing and comparing the insecticidal effects of antimalarial drugs, and the development of vaccines, in addition to the preparation of applicator plates and freeze-dried culture medium for the *in vitro* microassay to determine drug resistance. Further, it also provided possibilities to compared the effects of chloroquine

(CQ) and its two enantiomers [(+) -CQ, (−) -CQ], desethylchloroquine (D-CQ), and pyronaridine (PY) drugs. The results showed that the five drugs had similar inhibitory effects on CQ-sensitive strains, and D-CQ showed a poorer effect on the anti-CQ strain. PY was significantly better than the other four drugs. In the conventional method for screening antimalarial drugs *in vitro*, the *in vitro* metabolic system consisted of rat liver microsomes and oxidized coenzyme I, which was added to detect common antimalarial drugs (CQ, PY, TCL cycloguanil, and Proguanil) activity. The results confirmed that the introduction of liver microsomes into the *in vitro* metabolic system could avoid the detection of compounds that have antimalarial effects. In addition, a method for the detection of sensitivity of *P. falciparum* to PY/sulfadoxine/pyrimethamine *in vitro* was also established based on microassay, which subsequently demonstrated a synergistic effect between PY and sulfadoxine/pyrimethamine.

5.2.2 The establishment and improvement of the sensitivity of the testing method for malaria in vitro

Due to the spread of CQ resistance among *P. falciparum* and poor treatment effect among falciparum malaria patients, which resulted in a rise in the number of cases and an increase in mortality, antimalarial drug resistance has become the first priority for malaria control. The above-mentioned *in vitro* microassay for testing the sensitivity of malaria parasites was a new technology with simple, fast, accurate, and patient-friendly advantages, but required preformed applicators and culture medium for on-site use. In 1979, the imitative CQ plate was successfully developed by the NIPD, and the applicator plate was improved to make it more suitable for field application in China. In 1980, a freeze-dried medium was developed for the convenience of on-site investigation, thereby establishing the sensitivity test for malaria parasites against drugs using the *in vitro* microassay method. In the same year, a resistance investigation workshop was held in Wanning County, Hainan Island, and the technology was trained to local health workers, which laid a foundation for the national drug resistance investigation.

5.2.3 Cloning and expression of the three repeats of a P. falciparum gene

In order to clone and express the three repeats 3R, 6R, and 9R of the *P. falciparum* (Pf) 11.1 gene product, the NIPD designed primers to amplify the three repeats from the genomic DNA of cultured *P. falciparum* 3D7

strain. The PCR product was cloned into the pT7 vector for bidirectional sequencing. The sequencing results were analyzed by GENETYX-MAC software. The amplified fragment was subcloned into pET32a(+) or pET32b(+) and induced by isopropyl-β-D-thiogalactoside (IPTG) to express recombinant proteins in *Escherichia coli* BL21. The 3R, 6R, and 9R fragments were successfully amplified by PCR, and the sizes were 552, 630, and 444 bp, respectively. The sequencing results showed that the Pf11.1 gene of 3D7 strain had four 3AA and one 6AA repeat units more than that of Palo Alto strain. The identities of the 3R and 6R fragments of the two *Plasmodium* strains were 92.8% and 95.1%, respectively. The amplified 9R fragment contained 13 9AA repeat units. Three recombinant proteins were expressed in the BL21 strain with relative molecular masses (Mr) of 45,000, 60,000, and 42,000, respectively.

5.2.4 Preparation of monoclonal antibodies specific to the lactate dehydrogenase of P. falciparum

By cloning and expressing the lactate dehydrogenase gene of *P. falciparum* and immunizing BALB/c mice with the expressed recombinant protein, the NIPD was able to prepare monoclonal antibodies via a hybridoma technique. Subsequently, the prepared monoclonal antibodies were determined by their subclasses and titres, and their specificity was analyzed by western blotting. The lactate dehydrogenase gene of *P. falciparum* was successfully cloned and expressed. The monoclonal antibody was prepared by using recombinant lactate dehydrogenase protein as the immunogen source. A total of 15 strains were screened for effective secretion titre at 1:6400–1:51,200 with the antibody subclass being IgG1 or IgG2. All antibodies recognized the *P. falciparum* protein Mr 33000 component and did not cross-react with the red blood cell components of non-malarial fever patients in the non-endemic areas.

5.3 Research on *P. vivax* biology

Researchers at NIPD built long-term collaborations with the staff in certain regions of China, including Henan, Yunnan, and Hunan, in order to investigate the relationship between the incubation and recurrence of *P. vivax*, the latency and latent period of *P. vivax*, and perform experimental research and on-site observation of the viability of gametophytes. The results further proved that the incubation period of *P. vivax* in China was of two types—the short incubation period was 10–28 days, and the long incubation period was 228–443 days. The length of the incubation period was related to the

amount of sporozoite inoculation, which can be artificially controlled from long-to-short or from short-to-long. In the temperate regions of China with four distinct seasons, there were no long-latency or short-latency *P. vivax* strains with stable biological characteristics. By observing the treatment of vivax malaria patients with CQ in southern Anhui for one year, 95.2% of the cases relapsed, but the length of the latent period was different, ranging from 50 to 100 days in the short period and 160 to 300 days. The time limit for recent recurrence and long-term recurrence was about 3 months, instead of 8–10 weeks as determined by foreign scholars. For the long incubation period vivax malaria, there was a recent recurrence but no long-term recurrence, and the number of recurrences was shorter during the incubation period. The short-term was only found during the long-latency period of malaria and recurrence, which can roughly distinguish between two different types of vivax malaria and its composition ratio. It is more credible to use the median latency to estimate the length of the incubation period.

The periodic study of gametophytic viability based on the observation of *P. vivax* cases revealed that *P. vivax* needs to pass through several schizontial proliferation cycles after completing tissue development into the bloodstream in order to meet the infection threshold in mosquitoes for infection transmission. Therefore, the primary infection case with a longer incubation period is more likely to cause infection after being bitten by a mosquito vector. In the study of the growth and decline of gametophytic infectivity during the whole course of vivax malaria infection, it was found that artificially infected cases of vivax malaria, if temporarily not treated after clinical onset, were infectious to mosquito vectors throughout the course of the disease, but the duration of high infection status was very short. The reason is that the infection period of the *P. vivax* gametophyte has periodic fluctuations. The peak of mosquito infection occurs with large trophozoites in the peripheral blood of patients, while low infection occurs with mature schizonts and immature trophozoites in the blood. These findings provided a basis for future research on how to select blood donor patients and the amount of mosquitoes needed for infection experiments.

In addition, the mechanism of vivax malaria recurrence is a long-standing problem that needs to be solved. The NIPD used the non-human primate malaria model of *P. cynomolgi–Anopheles stephensi–Macaca rhesus* system to conduct experimental studies regarding malaria recurrence. After *R. macaques* were infected with *P. cynomolgus* sporozoites, serial sections of tissues were taken from different parts of the liver on day 8 post-infection. A large number of intrahepatic schizonts at different stages

of development were found via microscopy, indicating that the development of *P. cynomolgus* in the liver tissue was asynchronous with the schizont matured and released merozoites into the blood circulation. The intracranial merozoites disappeared when treated with PY and M8132; however, the ring stage was present in the blood, but without other *Plasmodium* periods. This is because the mature intrahepatic schizonts released merozoites into the blood, and this time period could last for more than 50 days. After the disappearance of the drug effect, the delayed release of intrahepatic merozoites were able to repeatedly proliferate in the blood, causing a recent recurrence. When intrahepatic merozoites are released into the blood circulation to form parasitemia, administration of PY (6 mg/kg) combined with artemether (10 mg/kg) and CQ (20 mg/kg) could effectively remove the parasite and prevent recrudescence, seeing of recent recurrences indeed reliable. Rhesus monkeys were infected with $(50–55) \times 10^4$ *P. cynomolgi* sporozoites, and the pre-existing period was 8–9 days, which was not affected by the number of spores inoculated. However, the recurrence status varied. The monkeys inoculated with 55×10^4 sporozoites relapsed 3 times, while the monkeys inoculated with 11×10^3 sporozoites relapsed only once, and monkeys inoculated with 50 and 5500 sporozoites had no recurrence within 200 days. The number of sporozoites was related to the frequency of recurrence. The experimental results showed that the real recent recurrence was observed when ruled out the relapse. The results of this study provided a biological experimental basis for the recent recurrence mechanism.

6. Biological investigations on malaria vectors

Anopheles mosquitoes are the main vector for malaria. There are 3 genera and more than 400 species in the world, and about 35 of them have medical importance. More than 60 species of *Anopheles* mosquitoes are known in China. It is generally believed that the main malaria vectors include four species, namely *Anopheles minimus*, *Anopheles dirus*, *Anopheles sinensis*, and *Anopheles anthropophagus* (Fig. 4) (Feng et al., 2017).

An. sinensis is distributed in all provinces (autonomous regions and municipalities directly under the Central Government), except Qinghai and Xinjiang. North of 34° north latitude, *An. sinensis* is an important vector for malaria and filariasis in the vast plain areas, especially in the rice-planting areas, and is the main vector for *vivax* malaria. *An. sinensis* is a semi-domestic mosquito species, and 40%–75% can be found indoors. This mosquito species can feed on bovine and human blood, and it has a strong outside-feeding

Fig. 4 The four main malaria vectors, e.g., *An. sinensis*, *An. anthropophagus* (or *An. lesteri*), *An. minimus*, *An. dirus*, in China.

habit in the plain rice areas, mountains, and hills. By contrast, it prefers inside-feeding habits in rice-planting areas. The peak of blood-sucking activity at night is one hour before and after midnight. In most parts of China, population breeding season lasts from July to September, and the peak of human-biting is from July to August. Female mosquitoes mostly lay eggs before midnight, and mosquito eggs cannot tolerate extreme low or high temperatures and will die at freezing temperatures or temperatures above 40°C. The development of the entire larval stage is 8.3 days in July and August, and 9–10 days before June or after September. Larvae have a wide adaptability to the environment, but tend to prefer a water body around a shady place. Since the 1950s, many irrigation rivers in the low-lying areas of the Huanghuai Plain tend to be the main breeding ground for *An. sinensis* because these rivers usually have a very slow water flow and are overgrown with vegetation. The larvae can survive at low temperatures, and a few larvae can even survive at 3°C. Although the infection rate of *An. sinensis* to *Plasmodium* is relatively low, it has a large population. Thus, there are many opportunities for mosquito exposure, and it plays an important role in malaria transmission, especially in some areas with poor

mosquito-repellent conditions and with outside-sleeping habits during the summer. Therefore, the mosquito is the recognized as the main or only vector for maintaining low prevalence of malaria in most plain areas of mainland China.

An. anthropophagus, which used to be a subspecies of *Anopheles lesteri*, is a relative species of *An. sinensis*. It used to be recognized as a subspecies of *An. sinensis* or *An. anthropophagus* around 1975. In 2008, it was revised by an entomologist and was regarded as a synonym of *An. lesteri*. So far, it has been confirmed that *An. Anthropophagus* is distributed in more than 300 counties in 18 provinces (autonomous regions, municipalities directly under the Central Government) and the Hong Kong Special Administrative Region (Liu, 1990). *An. anthropophagus* has been found with a distribution ranging from 102°20′ to 121°30′ east longitude and 20°00′ to 42°10 north latitude. It is the main vector for falciparum malaria. *An. anthropophagus* is a semi-domestic mosquito species and also a facultative feeder. The peak season is from July to September in most areas of China. In some areas, there may be a second density peak in late October. This species is often distributed in the same area as *An. sinensis* (sympatric distribution). The population density of *An. anthropophagus* is generally not as high as *An. sinensis*, but it can be act as the dominant species in human houses in some areas. Nighttime blood-sucking activities begin around half an hour to two hours after sunset, and the peak of activity lasts from midnight until dawn (Xia et al., 2003). Larvae are found in rice fields, but prefer to be born in water bodies with plant or water bodies with water seepage. While its larvae are symbiotic with *An. sinensis*, the density of *An. sinensis* is higher in the early and middle stages of rice growth, and the density of *An. anthropophagus* larvae increases with the growth of rice. The vector competence of *An. anthropophagus*, especially for falciparum malaria, is far stronger than that of *An. sinensis*. Its salivary gland infection rate is quite high, generally above 0.5%, and its distribution is closely related to the prevalence of malaria. In 1984, through a quantitative survey of entomology and parasitology in Mu'ai Township, Qilian County, Sichuan Province, the vector competence of *An. sinensis* and *An. anthropophagus* was compared. The local malaria cases estimated to be transmitted by *An. anthropophagus* were about 94.3%, while transmission by *An. sinensis* was about 5.7%, about 20 times the former. In recent years, researchers from the NIPD have confirmed that there is *An. anthropophagus* in Faku County, Liaoning Province at 42.5° north latitude, but its ecological habits are obviously different from those south of 33°, which has caused widespread concern and research interest.

An. minimus is distributed in the hilly areas of China south of 33° north latitude, especially south of 25° north latitude. In recent years, classification studies have shown that *An. minimus* has multiple types, such as A, B, and C (Lu, 1997). This mosquito is the main vector of malaria in the mountainous and hilly areas in the south China, and is also one of the most important vectors of malaria in southeast Asia. It is also the vector of filariasis in Hainan Province. *An. minimus* prefers indoor habitats, and the habitats during the day are mainly human houses and barns. The proportion of *An. minimus* in Hainan Island during the day or night is about 60% in the mountainous area and about 40% in the hilly area. The proportion of *An. minimus* inhabiting inside of the net during the day is about 42%–66%, and at night it is about 42%–88%. The density of *A. minimus* begins to rise sharply around March, peaking from April to June before the rainy season and then gradually decreases. A small density peak appears again in some areas from September to October. The larvae are mostly found in ditches where the water quality is good, and the flow of water on the shore is slow. During winter in southern China, larvae can survive in the breeding sites. In an environment of 22–25°C, it takes 18–21 days to develop from an egg to an adult. In Hainan Island, the mosquito tends to prefer human blood (85%), while in Yunnan, Guizhou, Guangxi, and other provinces (regions), the local *An. minimus* sucks both human and bovine blood. According to the geographical observation, the farther north, the smaller the proportion of *An. minimus* that prefers human blood. The blood-sucking activity of female mosquitoes at night starts 1 h after sunset and lasts all night with the peak around midnight. However, in the special ecological environment of the beach in Wenchang County, Hainan, the peak of blood-sucking activity can occur 1–2 h before dawn. *An. minimus* is the main malaria vector in the southern malaria area of China. Its key role in malaria transmission is not only reflected in the high natural infection rate of salivary glands, but also in the area distribution of salivary gland positive infection rate and its duration in the malaria transmission season. There have been positive salivary gland infections in Hainan Island every month in the past (50 years ago).

An. dirus is a tropical jungle *Anopheles* mosquito mainly distributed in Hainan Province, west and south of Yunnan, and at the southwestern border of Guangxi (Xu and Feng, 1975). This mosquito species is only found in the forest and hilly areas of tropical rain forests or secondary mixed forests. It is a typical outdoor habitat mosquito species. It is generally believed that this mosquito species is an important vector for malaria transmission in the mountainous areas of central and southern Hainan Province.

However, the role of malaria transmission in other areas is still unclear. Historical studies have shown *An. dirus* prefers human blood (96.2%). Blood-seeking activity begins one hour after sunset, and while it peaks at around midnight, there is blood-sucking activity all night. The population peaks from July to October, and the average human-biting rate in undeveloped forest areas is 14.8%, and 2.03% in semi-development zones. The proportion of the composition of *An. dirus* is about 2%–10% in the hilly area, more than 10% in the mountainous area, and 50% in the isolated and isolated small villages in a few undeveloped forest areas. The main habitat of *An. dirus* larvae is small natural water that is shaded by trees or bushes. Due to strict requirements for the environment, the population is usually small. *An. dirus* is the main malaria vector in the mountainous forest area and mountainous area of Hainan Island. The infection rate of salivary gland fluctuates from 2.1% to 8.3%, and there are natural infections from April to November throughout the year. Because the *An. dirus* larvae must have a certain degree of shading, once the surrounding jungle is cut down and shade conditions disappeared, the *A. dirus* larvae will quickly disappear and subsequently reduce the spread of malaria.

7. Research and development of control tools, including diagnostics, drugs, and insecticides

7.1 Diagnosis

7.1.1 Etiological diagnosis

When compared with traditional methods, such as Wright's and Giemsa staining, both the fluorescent pigment acridine orange staining method and fluorescence microscopy are able to quickly detect *Plasmodium*, even at a low-density. Fluorescence microscopy is more efficient than ordinary microscopy. In a study conducted by the NIPD, the researchers first stained the blood film with fluorescein acridine orange and then examined it with fluorescence microscopy and then counterstained it with Gill's stain. The results of 179 primary school students showed that the detection rate of acridine orange method was 42.0% higher than that of Giemsa method. The acridine orange method was able to detect 11 blood films per hour on average, which is 1.8 times faster than the Giemsa method, which was able to detect four blood films per hour. In 1978, the detection efficiency of *P. falciparum* by the acridine orange method was compared in Hainan Island, where 119 people were investigated and 34 cases were detected (including 32 cases of falciparum malaria). The detection rate was 97% when

compared to Giemsa method (85%). Among the falciparum patients, 7 cases were detected with 5–10 parasites in the blood smears, and 9 cases with less than 5 parasites, indicating that the acridine orange method has a high accuracy for detection of the thin ring stage and fewer parasites. Fluorescein acridine orange was used to examine *Plasmodium gallinaceum* sporozoites in the salivary gland of *Aedes* with expected results. The nucleus of these sporozoites showed a punctate yellow-green fluorescence. Most of the sporozoites were mononuclear, but a few had dual nuclei. The cytoplasm was sickle-shaped with the sharp ends showing orange-red fluorescence, and the sporozoites had clear morphological structure, sporozoite activity was still visible after 2 h.

Researchers at the NIPD have established a polymerase chain reaction (PCR) to detect *P. falciparum*. The amplified products were analyzed by agarose gel electrophoresis to amplify a specific 492 bp DNA fragment. By contrast, DNA from *P. vivax*, *P. cynomolgus*, *P. yoelii*, *Plasmodium berghei*, and healthy human leukocytes did not amplify this product. This method can detect the lower limit of parasite density at 10 parasites/20 μL with a high sensitivity and specificity. PCR was used to detect 53 blood samples of falciparum malaria patients in Yunnan. The results were all positive, and the coincidence rate with microscopy was 100%. At the same time, 8 samples of vivax malaria and 12 healthy blood samples were tested, all of which were negative, further indicating that this method has a high sensitivity and specificity. A rapid diagnosis using a malaria colloidal gold immunochromatographic strip method was established, and 54 blood samples of patients with falciparum malaria were tested, of which 51 were positive. The detection rate with this method was 94.4%. There were also 110 samples of vivax malaria tested, among which 102 were positive, and the detection rate was 92.7%, which also presented with high sensitivity and specificity.

7.2 Serological diagnosis

7.2.1 Indirect fluorescent antibody test (IFAT)

In 1976, serum antigens were detected in 126 cases of vivax malaria patients in the northern malaria area of northern Fujian Province using *Plasmodium cynomolgus* monkey (*P.c*) antigen. The antibody titres $\geq$1:10 and $\geq$1:20 were 73.8% and 60.7%, respectively, for those under 15 years old and 81.0% and 69.0%, respectively, for those over 16 years old. In 1978, *P.c* antigen was used to detect serum antibodies in 50 patients with falciparum malaria in Suining County, Jiangsu Province. The positivity rate of the antibody titre $\geq$1:20 was 98.0%. Also, 52 cases of falciparum malaria in Hainan Island

were detected using the serum antibodies from each patient, and the positivity rate of the antibody titre ≥1:20 was 100%. In 1983, the serum antibodies of 35 patients with falciparum malaria were detected by *in vitro* cultured *P. falciparum* and *P.c.* antigen. The positive rates were 100% and 97.1%, respectively. However, the geometric mean titre reciprocal (GMRT) of positive individuals detected by *falciparum malaria* (*P.f*) antigen was 1445.4, which was significantly higher than 186.5 of *P.c* antigen. The positive rate of the serum antibodies in 38 cases of vivax malaria was 89.5% and 100%, respectively. The positive GMRT of P.f antigen was 40.6, which was significantly lower than 215.9 by *P.c* antigen, indicating that the same antigen was superior to the heterologous antigen, and the *P.c* antigen was superior to the P.f antigen for detecting the vivax malaria antibody. In Hainan Island and Yunnan Xishuangbanna, where *An. minimus* almost disappeared, serological investigation confirmed that malaria has been controlled for many years. In places where *An. minimus* and *An. sinensis* do not exist, the antibody level of the population indicated that malaria was still in transmission. Five different types of populations were investigated horizontally using *P.f and P.c* antigens. The results of the serological assay were consistent with the blood smear test, indicating that the two antigens are able to be used in the IFAT to investigate the prevalence of *falciparum* and *vivax* malaria. Subsequently, a three-year longitudinal survey was conducted on three mixed epidemic areas of falciparum and vivax malaria in Anhui Province, where different antimalarial measures were applied. As a result, the serum antibody positive rate, antibody titre, and malaria parasite rate showed a parallel relationship in reflecting malaria prevalence levels. The incidence and recovery rate of malaria parasitemia in the malaria transmission season in three regions, the positive rate and recovery rate of antibodies detected by *P.f* and *P.c* antigen, and their magnitude and trends were associated with the intensity of falciparum and vivax malaria transmission and the measures taken against malaria. Two years of horizontal surveys were conducted in 1983–84 in three malaria areas with different antimalarial measures and compared with the longitudinal surveys. The results of the two surveys were roughly the same. Both of them can be used to investigate the prevalence and changes of falciparum and vivax malaria in the region, and in turn, to evaluate the effects of antimalarial measures. In recent years, malaria epidemic in the Yangtze River Three Gorges reservoir area was accessed by the IFAT, and result showed a certain extent of malaria infection existed in this area (Duo-Quan et al., 2009).

7.2.2 Enzyme-linked immunosorbent assay (ELISA) for malaria epidemiology survey

In 1978, by using *P.c* antigen to prepare antigen, and labelling it with alkaline phosphatase anti-human IgG, researchers from the NIPD detected 77 dry blood drops from patients with falciparum malaria and found that the positive rate was 96.1%. Moreover, 85 cases of dry blood drops from patients with vivax malaria were analyzed by this method, and the positivity rate was 98.8%. Subsequently, by using *in vitro* cultured *P.f* was used as antigen, 307 malaria positive dry blood drops were collected and analyzed via this method. The positivity rate was 90.23%, and the positive coincidence rate of falciparum malaria was 94.29%, and the positive coincidence rate of *vivax* malaria was 84.85% (Gao et al., 1991; Zhang et al., 1998). The false positive rate of healthy people in non-malarial areas was only 1.2%. A subsequent ELISA was used to conduct a sero-epidemiological survey of residents in two types of areas—one area was a malaria endemic area and the another area was where a malaria outbreak occurred that was under control in Hainan Island. The researchers took fresh blood samples or dry blood drops for ELISA. The antigen was a soluble antigen cultured *in vitro* from *P. falciparum*, and the enzyme conjugate was anti-human IgG-HRP. The results showed that the infective rate of the malaria parasite in the former category area was 9.8%, and the ELISA positive rate was 35.2%. The antibody level of the residents increased with age, while the infection rate decreased due to the increase in age. In the latter area, the ELISA positive rate of residents was 72.2% during the second year after the outbreak, which was significantly higher than that of the former category. In the fourth year, although the infection rate in the residents did not change significantly, the average antibody level and ELISA positive rate significantly decreased. The average antibody level in all age groups under 25 recovered, indicating that malaria re-emergence may occur once there is a source of infection.

7.2.3 The establishment of a single epitope monoclonal antibody against P. falciparum

Researchers in the NIPD selected HRP-II peptides with 6–9 amino acids, which represents the antigen trait of *P. falciparum* protein. After artificial synthesis and determination of its purity, BALB/c mice were immunized with such peptides. Four hybridoma cells against a single epitope of *P. falciparum* HRP-II were obtained by spleen implantation and hybridoma technology. In this test, the carrier protein (KLH) was first adsorbed on the ELISA plate,

and the nine peptides were covalently cross-linked onto the carrier protein by the cross-linking agent in order to increase the adsorption and reduce the steric hindrance, which improves the detection sensitivity. This experiment also solved the adsorption problem of small molecule peptides in ELISA assays. This has potential benefits for an in-depth study of malaria diagnosis, in addition to the structure and function of HRP-II.

7.2.4 DNA probes for the diagnosis of P. vivax

Blood samples from patients with falciparum malaria were positive for dot-hybridization with a 0.24 kb DNA probe specific for *P. vivax*, and those with negative on-site microscopy were screened by Southern blotting. The results showed that the positive reaction of the dot-hybridization reaction was also positive. The experiments confirmed and concluded that some patients with falciparum malaria and some fever patients with negative microscopy may also infected be with *P. vivax*. It is further proved that the *P. vivax* DNA probe could not only detect the presence of *P. vivax* infection in patients with falciparum malaria, but could also detect *P. vivax* in fever patients with negative microscopy. This is important for identifying whether a patient is a mixed infection, as well as for screening asymptomatic carriers in an endemic area.

7.3 Drugs

In the field of antimalarial drug research, there was only one chicken malaria model in China when the People's Republic of China was founded. Due to the need of improved research work, the NIPD established a variety of animal models to study malaria pathogenesis, in addition to animal models for antimalarial screening and pharmacological research for the supply of antimalarial drugs and other research Institutes.

7.3.1 Established malaria animal model

Avian malaria parasites were introduced from India during the Anti-Japanese War. *Aedes albopictus* is highly sensitive to chicken *Plasmodium* (*P. gallinaceum*) and can be used to study antimalarial drugs that inhibit sporozoite development in mosquitoes. The extraerythroid stage of *P. gallinaceum* can be found in tissues such as the brain, lung, spleen, liver, heart, and kidney, and can be used to study the development of drugs aiming to eradicate vivax malaria. In 1954, the Indian strain of *P. berghei* was introduced from India, and the Soviet strain of this parasite was later introduced from the Soviet Union. It can be used to screen antimalarial

drugs for *erythrocytic stage* when *P. berghei* is infected in the blood of mice. Since drug resistance research was needed, a 52-fold resistant strain of *P. berghei* was developed. In this way, researchers in the NIPD induced the anti-artemisinin *P. berghei* strain with 13-fold resistance, the anti-pyronaridine with 300-fold resistance, and the resistance index of 197.2 multi-resistant to chloroquine and pyronaridine *P. berghei* strains. In order to establish a murine malaria model for screening etiological and curative drugs, *Anopheles stephensi* was introduced from London in 1973, and the Yes265 strain of *P. yoelii* was introduced from Paris in 1976, thereby establishing the *P. yoelii–An. stephensi* system. In 1973, *Plasmodium knowlesi* was introduced from the United Kingdom to establish a rhesus monkey model to test the effects of various compounds on the erythrocytic schizont. Later, one kind of monkey *Plasmodium* strain was introduced from Vietnam and identified as *P. cynomolgus* by experimental observation. The NIPD has thus established a comprehensive non-human primate malaria model consisting of *P. cynomolgus–An. stephensi*–rhesus monkey. *P. cynomolgus* is similar to *P. vivax* and is easier to maintain in the laboratory. This model can be used for the development of drugs to kill schizonts during the blood stage, as well as for etiological prophylaxis and radical drug development. Rhesus monkeys are sensitive to *P. cynomolgus* in Guangxi and other places. In monkeys infected with *P. cynomolgus*, the parasitemia was temporarily negative when treated with CQ (5 mg/kg) via the intranasal route, but recrudescence occurred soon. The parasitemia totally disappeared with a single dose of 40 mg/kg CQ. Primaquine (6 mg/(kg*d)) with CQ (20 mg/(kg*d)) for 33 days cured the infected monkeys inoculated with sporozoites. Based on these data, it was possible to test if other compounds were better than CQ in killing erythrocytic schizonts. Any drug having exo-erythrocytic effects can be tested and compared with the efficacy of primaquine in the monkey malaria model. Thus, a murine malaria model and monkey malaria model have been established, and a system for screening antimalarials has been formed. Chicken *Plasmodium*, murine *Plasmodium,* and monkey *Plasmodium* were preserved in the laboratory for a long time by means of blood transfusion and sporozoite seed transfer. At the beginning, a cryopreservation method was used to protect the blood containing *Plasmodium* with 15% glycerol phosphate buffer and stored in dry ice and acetone. In recent years, 24% dimethyl sulfoxide has been used to protect the activity of the parasites, and the stock is stored in liquid nitrogen to maintain the infection activity of parasite during the erythrocytic stage. In 1980, a cryopreservation

method for the sporozoites of *P. berghei* in whole mosquitos and liver tissue without adding any protective agent was established. In addition, laboratory breeding and breeding methods for *Ae. albopictus*, *Ae. Aegypti, An. sinensis*, *An. stephensi*, and *Culex pipiens pallens* have been established to supply mosquitoes for experimental study.

7.3.2 The development of new antimalarial drugs

CQ resistance has become increasingly widespread in the world, and there is an urgent need for high-efficiency, low-toxic therapeutic new drugs without cross-resistance to CQ both at home and abroad. According to the requirements of the National Leading Group for Malaria Prevention and Treatment Drugs, the NIPD undertook research on new antimalarial drugs since the NIPD was founded. In 1964, a new compound, bispyroquine was synthesized. Animal studies proved that it had an antimalarial effect, and the clinical trial was carried out. As a result, the new drug was effective for the treatment of *P. vivax*, but the trial was stopped due to certain cross-resistance with CQ. The new compound, pyracrine, was synthesized in 1967, but it was eventually discontinued for toxicity reasons. At the end of 1969, another new compound, pyracrine phosphate, was designed and synthesized. It was found that pyrrolidine was better than CQ in animal experiments, and the adverse reactions were lower than CQ, and there was no cross-resistance with CQ. Pyracrine phosphate can be taken orally, intramuscularly, or intravenously. It entered clinical trials in 1971 and passed the production process identification in 1979. It was registered in 1980 and received three approval numbers for pyrrolidine and its tablets and injections dosages were officially put on the market. Since 1990, these tablets and injections have been included in the Pharmacopoeia of the People's Republic of China. It is the only chemical synthetic antimalarial drug created in China and included in the Pharmacopoeia. The related research work won the National Science Conference Award (1978) and the National Invention Third Prize (1985).

For the application research of malaria treatment and preventive drugs, it was necessary to explore a plan to cure children with *P. vivax* and to facilitate mass drug administration (MDA). From 1961 to 1963, patients with parasitemia under 15 years of age were selected for treatment during malaria transmission using primaquine 210 mg (7 days), 180 mg (8 days), and 120 mg (4 days) with a double course of treatment and primaquine 210 mg (7 days divided) with single course of treatment compared with CQ 600 mg double course and a no medication group as a control. The results demonstrated that all single-course regimens were not satisfactory, while the three groups

treated with the dual-treatment regimen achieved good results. The length of the two treatment intervals had no significant effect on the efficacy. From the perspective of efficacy, safety, economy, and simplicity, primaquine 180 mg (8 days) and primaquine 120 mg (4 days) were suitable for field MDA application. In addition, the NIPD also organized a special study to evaluate the efficacy of CQ and chlorhexidine, chlorpyridinium chloride, and pyronaridine for the treatment of vivax malaria. For the evaluation of preventative drugs, 50 mg of pyrimethamine combined with 22.5 mg of primaquine 22.5 was tested in 1965 in the Daishan Township, Wuxing County, Zhejiang Province, and the drug was administered once every two weeks for a total of nine times. During the period of preventative medication, the incidence of malaria among residents decreased by 81.8% as compared with the same period of the previous year, and the incidence of vivax and falciparum malaria decreased by 91.8% and 70.1%, respectively. During the medication, 32 cases of falciparum malaria and 10 cases of vivax malaria occurred. The average interval time between falciparum malaria and the last preventive medication was 9.8 days, and the average interval for vivax malaria was 12 days. These results indicated that regardless of the epidemic area of vivax malaria or the mixed epidemic area of falciparum and vivax malaria, the use of pyrimethamine 50 mg for 10–15 days was effective. When chlorpyrifos used to prevent malaria, the results were poor.

The studies on CQ resistance and its geographical distribution in China showed that CQ resistance was first confirmed in Yunnan Province in 1972 and then in Hainan Island and Guangdong Province in 1974 and then Ningming County in Guangxi in 1977. In 1980, the NIPD collaborated with eight provinces (regions), including Guangdong, Guangxi, Guizhou, Guizhou, Guizhou, Jiangsu, Henan, and Anhui to carry out a large-scale investigation on the CQ resistance to *P. falciparum*. By 1984, the baseline of geographical distribution and CQ resistance status in the above areas were identified. The results showed that there was high CQ resistance in Hainan Island and southern Yunnan Province; more than 80% of cases were CQ-resistant to *P. falciparum* in Hainan Island epidemic areas, and more than 85% of cases were CQ-resistant to *P. falciparum* in Yunnan Province. The researchers also identified CQ resistance in Guangxi and Anhui, but the degree of resistance was low. The CQ resistance to *P. falciparum* in Henan, Guizhou, and Jiangsu was at the initial stage.

In order to reduce CQ resistance in falciparum malaria, from 1981 to 2003 the sensitivity of *P. falciparum* to CQ in Hainan and Yunnan was determined by an *in vitro* four-week micro-method. The results showed that the

resistance rate of the *in vitro* assay in Ledong County of Hainan decreased from 97.9% in 1981 to 26.7% in 1997 ($P<0.01$), and the average concentration for the complete inhibition of schizont formation was reduced from 10.46 ± 7.14 to 1.63 ± 1.47 pmol/μL ($P<0.01$) in the blood. Moreover, the *in vivo* method determined that the resistance rate decreased from 84.2% in 1981 to 18.4% in 1997 ($P<0.01$), and the RIII (Resistance 1) in the resistant cases decreased from 53.1% to 14.3% ($P<0.01$). In 2001, the resistance rate as determined by the *in vitro* method in Yaliang Township of Sanya City was 59.8%, and the average concentration of needed to inhibit schizont formation was 3.56 ± 2.12 pmol/μL in the blood. In 2003, the resistance rate in Fuhu Township of Ledong County was 62.5%, and the RI, RII, and RIII (Resistance 1–3, respectively) accounted for 50%, 30%, and 20% of the resistant cases, respectively. The *in vitro* resistance rate of CQ in Mengla County of Yunnan Province decreased from 97.4% in 1981 to 77.8% in 1999 ($P<0.01$), and the average drug concentration that completely inhibited the formation of schizonts was reduced from 17.2 ± 12.6 to 4.4 ± 3.1 pmol/μL ($P<0.01$). In 2002, Jinghong County, Yunnan Province, demonstrated that the resistance rate was 70.4% as determined by the *in vitro* assay, and the average concentration of inhibition of schizont formation was 4.0 ± 3.3 pmol/μL. The results showed that *P. falciparum* presented with some degree of reversible resistance to chloroquine. After CQ resistance was produced, if CQ was stopped in time, *P. falciparum* was able to gradually restore sensitivity to CQ without drug pressure. However, the recovery rate was slow. For example, in Hainan Province, 20%–60% of the cases of CQ resistance have been reduced for more than 20 years; however, there is still some resistance to CQ.

In order to clarify the sensitivity of *P. falciparum* to commonly used antimalarial drugs in China, various malaria drug applicator plates were successfully developed to monitor the sensitivity of the malaria parasite to antimalarial drugs, such as piperaquine, pyronaridine, artemether, and artesunate, on the basis of successful imitation and improvement of chloroquine assay plate From 1986 to 1992, the sensitivity of *P. falciparum* to nine antimalarials was determined in Hainan and Yunnan provinces. As a result, no resistant cases were found for mefloquine and quinine, but resistant cases were found for all seven other drugs. Of note, high resistances to CQ, piperaquine, and acetaminophen were found, and 8.3% of the cases were resistant to pyronaridine. Moreover, a few cases had decreased sensitivity to artemisinin. After CQ was stopped or reduced in Hainan and Yunnan provinces, the sensitivity of *P. falciparum* to CQ recovered, while the resistance to piperaquine increased. The above results

provided a scientific basis for the development of anti-malaria planning and rational selection of antimalarial drugs. In order to rationally apply the existing antimalarial drugs to delay the emergence of resistance, the Ministry of Health commissioned the NIPD to establish a principle for antimalarial use in China in 2000, and divided the antimalarial drugs used in China into first, second, and third-line drugs. CQ and piperaquine were the first-line drugs for vivax and falciparum malaria endemic areas sensitive to CQ and piperazine. Pyronaridine and artemisinin were second-line drugs for areas with moderate-to-high CQ and piperaquine resistance. Pyronaridine or artemisinin drugs and other antimalarial drugs were used as third-line drugs in areas where the efficacy of second-line drugs was not obvious.

7.4 Insecticide resistance

In the early 1960s, *An. sinensis* was very sensitive to organochlorine insecticides before the widespread use of pesticides in various places. The NIPD performed research on the sensitivity of *An. sinensis* to DDT, benzene hexachloride (BHC), organophosphorus pesticides and other insecticides (Cheng et al., 1995; Li et al., 1989; Wang et al., 2013; Wang, 1999; Wu et al., 1993). From 1978 to 1982, a survey of 62 points in 15 provinces (cities) showed that, except for Luodian, Guizhou Province, *An. sinensis* had different degrees of resistance to DDT across all areas, 34 of which (54.8%) had significant resistance, and the LC50 was >2%. Moreover, 27 points had initial resistance. In the late 1960s, *An. sinensis* was generally resistant to BHC. According to a survey of 57 points in 11 provinces (cities), *An. sinensis* in 49 points (86%) showed resistance to BHC, mostly initial resistance. With the restriction on the use of BHC, and eventual total ban on its production and use, the mosquito resistance to BHC has demonstrated a significant downward trend. In the suburbs of Shanghai, the LC50 was 1.87% in 1973, an increase of 86 times from 1961, but it fell to 1.20% and 0.3% in 1977 and 1979, respectively. The LC50 of BHC was at 0.99% in 1977 and fell to 0.17% in 1982 in Zhengzhou and Henan Province, which was close to the sensitive level. Since the 1960s, organophosphate insecticides have been widely used in the prevention and control of agricultural pests in China, but they are rarely used as a malaria vector control. According to the results of the 1979–83 survey, *An. sinensis* resistance to organophosphate insecticides was widespread and very severe in the southeastern region, while resistance to organophosphate was limited and relatively slight in the western region. Resistance to chlorpyrifos occurred

only in the east, and resistance to malathion was mostly in the east and central China. This may be related to the widespread use of organophosphorus pesticides in the local area to control agricultural pests. In 1963, the sensitivity investigation of *An. minimus* to DDT and BHC in Taishan and Shangxiachuan of Guangdong showed that the LC50 of DDT was 0.044%, and the LC50 of BHC was 0.005%. This was similar to the reported LC50 of DDT in Taiwan, which was 0.09%, and the LC50 of BHC was 0.0066% in Indonesia, which can be regarded as the normal sensitivity of the mosquito to DDT and BHC. In southern China, organochlorine insecticides were mostly used for residual indoor spraying to prevent *An. minimus*. Since 1977, the results of various surveys have shown that this species of mosquito has reduced sensitivity to DDT and BHC. The LC50 of *An. minimus* in Guangdong was 0.077%–1.46%, while it was 0.3633%–0.7619% and 0.495% in Yunnan and Guizhou, respectively. By contrast, the LC50 was 0.05%–0.155%, 0.0043%–0.2172%, and 0082% in the above three regions, respectively. Compared with the surveillance data in 1963, the sensitivity to DDT and BHC decreased by 33 times and 43 times, respectively. However, this is not enough to influence the effect of residual spraying.

8. Future perspectives

Since the founding of the People's Republic of China, malaria has been regarded as a critical infectious disease requiring prevention and control. Different prevention strategies have been proposed based on malaria epidemic characteristics and degree of severity in different periods, and as such, remarkable achievements have been made. The annual incidence of malaria fell from 122.9/10,000 (6.97 million cases) in 1954 to 0.06/10,000 (7855 cases) in 2010. A solid foundation has been built such that China could move from "malaria prevention" to "malaria elimination." In the past 30 years, China's economy has developed rapidly, society has been stable, the government has paid attention to the people's livelihood, and a sound grassroots public health service system has been established, all of which provides favourable conditions for the elimination of malaria in rural areas. China was once a country with a serious epidemic of falciparum malaria. However, after a vigorous prevention and control programme, there have been no new indigenous cases of falciparum malaria in provinces since 2010, except for Yunnan Province. In addition, the island-type Hainan Province and central Jiangsu, Anhui, and Henan provinces have eliminated malaria and accumulated successful experience. Since the launch of the national malaria

elimination programme (NMEP), the national work to eliminate malaria has progressed smoothly. China has been listed by the World Health Organization (WHO) as one of the 21 countries in the world to achieve the goal of eliminating malaria by 2020. The "1-3-7" work model for malaria elimination has also been officially written into the WHO's technical documents for global application. Since May 2010, the former Ministry of Health and other 13 ministries and commissions have issued the "China Malaria Elimination Action Plan (2010–20)." Since then, various local departments have carefully organized and implemented malaria prevention and control work. Of note, in 2017, China reported no indigenous malaria cases for the first time nationwide. Although the malaria prevention and treatment work in China has made significant progress and possesses the conditions and foundation for malaria elimination, there is still a long way to go to eliminate malaria eternally. At present, the challenges for eliminating malaria and consolidating results are mainly reflected in the aspects discussed below.

8.1 Malaria retransmission risk

At present, China is in a critical period to eliminate malaria. Given the fact that the source of infection (imported cases from abroad) has not been completely eliminated and that the malaria vector still exists, in addition to the Yunnan border area being a hot spot and focus for malaria elimination in China, malaria transmission conditions still exist in these areas (Zhang et al., 2016). If malaria transmission conditions cannot be discovered and removed in time, then China will still face the risk of retransmission and recovery of the epidemic. In addition, the larger number of imported cases from abroad will pose a huge challenge and threat to the prevention and elimination of malaria in China, which also faces the risk of local retransmission caused by imported malaria. Therefore, it is necessary to maintain the continuity of malaria surveillance, ensure adequate professionals, sustain investment on prevention and treatment, provide policy support, and manage support to effectively prevent the resurgence of local infections and transmission in China (Shi et al., 2017).

8.2 Maintenance of post-malaria elimination surveillance and response capacity

China has built a nationwide network epidemic reporting system and comprehensive monitoring network and has developed feasible monitoring indicators. In recent years, the malaria epidemic has dropped to a low level in

China, and as a result, the number of professionals engaged in malaria prevention and treatment has been seriously deficient. In some areas, there are no departments specializing in malaria prevention and treatment. The building capacity of the disease control institutions and construction of the talent team are still very weak, and there is not enough financial support. The relevant applied research and development of early and sensitive diagnostic tools to discover and dispose of local active epidemics and respond to unexpected imported malaria cases are also seriously lagging behind. In order to achieve the goal of eliminating malaria as scheduled and to ensure the implementation of various technical measures to eliminate malaria, capacity building is a top priority in the elimination of malaria. Therefore, it is necessary to further strengthen institutional construction and technical training, and build a professional team that can implement various measures in the process of eliminating malaria and long-term monitoring after malaria elimination. Only after these foundations are consolidated can the goal of eliminating malaria be achieved as scheduled.

8.3 Long-term mechanism for managing imported malaria cases and mobile population

With the advancement of global integration, the number of cases imported by migrants and foreign workers has been rising (Feng et al., 2014a). Since 2017, all the malaria cases have been imported malaria cases from abroad. However, at present, there are many weak points in the prevention and treatment of imported malaria. First, because the management of imported malaria involves many departments, effective management mechanisms have not been established or improved, and as such, various management measures cannot be implemented well. Second, the mobile population generally lacks the awareness, knowledge, and conditions of malaria protection, active consultation and standardized treatment. It is easy to get malaria when working or traveling in high malaria areas abroad, and after infection, these infected patients cannot seek timely treatment and obtain effective antimalarial treatment. A large number of imported malaria cases will likely occur in non-malaria endemic areas in China, where medical staff generally lack the awareness, diagnosis, and treatment skills of malaria, as well as the necessary diagnostic equipment and effective drug treatment, which may often lead to misdiagnosis or missed diagnosis and cannot effectively deal with the occurrence of falciparum malaria cases or epidemics, resulting in increased disease burden and even death of patients (Feng et al., 2014a).

8.4 Malaria diagnosis, drug resistance, and insecticide resistance issues

The conventional blood smear for fever patients has low sensitivity and is prone to missed diagnosis (Zhao et al., 2018). The rapid diagnosis test (RDT) for malaria has difficulty in identifying parasite species and provides false negatives in patients with a low-density parasite burden. These shortcomings cannot meet the requirements for the identification of malaria patients. Therefore, it is urgent to develop a new, sensitive, and specific malaria detection technology that can efficiently detect malaria infection sources in a timely manner. It has been reported that the sensitivity of *P. falciparum* to artemisinin has begun to decrease, and this resistance on emerged on the Thai-Myanmar border. A CQ-resistant strain of *P. vivax* has already evolved and begun to spread within Southeast Asia. Once these drug-resistant strains disseminate into China, they will inevitably affect the implementation and completion of our malaria elimination (Chen et al., 2010). However, the conventional in vivo drug sensitivity surveillance method is difficult to implement in China due to a few, scattered cases and cannot meet the higher requirements of the resistance test. The research on new technology for the detection of malaria parasite resistance based on resistance gene detection has become one of the key technical problems in malaria elimination in China. Also, with the long-term use of pesticides, it has been found that a variety of Anopheles mosquitoes have developed resistance worldwide (Dai et al., 2015; Feng et al., 2015b). This is an increasing trend, which poses a serious threat to the current stage of malaria prevention and control. Therefore, other new vector control strategies should be developed to mitigate the impact of pesticide resistance on global health.

9. Conclusion

Malaria, a disease that seriously endangers the health of the people, is currently at the stage of elimination. It is another disease that will be eliminated, following the eradication of smallpox and filariasis in China. It will be a milestone in the history of disease prevention in China, as well as the world. According to current work progress and overall plan to eliminate malaria, the goal of eliminating malaria nationwide by 2020 is fully achievable. However, the elimination of malaria is a complex and social system related project involving a wide range of factors with many influencing elements. Therefore, we should conscientiously implement the principles of

scientific prevention, adaptation to local conditions and providing guidance to different strata, and adherence to the working mechanism of government taking the leadership, in cooperation with different departments, and participation of the whole society. We should also continuously improve science and technology on malaria control and surveillance tools, strengthen international cooperation and information exchanges on malaria elimination, make full use of domestic resources to consolidate the achievements gained in the national malaria elimination programme, and strive to achieve the national goal of eliminating malaria on schedule. At the same time, after the elimination, we should continue to focus on migrated people who return from malaria endemic areas of Africa and Southeast Asia, strengthening the surveillance and response systems to prevent introduction of imported malaria cases or foci, and maintaining malaria surveillance and response capabilities both in laboratory and epidemiological works. Through closely multi-sectoral cooperation between health and other departments, such as departments of commerce, tourism, customs and so one, we will effectively perform joint surveillance and control work to prevent secondary transmission caused by imported malaria.

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Competing interests

The authors declare that they do not have competing interests.

Authors' contributions

X.F., Z.X., X.N.Z., S.S.Z. conceived the study; X.F., Z.X., J.F., L.Z., H.Y., wrote the first version of the chapter and revised the chapter; X.F., Z.X., L.T., X.N.Z., S.S.Z. revised the chapter and gave approval of the version to be published. All the authors read and approved the final version of the chapter.

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